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Preface

This book is the Proceedings of the 10th International Symposium on "Plant-Soil Interactions at Low pH" (10th PSILPH) held in Putrajaya, Malaysia from June 25 to 28, 2018. At the meeting of the international steering committee of the 8th Symposium on Plant-Soil Interactions at Low pH held in Bangalore, India in 2012, it was decided that the 10th PSILPH would be held in Malaysia. The 10th PSILPH is organized by the Faculty of Agriculture, Universiti Putra Malaysia (UPM) in collaboration with the Malaysian Society of Soil Science (MSSS) and Malaysian Agricultural Research and Development Institute (MARDI) and Department of Agriculture (DOA) Malaysia. The theme of the symposium is "Achieving Sustainable Food Production on Acid Soils" with the main objective of addressing issues related to sustainable food production on soils with low pH and at the same time achieving environmental sustainability.

In tropical regions around the world, soils with low pH are very common due to high temperature and rainfall they receive all year round. Low pH soils can cause injuries to the plant root systems, inhibiting overall plant growth. Agriculture and agronomic practices are continuously being developed and improvised to overcome the problems that growers are facing without neglecting the effects to the environment.

The 10th PSILPH encompass 7 theme which are Genetics and breeding of crops, Soil-microbe-plant interactions, Physical and chemical properties of low pH soil, Soil fertility, chemistry, amelioration and remediation of low pH soils, Physiological and molecular mechanisms of plant, Sustainable management of plantation and other crops and Environmental monitoring of low pH soil. A total of 111 papers have been submitted which include 4 plenary, 7 keynote, 33 oral and 67 poster presentations.

The 10th PSILPH aims to gather researchers, scientists, experts and academicians in the field of soil science, plant physiology and others to share and discuss the latest research findings and thoughts on current status of agriculture production and practices in low pH soil, thus, ensuring food security and environmental sustainability.

Che Fauziah Ishak, UPM (Chairperson)
Mohd Izuan Effendi Halmi, UPM
Daljit Singh Karam, UPM
Muhammad Firdaus Sulaiman, UPM
Annur Bin Mohd Razib, UPM

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PL-1

Crop adaptation to acid soils: Common features between aluminum resistance and tolerance to phosphorous deficiency

***Leon Kochian*[1], *Laiane S. Maciel*[2], *and Jurandir V. Magalhaes*[2]**

[1]Global Institute for Food Security, University of Saskatchewan, Saskatoon, Canada
[2]Embrapa Maize and Sorghum, Rod. MG 424, Km 65, Sete Lagoas – MG, 35701-970, Brazil
; Leon.Kochian@usask.ca

Crop production on acid soils is limited by a number of abiotic stresses, with the two most important usually being aluminum (Al) toxicity and phosphorus (P) deficiency due both to low soil P levels and high fixation of P on the surface of clay minerals, which reduces the bioavailability of soil P to plant roots on acidic, tropical soils. Advances in our understanding of the physiological and genetic mechanisms that govern plant Al resistance have led to the identification of multiple Al resistance genes, both in model plant systems such as Arabidopsis and in crop species. It has long been known that Al resistance has an indirect, beneficial effect on global crop adaptation to acidic soils. This positive effect happens because the root systems of Al tolerant plants show better development in the presence of soil ionic Al^{3+} and are, consequently, more efficient in absorbing sub-soil water and mineral nutrients. This global effect of Al resistance on crop production, by itself, warrants intensified efforts to develop and implement, on a breeding scale, modern selection strategies to profit from the knowledge of the molecular determinants of plant Al tolerance. Recent studies also suggest that Al resistance can exert pleiotropic effects on P acquisition, potentially expanding the role of what we previously thought were solely Al resistance proteins on crop adaptation to acid soils. This could happen via membrane transporters involved in plant adaptation to acid soils. Some of these transporters mediate root organic acid exudation and others mediate root uptake of iron, with these transporters working together with other proteins within the root in a joint, iron-dependent interplay between Al resistance and enhanced P uptake via changes in root system architecture. Very recent research findings suggests these interactions to be part of a P deficiency stress response, suggesting that this mechanism could have evolved in crop species to improve global adaptation to acid soils. Should this pleiotropism prove functional in crop species grown on acidic soils, molecular breeding based on Al resistance genes may have a much broader impact on crop performance than previously thought. To explore this possibility, this talk will review the potential impact of Al resistance proteins on the dynamics of root-mediated solubilization and uptake of soil P. This will be used to provide the foundation to discuss this pleiotropy as a genetic linkage between Al resistance and P efficiency. The talk will then be concluded by exploring what may be needed to enhance the utilization of Al resistance genes to enhance both resistance to toxic Al in the soil and improve P acquisition, thus improving crop yields on acid soils.

PL- 2

Plant-microbial interactions in low pH soils: Options for maximising soil biological fertility

***Abbott, Lynette K*[1]**
[1]UWA School of Agriculture and Environment and UWA Institute of Agriculture, The University of Western Australia
; lynette.abbott@uwa.edu.au

This presentation highlights selected case studies of key plant-microbe interactions in agricultural soils influenced by low pH, with pasture grazed by sheep in south-western Australia used as a model agricultural system. Effective management of agricultural soils influences their chemical and biological fertility. This occurs in a pedological context that dictates the primary characteristics of soil physical fertility. The first case study considers plant-microbial interactions in the rhizosphere. The second extends rhizosphere interactions to include the ubiquitous arbuscular mycorrhizal fungi that bridge the soil-plant interface. The third case study highlights the role of low pH in influencing symbiotic nitrogen fixation with pasture legumes, and the final case study highlights plant-pathogen interactions in acid soils. Identifying mechanisms underlying interactions between plants and soil microorganisms at low pH can be challenging (Robson and Abbott 1989).

Increasing interest in use of alternative fertilisers and biostimulants (Abbott et al. 2018) has potential to influence, positively or negatively, effective functioning of rhizosphere communities in pastures. As an example, rhizosphere microbial responses to application of fertilisers of different solubility was plant-dependent; bacterial OTU richness was correlated with the pH of rhizosphere soil for subterranean clover but not for annual ryegrass (Mickan et al. 2018).

Arbuscular mycorrhizal fungi are known to differ in their responses to low soil pH (e.g. Porter et al. 1987a, Porter et al. 1987b), even at the same location where soil pH is highly variable. Low pH may influence some stages of the life cycle of arbuscular mycorrhizal fungi but not others. Even if there is no affect of acidity on colonisation of roots by arbuscular mycorrhizal fungi, low pH may influence the relative abundance of taxa of mycorrhizal fungi present in these soils. However, the overall symbiotic function may or may not be affected by changes in relative abundance of the fungi. This needs to be considered when deciding whether or not there is a likely practical benefit of introducing inoculant arbuscular mycorrhizal fungi (Hart et al. 2017).

Nodulation of pasture legumes can be significantly influenced by low pH (e.g. Cheng et al. 2002). Furthermore, symbiotic nitrogen fixation can be limited by genetic instability of root nodule bacteria but this differs among bacterial taxa (e.g. some bacteria that nodulate *Medicago* spp. can be more genetically unstable than bacteria that nodulate *Ornithopus spp.*). Many years of investigations of mechanisms underlying successful nitrogen fixation in legume-based pastures of south-western Australia illustrate a comprehensive case study relevant to management of pastures in low pH soils (e.g. Howieson and Ewing 1989).

Consortia of root pathogens can influence plant production in acid soils (e.g. McNish 1986). Interactions between plant nutrition, plant growth, and susceptibility to root disease have been well-studied. Management practices that ameliorate root disease may have either direct or indirect impacts on plant pathogens associated with pastures.

CONCLUSION

Understanding processes that lead to beneficial interactions between plants and soil microorganisms at low pH is essential for determining appropriate options for soil management (Robson and Abbott 1989). Success in selecting management practices that support beneficial rather then detrimental interactions between plants and soil microorganisms at low pH can be facilitated by knowledge of microbial life history strategies. All of the plant-microbe interactions mentioned above must to be managed simultaneously in low pH pasture soils, and this ensures soil biological fertility is held in balance with soil chemical and physical fertility (Abbott and Murphy 2003). The challenge is to avoid risks associated with ameliorative practices suited to one category of plant-microbe interaction that may have sub-optimal outcomes for other plant-microbe interactions.

ACKNOWLEDGEMENTS

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REFERENCES

Abbott, L.K., Murphy, D.V., 2003. Soil Biological Fertility: A Key to Sustainable Land Use in Agriculture, Kluwer Academic, The Netherlands (Reprinted Springer, 2007) pp 264

Abbott, L.K., Macdonald, L.M., Wong, M.T.F., Webb, M.J., Jenkins, S.N., Farrell, M., 2018. Potential roles of biological amendments for profitable grain production – A review. Agriculture, Ecosystems and Environment **256:**34-50

Howieson, J.G., Ewing, M.A., 1989. Annual species of *Medicago* differ greatly in their ability to nodulate on acid soils. Australian Journal of Agricultural Research **40:** 843-850

McNish, G.C., 1986. Effects of fumigation on soil nitrogen, plant nitrogen and root disease incidence in wheat at Wongan Hills, Western Australia. Australian Journal of Soil Research **24:** 81-93

Mickan, B., Alsharmani, A., Solaiman, Z. M., Abbott, L.K., 2018. Plant-dependent soil bacterial responses following amendment with mineral and chemical fertilisers are driven by soil pH. (Poster, 10th-PSILPH2018)

Hart, M., Antunes, P., Chaudhary, B., Abbott, L., 2017. Fungal inoculants in the field – is the reward greater than the risk? Functional Ecology **32:** 126-135

Porter, W.M., Robson, A.D., Abbott, L.K., 1987a. Factors controlling the distribution of VA mycorrhizal fungi in relation to soil pH. Journal of Applied Ecology **24:** 663-672

Porter, W.M., Robson, A.D., Abbott, L.K., 1987b. Field survey of the distribution of VA mycorrhizal fungi in relation to soil pH. Journal of Applied Ecology **24:** 659-662

Cheng, Y., Howieson, J.G., O’Hara, G.W., Watkin, E.L. J., Souche, G., Jillard, B., Hingsinger, P., 2004. Proton release by roots of *Medicago murex* and *Medicago sativa* growing in acidic conditions, and implications for rhizosphere pH changes and nodulation at low pH. Soil biology and Biochemistry **36:** 1357-1365

Robson, A.D., Abbott, L.K., 1989. The effect of soil acidity on microbial activity in soil. In: Soil Acidity and Plant Growth (Ed. A.D. Robson). Academic Press, Sydney, pp139-165

PL-3

Managing phosphorus under acid soils environment

Mohamed M Hanafi[123]
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia,
[2]Laboratory of Climate-Smart Food Crop Production, Institute of Tropical Agriculture and Food Security (ITAFoS), Universiti Putra Malaysia.
[3]Laboratory Science and Technology, Institutes of Plantation Studies (IPS), Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia.
; mmhanafi@upm.edu.my

ABSTRACT
Most Malaysian soils are acidic and low in phosphorus (P) status and therefore all crops require an addition of P fertiliser to achieve a reasonably good yield target. Since no phosphate mineral deposits are found in Malaysia, the P sources are mostly imported from various countries. Importation of this P source for agricultural production will continue to rise in the future due to an expansion of the agriculture sector and the sector's status as the third engine economic growth. A majority of the P sources imported are in the form of ground PR. This PR is used as direct application fertiliser for perennial and annual crops production because it is cheaper source of P per unit compared to the water soluble P sources. The performance of various PR sources depends on the characteristics of PR and soil, types of crops, and environmental conditions. Thus, evaluation of P sources in a real field situation is very important to determine the best source of PR for any soil-crop-environmental combination. Judicious selection of P sources should be based on the most cost effective using RAE in that particular situation. However, directly applied PR sources are not effective at all in some situations. Then, there is a need to improve their agronomic effectiveness which would make the PR sources more economically attractive to farmers. In this presentation, the following strategies, such as physical, chemical (acidulation and partial acidulation), and biological approaches (micro-organisms, macro-organisms, crop/pasture improvement, and crop mixtures) will be discussed. This strategy may be used individually and/or in combination.

PL-4

Regulation and mathematical modeling of essential nutrient transport

Toru Fujiwara[1]
[1]Graduate School of Agricultural Life Sciences, The University of Tokyo, Tokyo, Japan
; atorufu@mail.ecc.u-tokyo.ac.jp

For the growth of crops in acid soils, it is important to maintain uptake of essential nutrients from soil. Low pH affects availability of nutrients in soils and also membrane potentials important for secondary active transport of nutrients. In this presentation I would like to introduce the case of boron transport. We have been studying boron transport mechanisms mainly in Arabidopsis and identified several major transporters (Takano et al., 2002, 2006, Tanaka et al., 2008, Miwa et al., 2013, 2014) and characterized physiological roles of the transporters. We have been successful in generating transgenic plants tolerant to low- or high-boron conditions (Miwa et al 2006, 2007, Uraguchi et al 2014). Mathematical modeling of boron transporters allowed to predict the boron distribution within the cell.

As the availability of boron varies widely, regulation of transporter expression is a key element. We have demonstrated that plants regulate boron transport process by regulating abundance of transporter proteins (Takano et al 2005, 2010, Nakagawa et al 2007, Miwa et al 2013). We recently identified a novel mechanism to regulate transporter expression in response to boron. *Arabidopsis thaliana NIP5;1*, a boric acid transporter required for efficient uptake of boron from soil, is expressed to high level under low B conditions. We found that ribosome is involved in the regulation of *NIP5;1* mRNA accumulation in response to boron (Tanaka et al. 2016). B-dependent regulation of *NIP5;1* transcript accumulation is regulated mainly through B-dependent mRNA degradation (Tanaka et al. 2011) and this degradation is regulated through boron dependent ribosome stall at AUGUAA sequence in the 5'UTR of the gene. Highlighting the importance of translational regulation. Ribosome profiling suggested that regulation patterns in response to boron condition are almost independent between the transcript accumulation and translational regulation.

In this presentation I would also like to mention about the mathematical modeling of boron transport. We have also established mathematical model to understand the overall regulation systems of boron transport in roots incorporating spatial patterns (Shimotono and Sotta et al 2015) and boron dependent regulation (Sotta et al 2017). These mathematical modeling will be useful for comprehensive understanding of boron transport processs and regulation in roots.

REFERENCES

Sotta et al. Rapid transporter regulation prevents substrate flow traffic jams in boron transport.
eLife **6:**e27038 **(2017)**

Tanaka et al. The Minimum Open Reading Frame, AUG-Stop, Induces Boron-Dependent Ribosome Stalling and mRNA Degradation ***Plant Cell*** **28:**2830-2849 **(2016)**

Shimotohno and Sotta et al. Mathematical modeling and experimental validation of the spatial distribution of boron in the root of *Arabidopsis thaliana* identify high boron accumulation in the tip and predict a distinct root tip uptake function. ***Plant Cell Physiol.*** **56:**620-630 **(2015)**

Uraguchi et al Generation of boron-deficiency-tolerant tomato by overexpressing an *Arabidopsis thaliana* borate transporter AtBOR1. ***Front Plant Sci***.**5:**125. **(2014)**

Miwa et al. Arabidopsis thaliana BOR4 is upregulated under high boron conditions and confers tolerance to high boron. ***Soil Science and Plant Nutrition*** **60:** 349-355 **(2014)**

Hanaoka et al. OsNIP3;1, a rice boric acid channel, regulates boron distribution and is essential for growth under boron-deficient conditions. ***Plant Journal*** **78:** 890-902 **(2014)**

Miwa et al. Roles of BOR2, a Boron Exporter, in Cross Linking of Rhamnogalacturonan II and Root Elongation under Boron Limitation in Arabidopsis ***Plant Physiology*** **163:** 1699–1709 **(2013)**

Tanaka et al. Roles of Pollen-Specific Boron Efflux Transporter, OsBOR4, in the Rice Fertilization Process ***Plant Cell Physiol***. **54:**2011-2019 **(2013)**

Tanaka et al. Boron dependent degradation of *NIP5;1* mRNA for acclimation to excess boron conditions ***Plant Cell*** **23:** 3547–3559 **(2011)**

Takano et al. Polar localization and degradation of Arabidopsis boron transporters through distinct trafficking pathways. ***Proc. Natl. Acad. Sci. U.S.A.*** **17**, 5220-5225 **(2010)**

Tanaka et al. NIP6;1 is a boric acid channel for preferential transport of boron to growing shoot tissues in *Arabidopsis thaliana*. ***Plant Cell*** **20:**2860-2875 **(2008)**

Miwa et al. Plants tolerant of high boron levels. ***Science*** **318:**1417 **(2007)**

Nakagawa et al. Cell-type specificity of the expression of Os *BOR1*, a rice efflux boron transporter gene, is regulated in response to boron availability for efficient boron uptake and xylem loading. ***Plant Cell*** **19:** 2624–2635 **(2007)**

Miwa et al. Improvement of seed yields under boron-limiting conditions through overexpression of BOR1, a boron transporter for xylem loading, in *Arabidopsis thaliana*. ***Plant J.*** **46:** 1084-1091 **(2006)**

Takano et al. The Arabidopsis major intrinsic protein NIP5;1 is essential for efficient boron uptake and plant development under boron limitation. ***Plant Cell*** **18:** 1498-1509 **(2006)**

Takano et al. Endocytosis and degradation of BOR1, a boron transporter of *Arabidopsis thaliana*, regulated by boron availability ***Proc. Natl. Acad. Sci. USA*** **102:** 12276–12281 **(2005)**

Takano et al. *Arabidopsis* boron transporter for xylem loading. ***Nature*** **420:** 337-340 **(2002)**

K-1

Regulation of aluminum tolerance in barley

Jian Feng Ma[1]*, Miho Fujii-Kashino[1], Naoki Yamaji[1], Miki Yamane[1], Daisuke Saisho[1], and Kazuhiro Sato[1]
[1]Institute of Plant Science and Resources, Okayama University, Chuo 2–20–1, Kurashiki 710–0046, Japan,
; maj@rib.okayama-u.ac.jp

INTRODUCTION

Barley (*Hordeum vulgare* L.) is one of the most Al-sensitive cereal crops. However, several studies have reported that there is a wide genotypic variation in Al tolerance within barley species. Different from rice, whose Al tolerance is controlled by multiple genes, Al tolerance of barley is only controlled by a major gene on chromosome 4H, HvAACT1 (Al-activated citrate transporter 1) (Furukawa et al., 2007), a member of multidrug and toxic compound extrusion (MATE) family. It functions to mediate citrate release from the roots to detoxify Al externally in the rhizosphere. The expression of *HvAACT1* is not affected by Al, but its expression level is constitutively higher in the Al-tolerant accessions than the Al-sensitive accessions (Furukawa et al., 2007).

The differential expression of *HvAACT1* in different barley accessions has been linked to the insertion of a 1023 bp CACTA-like transposon at the upstream of HvAACT1 in some Al-tolerant accessions in East Asia (Fujii et al., 2012). This insertion resulted in enhanced expression level and altered location of HvAACT1 from mature root region to the root tips (Fujii et al., 2012). Here, we report a novel mechanism regulating HvAACT1 expression in European barley accessions, which is different from those in East Asia.

MATERIALS AND METHODS

Invers PCR was performed to identify unknown genome sequence of *HvAACT1*. To examine the presence of insertions in different barley accessions, genome PCR was performed using Quick taq® HS DyeMix (TOYOBO). To determine the transcriptional start site of HvAACT1 in FM404, 5’-RACE was performed by GeneRacer kit (Thermo Fisher Scientific). Tissue-dependent expression of HvAACT1 ORF and 5’-UTR of the roots (cvs. Golden promise and FM404) was determined by absolute quantitative real-time PCR using standard curve method in different root segments. The promoter activity was performed by introducing the GFP-fused constructs into onion epidermal cells by Helios® Gene Gun system (BIO-RAD). The methylation was investigated by bisulfite sequencing. A panel of 274 domesticated barley representative of global barley diversity and 289 wild barley accessions were used to investigate the presence of the MRL insertion.

RESULTS AND DISCUSSION

FM404 is an Al-tolerant accession developed in Brazil. We found that this accession showed high expression of *HvAACT1*, but did not contain 1-kb insertion reported in the promoter region. We therefore compared the upstream sequence of *HvAACT1* between FM404 and Morex, an Al-sensitive cultivar. With help of inverse PCR, we were able to detect an insertion of at least 15.3 kb in the upstream of *HvAACT1* in FM404 (Fig. 1). This insertion did not show any similarity with the 1-kb insertion reported before (Fujii et al., 2012). BLAST search showed that this insertion contained multiple retrotransposon-like sequence with long terminal repeat (LTR) (designated to MRL insertion thereafter).

To identify promoter of *HvAACT1* in FM404, transcriptional start site (TSS) of *HvAACT1* was investigated by 5'-RACE. As a result, multiple TSS were detected at the MRL insertion region; 6.6 kb and 7.2 kb upstream from translational start site of *HvAACT1* in FM404, yielding three different types of transcript variants. A transient promoter assay showed that the MRL insertion functions as a promoter. Among 289 wild barley (*Hordeum vulgare* ssp. spontaneum (K.Koch) Thell) and 274 cultivated barley (*H. vulgare* ssp. vulgare) collections from different regions of the world, the insertion was detected in two wild barley accessions and 26 cultivated accessions. These cultivated accessions were distributed in European area only. However, among them, only 9 cultivated accessions showed higher expression of *HvAACT1* as FM404.

To investigate the novel mechanism regulating HvAACT1 expression, we compared methylation level of FM404 with two cultivated barley (SV222 and SV239) and two wild barley accessions (OUH601 and ICWB181646 (BCSP127)), which contain the MRL insertion but show low *HvAACT1* expression. The GC rich region was predicated to be located at the upstream of UTR-5 region in FM404; 2 kb from the common promoter. Comparison of methylation in these regions indicated that 97.8% of whole cytosine was unmethylated in FM404. By contrast, SV222 and SV239 and two wild barley showed much higher rate of methylation including CG, CHG and CHH compared with FM404.

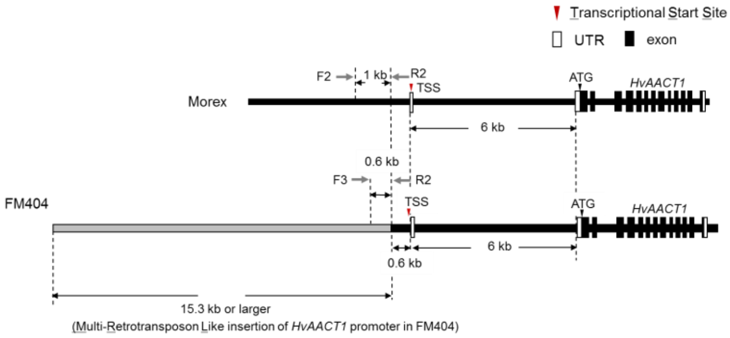

Fig. 1 Sequence comparison of *HvAACT1* genome between Al-sensitive (Morex) and Al-tolerant (FM404) barley accessions

CONCLUSION

Both multiple retrotransposons insertion and demethylation are required for enhancing *HvAACT1* expression. Furthermore, our results indicate that barley in East Asia and Europe has developed independent but equivalent strategies to cope with Al toxicity in acid soils.

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REFERENCES

Fujii, M., Yokosho, K., Yamaji, N., Saisho, D., Yamane, M., Takahashi, H., Sato, K., Nakazono, M., Ma, J. F. (2012). Acquisition of aluminium tolerance by modification of a single gene in barley. Nature Commun **3**:713.

Furukawa, J., Yamaji, N., Wang, H., Mitani, N., Murata, Y., Sato, K., Katsuhara, M., Takeda, K., Ma, J. F. (2007). An aluminum-activated citrate transporter in barley. Plant Cell Physiol. **48**:1081-1091.

K-2

A magnesium dechelatase gene is involved in low magnesium- induced leaf chlorosis and magnesium remobilization in rice.

***Zhi Chang Chen*[1], *Yu Yang Peng*[1] *and Jian Li*[1]**
[1]Root Biology Center, Fujian Agriculture and Forestry University, Fuzhou, 350002, China
; zcchen@fafu.edu.cn

INTRODUCTION

Magnesium (Mg) is the second most abundant cation in plants, which is essential for numerous physiological and biochemical processes (Maguire and Cowan, 2002). Mg deficiency is a growing problem in agriculture, particularly in acidic soils with low cation exchange capacity (Cakmak, 2013). In general, the typical symptoms for Mg deficiency are not only growth retardation, but also leaf interveinal chlorosis. Since Mg is relatively mobile in plants, the first visual deficiency symptoms appear from older leaves. However, recently developed views consider young mature leaves are more vigorous in Mg remobilization than other tissues, which leads to more evident leaf chlorosis in young mature leaves (Chen et al., 2018). In this study, we compared transcriptomic profiles of different leaves between rice exposed to low and high Mg concentrations. We found that a chloroplast Mg-dechelatase gene *OsSGR* is involved in the low Mg-induced leaf chlorosis and Mg remobilization.

MATERIALS AND METHODS

Two-week-old rice seedlings were hydroponically grown in 1/2 Kimura nutrient solution containing 5 µM (low Mg) or 250 µM (high Mg) of $MgCl_2$ for different days. Mg concentration in the plant tissues was determined by ICP-MS. The chlorophyll content (SPAD value) was measured using a chlorophyll meter (SPAD-502 Plus). The photosynthetic parameters were measured by a portable photosynthesis system (LI-6800, Li-Cor). *ossgr* mutants were generated by CRISPR/Cas9 technique. The RNA-seq was conducted using Illumina HiSeqTM 2500 by Novogene Biotechnology Co. (Beijing, China).

RESULTS AND DISCUSSION

Young mature leaves are more susceptible to Mg deficiency

Phenotypic observation showed that low Mg caused leaf chlorosis (SPAD value) was more serious in young mature leaves (L2, L3, L4), whereas hardly be observed in the older (L5, L6) and the newest leaves (L1) (Fig. 1). Consistent with the phenotype, net photosynthesis rate, stomatal conductance, transpiration rate and Mg concentrations in young mature leaves were much lower under low Mg condition. These results reveal that low Mg initially triggers the chlorosis in young mature leaves, but not in the older and new developing leaves in rice.

Mg deficiency triggers chlorophyll degradation by up-regulating the expression of *OsSGR* in young mature leaves

To investigate the molecular mechanisms underlying Mg deficiency in plants, we carried out Mg-responsive transcriptomic analysis of each leaf in rice through high-throughput RNA sequencing. Our data revealed that many metabolic processes and signaling pathways have been affected by low Mg stress. Among them, a Mg-dechelatase gene *OsSGR* was remarkably up-regulated by low Mg, particularly in the young mature leaves (L2, L3, L4). Therefore, low Mg induced leaf chlorosis in young mature leaves could be due to chlorophyll degradation by an active defense mechanism.

Knockout of *OsSGR* in rice decreases photosynthesis and Mg remobilization in young mature leaves

No significant differences were observed between wild-type and two *ossgr* mutants during the vegetative growth period under high Mg condition. When transferred to low Mg condition, young mature leaves of the wild-type leaves turned yellow, whereas those of the mutants were still green (Fig. 1). However, photosynthetic parameters including the net photosynthetic rate, transpiration rate and electron transfer rate were remarkably lower in mutants than wild-type rice, suggesting that *OsSGR* is required for preventing leaf from low Mg induced photodamage. Besides, mutants accumulated higher Mg in mature leaves than the wild-type rice, suggesting that Mg recycling requires *OsSGR*-regulated Mg dechelation from chlorophyll in young mature leaves.

CONCLUSION

Our results reveal that Mg deficiency triggers leaf chlorosis and Mg remobilization in young mature leaves by up-regulating a Mg-dechelatase gene *OsSGR* in rice. We conclude that low Mg induced chlorosis in young mature leaves is one possible strategy for plants to overcome Mg shortage by recycling chlorophyll Mg from mature leaves.

ACKNOWLEDGEMENTS

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REFERENCES

Cakmak, I. 2013. Magnesium in crop production, food quality and human health. Plant and Soil **368:** 1-4.

Chen, Z.C., Peng, W.T., Li, J. and Liao, H. 2018. Functional dissection and transport mechanism of magnesium in plants. Seminars in Cell & Developmental Biology **74:** 142-152.

Maguire, M.E. and Cowan, J.A. 2002. Magnesium chemistry and biochemistry. BioMetals **15:** 203-210.

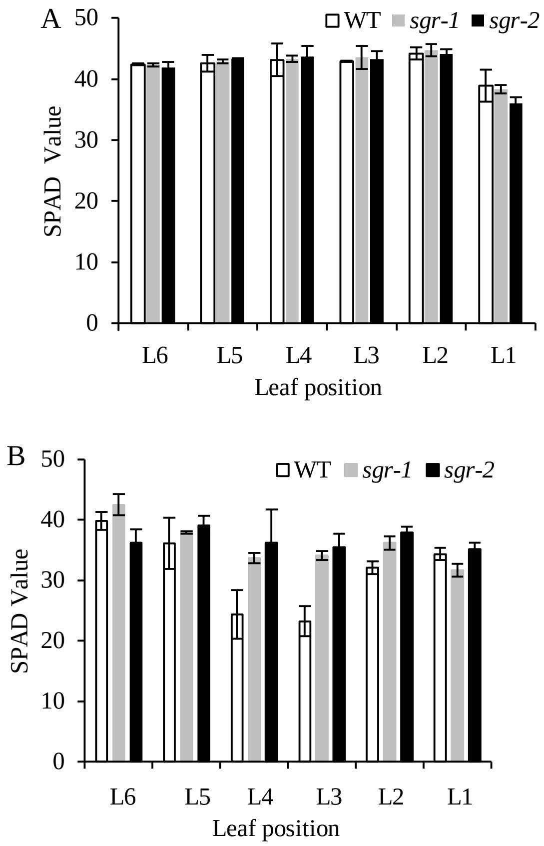

Figure 1. The SPAD value of each leaf (L6 to L1, old to young) in rice under high Mg (A) and low Mg (B) conditions

K-3

Unravelling the organic anion exudation from different root types of aluminium-resistant wheat and its effect on the root colonizing microbiome

***Akitomo Kawasaki*[1], *Paul G. Dennis*[2], *Emmanuel Delhaize*[1], *Alan E. Richardson*[1], *Ulrike Mathesius*[3], *Matthew Gilliham*[4], *Michelle Watt*[5] *and Peter R. Ryan*[1]**
[1]CSIRO Agriculture and Food, Canberra, ACT 2601, Australia
[2]School of Earth and Environmental Sciences, Faculty of Science, The University of Queensland, St Lucia, QLD, Australia
[3]Research School of Biology, Australian National University, Canberra, ACT, Australia
[4]School of Agriculture, Food and Wine, University of Adelaide, Glen Osmond, SA, Australia
[5]Forschungszentrum Jülich GmbH, Jülich, Germany
; peter.ryan@csiro.au

INTRODUCTION

One of the major factors limiting plant production in acid soils is aluminium (Al3+) toxicity. Al3+-resistant genotypes of wheat release malate and citrate anions from their roots which protect the sensitive root apices from the toxic Al3+ cations [1]. Whilst these exudates have been measured from the seminal roots of young (4-5 day old) hydroponically-grown wheat seedlings, it is unknown whether similar exudates occur from nodal and lateral roots of older plants. These experiments are more difficult to perform because older plants are prone to colonisation by microbes which can affect the detection of exudates and also it is technically challenging to maintain wheat in axenic conditions for many weeks. This study describes the development of a novel fully-enclosed hydroponic growth chamber that can grow wheat for more than four weeks in sterile conditions. Using this technique we were able to characterise the malate and citrate exudation from various root types of Al-tolerant wheat genotypes. Moreover, we used near-isogenic lines (NILs) varying in organic anion exudation, to test how these exudates influenced the microbiome around different root types grown in contrasting soils.

MATERIALS AND METHODS

A novel fully-enclosed and sterile hydroponic system was developed that was comprised of two compartments. The top compartment covered the shoots and the bottom compartment contains the roots and nutrient solution. The genetic materials included wheat NILs (NIL_Null and NIL_Citrate). Both lines possessed the major Al-resistance gene *TaALMT1* and therefore released malate from the seminal roots when treated with Al. However the lines possess different alleles of *TaMATE1B* and therefore only one line (NIL_Citrate) released citrate from the root apices (confirmed in seminal roots). The plants were cultivated in the hydroponic system for 28 days with regular changes of nutrients. Malate and citrate exudation from the seminal, nodal and lateral roots were measured using excised root apices

(5 mm) in CaCl2 solution, with (+Al treatment) and without (Ctrl treatment) Al3+. Relative expression of the *TaALMT1* and *TaMATE1B* genes were also quantified in each root type.
The microbiome on the tip and base of each root type was measured in three different soils (acid and Al-toxic, acid but non Al-toxic, and non acid soils). The microbiome was characterised with Illumina sequencing by targeting the bacterial 16S rRNA genes.

RESULTS AND DISCUSSION

Malate and citrate exudation from seminal, nodal and lateral roots of mature wheat plants were characterized for the first time. Al3+-activated malate efflux was detected from all root types at 2-3 nmol root apex-1 h-1 which is similar or greater than previous reports [2]. Exudation was similar from the seminal and nodal roots but the lateral roots released less malate, perhaps due to their smaller diameters. A constitutive release of citrate was measured from the seminal and nodal roots (~80 pmol root apex-1 h-1) but no citrate release was detected from the lateral roots. Since lateral roots can occupy more than 95% of total root length in cereal crops [3] these results are consistent with the minor role of citrate release in Al3+ resistance compared to malate release [1].
The microbial communities colonizing the rhizosphere and root surfaces can affect plant growth and the exudates from roots are important for shaping these communities [4]. We tested how root exudates influenced the microbiome structure around different root types of the wheat NILs grown in contrasting soils. These results are being analysed and will be presented at the conference.
Al-resistance in wheat involves the release of malate and citrate but, prior to this study, this response has only be reported and measured on the seminal roots of young seedlings. Development of a novel hydroponic system enabled us to cultivate wheat plants axenically for 28 days without any detectable contamination. This allowed us to measure organic anion efflux from the different roots types of more mature plants. Al3+-activated malate release was detected from the seminal, nodal and lateral roots whereas citrate efflux was only detected from the seminal and nodal roots. The results will also determine how these fluxes affect the microbiome around the different root types. This information may help us understand how the microbiome can be manipulated in the future to benefit plant performance.

REFERENCES

Ryan PR, Raman H, Gupta S, Horst WJ, Delhaize E: A second mechanism for aluminum resistance in wheat relies on the constitutive efflux of citrate from roots. *Plant Physiol* 2**009**, **149:**340-351.

Ryan PR, Delhaize E, Randall PJ: Characterization of Al-stimulated efflux of malate from the apices of Al-tolerant wheat roots. *Planta* **1995, 196:**103-110.

Ito K, Tanakamaru K, Morita S, Abe J, Inanaga S: Lateral root development, including responses to soil drying, of maize (*Zea mays*) and wheat (*Triticum aestivum*) seminal roots. *Physiol Plant* **2006, 127:**260-267.

Sasse J, Martinoia E, Northen T: Feed your friends: Do plant exudates shape the root microbiome? *Trends Plant Sci* **2018, 23:**25-41.

K-4

The mechanism underlying regulation of internal P reutilization by different nitrogen forms in rice in acid soils

***Ren Fang Shen*[1], *Xiao Fang Zhu*[1], *Chun Quan Zhu*[1]**
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China
; rfshen@issas.ac.cn

INTRODUCTION

Acid soils cover about 50% of the world's potentially arable soils (Kochian et al, 2004), and besides aluminium (Al) toxicity, suboptimal levels of phosphorous (P) is also the major limiting factor for the productivity of acid soils (von Uexkull and Mutert, 1995; Kochian et al, 2004). To enhance the absorption of P under conditions of P starvation and maintain internal P homeostasis, plants are able to remodel the architecture and morphology of their root systems (Fitter et al., 2002; Lynch and Brown, 2001); secrete protons, organic acids, and phosphatases from their roots (Noriharu et al., 1990, Hinsinger 2001, Vance 2008); alter their metabolic pathways; increase their ribonuclease (RNase) activity; transfer P from older to younger organs (Liang et al., 2014); and reutilize cell-wall P under conditions of P starvation (Zhu et al., 2015).

Besides P, nitrogen (N) is also a pivotal macronutrients for plant growth, and NH_4^+ and NO_3^- are the two major N sources that are taken up by plant roots (Marschner, 1995; Falkengren-Grerup et al., 2000). Zeng et al. (2012) demonstrated that in rice the increased P uptake in the presence of NH_4^+ instead of NO_3^- is due to increased activity of the plasma membrane H^+-ATPase. However, whether these two different forms of nitrogen affect P remobilization in rice under P-deprivation conditions remains still unclear. Here, we uncovered the mechanism underlying regulation of internal P reutilization by different nitrogen forms in rice in acid soils.

MATERIALS AND METHODS

Rice (*Oryza sativa*) spp. *japonica* 'Nipponbare' ('Nip') and *indica* 'Kasalath' ('Kas') were used in this study. After seeds were surface-sterilized, they were cultivated in full-strength nutrient solution for 7d according to Zhu et al. (2015). Then, uniform seedlings were planted in 1.5-L pots (10 seedlings per pot) with the following treatments: +P+NH_4^+, –P+NH_4^+, +P+NO_3^-, –P+NO_3^-, –P+NH_4^++c-PTIO, –P+NO_3^-+c-PTIO, +P+1mM NO_3^-, +P+0 mM NO_3^-, +P+ 0 mM NO_3^-+SNP, +P+ 1 mM NO_3^-+SNP, –P+1 mM NO_3^-, –P+0 mM NO_3^-, –P+ 0 mM NO_3^-+SNP and –P+ 1 mM NO_3^-+SNP. For NH_4^+ and NO_3^- treatments, 1 mM NH_4Cl and different concentrations of $NaNO_3$ were applied to the nutrient solution. The final concentration of SNP and c-PTIO were 2.5 μM and 10 μM, respectively. The pH was adjusted to 5.5 with 5 mM MES. The soluble P in rice were measured as described by Zheng et al. (2009). Extraction of cell walls and cell wall pectin were carried out according to Zhu et al. (2015). Pectin content was assayed according Blumenkrantz and Asboe-Hansen (1973). Cell wall P content was conducted according to Zhu et al. (2015). The accumulation of endogenous nitric oxide (NO) in rice roots was measured according to Besson-Bard et al. (2009). Extraction and analysis of pectin methylesterase (PME) activity were carried out in reference to Zhu et al. (2012). Quantitative real-time PCR analysis was in reference to Zhu et al (2016). Each experiment was repeated at least three times, and one set of data is shown in the Results. Data were analyzed by one-way ANOVA and the mean values were compared by Duncan's multiple range test. Different letters indicate that the mean values were statistically different at the $P \leq 0.05$ level.

RESULTS AND DISCUSSION

To investigate the effects of NH_4^+ and NO_3^- on the reutilization of P in rice roots, one *japonica* variety, 'Nipponbare' (Nip) and one *indica* variety, 'Kasalath' (Kas) were used. After grown for seven days in the nutrient solution with NH_4^+ or NO_3^- as the sole N source under P sufficient (+P) or P deficient (-P) conditions, more soluble P were found under -P conditions when grown with NH_4^+ as the N source, irrespectively in Nip and Kas (Fig. 1), suggesting that NH_4^+ may stimulate P reutilization in both rice cultivars. Then, to investigate the relationship between NO_3^- and P in rice, plants were grown for 7 d in +P or –P nutrient solutions with one of four NO_3^- concentrations (0, 0.1, 1 and 10 mM). It is interesting that in the –P condition, P content was significantly higher in plants grown with 0 mM or 0.1 mM NO_3^- nutrient solution compared with 1 mM or 10 mM NO_3^- (Fig. 2), implying NO_3^- and P content in rice may possess a close relationship with each other in rice.

As about 50% of the total root P is accumulated in the root cell walls of rice, and pectin contributes to the different P reutilization efficiency in Kas and Nip (Zhu et al., 2015), P retention in the cell walls and the pectin content were measured. As shown in Fig. 3A and 3B, less P were accumulated in the cell walls of both rice cultivars when NH_4^+ was applied as the sole N source under P-deficient conditions, in company with the increment of the pectin content (Fig. 3C and 3D), suggesting that the reduction of P in the NH_4^+-treated cell wall maybe related to the increment of the cell wall pectin content. Moreover, the negative charges of the cell wall pectin was generated by the activity of PME, and as shown in Fig. 4, the PME activity was significantly higher after NH_4^+ treatment than after NO_3^- treatment under different P conditions, indicates that NH_4^+ treatment may enhance negative charges in the cell wall. Then, to clarify whether NO_3^- affects rice cell wall P reutilization, rice root cell walls were extracted for further study. As shown in Fig. 5, less P was deposited in the cell walls of rice roots when NO_3^- was absent from the –P nutrient solution compared with the addition of 1 mM NO_3^-, in company with the higher pectin content and the PME activity, indicating that NO_3^- inhibited the remobilization of cell wall P through repressing the pectin content and PME activity under –P conditions.

Because nitrogen form can affect endogenous NO content (Chen et al. 2010), and NO is involved in P deficiency (Wang et al., 2010), we hypothesized a direct relationship between nitrogen form (NH_4^+ or NO_3^-), P condition (+P or -P) and NO production. NH_4^+ significantly induced the NO production independent of P conditions (Fig. 6), while after the addition of the NO scavenger (c-PTIO), the fluorescence associated with the presence of NO was decreased (Fig. 7), in company with the elimination of the NH_4^+-induced increment of root and shoot soluble P content (Fig. 8A and 8B) and the elimination of the difference of root cell wall P between NH_4^+ and NO_3^- treatment (Fig. 8C) under -P conditions, indicates that NO plays an important role in the NH_4^+ -regulated reutilization of cell wall P in rice root. Notably, the addition of 1 mM NO_3^- to the nutrient solution significantly decreased the NO content compared with the 0 mM NO_3^- treatment in either the +P or –P condition (Fig. 9), indicating that the existence of NO_3^- in nutrient solution inhibited NO accumulation in rice root. To further clarify whether NO is involved in the reutilization of P in root cell walls, the NO donor sodium nitroprusside (SNP) was applied to the nutrient solution. SNP increase soluble P concentration in rice under different NO_3^- treatments, and the increment of soluble P even without exogenous P implied that NO is important for the P remobilization (Fig. 10). After addition of SNP to the nutrient solution, cell wall P content was significantly decreased, and the activity of root cell wall PME was significantly increased (Fig. 11). An increase in pectin content was induced only in the

1 mM NO_3^- condition, suggesting that there might be a threshold level of NO needed to stimulate pectin synthesis in rice root; notably, both high and low NO contents inhibited the synthesis of pectin.

To determine whether NH_4^+ influence the translocation of P from roots to shoots, expression of genes that involved in P translocation from roots to shoots were analyzed by quantitative RT-PCR. Under P-sufficient conditions, there were no significant differences between NH_4^+ and NO_3^- treatment, except for *OsPT2* in the Nip cultivar, which showed higher expression in NH_4^+ treatment than NO_3^- treatment (Fig. 12). Interestingly, under -P conditions, NH_4^+ strongly induced *OsPT2* expression in both Nip and Kas (Fig. 12A and 12D), indicating that *OsPT2* may be involved in the NH_4^+ alleviated P deficiency.

CONCLUSION

P deficiency is a major limiting factor for crop production in acid soil, thus, accumulating evidences have focused on the importance of the P reutilization in acid soil. NH_4^+ and NO_3^- are two major sources of inorganic N for rice, however, they exhibited different effect on P reutilization in rice. NH_4^+ positively regulates pectin content and activity of PME in root cell walls and upregulated the expression of phosphate transporter gene *OsPT2* under -P conditions, thereby increased the remobilization of P in rice root cell wall and increased the translocation of soluble P from rice root to shoot, finally showed a higher soluble P content in rice; while the present of NO_3^- significantly decreased the reutilization of cell wall P by inhibited the pectin content and the activity of PME in root cell walls, thereby decreased the soluble P content in rice under P deficient condition. In addition, the signal molecule NO was involved in the NH_4^+ stimulated cell wall P remobilization and NO_3^- depressed cell wall P reutilization, suggesting NO plays important roles in the regulation of internal P reutilization by different nitrogen forms in rice in acid soil that under P deficient condition. In conclusion, NH_4^+ facilitated the reutilization of cell wall P through signal molecule NO to regulate the cell wall pectin content, however, the NO_3^- displayed a reverse process.

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REFERENCES

Ae N, Arihara J, Okada K, Yoshihara T, Johansen C. 1990. Phosphorus uptake by pigeon pea and its role in cropping systems of the Indian subcontinent. *Science,* **248**: 477-480.

Besson-Bard A, Gravot A, Richaud P, Auroy P, Duc C, Gaymard F, Taconnat L, Renou JP, Pugin A, Wendehenne D. 2009. Nitric Oxide Contributes to Cadmium Toxicity in Arabidopsis by Promoting Cadmium Accumulation in Roots and by Up-Regulating Genes Related to Iron Uptake. *Plant Physiology,* **149**: 1302-1315.

Blumenkr.N, Asboehan.G. 1973. New method for quantitative determination of uronic acids. *Analytical Biochemistry,* **54**: 484-489.

Chen WW, Yang JL, Qin C, Jin CW, Mo JH, Ye T, Zheng SJ. 2010. Nitric oxide acts downstream of auxin to trigger root ferric-chelate reductase activity in response to iron deficiency in Arabidopsis. *Plant Physiology,* **154**: 810-819.

Falkengren-Grerup U, Mansson KF, Olsson MO. 2000. Uptake capacity of amino acids by ten grasses and forbs in relation to soil acidity and nitrogen availability. *Environmental and Experimental Botany,* **44**: 207-219.

Fitter A, Williamson L, Linkohr B, Leyser O. 2002. Root system architecture determines fitness in an Arabidopsis mutant in competition for immobile phosphate ions but not for nitrate ions. *Proceedings Biological Sciences,* **269**: 2017.

Hinsinger P. 2001. Bioavailability of soil inorganic P in the rhizosphere as affected by root-induced chemical changes: a review. *Plant and Soil,* **237**: 173-195.

Kochian LV, Hoekenga OA, Pineros MA. 2004. How do crop plants tolerate acid soils? - Mechanisms of aluminum tolerance and phosphorous efficiency. *Annual Review of Plant Biology,* **55**: 459-493.

Liang CY, Wang JX, Zhao J, Tian J, Liao H. 2014. Control of phosphate homeostasis through gene regulation in crops. *Current Opinion in Plant Biology,* **21**: 59-66.

Lynch JP, Brown KM. 2001. Topsoil foraging - an architectural adaptation of plants to low phosphorus availability. *Plant and Soil,* **237**: 225-237.

MARSCHNER, H. 1995. Mineral Nutrition of Higher Plants (2nd edn.) Academic. London.

Vance CP. 2008. Plants without arbuscular mycorrhizae. **7**: 117-142.

Vonuexkull HR, Mutert E. 1995. Global extent, development and economic impact of acid soils. *Plant and Soil,* **171**: 1-15.

Wang BL, Tang XY, Cheng LY, Zhang AZ, Zhang WH, Zhang FS, Liu JQ, Cao Y, Allan DL, Vance CP, Shen JB. 2010. Nitric oxide is involved in phosphorus deficiency-induced cluster-root development and citrate exudation in white lupin. *New Phytologist,* **187**: 1112-1123.

Zeng HQ, Liu G, Kinoshita T, Zhang RP, Zhu YY, Shen QR, Xu GH. 2012. Stimulation of phosphorus uptake by ammonium nutrition involves plasma membrane H+ ATPase in rice roots. *Plant and Soil,* **357**: 205-214.

Zheng LQ, Huang FL, Narsai R, Wu JJ, Giraud E, He F, Cheng LJ, Wang F, Wu P, Whelan J, Shou HX. 2009. Physiological and Transcriptome Analysis of Iron and Phosphorus Interaction in Rice Seedlings. *Plant Physiology,* **151**: 262-274.

Zhu XF, Lei GJ, Jiang T, Liu Y, Li GX, Zheng SJ. 2012. Cell wall polysaccharides are involved in P-deficiency-induced Cd exclusion in Arabidopsis thaliana. *Planta,* **236**: 989-997.

Zhu XF, Wang ZW, Wan JX, Sun Y, Wu YR, Li GX, Shen RF, Zheng SJ. 2015. Pectin enhances rice (*Oryza sativa*) root phosphorus remobilization. *Journal of Experimental Botany,* **66**: 1017-1024.

Zhu XF, Zhu CQ, Zhao XS, Zheng SJ, Shen RF. 2016. Ethylene is involved in root phosphorus remobilization in rice (*Oryza sativa*) by regulating cell-wall pectin and enhancing phosphate translocation to shoots. *Annals of Botany,* **118:** 645-653.

K-5

Weathered soils in Southeast Asia can sustain oil palm production in the long run

***Shamshuddin, J.*[1], *C.I. Fauziah*[1] *and O. Radziah*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 4300 Serdang, Selangor, Malaysia
; shamshud@upm.edu.my

INTRODUCTION

Among the most common soils available for crop production in the upland areas of Southeast Asia are highly weathered soils classified as Ultisols and Oxisols. The former are soils having argillic diagnostic horizon (Bt) with low pH, CEC, but high in Al, while the latter have oxic diagnostic horizon (Bo) with low pH, CEC and also high in Al. These soils are mainly utilized for oil palm cultivation, especially those found in Malaysia and Indonesia. This paper will explain or tries to find out if the fertility the soils is able to sustain oil palm growth and/or production in the long run.

Charge generation in highly weathered tropical soils

Ultisols and Oxisols are strongly leached and highly weathered, whose mineralogy dominated by kaolinite and oxides of Fe and/or Al. The pH of Ultisols is close to 4, while that of the Oxisols is moving towards 5, which are related to the difference in the chemical and/or mineralogical properties of the soils. The soils have both negative and positive charges, which are either permanent (Q_P) or variable (Q_V) in nature, with the amount dependent on soil mineralogy. These charges are respectively developed by the process of isomorphic substitution (called permanent) or adsorption/desorption of potential determining ions (H^+ and OH^{-1}) (called variable). It can be assumed that total charge (Q_T) in a soil is the sum of Q_P and Q_V, with the latter being represented by this equation:

$Q_V = k\ (pH_o - pH)$; pH_o is the point of zero charge, while k is a constant

That means Q_V can be manipulated by changing pH_o and/or increasing soil pH. It is known that soil pH_o can be decreased by applying organic matter. Applying ground basalt onto the soil can both lower its pH_o and increase pH. As such, the CEC of soils in an oil palm plantation in Southeast Asia can be increased by agronomic manipulation.

Fertilizer application in oil palm plantations

Fertilizer application is about 60% of the total cost of palm oil production. The fertilizers so applied to keep oil palm in the plantation going are ammonium sulfate [$(NH_4)_2SO_4$], rock phosphate (Ca_3PO_4), Muriate of Potash (KCl) and kieserite ($MgSO_4.H_2O$). S and Ca will be concomitantly available when N- and P-fertilizers are applied. Ca and Mg can also be made available if ground magnesium limestone is applied to increase soil pH to enhance oil palm growth. Continuous application of ammonium sulfate onto soil would lower its pH. However, the decrease in soil pH can be somewhat offset by the specific adsorption of SO_4^{2-} ions on the surface of oxides of

Fe. Fortunately, oil palm is an acid tolerant plant species that can survive even at the soil pH of 4.3, and it can also withstand the problem caused by Al^{3+} activity of about 100 µM. So, at the soil pH of Ultisols and Oxisols of 4-5, oil palm in the plantation can still be productive.

Enhancement of soil productivity

When SO_4^{2-} ions are adsorbed onto the surface of Fe Oxides, soil pHo is slightly decreased. It is also known that pHo is decreased by ground basalt application. Besides, application of ground basalt onto a highly weathered soil would increase its pH in the long run. The decrease in pHo and/or increase in soil pH would eventually result in the increase of cation exchange capacity. Hence, agronomic practice involving application of ground basalt in combination with organic matter improves the productivity of soils in oil palm plantations. Addition of Ca via agronomic means further enhances soil productivity. Ca is, to a certain extent, able to reduce the negative effect of Al^{3+} toxicity. Some of the SO_4^{2-} ions released by the fertilizers react with Al^{3+}, forming non-toxic $AlSO_4^+$. This in a way reduces the toxic Al^{3+} in the soil solution. The presence of oxides of Fe and/or Al in the highly weathered soils would reduce P availability for oil palm requirement via the formation of insoluble Fe-Al-phosphate. But this problem can be reduced by the use of special microbes called phosphate-solubilizing bacteria.

CONCLUSION

Highly weathered soils of Southeast Asia are naturally infertile, but they can be used to grow oil palm sustainably provided that they are properly managed by special soil/agronomic means. To keep the oil palm in the plantation productive, sufficient amounts of NPKMg fertilizers have to be regularly applied. The subsequent chemical reactions taken place in the soils will be able to maintain soil fertility that sustain oil palm production in the long run.

K-6

Regulation of STOP1-mediated aluminum tolerant mechanisms in Arabidopsis roots

[1]Hiroyuki Koyama** and* ***[1]Yuriko Kobayashi

[1]Applied Biological Sciences, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan

; koyama@gifu-u.ac.jp

Several plant species evolved active mechanisms for protecting root tip from aluminum (Al) rhizotoxicity. Previous research in Arabidopsis identified typical active Al tolerant mechanism, which involves Al inducible malate excretion. The inducible excretion of malate is concomitant with Al-inducible expression of *ALMT1* encoding Aluminum-Activated Malate Transporter 1. The gene is inducible by Al with a wide dynamic range of fold-induction (1-100 fold), while it is activated the transcription by very low concentration of Al in the solution (Kobayashi et al., 2013). These characteristics of Al-inducible expression could be enabled by the complex regulatory mechanism by multiple transcription factors. In fact, a previous integrated study of promoter-bioinformatics and *in planta* reporter assay identified involvement of several transcription factors, including a zinc-finger transcription factor (STOP1; SENSITIVE TO PROTON RHIZOTOXICITY 1) and CAMTAs. Combination of these multiple transcription factors could explain wide-dynamic range of Al-inducible *AtALMT1*'s expression (Tokizawa et al., 2015). Recent studies have identified several molecular mechanisms underlying early Al activation mechanisms. In this topic, I will introduce STOP1-mediated Al signaling and activation of transcription of Al inducible genes in Arabidopsis roots, including *AtALMT1*.

1. Early Al inducible gene expression regulated by STOP1.

Aluminum activates various Al-tolerant mechanisms, while the processes are likely regulated by in quick and sensitive manner. Sensitive and quick Al activation was first identified for the Al-activated malate excretion in wheat, which is explained by the direct activation on the pre-existing ALMT1 protein in the PM (plasma membrane) by Al. Later, similar quick Al activation for the gene expression was identified in Arabidopsis, including critical Al tolerance genes *AtALMT1*. Al induces *AtALMT1* expression within 30 min, which is concomitant with accumulation of STOP1-protein in the nuclei. Since the promoter of *AtALMT1* carries binding site of the STOP1, it suggested that STOP1 directly activates transcription through accumulation to the nuclei. This possibility was supported by similar quick activation of the transcription of other genes that carry STOP1-binding sites in their promoters. Interestingly, the STOP1-binding sites are likely consisted of 15bp < and observed in the promoters of these quickly responsive genes to Al. This sequence (i.e. STOP1-binding site in *AtALMT1*) was found in the Al-inducible MATE in pigeon pea, which shows quick response to exogenous Al. Al-inducible expression of *AtMATE* and *ALS3* are STOP1-dependent, but they require longer period (about 2 hours) of Al-inducible expression. These genes would be regulated by other transcription factors whose transcription are regulated by STOP1.

2. Al sensing process of STOP1-regulated genes

AtALMT1 expression is inducible by non-toxic level of Al (*i.e.* submicromolar of $\{Al^{3+}\}_{PM}$, and which is not toxic for the dysfunctional mutant of *AtALMT1*). It indicates that Al-inducible *AtALMT1*-expression is regulated by very sensitive Al-sensing mechanisms, which allows to protect the Al-sensitive root tip sufficiently from Al toxicity. This process may involve accumulation of STOP1 to nuclei since binding of STOP1 to the promoter is critical for transcription of *AtALMT1*. Previous studies suggested that Al-inducible expression of *AtALMT1* involves protein phosphorylation/diphospholylation processes. In fact, several inhibitors (e.g. protein kinase inhibitors) suppress Al-inducible *AtALMT1* expression at early (2 hours) and late (6-12 hours) phases. On the other hand, some of such inhibitors also inhibit Al-activated malate excretion of AtALMT1. It suggests that some of Al-signaling pathways are shared by both Al-inducible transcription and activation of AtALMT1-proteins. On the other hand, STOP1-orthologues often failed to complement Al-tolerance by activation of *AtALMT1* expression during *in planta* complementation assay in Arabidopsis, while they could activate transcription of several low-pH tolerance genes (*e.g.* PGIP1). It suggests that STOP1-protein carry some domains that are essential for activating Al-inducible genes.

REFERENCES

Kobayashi, Y., Kobayashi, Y., Watanabe, T., Shaff J. E., Ohta, H., Kochian, L. V., Wagatsuma, T., Kinraide, T. B. & Koyama (2013). Molecular and physiological analysis of Al^{3+} and H^{+} rhizotoxicities at moderately acidic conditions. Plant Physiol. 163: 180-92

Kusunoki K, Nakano Y, Tanaka K, Sakata Y, Koyama H and Kobayashi Y (2017). Transcriptomic variation among six Arabidopsis thaliana accessions identified several novel genes controlling aluminium tolerance. (2017) Plant Cell Environ. 40: 249-263.

Tokizawa M, Kobayashi Y, Saito T, Kobayashi M, Iuchi S, Nomoto M, Tada Y, Yamamoto Y. Y & Koyama, H (2015). SENSITIVE TO PROTON RHIZOTOXICITY1, CALMODULIN BINDING TRANSCRIPTION ACTIVATOR2, and other transcription factors are involved in *ALUMINUM-ACTIVATED MALATE TRANSPORTER1* expression. Plant Physiol. 167: 991–1003

K-7

Mitigating soil acidity for agricultural sustainability in the humid tropics of Micronesia

[1]Mohammad H. Golabi, [2]Ferdinand Galsim, [2]Clancy Iyekar, and [3]Chieriel S. Desamito
[1]Professor, College of Natural and Applied Sciences, University of Guam, Mangilao, Guam-USA.
[2]Research Associates, College of Natural and Applied Sciences, University of Guam
[3]Graduate student, College of Natural and Applied Sciences, University of Guam
; mgolabi@triton.uog.edu ; m_golabi@hotmail.com

Soil acidity and consequently soil fertility decline are among the major forms of soil degradation that adversely affect sustainable agriculture in Guam and other islands in Micronesia. Some of the causes of soil acidity are inherited from the weathered volcanoclastic parent material, however, use of acid forming fertilizers are also to be blamed for the low pH soils in some farming areas. In the humid tropics however, soil acidity is aggravated by leaching of 'basic' forming 'cations' due to area's high rainfall both in term of frequency as well as intensity. Acidic soils are therefore deficient in essential plant nutrients such as K, Ca, Mg, and Mo, mainly due to excessive leaching of these nutrients.

Furthermore, in the low pH (below 5) soils, aluminum (Al) as well as manganese (Mn) are soluble in water, making them the dominant ions in the soil solution. The excess aluminum in low pH soils injures the root apex thus inhibits the root elongation and its development. The poor root growth therefore, leads to reduced water and nutrient uptake, consequently crops grown on these low pH soils are subject to low yield and overall crop performance.

Among the approaches to reduce soil acidity and raise the pH of these soils is the application of agricultural limes for ameliorating soil acidity. However, the cost and even sometimes the availability of liming material can be a limiting factor depending on the geographical locality of farming communities. Alternatively, the application of organic material such as compost as well as 'biochar' are used to balance the pH and improve the overall quality and the health of these soils. On the other hand, increased soil organic matter sequesters the aluminum ions thereby, avoiding its toxic effect on crops growing on these acidic soils. The use of organic fertilizers such as farm yard manure, compost, green manure and available biomass of leguminous trees/bushes are viable alternatives to inorganic fertilizers for improving soil fertility. Furthermore, organic materials (i.e., compost), in addition to being nutrient sources, they also improve the physical and chemical properties of the soil thus, enhancing the water holding capacity, CEC as well as the biological activities of these soils. The best result however, is obtained when organic and inorganic fertilizers are applied together. Many research findings have proved that combined application of organic and inorganic fertilizer produce superior crop yield as compared to either sources applied alone.

In some areas of the tropics, farmers are shifting towards producing crops which are more tolerant to soil acidity. However, use of acid tolerant varieties have their own merits and limitation causing the practice to be unsustainable and more than often non-profitable. For agricultural sustainability and for maintaining the environmental quality however, the integrated application of composted animal and green manure with the combination of inorganic fertilizers may have a significant impact on soil fertility hence, improved crop productivity.

In our soil program at the University of Guam, we are evaluating the use of organic material as alternative to synthetic fertilizers on highly weathered acidic soils of southern Guam. These soils are formed from the weathered volcanic rocks saturated with aluminum and iron oxides found in the mountainous regions of southern Guam. As a comparison, we are also evaluating the use of organic material as alternative to synthetic fertilizers on the alkaline soils formed from the weathered limestone rocks in northern Guam. Our goal is to develop management strategies for using composted organic material for improving crop production while conserving resources and preserving environmental quality. In our pilot projects compost is being produced from wood chips mixed with animal manure, chicken litter, food and other organic wastes available locally. Matured compost is then applied on the fields (southern as well as in northern Guam) at the rate of; 0, 30, 60 and 90 tons per acre as soil amendment. Corn is planted and monitored for growth performance and yield evaluation in both cases. Additionally, the application of 'biochar' as the 'other' soil amendment, is being evaluated not only for improving soil quality and balancing the soil pH, but also to 'sequester' the carbon in the soil biota thus, reducing the amount of CO_2 emission from the soil surface upon disturbances. Toward these goals, we are evaluating the effect of 'biochar' application not only as soil amendment, but also for improving the soil carbon storage capacity facilitated by 'sequestration' potential of amending 'biochar' on north as well as the southern Guam soils.

Thus, this paper, narrates the key findings as well as the methodology that illustrate the effect of land application of composted organic wastes on organic matter content, soil pH and, other soil indices, focusing on soil quality improvement for agricultural sustainability while maintaining the integrity of the island's environment. In this presentation we will also report the result of the land application of 'biochar' on the dynamics of soil carbon content and the soil storage capacity of highly weathered acidic soils of southern Guam as well as soil formed from limestone parent material in the north.

O-1

Cell wall pectin and its methyl-esterification in transition zone determine al resistance in cultivars of pea (*Pisum sativum*)

***Li Xuewen*[1], *Xiao Hongdong*[1], *Yu Min*[1]**
[1]Department of Horticulture, Foshan University, Foshan, 528000, Guangdong, China
; yumin@fosu.edu.cn

INTRODUCTION

Aluminum (Al) toxicity is a major limiting factor for plant growth and development in acid soils. Cell wall is the first target of Al accumulation. Pectin, a major component of cell wall, is processed by PME after its production and release to apoplast, it is thus hypothesized that cell wall pectin and pectin methylesterases (PME) may be responsible for Al sensitivity in root transition zone and Al resistance in cultivars of pea. The objective was to disclose the significance of pectin content and its degree of methyl esterification in determining Al resistance in different cultivars.

MATERIALS AND METHODS

According to Yang et al. (2008) method, pea (*Pisum sativum* L) seeds were germinated and cultured, and treated with 15 or 30µM $AlCl_3$ solution (25 µM H_3BO_3, 0.5 mM $CaCl_2$, pH 4.5) for 24 h. Root length determined using WinRhizo-Pro software. Selected lateral roots of the seedlings were stained with morin, and roots were observed under fluorescent microscope directly, while free-hand sections were observed under a Laser-Scanning Confocal Microscope. The measurement of Al concentration by ICP-AES. The pectin content and PME activity was measured by the colorimetric method, modified from Yang (2008).

RESULTS AND DISCUSSION

1 Different Al resistance in cultivars of pea

Root elongation was inhibited by Al toxicity in both cv Onward and cv Sima, but relative root elongation in cv Onward was higher than that in cv Sima (Fig. 1A). Meanwhile there was significantly less Al accumulation in cv Onward compared to cv Sima (Fig. 1B).

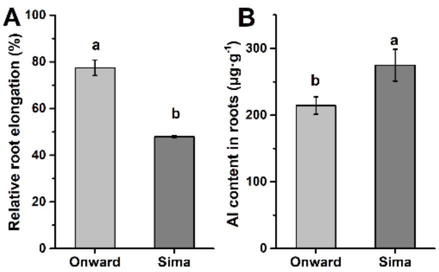

*Fig. 1 Effect of Al application on root elongation (A) and Al content (B) in different cultivars of pea

2. Al content in different root segments.

Content of Al was higher in cv Sima than that in cv Onward at 0-1.0 mm and 1.0-2.5 mm root segment, and there was a significant difference in the cell wall (Fig. 2B).

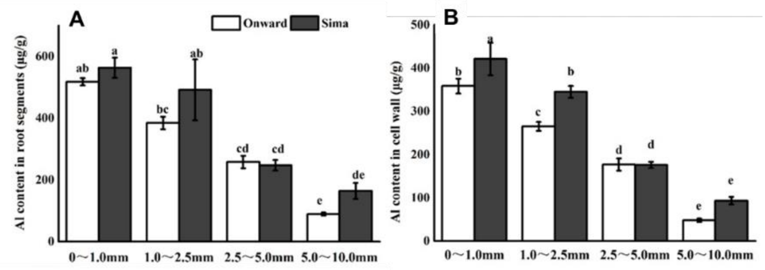

*Fig. 2 Al content in root segments (A) or cell wall (B) in different cultivars of pea

3. Morin staining

Roots showed stronger fluorescence at 0-3 mm root tips (Fig. 3A and Fig. 3D) than the other root segments (Fig. 3B-C and Fig. 3E-F) in both cv Onward and cv Sima. There was brighter fluorescence at 1.0-2.5 mm root, the transition zone, in cv Sima compared with cv Onward (Fig. 3A and Fig. 3D, Fig. 3H and Fig. 3L).

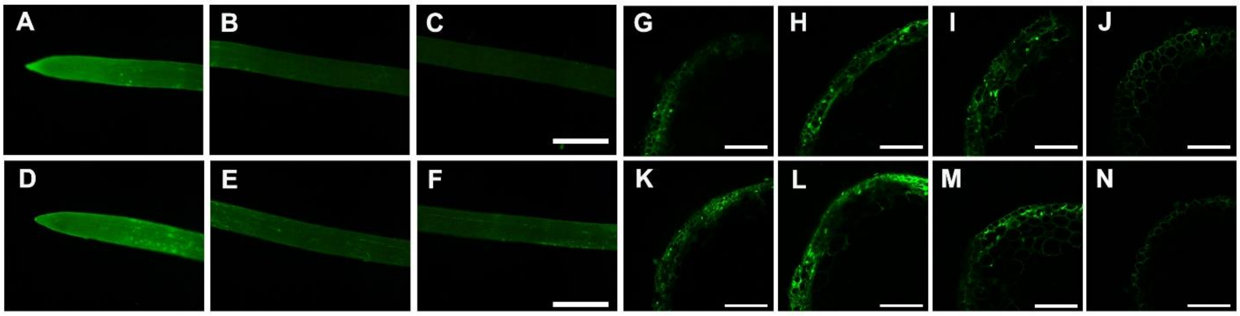

*Fig. 3 The distribution of Al indicated by morin (green fluorescence) stain. Cv. Onward: A, B, C, G, H, I, J; Cv. Sima: D, E, F, K, L, M, N. Meristem (G, K), transition zone (H, L), elongation zone (I, M) and mature zone (J, N).

4. Effect of Al treatment on pectin

Under Al toxicity, the content of pectin in the transition zone is significantly higher in cv Sima (Fig. 4A). and its degree of pectin methyl-esterification was lower, compared with cv Onward (Fig. 4B), leading to a higher un-esterified pectin content (Fig. 4C). This might be caused by the difference of PME activity, which was 2.2 folds in cv Sima than in cv Onward.

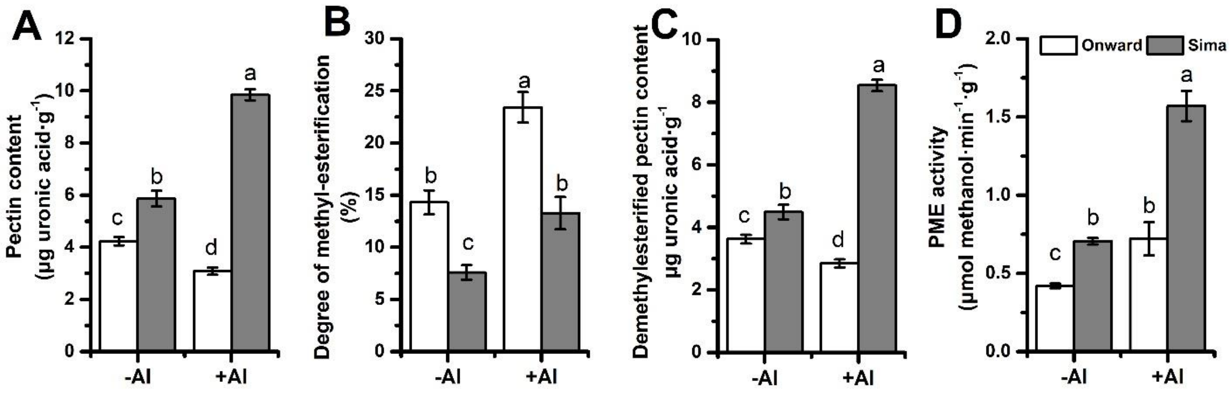

*Fig. 4 Cell wall pectin properties. A. Cell wall pectin content. B. the degree of pectin methyl-esterification. C. The demethylesterified pectin content. D. PME activity.

CONCLUSION

In the transition zone of Al-sensitive cultivar, which is the most sensitive zone to Al, the most prominent to Al-induced pectin increased, and the higher PME activity resulted in higher content of demethylesterified pectin and higher Al accumulation in cell wall and cytosol. Therefore, the transition zone contributes, at least in part, to the differential Al resistance among cultivars.

ACKNOWLEDGEMENTS

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REFERENCES

Yang, J., Li, Y., Zhang, Y., and Zhang, S. (2008). Cell wall polysaccharides are specifically involved in the exclusion of aluminum from the rice root apex. Plant Physiol. **146,** 602-611

O-2

Growth promoting characteristics of salt-tolerant rhizobacteria isolated from paddy field in northern coastal saline areas of Malaysia

***Rakiba Shultana*[1], *Radziah Othman*[1], *Ali Tan Kee Zuan*[1] *and Mohd Rafii Yusop*[2]**
[1]Department of Land Management, Faculty of Agriculture,
[2]Institute of Tropical Agriculture and Food Security, Universiti Putra Malaysia, 43400 UPM Serdang, Malaysia
; rakiba_83@yahoo.com

INTRODUCTION

Bacterial inoculation such as plant growth-promoting rhizobacteria (PGPR) is an effective and eco-friendly method to improve plant stress tolerance. Several reports have shown that PGPR effectively improve growth of a wide range of agricultural crops under environmental stress conditions (Timmusk et al., 2015; Zahid et al., 2015). In addition, the ability of PGPR to serve as bio-fertilizer or phyto-stimulator helps in maintaining the soil fertility, thereby providing a promising alternative to chemical fertilizers and pesticides for sustainable agriculture (Majeed et al., 2015). Considering this beneficial interactions, a study was undertaken to isolate, characterize and identify the salt-tolerant PGPR isolated from Malaysian coastal saline rice ecosystem.

MATERIALS AND METHODS

Soil samples were collected from salt-affected rice field at seven locations at Kuala Muda, Kedah, Malaysia. Isolation of rhizospheric, non-rhizospheric and endophytic bacteria were undertaken using the method as described by Somasegaran and Hoben (1985) and Tan *et al.*, (2014). These bacteria were screened for salinity tolerance upto 2M of NaCl and plant growth-promoting traits namely biological nitrogen fixation, phosphate and potassium solubilizations, and phytohormone production. Bacterial EPS production was measured following the method of Hong *et al.* (2017). Bacterial flocculation expressed as floc yield was carried out following the protocols of Sadasivan and Neyra (1985). Sodium uptake by the bacteria from the salt amended media were measured following the method described by Damodaran *et al*. (2013) with some modifications. The data were analyzed statistically using Analysis of Variance (ANOVA) by SAS 9.4. Means were compared by Tukey (HSD) test at a probability level of 0.05.

RESULTS AND DISCUSSION

A total of 43 bacterial strains were isolated from the salt-affected rice field. Biochemical tests showed that among the strains, five potential strains can tolerate higher amount of NaCl (2M) through exhibiting salt tolerant mechanisms like the production of exopolysaccharide and flocculation which helps them to uptake sodium ion from the media. Significantly highest amount of exopolysaccharide production was recorded by UPMRB9, UPMRE6 and UPMRG1. The highest amount of flocculation which is expressed as floc yield was produced by the isolates UPMRA4, UPMRB9, UPMRE6 and UPMRG1. However, the isolates UPMRG1 exhibited the highest intake of sodium (17.73 mg L^{-1}) from the media. These bacterial strains were further tested for the production of plant growth promoting traits. The highest amount of phosphate was solubilized by the isolate UPMRE3 followed by UPMRE6 and UPMRG1 through the production of organic acid and as a result media pH was reduced. The isolates UPMRE6 solubilized the highest amount of potassium. The highest amount of indole acetic acid (IAA) production was recorded by the isolate UPMRB9 followed by UPMRA4 and

UPMRG1. All isolates showed positive indication for biological nitrogen fixation in N-free malate medium except for UPMRA4 strain (Table 1).

Table1: Salt tolerant and plant growth promoting traits by the selected bacterial strains

Bacterial strains	NaCl (2M)			AvailableP (µg mL^{-1})	Available K (mg L^{-1})	IAA (µg mL^{-1})	Nitrogen fixation
	EPS production (g L^{-1})	Floc yield (g L^{-1})	Na uptake (mg L^{-1})				
UPMRA4	**4.26bC**	**16.87a**	**7.14bc**	22.11c	5.72de	12.48a	-
UPMRB9	**19.21a**	**19.92a**	**12.30b**	35.53b	6.90cd	16.33a	+
UPMRE3	**7.10b**	**9.69b**	**4.91c**	41.50a	13.82b	7.13b	++
UPMRE6	**20.51a**	**19.67a**	**6.48c**	40.49a	15.79a	7.0b	+
UPMRG1	**21.73a**	**20.13a**	**17.73a**	40.14a	7.76c	13.06a	+

Means having same letter(s) in a column do not differ significantly at ≥ 0.05 level by Tukey test

CONCLUSION

Several promising bacterial strains were isolated from salt-affected rice field and were observed to have several plant growth-promotional traits and abilities to tolerate soil salinity. Five potential salt-tolerant PGPR isolates were selected for subsequent test due to having the positive ability to fix atmospheric nitrogen, phosphate and potassium solubilization and phytohormone production. These strains were able to survive saline conditions due to the production of exopolysaccharide and flocculation which help them to bind and take up sodium ion from the saline media. The study revealed a possible new and beneficial biofertilizer sources to promote growth and yield of rice in salt affected areas in a sustainable and environmental-friendly approach.

ACKNOWLEDGEMENTS

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REFERENCE

Hong B.H., Joe M.M, Selvakumar G., Kim K.Y, Choi J.H. and Sa T.M. 2017.Influence of salinity variations on exocellular polysaccharide production, biofilm formation and flocculation in halotolerant bacteria.Journal of Environmental Biology. **38**: 657-664.

Majeed A., Abbasi M. K., Hameed S., Imran A., Rahim N. 2015. Isolation and characterization of plant growth-promoting rhizobacteria from wheat rhizosphere and their effect on plant growth promotion. Front. Microbiol. **6**:198.

Sadasivan L. and Neyra C.A. 1985. Flocculation in Azospirillumbrasilense and Azospirillumlipoferum: Exopolysaccharides and cyst formation. J. Bacteriol. **163**:716-723.

Somasegaran P. and Hoben H.J. 1985. Methods in legume-rhizobium technology. University of Hawaii NifTAL Project and MIRCEN

Tan K.Z., Radziah O., Halimi M.S., Khairuddin A.R., Habib S.H. and Shamsuddin Z.H. 2014. Isolation and characterization of rhizobia and plant growth-promoting rhizobacteria. **9**(3), 342–360

Timmusk S., Kim S.B., Nevo E., Abd El Daim I., Ek B., Bergquist J. 2015. Sfp-type PPTase inactivation promotes bacterial biofilm formation and ability to enhance wheat drought tolerance. Front. Microbiol. **6**:387.

Zahid M., Abbasi M. K., Hameed S. and Rahim N. 2015. Isolation and identification of indigenous plant growth promoting rhizobacteria from Himalayan region of Kashmir and their effect on improving growth and nutrient contents of maize (Zea mays L.). Front. Microbiol. **6**:207

O-3

Effect of organic amendments and microbial inoculant on the nutrient balance and productivity of sugarcane grown in an acid Typic Hapludand

***Ryan T. Sarimong*[1], *Pearl B. Sanchez*[2], *Erlinda S. Paterno*[2], *Rodrigo B. Badayos*[2], *and Pompe C. Sta. Cruz*[2]**
[1]Capiz State University, Burias, Mambusao, Capiz, Philippines
[2]University of the Philippines Los Baños, Laguna, Philippines,
; ryansoiler4@yahoo.com

INTRODUCTION

The sugar industry is one of the key players in the Philippine economy as indicated by its contribution to national output, employment and export earnings. In order to maximize the productivity of sugarcane farms, fertilizer has always been an inevitable input which supply additional nutrients to the soil in order to meet the crop nutritional needs. As pointed by Padilla-Fernandez and Nuthall (2009), better use of available inputs by rationalizing the use of NPK, especially N fertilizer could increase output of sugarcane farmers. Under sugarcane monoculture, Meyer et al. (1996) considered loss of soil organic matter as a major contributor to soil degradation. As also indicated in NAP (2004), intensive cultivation of upland areas in the Philippines without addition of nutrients and organic matter to the soil is considered to be a contributor to the widespread occurrence of acid upland soils. These processes occurring naturally or human-induced operate singly or in combination diminish the quality of the soil resource and thus lower the current or future capacity of the soil to produce goods or services. Hence, this study was undertaken to optimize the use of organic amendments such as mudpress and bagasse ash, microbial inoculant and inorganic fertilizer for sugarcane production in an acid Typic Hapludand.

MATERIALS AND METHODS

A field experiment in Randomized Complete Block Design was established in an acid Guimbalaon sandy clay loam classified as Typic Hapludand (Carating et al., 2014) in Isabela, Negros Occidental (10^0 10.101' N, 122^0 59.223' E). Twelve treatments were employed including no fertilization, with full fertilization using inorganic fertilizer, and with full fertilization + lime. The recommended N rate (RR_N) was reduced to 75, 50 and 25% with subsequent application of mudpress to satisfy the full N recommendation. Bagasse ash and microbial inoculant were likewise used to supplement the nutrient sources. 'Phil 2004-1011' sugarcane variety was planted in 20m x 8m plots. The cultural and management procedures for sugarcane production specified in the Sugarcane Production Manual of SRA-OPSI (2004) were followed. Cane and sugar yield, return on investment and NPK balance were determined and analyzed to come up with a sustainable fertilization program.

RESULTS AND DISCUSSION

Table 1 shows that reducing the recommended N rate (RR_N) with subsequent application of mudpress (MP), bagasse ash (BA) and microbial inoculant (MI) produced the highest cane yield. Likewise, reducing RR_N up to 50% with the application of mudpress, with or without bagasse ash and microbial inoculant improved sugar yield comparable to lime application. Better return on investment was obtained from the application of 50% RR_N from inorganic fertilizer and 50% RR_N from mudpress. Combined application of inorganic fertilizer, mudpress, bagasse ash and microbial inoculant also resulted to a net positive actual NPK balance (Table 2).

Table 1. Cane yield, sugar yield and return on investment of sugarcane grown in an acid Typic Hapludand.

Treatments	Cane Yield (TC ha^{-1}) **	Sugar Yield (LKg ha^{-1})**	Return on Investment (%)
T1 - Control (no fertilizer application)	69.73e	126.41d	60.75
T2 - RR_N IF	93.27d	185.58c	52.25
T3 - RR_N IF + lime	99.39cd	208.10bc	33.68
T4 - 75% RR_N IF: 25% RR_N MP	105.08bcd	220.81ab	71.11
T5 - 75% RR_N IF: 25% RR_N MP + BA	105.39bcd	218.74ab	64.79
T6 - 75% RR_N IF: 25% RR_N MP + BA + MI	114.39ab	235.34ab	48.71
T7 - 50% RR_N IF: 50% RR_N MP	107.76abc	226.74ab	72.55
T8 - 50% RR_N IF: 50% RR_N MP + BA	110.04abc	232.25ab	70.59
T9 - 50% RR_N IF: 50% RR_N MP + BA + MI	117.09ab	240.13a	49.44
T10 - 25% RR_N IF: 75% RR_N MP	110.95abc	216.89ab	63.17
T11 - 25% RR_N IF: 75% RR_N MP + BA	116.85ab	222.57ab	60.68
T12 - 25% RR_N IF: 75% RR_N MP + BA + MI	120.24a	235.83ab	45.10

***=highly significant; CV (cane yield) = 6.61; CV (sugar yield) = 6.79; means having same letter are not significantly different at the 5% level by DMRT.*

Table 2. NPK balance of sugarcane production grown in acid Typic Hapludand.

Treatments	Nutrient balance (kg ha^{-1})		
	N	P	K
T1 - Control (no fertilizer application)	-158.28	-0.36	-13.34
T2 - RR_N IF	815.91	1.45	86.31
T3 - RR_N IF + lime	673.80	6.22	107.61
T4 - 75% RR_N IF: 25% RR_N MP	828.15	5.54	98.81
T5 - 75% RR_N IF: 25% RR_N MP + BA	830.13	3.50	86.84
T6 - 75% RR_N IF: 25% RR_N MP + BA + MI	992.64	7.85	76.49
T7 - 50% RR_N IF: 50% RR_N MP	624.28	7.48	116.73
T8 - 50% RR_N IF: 50% RR_N MP + BA	545.95	5.41	67.07
T9 - 50% RR_N IF: 50% RR_N MP + BA + MI	944.93	7.47	90.94
T10 - 25% RR_N IF: 75% RR_N MP	578.28	2.12	65.55
T11 - 25% RR_N IF: 75% RR_N MP + BA	472.20	3.83	77.21
T12 - 25% RR_N IF: 75% RR_N MP + BA + MI	837.49	3.56	65.94

CONCLUSION

In conclusion, the combined use of inorganic, organic and biofertilizers can improve cane and sugar yield and enhance soil nutrient balance. Integrated use of inorganic and organic fertilizers can increase economic efficiency.

REFERENCES

Carating, R. B., Galanta, R. G., and C. D. Bacatio. 2014. The Soils of the Philippines. World Soils Book Series, Springer Science. 346 pp.

Meyer, J. H., Van Antwerpen, R. and E. Meyer. 1996. A review of soil degradation and management research under intensive sugarcane cropping. Proc S Afr Sug Technol Ass **70:** 22-28.

NAP. 2004. The Philippine National Action Plan to combat desertification, land degradation, drought, and poverty for 2004-2010. Retrieved from www.unccd.int. on January 25, 2016.

Padilla-Fernandez, M. D. and P. L. Nuthall. 2009. Technical efficiency in the production of sugarcane in Central Negros area, Philippines: an application of data envelopment analysis. J. ISSAAS Vol. 15 No**. 1:** 77-90.

SRA-OPSI. 2004. Sugarcane Production Manual. Sugar Regulatory Administration, La Granja Agricultural Research and Extension Center, La Carlota City, Negros Occidental. 205 pp.

O-4

Interaction between oppositely charged soil particles and its effect on soil natural acidification in variable charge soils

***Li J.Y.*, *R.K. Xu*[1], *and K.Y. Deng*[1]**
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, 21008, Nanjing, China
; jyli@issas.ac.cn

INTRODUCTION

Variable charge soils are important resources in tropical and subtropical regions. They are acid and contain a plenty of Fe and Al oxides due to intensively development under highly weathering and leaching. Soil acidity properties are closely related with their electrochemical properties. Iron and Al oxides can decrease the effective negative charge (represented by effective CEC, ECEC) of soils by overlapping of electric double layer between oppositely charged colloidal particles and coating on the silicates (Li et al., 2009; Wang et al., 2011). Thus we hypothesize that Fe and Al oxides may decrease the effective negative charge of soils and thus inhibit soil natural acidification. The objectives of this study was to investigate: (i) the effect of different species of Fe and Al oxides on soil charge and acidity properties during the natural acidification process; and (ii) the mechanisms of Fe and Al oxides for inhibiting soil natural acidification.

MATERIALS AND METHODS

First, more than sixty variable charge soils were collected little manual disturbance but with different location, parent materials, depth in South China. The relationships between soil acidity properties, content of free Fe and Al oxides, CEC, ECEC were investigated (Li et al., 2012). Then, different free Fe and Al oxides, kaolinite, Alfisol, and Fe and Al oxide coated kaolinite or Alfisol were prepared. Moreover, electro-dialysis was used to stimulate soil natural acidification. Kaolinite (or Alfisol) mixed or coated with different species and amount of free Fe and Al oxides were subjected to electro-dialysis. The charge and acidity properties, such as the content of positive and negative charge, pH, CEC, ECEC, exchangeable acidity of all samples before and after electro-dialysis were measured (Li and Xu, 2013a and 2013b; Li et al., 2014). After the adsorption of natural organic matter (NOM), these Fe and Al oxides were also used to test the effect of NOM on the interaction between oppositely charge particles and its effect on soil acidification.

RESULTS AND DISCUSSION

Electro-dialysis is a very efficient method to simulate soil acidification naturally in variable charge soils. The higher the content of free iron oxides, the lower the soil acidity of the natural variable charge soils. When soils were electro-dialyzed to induce them reached their ultimate acidification state, soil CEC and free Fe_2O_3 content were two important factors in determining the soil acidity. Soil pH was negatively correlated with CEC and positively correlated with free Fe_2O_3. These results suggested that soil with higher content of free Fe_2O_3 displayed weaker acidity in the ultimate acidification state, which was consistent with the results observed in the untreated samples of the variable charge soils. Further study indicated when Fe and Al oxides just mixed with kaolinite or Alfisol, Fe oxides could inhibit the acidification of kaolinite and Alfisol during electro-dialysis through the overlapping of diffuse double layers between negatively charged kaolinite or Alfisol and positively charged Fe oxides. While aluminum oxides decreased the effectively negative charge and acidity of electro-dialyzed kaolinite and Alfisol through the overlapping of electric double layer and coating on the silicates. Coating of iron or aluminum oxides showed the same mechanism as simply mixed aluminum oxides to decrease the effectively negative charge and acidity of electro-dialyzed silicates and soils. The inhibiting effect intensified with the increasing content and quantity of positive charge of Fe and Al oxides. The adsorption of NOM decreased the quantity of positive charge of Fe and Al oxides and thus reduced their effect on soil natural acidification.

CONCLUSION

Iron or aluminum oxides can inhibit the natural acidification of variable charge soils through the overlapping of electric double layer between oppositely charged colloidal particles and coating on the silicates. The effect of overlapping of electric double layer intensify with the amount and their content of positive charge of Fe and Al oxides. The adsorption of natural organic matter decreases the positive charge of Fe and Al oxides and thus reduces their inhibiting effect on natural acidification in variable charge soils.

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REFERENCES

Li, J.Y., R.K. Xu, and K.Y. Deng. 2014. Coatings of Fe/Al hydroxides inhibited acidification of kaolinite and an alfisol subsoil through electrical double-layer interaction and physical blocking. Soil Science **179:** 495-502

Li, J.Y., and R.K. Xu. 2013. Inhibition of acidification of kaolinite and an Alfisol by iron oxides through electrical double-layer interaction. Soil Science **178(1):** 37-45

Li, J.Y., and R.K. Xu. 2013 Inhibition of acidification of kaolinite and an Alfisol by aluminum oxides through electrical double-layer interaction and coating. European Journal of Soil Science **64:** 110-120

Li, J.Y., R.K. Xu, and H. Zhang. 2012. Iron oxides serve as natural anti-acidification agents in highly weathered soils. Journal of Soils and Sediments **12:** 876-887

Wang Y.P., R.K. Xu, and J.Y. Li. 2011. Effect of Fe/Al oxides on desorption of Cd^{2+} from soils and minerals as related to diffuse layer overlapping. Soil Research **49**: 231–237

Li, S.Z., R.K. Xu, and J.Y. Li. 2009. Interaction of electrical double layers between oppositely charge particles in variable-charge soils as related to salt adsorption. Soil Science **174:** 27-34

O-5

Physico-chemical characteristics, suitability assessment, and constraints analysis of major soil series grown to sugarcane in Negros occidental, Philippines

***Clea Anne V. Corsiga*[1], *Rodrigo B. Badayos*[2], *Pearl B. Sanchez*[3], *Erlinda S. Paterno*[3], *and Pompe C. Sta. Cruz*[3]**
[1]Visayas State University-Villaba, Cagnocot, Villaba, Philippines
[2]Philippine Sugar Research Institute, Makati City, Philippines
[3]University of the Philippines Los Baños, College, Los Baños, Laguna, Philippines
; vallejera_19@yahoo.com;

INTRODUCTION

Negros Occidental, known as the "Sugarbowl of the Philippines", were the lifeblood of the economy is on sugar industry (Discover World Ventures, Inc., 2018) and as the major sugar producer, contributes to more than half of the country's total sugar production.

The efficient and sustainable use of land involves matching site conditions with the specific requirements and potential impacts of different land uses. Lands that are not physically capable of supporting specific landuse might incur significant costs to the environment and society. Suitability analysis is of great help in assessing and determining the appropriateness of a given area for a particular use. The fundamental premise of suitability analysis is that each aspect of the landscape has intrinsic characteristics that are in some degree either suitable or unsuitable for the activities being planned. It is used extensively to find and maintain better cropland as well as ensure proper crop rotation. Hence, in the process, it helps to ensure that land resources are used in the most productive and sustainable ways because different crops require different land types and growing conditions. Consequently, the productive potential of soil is limited by its inherent constraints. Identifying and managing these constraints is fundamental to sustainable production systems. Thus, this study aimed to determine the physical and chemical characteristics of major soil series grown to sugarcane; and assess its suitability and identify possible constraints of the soils for sugarcane production.

MATERIALS AND METHODS

Survey, Assessment, and Selection of the Study Sites

A comprehensive assessment was conducted on sugarcane areas in the province. Topographic, geologic, and soil maps as well as other publications, were used as materials in identifying the sampling sites. Out of the nine (9) soil series planted to sugarcane in Negros Occidental, five (5) soil series (Guimbalaon, Luisiana, Isabela, San Manuel, Silay), were comprehensively assessed and utilized in this study since these are the most widespread soil series found in the province.

Soil Collection and Laboratory Analysis

Twenty soil samples were collected from representative sites at depths of 0-30 and 30 – 60cm and were processed and analyzed in the laboratory for soil pH, total N, available P, exchangeable bases, percent organic carbon, extractable Fe, exchangeable Al, particle size distribution, and cation exchange capacity following standard procedures.

Suitability Evaluation and Constraint Analyses

Soil and climatic characteristics of the five soil series (Guimbalaon, Isabela, Luisiana, San Manuel, Silay) were matched with the criteria set by Sys, Ranst, Debaveye and Beernaert (1993) for sugarcane requirements to determine their suitability classes either as: S1 (highly suitable); S2 (moderately suitable); S3 (marginally suitable); N1 (marginally not suitable); and N2 (permanently not suitable). Nevertheless, subclasses reflect the kinds of limitation due to topography (t), soil wetness (w), soil physical properties (s), soil fertility (f), and climate (c). Suitability and Constraints Maps were made after that.

RESULTS AND DISCUSSION

Soil Characteristics of Major Soil Series Grown to Sugarcane

Isabela series obtained the highest value on pH, total N, percent organic carbon, and available P among other soil series. Exchangeable K was found to be highest in Guimbalaon series while exchangeable Na, Ca, Mg as well as extractable Fe and cation exchange capacity were highest in San Manuel series. Exchangeable Al was highest in Luisiana series as compared to other soil series. However, Luisiana soil series was also found to be lowest in most of the chemical properties evaluated in the soil (pH in both in H_2O and $CaCl_2$, percent total N, exchangeable K, Na, Ca, and Mg, CEC, and extractable Fe). Silay series also obtained the lowest values in percent organic carbon and available P while exchangeable Al as the lowest in Isabela series.

Suitability Evaluation of Major Soil Series Grown to Sugarcane

Guimbalaon, Isabela, Luisiana, San Manuel, and Silay were classified as marginally suitable (S3) however; a limitation for sugarcane production varies in each soil series.

Crop Constraints Analysis of Major Soil Series Grown to Sugarcane

Topography and wetness became the severe constraints for most of the soils. Isabela, San Manuel, and Silay series have problems with wetness due to its seasonal flooding occurrences while Guimbalaon and Luisiana series have a constraint on topography because of its rolling to hilly to a mountainous topographic position. Climate (relative humidity) was observed to be the common factor for all soil series which moderately limit production of sugarcane since it exceeded the maximum requirement of humidity needed by the crop. Other factors noted were physical soil characteristics and fertility, however, the limitation is moderate and manageable.

CONCLUSIONS

Isabela soil series have the highest value on pH, total N, percent organic C, and available P; exchangeable K in Guimbalaon series; exchangeable Na, Ca, and Mg, extractable Fe, and CEC in San Manuel series; and exchangeable Al in Luisiana series. Luisiana soil series found to be low in most of the chemical properties evaluated in the soil (pH in both in H2O and CaCl2, percent total N, exchangeable K, Na, Ca, and Mg, CEC, and extractable Fe while Silay series were low in percent organic carbon, available P and Isabela series for exchangeable Al. Guimbalaon, Isabela, Luisiana, San Manuel, and Silay soil series were classified as marginally suitable (S3) for sugarcane production although, soil constraints varied across soil series. Topography and wetness became the severe constraints for most of the soils however, limitation for fertility and physical soil characteristics were considered moderate and manageable. Isabela and Silay series have problems on drainage but the former have constraints on soil texture while the latter on low organic carbon which is manageable than the other.

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REFERENCES

Discover World Ventures, Inc. (2018). Retrieved from https://www.discoverworld.com/Philippines/Negros-Occidental:In-depth

Sys, C., Van Ranst, E., Debaveye, J., and Beernaert, F. Land Evaluation. Part III: crop requirements. Agricultural Publications n° 7, GADC, Brussels, Belgium, **1993,** 191 p.

O-6

Mechanisms for increasing soil pH buffering capacity by application of organic amendments

***Renkou Xu*[1], *Shi, R. Y.*[1] *and Pan, X. Y.*[1]**
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Science, Nanjing 210008, China
; rkxu@issas.ac.cn

INTRODUCTION

Soils in tropical and subtropical regions normally have low pH buffering capacity (pHBC) due to high content of kaolinite and low content of organic matter (Xu et al. 2012), and thus soil acidification is easy to occur in these regions. Increase in organic matter content and application of organic amendments can increase soil pHBC and thus should slow down soil acidification. However, the mechanisms for increasing soil pHBC by application of organic amendments were not well understood. The objectives of this study were: (1) to investigate the effects of crop straw biochars and humic acid on soil pHBC; (2) to elucidate the mechanisms for increasing soil pHBC by application of organic amendments; (3) to confirm the increase in resistance of soils to acidification due to the increase in pHBC by organic amendments.

MATERIALS AND METHODS

Two Ultisols used in this study were collected from Yingtan, Jiangxi Province (28°14' N, 116°55' E; 28°12' N, 116°56' E). All soil samples were taken from top layer (0-15 cm), air-dried and then ground to pass through a 2 mm sieve for incubation experiments and a 0.25 mm sieve for measurement of pHBC and soil basic properties. Ultisol-1 and Ultisol-2 were derived from Tertiary red sandstone and Quaternary red earth, respectively. Soil pH was 4.96 and 4.78; soil organic matter content was 8.1 and 14.9 g/kg; soil CEC was 6.34 and 12.4 cmol/kg for both Ultisols, respectively. Two biochars prepared from straws of corn and peanut at 400°C were used and their characteristics were shown in Table 1. A humic acid extracted from coal was obtained commercially.

Table 1 Basic properties of biochars derived from straws of corn and peanut at 400°C

Biochar	pH	Alkalinity	CEC	Functional group
		---------------------------cmol/kg---		
Corn straw	9.19	148.1	132.5	155.0
Peanut straw	10.28	321.3	146.8	194.4

Soil samples were mixed with biochars, humic acid and $Ca(OH)_2$, wetted with deionized water to 70% of the field water holding capacity of the soils and then incubated for 30 days. Soil pHBC was determined by acid-base titration technique (Xu et al. 2012). Simulated acidification experiment was conducted by the addition of various amounts of HNO_3 to examine the effect of biochars and humic acid on soil resistance to acidification (Shi et al. 2017). Soil with $Ca(OH)_2$ was set as control.

RESULTS AND DISCUSSION

Incorporation of two biochars at 3% level increased not only soil pH but also soil pHBC for both Ultisols. Soil pH of the Ultisol derived from Tertiary red sandstone was increased from 4.96 for control to 6.23 and 7.83 for the treatments with corn and peanut straw biochars. Similarly, soil pH was increased from 4.78 for control to 5.38 and 6.32 for the treatments with both biochars for Ultisol derived from Quaternary red earth. Peanut straw biochar induced more increase in soil pH due to its greater alkalinity compared with corn straw biochar. Soil pHBC was increased from 12 mmol/kg·pH for control to 15.3 and 21.3 mmol/kg·pH for the treatments with both biochars for the former Ultisol. Soil pHBC was increased from 26.4 mmol/kg·pH for control to 32.1 and 33.2 mmol/kg·pH for the treatments with both biochars for the latter Ultisol. Peanut straw biochar also induced more increase in soil pHBC for both Ultisol than corn straw biochar. This may be due to the greater CEC and more functional groups on the biochar than these of corn straw biochar. Incorporation of humic acid also increased soil pHBC. Soil pHBC of the Ultisol derived from Tertiary red sandstone was increased from 9.0 mmol/kg·pH for control to 12.2, 14.6 and 18.8 mmol/kg·pH at addition levels of 1%, 2% and 5%, respectively.

Results of simulated acidification experiment indicated that incorporation of biochars and humic acid inhibited soil acidification and thus increased soil resistance to acidification. Peanut straw biochar showed greater inhibition on soil acidification than corn straw biochar, which was consistent with the increase in soil pHBC induced by both biochars.

The weak acid groups on the biochars/humic acid and organic anions from the dissociation of these weak acid groups constructed a strong pH buffering system, which was mainly responsible for the increase in pHBC and resistance to acidification of these soils by the biochars and humic acid. This mechanism was supported by the decrease in soil effective cation exchange capacity with decreasing soil pH (Fig. 1). The anions of weak acid groups combined with H^+ to form neutral molecular, which led to the decrease in negative charges on soil surface. ATR-FTIR analyses indicated that the peak intensity at 1700 cm^{-1} for –COOH on the biochars increased with decreasing pH, while the opposite trend was observed for peak intensity at 1375 cm^{-1} for $–COO^-$, which confirmed the contribution of protonation of organic anions on the biochars to the increase in soil pHBC.

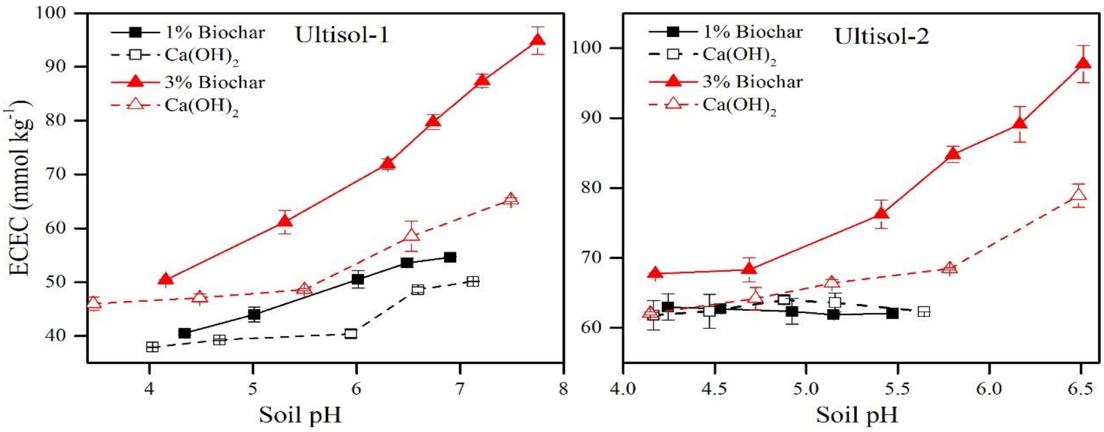

Fig. 1 The changing trends of effective cation exchange capacity (ECEC) in the Ultisols with peanut straw biochar and $Ca(OH)_2$ during simulated acidification.

CONCLUSIONS

Incorporation of crop straw biochars and humic acid increased soil pHBC and thus soil resistance to acidification. The main mechanism for increasing pHBC by these organic amendments was: the anions of weak acid groups on the amendments combined with H^+ to form neutral molecular.

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REFERENCES

Shi, R. Y., Z. N. Hong, J. Y. Li, J. Jiang, M. A. Baquy, R. K. Xu, W. Qian. 2017. Mechanisms for increasing the pH buffering capacity of an acidic Ultisol by crop straw derived biochars. Journal of Agricultural and Food Chemistry **65:** 8111-8119.

Xu, R. K., A. Z. Zhao, J. H. Yuan, J. Jiang. 2012. pH buffering capacity of acid soils from tropical and subtropical regions of China as influenced by incorporation of crop straw biochars. Journal of Soils and Sediments **12:** 494-502.

O-7

Response of soil clay minerals to soil acidification: field study of long-term fertilization during 1990 to 2013

***Tao, L.1,* Li, F.B.[1], Feng, X.H.[2], Gu, L.L.[2], Wang, B.R.[3], Wen, S.L[3]. and Xu, M.G.[3]**

[1]Guangdong Key Laboratory of Agricultural Environment Pollution Integrated Control, Guangdong Institute of Eco-Environmental Science & Technology, Guangzhou 510650, P.R. China

[2]Key Laboratory of Arable Land Conservation (Middle and Lower Reaches of Yangtze River), Ministry of Agriculture, College of Resources and Environment, Huazhong Agricultural University, Wuhan 430070, P.R. China

[3] National Engineering Laboratory for Improving Quality of Arable Land, Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing 100081, P.R. China

; taoliang@soil.gd.cn

INTRODUCTION

Soil acidification is commonly associated with plant nutrients deficiency, Al toxicity, and reduced biodiversity and productivity (Blake et al., 1994). Soil acidification, which carries the characteristics of (a) increasing the soil acidity, or decreasing the pH, (b) decreasing the base saturation, (c) unbalancing the availability of elements in the root environment, and (d) decreasing the acid-neutralizing capacity of the soil, is a natural process and it can be accelerated by human activities (Bolan et al., 2003; Breemen, 1991). Numerous studies have focused on the impact of soil acidification on agricultural production, soil organisms and Al toxicity. Nevertheless, investigations of the influences of soil acidification on soil clay minerals are rare. In this study, soil samples from five long-term fertilization areas were collected from the Qiyang Red Soil Experimental Station in Hunan Province in subtropical China. The clay minerals of these samples were qualitatively and quantitatively analyzed. The objective of this study was to investigate the influence of soil acidification caused by long-term fertilization on the phase transformation of clay minerals.

MATERIALS AND METHODS

The research site is located at the Qiyang Red Soil Experimental Station of Chinese Academy of Agricultural Science in Hunan Province (26°45'12"N, 111°52'32"E, 120 m altitude) (Cai et al., 2014). Five fertilization treatments including: (1) no fertilizer (Blank); (2) chemical N fertilizer only (N); (3) chemical N, P, and K fertilizers (NPK); (4) chemical N, P, and K fertilizers plus manure (NPKM); and (5) manure only (M) were applied from November 1990 to 2013. Soil samples were collected from the topsoil (0 to 20 cm) and the subsoil (20 to 40 cm) in these five fertilization areas after the crops were harvested using a soil auger (5 cm in diameter).

Soil colloids were collected using natural sedimentation and the centrifugation method with modification. After that, Mg-glycerin saturated oriented sheets and K saturated oriented sheet were prepared. The clay minerals were identified using X-ray diffraction (XRD). After that, qualitative and quantitative analyses of the intergradient minerals were conducted.

RESULTS AND DISCUSSION

After 23 years of fertilization, the soil pH decreased from 5.70 in 1990 to an of average 5.04 ± 1.09 in 2013 (n=5). In detail, from 1990 to 2013, the average acidification rate (ΔpH) was approximately -0.03E-1 y^{-1} (blank), -0.83E-1 y^{-1} (N), -0.73E-1 y^{-1} (NPK), -0.24E-1 y^{-1} (NPKM), and +0.27E-1 y^{-1} (M), respectively.

Compared with these above treatments, long-term fertilization using chemical fertilizer containing N accelerates soil acidification, whereas manure fertilizer could mitigate soil acidification.

Qualitative and quantitative analysis of clay minerals

The clay mineral after cation saturation by magnesium and potassium, glycerol solvation and heat treatments (300 °C and 550 °C) were identified by XRD with Cu Kα radiation. The obtained results indicate the existence of kaolinite, hydromica, vermiculite and 1.4 nM intergradient minerals as well as the exclusion of montmorillonite and chlorite in the clay minerals.

Besides, semi-quantitative analysis of clay minerals was conducted to investigate the impact of long-term fertilizations on soil clay minerals, and the results are shown in Figure 1. In the topsoil (0~20 cm) and in the subsoil (20~40 cm), the proportion of kaolinite (0.7 nM), hydromica (1.0 nM), and 1.4 nM intergradient minerals were approximately 50%~60%, 20%~45%, and 3%~14%, respectively. The transformation of hydromica (1.0 nM) to kaolinite (0.7 nM) (Figure 1) illustrate the weathering of clay minerals under long-term fertilizations in Blank, maintaining the characteristic of the transformation of clay minerals from 2:1 type to 1:1 type. Compared with $Blank_{2013}$-topsoil, the proportion of 1.4 nM intergradient minerals and kaolinite under different treatments (N, NPK, NPKM, and M) showed decreasing trends, whereas the proportion of hydromica under different treatments showed increasing trends in the topsoil and the subsoil (Figure 1). The decreasing proportions of 1.4 nM intergradient minerals as well as the increasing proportion of hydromica indicate the transformation of clay minerals from expansive clay minerals to non-expansive clay minerals and illustrate the acceleration of soil aging and development under long-term fertilizations. Furthermore, the decreasing proportions of 1.4 nM intergradient minerals in long-term fertilizations containing K fertilizer (NPK and NPKM) were more than those values in long-term fertilizations without K fertilizer (N and M), indicating the enhancement of the transformation effect of 1.4 nM intergradient minerals accounting for the increase of K^+ in soil by K fertilizer.

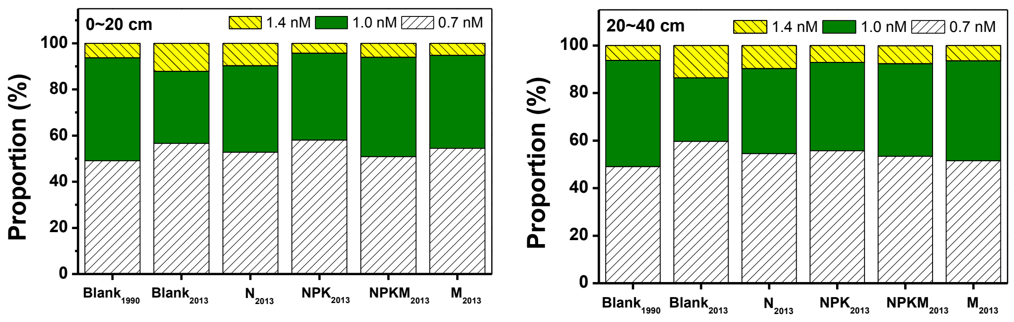

Figure 1 Proportion of kaolinite (0.7 nM), hydromica (1.0 nM), and 1.4 nM intergradient minerals in clay mineral.

CONCLUSION

Following 23 years (1990-2013) of long-term fertilization, chemical fertilizer accelerated and manure fertilizer decelerated soil acidification. Response to soil acidification, the clay minerals transforms from 2:1 to 1:1 type, illustrating the acceleration of soil aging and development under long-term fertilization.

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REFERENCES
Blake, L., A.E. Johnston, and K.W.T. Goulding. 1994. Mobilization of aluminum in soil by acid deposition and its uptake by grass cut for hay - a Chemical Time Bomb. Soil Use and Management **10**: 51-55.
Bolan, N.S., D.C. Adriano, and D. Curtin. 2003. Soil acidification and liming interactions with nutrientand heavy metal transformationand bioavailability. Advances in Agronomy **78:** 215-272.
Breemen, N.V. 1991. Soil Acidification and Alkalinization. Soil Acidity. Springer Berlin Heidelberg.
Cai, Z., Wang, B., Xu, M., Zhang, H., He, X., Zhang, L., Gao, S., 2014. Intensified soil acidification from chemical N fertilization and prevention by manure in an 18-year field experiment in the red soil of southern China. Journal of Soils and Sediments **15(2),** 260-270.

O-8

Variation in yield and phosphorus efficiency traits in tropical maize hybrids grown in low P acid soils of western Kenya.

Ouma E.O[1], Gudu S[1] and Ligeyo D.O[1]
[1]Rongo University, P.O. Box 103-40404, Rongo – Kenya; [2]Kenyan Agricultural and Livestock Research Organization, Kitale – Kenya,
; samgudu2002@yahoo.com ; evans.ouma13@gmail.com

INTRODUCTION

Low available phosphorus (P) remains a major limitation to maize (Zea mays L.) productivity in low P soils. The available P in most western Kenyan acid soils range between 2 to 5 mg P/kg soil which is far below the optimal range (10 to 15 mg P/kg soil) recommended for optimal crop productivity in this region (Kisinyo et al., 2013). This causes very low maize yields (0.5 -2 t/ha) (Ligeyo et al., 2014). Moreover, these soils have high P sorption (107-258 mg P kg) because of the predominant high clay fractions. In such soils, crops utilize only about 10-25% of the P fertilizer applied (Wissuwa et al., 1999) thereby negating the use of Pi fertilizers to maintain yields and crop production in this region. Selecting for P efficient cultivars is therefore very critical in enhancing crop productivity in this region. Phosphorus efficiency traits have been used to select superior maize cultivars for low P soils; however, the extent to which these traits vary under low or high P acid soils environments is little understood. This study was undertaken to determine the extent of genetic variation in P efficiency traits among selected Kenyan maize hybrids under low or high P soils and to select P efficient maize experimental hybrids for use in low P soils.

MATERIAL AND METHODS

A total of 32 experimental maize hybrids and 4 standard checks (efficient and inefficient) were evaluated for tolerance to low P in a replicated trial at four locations in western Kenya. All the sites are characterized by low pH of 4.5-4.8, with P levels ranging between 2.2- 4.4 mg P kg^{-1} of soil (Ouma et al., 2015). The experiment was laid out in a split plot arrangement in RCBD replicated three times. Main plot contained 2 levels of P (6 KgP/ha and 36 KgP/ha supplied as TSP) while the genotypes were randomized in the sub-plot. Each genotype was planted in a two-row plot measuring three meters long with inter and intra-row spacing of 0.75 m x 0.30 m respectively. All the plots were side-dressed using calcium Ammonium Nitrate (CAN) at the rate of 75 Kg N/ha. Data was collected on grain yield, stover yield and other P-efficiency traits determined. Data was analyzed using Genstat Version 18 (Payne et al., 2014) and means separated using Turkeys.

RESULTS AND DISCUSSION

Genotypic variation in maize yields and phosphorus efficiency traits in maize single crosses in acid soils

The low P treatment generally exhibited reduced performance relative to the corresponding high P treatment. Mean grain (GYD) and stover yields were significantly lower (2.49 & 7.28 t/ha) across the low P treatment compared to the high P treatment (4.78 & 9.79 t/ha). There was a big range (35-95%) for relative yield reduction (RYR) among the hybrids showing wide variation in genotypic grain yield performance among the hybrids. Under high P, highest and lowest hybrids yielded 6.41t/ha and 3.89 t/ha while under low P the yields attained were 3.27 and 1.58 t/ha respectively. Mean yield differences between highest and lowest hybrids were further evident across sites with Chepkoilel attaining 6.74 & 3.60 t/ha and Sega 2.12 & 0.89 t/ha respectively. Agronomic Efficiency (AE) of applied P was in the range of 22.7-72.9 kgkg-1 with a mean of 44.8 kgkg-1. Eighteen out of the 32 experimental hybrids exhibited AE above the locational mean (>44.8 kgkg-1 P). P acquisition efficiency (PAE) ranged from 0.06-0.2 KgP/Kgf with majority of the hybrids (57%) attaining values above the mean of the hybrids combined but with P utilization efficiency (PUE), 63% of hybrids exhibited values below the average of the hybrids (553.4 Kg/Kg). Application of higher P levels significantly increased the average grain P concentration (GPC) from 0.15% to 0.19% while that of stover P (SPC) from 0.03 to 0.06%. The increase in GPC & SPC due to high P application level observed in this study compare well with those of Hammond et al. (2009). Majority of the genotypes with low GPC in their tissues had higher PUE showing that they were able to utilize P better. From these findings, it can be suggested that selection for reduced GPC in maize lines may increasing phosphorus utilization. Most of the P sensitive hybrids exhibited low values for P-efficiency traits compared to the tolerant hybrids regardless of whether they were planted in low or high P conditions. Majority of the genotypes expressing higher phosphorus efficiency (PE) also showed higher PAE, PUE and higher GYD under low P implying a good correlation between these traits and hence they may be considered as alternative selection criteria for tolerance to low P in maize in acid soils. A total of 12 hybrids which were selected based on PAE and PUE across the four sites were also best grain yielders under low P conditions. In most cases, genotypes showing higher P efficiency traits (AE, PAE, PUE,) had higher grain yield production under low P supply showing that they may be considered as alternative selection criteria for tolerance to low P in maize.

CONCLUSION

A large genetic variation in P efficiency existed amongst the hybrids both at low P supply and in response to P application in Kenyan acid soils. The genetic variation demonstrates the potential for breeding cultivars with improved phosphorus efficiency with capacity to acquire and utilize applied inorganic Pi fertilizers more efficiently. This study has selected at least 12 experimental hybrids suitable for growing in low P soils of western Kenya.

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REFERENCE

Hammond J.P., Martin R. B., Philip J. W., Graham J. K., Helen C.B., Rory H., Mark C.M., Andrew M., Tracey O., William P. S., and Duncan J.G. 2009. Shoot yield drives phosphorus use efficiency in Brassica oleracea and correlates with root architecture traits. Journal of Experimental Botany, **7 (60):**1953–1968

Ligeyo D.O, E.Ouma, S.Gudu, P.O.Kisinyo, T. Matonyei, J.R. Okalebo and C.O. Othieno. (2014) Response of maize top cross hybrids to low phosphorous in acid soils of western Kenya. East. Africa. Forestry.J., **80(1)** 25-30.

Kisinyo P, Gudu S, Othieno C, Okalebo J, Ochuodho J, Agalo J, Ng'etich W, Opala R, Maghanga, J.K, Kisinyo J.A., Opile W.R, Ogola B.O. 2013a. Phosphorus Sorption and Lime Requirements of Maize Growing Acid Soils of Kenya. Sustainable Agriculture Research., **2(2):**116-123.

Parentoni, S.N., Souza JR., Alves, V.M.C.; Gama, E.E.G.; Coelho, A.M.; Oliveira, A.C. Guimaraes, P.E.O., Guimaraes, C.T., Vasconcelos, M.J.V., Pacheco, C.A.P., Magalhães, J.V., Meirelles, W.F.,Guimarães, L.J.M., Silva, A.R., Mendes, F.F., Schaffert, R.E. 2010. Inheritance and breeding strategies for phosphorus efficiency in tropical maize (Zea mays L.). Maydica., **55**: 1-15

Wissuwa, M. 2003. How Do Plants Achieve Tolerance to Phosphorus Deficiency? Small Causes with Big Effects. DOI: https://doi.org/10.1104/pp.103.02930

O-9

An overview of mitigation of Aluminium toxicity in acid soils by Biochar application

***Prakash Nagabovanalli*[1] *and Rajpal Shetty*[2]**
[1]Department of Soil Science and Agricultural Chemistry, UAS, GKVK, Bangalore 560065, INDIA
[2] Department of Plant Physiology, Faculty of Natural Sciences, Comenius University in Bratislava, Mlynska Dolina, Bratislava 4, Slovakia – 84215
; nagabovanalliprakash@rediffmail.com

Soil acidification exposes plants to toxic effects of aluminium (Al) and manganese (Mn) and further reduces the crop growth and yield drastically by causing deficiency of essential nutrients due to low pH conditions. Soil acidification is becoming a menace in intensively cultivated farmlands due to various cultivation practices and crucial to reclaim such toxic lands to enhance the arable land available for cultivation and sustainable crop production. (Uexkull and Mutert, 1995; Kochian et al., 2004)

Liming has been the prominent approach for ameliorating acid soils. Long term effects of liming on Al toxicity and sub-surface acidity is still an ambiguous area of research despite various studies on liming surface soil acidity (Jawad et al., 2014). The demand on alternate sustainable possibilities such as modification in suitable agronomic practices, crop selection, organic matter enrichment and bio residue application has been driving the scientific community ever since.

Recently, biochar application is considered as a novel approach in this field of study. Biochar as an amendment for acid and Al toxic soils and its effects on the critical properties of soil holds a great promise for the sustainable crop production in near future such as to create a carbon sink to mitigate global warming, increase soil water holding capacity and reduce emissions of NOx and CH_4, as well as to control the mobility of a variety of environmental pollutants, such as heavy metals, pesticides and other organic contaminants (Lehmann et al., 2006; Van Zwieten et al., 2010; Inyang et al., 2011).

The ameliorating effect of biochar is dependent on its pH value, pyrolysis temperature and feed stock materials. Biochar application may alleviate the Al toxicity in soil by multiple mechanisms (Qian and Chen, 2013; Zhao et al., 2015; Qian et al., 2016). It might be due to the liming effect of biochar or adsorption properties and the surface adsorption and co-precipitation of Al with silicate particles to fix Al in soil. Adsorption of Al by biochar might be dominated by the surface complexation of carboxyl groups with $Al\ (OH)^{2+}/Al(OH)_2^{+}$ rather than through electrostatic attraction of Al^{3+} with negatively charged sites. Increase in the rates of application of biochar has significantly decreased Al phytotoxicity in wheat due to increased precipitation and complexation of Al in a hydroponic system (Qian et al., 2013).

Application of wood, bamboo and rice husk biochar increased the pH, exchangeable Ca and Mg and available Si in acid soil and decreased the micronutrients like Mn and Fe suggesting their potential to decrease Al toxicity (Akshatha and Prakash, 2015; Rajpal and Prakash, 2016). Wood biochar application to acid soil significantly decreased soluble Al, exchangeable Al and exchangeable acidity (Rajpal and Prakash, 2016).

Biochar alleviates the Al toxicity in soil by Al adsorption in which the Al complexed with organic hydroxyl and carboxyl groups or the surface adsorption and co-precipitation with silicate particles (as $KAlSi_3O_8$) or both (Qian and Chen, 2013).

Hence, it can be concluded that the biochar amendment appears to be a novel approach with multiple mechanisms for Al detoxification in acidic soils.

REFERENCES

Akshatha, M. K., Prakash, N. B. 2015. Characterization of different biochars and their effect as a source of silicon on aerobic rice. *In* National Seminar on Developments in Soil Science: 2015, ***80th Annual Convention.*** December 5-8, Bangalore, India.

Inyang, M., Gao, B., Ding, W., Pullammanappallil, P., Zimmerman, A.R. And Cao, X., 2011, Enhanced lead sorption by biochar derived from anaerobically digested sugarcane bagasse. *Separ. Sci. Technol.*, **46**: 1950-1956.

Jawad, I. T., Taha, M. R., Majeed, Z. H. And Khan, T. A., 2014., Soil Stabilization Using Lime: Advantages, disadvantages and proposing a potential alternative. *Res. J. Appl. Sci. Eng. Technol.*, **8**(4): 510-520.

Kochian, L. V., Hoekenga, O. A. And Pineros, M. A., 2004, How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorous efficiency. *Ann. Rev. Plant Biol.*, **55**: 459-493.

Lehmann, J., Gaunt, J., Rondon, M., 2006, Bio-char sequestration in terrestrial ecosystems-a review. *Mitig. Adapt. Strat. Global Change*, **11**(2): 395-419.

Qian, L. And Chen, B., 2013, Dual role of biochars as adsorbents for aluminum: The effects of oxygen-containing organic components and the scattering of silicate particles. *Environ. Sci. Technol.*, **47**: 8759-8768.

Qian, L., Chen, B. And Hu D., 2013, Effective alleviation of aluminum phytotoxicity by manure-derived Biochar. *Environ. Sci. Technol.*, **47**: 2737-2745.

Qian, L., Chen. B. And Chen, M., 2016, Novel alleviation mechanisms of aluminium phytotoxicity via released biosilicon from rice straw-derived biochars. *Sci. rep.*, **6**: 29346.

Rajpal. S. And Prakash. N. B., 2017, Evaluation of biochar for management of aluminium toxicity in acid soil. *Commun. Soil Sci. Plant Anal.*, (In Press).

Uexküll, H. R. And Mutert, E., 1995, Global extent, development and economic impact of acid soils. *Plant Soil*, **171**:1-15.

Van Zwieten, L., Kimber, S., Morris, S., Chan, K.Y., Downie, A., Rust, J., Joseph, S. And Cowie, A., 2010, Effects of biochar from slow pyrolysis of papermill waste on agronomic performance and soil fertility. *Plant Soil.*, **327**: 235-246.

Zhao, R., Jiang, D., Coles, N. And Wu, J., 2015, Effects of biochar on the acidity of a loamy clay soil under different incubation conditions. *J. Soils Sediments*, **16**(9): 1-8.

O-10

Application of palm oil mill effluent sludges and soil properties improvement of an Entisol

Mohd Nizar, K.[123], Isharudin, M.I.[1] and Abd Jamil, Z[2]
[1] Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
43400 UPM Serdang, Selangor, Malaysia
[2] Institute of Agrobiotechnology, Universiti Sultan Zainal Abidin, Campus Besut,
22200 Besut, Terengganu, Malaysia.
[3] Felda Global Ventures Research and Development, PPP Tun Razak, 26400 Bandar Tun Abdul Razak Jengka, Pahang, Malaysia
; imi@upm.edu.my

INTRODUCTION

The Malaysian palm oil industry contributed 11% of the world's oils & fats production in 2014 and 27% of export trade (MPOC, 2015). Currently, Malaysia has almost 4.49 million hectares of oil palm under cultivation, producing 17.73 million tonnes of palm oil and 2.13 tonnes oil palm kernel oil in 2014 (MPOC, 2015). There were 416 palm oil mills engaged in processing the oil palms. Commonly, millions of tonnes of POME sludge discharge into open treatment pond systems in the context of affordable operation costs (Khairuddin et al. 2016). Indeed, continuing current practices could lead to environmental pollution with consequences for the human health. POME sludge was identified as high in organic matter content and has the potential to be applied as organic fertilizer in the plantation. Nutongkaew et al. (2014) stated that the POME sludge is high in moisture and nutrients content. According to Khairuddin et al. (2016), POME sludge is rich in carbon, nitrogen, ferrum and potassium. However, there was inadequate information about the application of POME sludge as an organic amendment in improving soil physico-chemical properties and pH such as Entisols.

MATERIALS AND METHODS

The studies were conducted at the research station of Universiti Putra Malaysia, Serdang, Malaysia (3.7562°N, 102.5611°E) from February 2015 until June 2016. Generally, this area is subject to a tropical climate with an average diurnal temperature of 31°C, rainfall of 2300 mm annually and relative humidity of 60–80%. The POME sludge obtained from Felda Palm Oil Industries Sdn Bhd, Bandar Tun Abdul Razak Jengka, Pahang, Malaysia was dried, powdered and sieved with a 2 mm sieve. In this experiment, Rasau series soil was collected from an oil palm plantation in Bandar Tun Abdul Razak Jengka. Based on USDA characteristics, the Rasau series soil is classified as an Entisol. Entisol is a poor quality soil of low nutrient properties and crop yields (Anda and Kurnia, 2010). It mainly consists of fine sandy clay loam with a low organic matter content, fine texture, poor grades, and friable structure, with a cation exchange capacity (CEC) of <5 $cmol^{(+)}/kg$ of soil (low base saturation) and low pH at 3.65 (Barzani et al., 2011). The soil samples were taken at a depth of 10 cm, dried, homogenized and passed through a 2 mm sieve for further processing. The POME sludge was prepared according to the standard methods following the procedures of wastewater (APHA, 2005) and soil analysis (Brady, 2008). Statistical analyses were conducted using SAS version 7 (2007) software. The results of the physico-chemical characteristic were evaluated using analysis of variance (ANOVA).

RESULTS AND DISCUSSION

Physico-chemical properties of POME sludge

Table 1 shows the treatments effect on total carbon in the soil. There was a significant difference ($p \leq 0.05$) in dumping pond (DP) sludge compared to all the treatments. The DP treatment showed the highest mean value of 20.8% compared to the other treatments. The mixing pond (MP) sludge showed low composition of organic carbon which could be attributed to the POME sludge decomposition process, still occurring through the microorganism's activities, and correlated to the Sludge Retention Time (SRT) of wastewater treatments practices (Bagdeli and Seilsepour, 2008). Consequently, the result from this study also showed that the application of organic amendment significantly improved the soil organic carbon content in the Rasau series soil. Total Nitrogen is important for crop growth and development. Table 1 shows the soil nitrogen composition that was significantly different ($p < 0.05$) in DP sludge compared to all the treatments. The total Nitrogen intake is an essential nutrient for cell growth (Ali et al., 2013). Ryals et al. (2014) stated that the composition process increases the N products from the organic matter and released available nutrients (N, P, and S) into the soil for plants uptake. Table 1 also shows the effect of treatments on the C/N ratio of all the treatments that was significantly different in MP sludge and the control compared to the other treatments. When the C/N ratio was stable, the decomposition process became faster and nutrients production by the microorganisms increased the fertility status of the soil. According to Baharuddin et al. (2010), the composting material was available when the POME sludge had reduced its C/N ratio to < 15. The DP sludge showed the highest pH (7.31) and was significantly different compared to the other treatments. The pH value was alkaline and suitable for plant growth such as cabbage, cauliflower, and thyme (Tchobanoglous et al., 1993). At this pH level, micronutrients such as iron, manganese, and phosphorus were also available (Jensen, 2010). The organic amendment application was able to improve the soil pH (Table 1). With the increase of organic matter, the soil recovers its natural buffer capacity; this means an increase of pH in acid soils. There were significant differences ($p < 0.05$) of CEC in the DP sludge and ALP sludge compared to the other treatments. The DP treatment showed the highest mean value of 0.30 meq/100g compared to the other treatments. There were significant differences between FP sludge, ANP sludge, MP sludge, and the control (Table 1). A high CEC value indicated that the soil was able to hold more cations. The organic amendment was associated with desirable soil properties including high water-holding capacity, CEC and low bulk density, fostering the beneficial micro-organisms (Doran, 2002; Drinkwater et al., 1998).

Table 1. The C, N, C/N ratio, pH and CEC characteristics of the POME sludge

Parameter	Treatment					
	DP sludge	ALP sludge	FP sludge	ANP sludge	MP sludge	Control
Total Carbon (%)	20.80a	18.32b	17.07c	16.15c	16.05cd	2.90e
Total Nitrogen (%)	3.82a	2.97b	2.02c	1.51e	1.24f	1.87d
C/N ratio	4.44a	5.16b	7.86c	9.69d	11.94e	11.05e
pH	7.31a	7.08b	6.42b	5.20c	4.12d	3.42e
CEC (meg/100g)	0.30a	0.28a	0.26b	0.24c	0.22d	0.20e

Note: MP sludge (Mixing ponds sludge), ANP sludge (Anaerobic ponds sludge), FP sludge (Facultative ponds sludge), ALP sludge (Algae ponds sludge) and DP sludge (Dumping ponds sludge)

Phosphorus was significantly ($p < 0.05$) higher in the DP sludge (8.60 mg kg^{-1}) than the ALP sludge (6.98 mg kg^{-1}), FP sludge (5.10 mg kg^{-1}), ANP sludge (4.59 mg kg^{-1}), MP sludge (2.86 mg kg^{-1}) and control (3.46 mg kg^{-1}) (Table 2). In this study, the DP and ALP sludge showed higher amounts of P than the other treatments which could help the maize growth through absorbance of P by the maize root. Batjes (2011) reported that P was an essential element in soil and existed in the form of organic phosphate that became part of the soil organic matter. In addition, the POME sludge waste was significantly higher ($p < 0.05$) in potassium in the DP (16.00 mg kg^{-1}) than the control (5.71 mg kg^{-1}). K was considered as an immobile nutrient in the soil. However, CEC was increased therefore it could help the K to mobilize in the POME sludge. The Mg concentration was highest in the DP sludge (5.32 mg kg^{1}) followed by ALP sludge (4.78 mg kg^{-1}). Moreover, Ca concentration was higher in DP sludge (26.29 mg kg^{-1}) than the control (18.17 mg kg^{-1}). This could be due to the higher Ca content of raw POME sludge from the dumping pond than the other treatments. The presence of Ca and Mg showed that the cation exchange capacity could be affected, simultaneously promoting the favorable soil structure.

For heavy metal properties, Table 2 showed the changes of Cu content in the various treatments. There were significant differences ($p < 0.05$) of Cu in the DP sludge (0.44 mg kg^{-1}), control (0.62 mg kg^{-1}) and MP sludge (0.79 mg kg^{-1}). Meanwhile, Cd showed no significant difference among the treatments. Indeed, the Pb and Ni content were significantly ($p < 0.05$) different between the DP sludge and control. Khairuddin et al (2016) reported that the concentrations of Cu, Cd, Pb and Ni were in accordance with the WHO/FAO standard and safe for human consumption. Heavy metals could affect the growth, morphology and metabolism of micro-organisms in bulk soils, through functional disturbance, protein denaturation or the destruction of the integrity of cell membranes (Igbinosa, 2015). Zeng et al. (2011) estimated that Cd, Pb and Zn contents were positively correlated with the content of organic matter in the soils.

Table 2. Nutrient and heavy metal characteristics of POME sludge

Parameter	Treatment					
	DP sludge	ALP sludge	FP sludge	ANP sludge	MP sludge	Control
P (mg kg^{-1})	8.60a	8.61a	6.98b	5.10c	4.59cd	2.85de
K (mg kg^{-1})	16.00a	14.66ab	14.65ab	10.87bc	9.35cd	5.71d
Mg (mg kg^{-1})	5.329a	5.33a	4.78b	3.50c	2.12d	0.40e
Ca (mg kg^{-1})	26.30a	24.10b	23.7bc	22.57cd	21.47d	18.17e
Fe (mg kg^{-1})	71.34a	65.57b	53.94c	45.69d	42.06e	39.11f
Cu (mg kg^{-1})	0.44c	0.47c	0.43c	0.66b	0.79a	0.62b
Cd (mg kg^{-1})	0.01a	0.01a	0.01a	0.01a	0.01a	0.01a
Pb (mg kg^{-1})	0.13b	0.14b	0.09b	0.15b	0.11b	0.21a
Ni (mg kg^{-1})	0.18b	0.10b	0.17b	0.18b	0.18b	0.34a

Note: MP sludge (Mixing ponds sludge), ANP sludge (Anaerobic ponds sludge), FP sludge (Facultative ponds sludge), ALP sludge (Algae ponds sludge) and DP sludge (Dumping ponds sludge)

CONCLUSION

The results from this study showed that the application of proper processing methodologies and techniques might transform this abundant material into beneficial materials such as organic fertilizer. The study revealed that the application of POME sludge in the acidic soil might improve soil fertility and nutrients availability for plant growth. Furthermore, the studies indicated the potential use of palm oil waste as an organic fertilizer with better pH, nutrients content and a low amount of heavy metals.

REFERENCES

Ali, H., Khan, E., & Sajad, M. A. 2013. Phytoremediation of heavy metals—concepts and applications. Chemosphere, **91(7),** 869-881.

Anda, M. and Kurnia, U. 2010. Restoring properties of artificially degraded Ultisols and Oxisols and the effect on crop yields under tropical conditions. Communications in soil science and plant analysis, **41(5),** 553-570.

APHA. 2005. Standard methods for the examination of water and wastewater, 20th edition. E.W. Rice, R.B. Baird, A.D. Eaton and L.S. Clesceri (eds.). Washington: American Public Health Association. p. 1200

Baharuddin, A.S., Lim, S.H., Md Yusof, M.Z., Abdul Rahman, N.A., Md. Shah,U.K., Hassan, M.A., Wakisaka,M., Sakai, K. and Shirai, Y. 2010. Effect Of Palm Oil Mill Effluent (POME) Anaerobic Sludge Based Compost Using Fourier Transform Infrared (FTIR) And Nuclear Magnetic Resonance (NMR) Analysis. African Journal Biotechnology, **10(41),** Pp. 2427-2436.

Batjes, N. H. 2011. Global distribution of soil phosphorus retention potential (No. 2011/06). ISRIC-World Soil Information.

Barzani, G., Ismail, B.S., Saujaul-Islam, M., Sahibin, A,R. and Ekhwan, T. 2011. The Physico-chemical properties of Four Soil Series in Tasik Chini, Pahang, Malaysia. Asian Journal of Earth Science, (2), 75-84.

Bigdeli, M., and M. Seilsepour. 2008. Investigation of Metals Accumulation in Some Vegetables Irrigated with Waste Water in Share Rey-Iran and Toxicological Implications. American-Eurasian Journal Agriculture & Environmental Science 4, no. **1**: 86-92.

Brady, Nyle C., and Ray R. Weil., 2008. The Nature And Property Of Soils, 14th Edition. Upper Saddle River, New Jersey: Prentice-Hall

Doran, J.W. (2002). Soil health and global sustainability: translating science into practice. Agriculture Ecosystem Environmental, **88:** 119-127.

Drinkwater, L. E., Wagoner, P., and Sarrantonio, M., 1998. Legume-based cropping systems have reduced carbon and nitrogen losses. Nature, **396(6708),** 262-265.

Igbinosa, E. O. (2015). Effect of cassava mill effluent on biological activity of soil mi-crobial community. Environmental monitoring and assessment, **187(7):** 418.

Jensen, T. L., 2010. Soil pH and the availability of plant nutrients. IPNI Plant Nutrition TODAY Fall.

Khairuddin, M. N., A.J. Zakaria, I.M. Isa, H. Jol, W.M.N.W.A. Rahman, and M.K.S. Salleh. "The Potential of Treated Palm Oil Mill Effluent (Pome) Sludge As An Organic Fertilizer." AGRIVITA, Journal of Agricultural Science 38, no. **2 (2016):** 142-154.

Malaysia Palm Oil Council (MPOC). (2015). An Overview of Malaysian Palm Oil in the Global Oils and Fats Scenario. Reviewed on 21 Nov 2017 at http://www.mpoc.gov.com

Nutongkaew, T., Duangsuwan, W., Prasertsan, S, and Prasertsan, P. 2014. Physico-chemical And Biochemical Changes During Composting Of Different Mixing Ratios Of Biogas Sludge With Palm Oil Mill Wastes And Biogas Effluent. Journal Mater Cycles Waste Manag. 16: 131-140

Ryals, R., Kaiser, M., Torn, M. S., Berhe, A. A., and Silver, W. L., 2014. Impacts of Organic Matter Amendments On Carbon And Nitrogen Dynamics In Grassland Soils. Soil Biology and Biochemistry, 68, 52-61.

SAS Intstitute Inc. 2007." SAS/STAT software version 9." SAS Institute, Inc. Cary, NC, USA.

Sharma, J. and R. Singh., 2001. Effect of nutrients supplementation on anaerobic sludge development and activity for treating distillery effluent. Bioresource Technology **79 (2):** 203-206.

Tchobanoglous, G. (1993). Integrated solid waste managementengineering principles and management issues (No. 628 T3).

Zeng, F., Ali, S., Zhang, H., Ouyang, Y., Qiu, B., Wu, F., and Zhang, G. (2011). The influence of pH and organic matter content in paddy soil on heavy metal availability and their uptake by rice plants. Environmental pollution, **159(1):** 84-91.

O-11

Increasing subsoil pH through addition of lucerne (*Medicago sativa* L.) pellets in the surface layer of an acidic soil

Hoang Han Nguyen[1,2]*, J. Sergio Moroni[1], Jason Condon[1], Alek Zander[1] and Guangdi Li[3]
[1]Graham Centre for Agricultural Innovation (Charles Sturt University and NSW Department of Primary Industries)
[2]Department of Agriculture and Rural Development of Soc Trang province, Vietnam; [3]NSW Department of Primary Industries, Australia
; hnguyen@csu.edu.au

INTRODUCTION

Soil acidity is a major agricultural problem around the world due to the deleterious effects of aluminium (Al^{3+}) and/or manganese (Mn^{2+}) and/or nutrient deficiency on crop growth. Raising soil pH is the most common strategy to improve conditions of acidic soils. It is relatively simple to increase pH in the surface soil layer (0-10 cm) by applying soil amendments such as lime ($CaCO_3$). However, acidity in subsoil layers (at depths greater than 10 cm) is difficult to correct due to the relatively low solubility of liming materials and the slow movement of the amendments down the soil profile. Organic amendment has been shown to increase soil pH at below the amended layer due to the movement of alkaline compounds (Butterly *et al.*, 2013; Cassiolato *et al.*, 2000; Cassiolato *et al.*, 2002; Miyazawa *et al.*, 2002; Wang *et al.*, 2016). For this work, it was hypothesised that this effect may be influenced by the particle size of the organic amendment and that greater solubility of organic compounds, compared with lime, which may be more effective in ameliorating acidic soil below the depth of placement or incorporation.

MATERIALS AND METHODS

Soil and amendments. Acid soil was collected in 10 cm increments from a 40 cm soil profile at Bethungra, NSW, Australia (34°38'S, 147°49'E). The soil was sieved through a 9.5 mm sieve, mixed and air-dried. The pH($CaCl_2$) was 4.82, 4.18, 4.12 and 4.62 of soil layers 0-10, 10-20, 20-30 and 30-40 cm, respectively. Lime and lucerne pellets with different sizes (LP1: ~20 mm, LP2: ~10 mm, LP3: ~5 mm and LP4: ~1-2 mm) were applied as inorganic and organic amendments, respectively.

Pot construction. Plastic pots (50 cm length) were built using 10 cm diameter polyvinyl chloride (PVC) pipes. The pots were capped at one end. A plastic lining (plastic bag, 9.8 cm ID), sealed in one end was inserted in the PVC column to prevent drainage. The soil profile was re-constructed by filling each column with soil layers to obtain 47 cm soil height (17 cm of soil layer 30-40 and 10 cm of the other soil layers). Lucerne pellets and lime were ameliorated in 5 and 10 cm of the surface soil column (layer 0-10) with rates of 15 t/ha and1.25 t/ha, respectively. A few small white plastic beads were placed at around the edge of each layer to identify soil layers upon sampling. Seven wheat seeds (cv. Dart) were sown and seedlings were thinned to 4 plants per pot at 4 days after sowing (DAS). Pots were watered to field capacity and rewatered every 2-3 days. The columns were placed in a climate controlled glasshouse at CSU.

Experimental design: The experiment was conducted as a Randomized Complete Block (RCB) design with 10 treatments (Control (nil), lime (L), LP1, LP2, LP3, LP4, L+LP1, L+LP2, L+LP3 and L+LP4) x 4 replicates x 4 sampling times.

Data collection: Shoot dry weight (SDW), root dry weight (RDW) and soil samples were destructively collected at 14, 21, 28 and 35 DAS. Shoots and roots were oven-died at 60^{o}C (96 hrs) before weighing. Soil samples were collected at depths of 2.5, 7.5, 11.0, 13.0, 17.5, 22.5, 27.5, 32.5 and 40.0 cm with a 2 cm ID core, placed in the plastic bags, oven-dried at 40^{o}C (72 hrs), ground and sieved through 2 mm sieve for pH analysis (pH(1:5 0.01M $CaCl_2$)).

Statistical analysis: Analysis of variance was conducted by GenStat v.18. The LSD at p=0.05 was computed and used for comparing the effects of treatments.

RESULTS AND DISCUSSION

Plant growth

At the early stages, the addition of lucerne pellets affected plant growth negatively. Shoot dry weight and RDW of plants grown in lucerne pellets treated soils were significantly lower than that planted in control soil up to 28 DAS (Figure 1). The finest particle size reduced wheat growth more than the other particle sizes as SDW and RDW of treatments with LP4 were the smallest (Figure 1). At 35 DAS, however, plant growth in organic amendment treatments were not significantly different compared with Nil or lime treatments. Moreover, SDW and RDW of wheat plants grown in treatments with lime and lucerne pellet ~5 mm were the highest (Figure 1).

a b

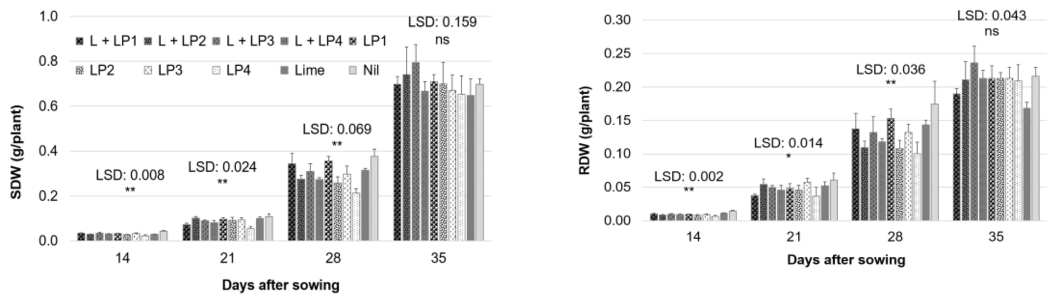

Figure 1. Shoot (a) and root (b) dry weight of wheat plant over harvesting times

pH changes at soil depths

Organic amendment increased soil pH not only at the placement but also at soil depths. Results indicated that at all sampling times, lucerne pellets increased soil pH throughout soil columns but lime did not (Figure 2). Similarly, Butterly *et al.* (2013) showed that organic matter such as chick pea and canola increased soil pH at soil layers below placement under field condition.

Lime addition increased soil pH at depths greater than that of lucerne pellets alone (Figure 2). When lime was applied, lime alleviated acidity at amended layer allowing more available of soluble alkalinity of lucerne pellets moving down the soil depths. Wang *et al.* (2016) also suggested that incorporation of lime and organic matter had a greater increase in soil pH at depths for a subsoil acidity of tea garden soil. Moreover, soluble alkalinity of organic matter may increase solubility of lime to move down the soil profile (Cassiolato *et al.*, 2000; Cassiolato *et al.*, 2002; Miyazawa *et al.*, 2002). Therefore, combination of lime and organic amendments on the surface soil may alleviate subsoil acidity without incorporation at depths.

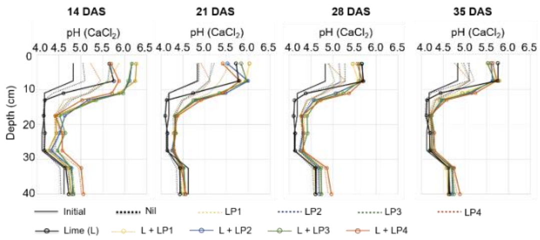

Figure 2. Soil pH changes throughout soil column over sampling times

CONCLUSIONS

Organic matter increased soil pH at layers below the placement, but lime did not. When applied with lime, the lime alleviated acidity in the topsoil, thereby allowing more soluble alkalinity from organic matter to move downwards and thus increased pH at depths greater. The finer the particle size the greater increased in soil pH at depths. However, the finest particle affected plant growth negatively. We concluded that the incorporation of lime and lucerne pellets with size ~5 mm was the best amelioration strategy to increase subsoil pH from 10-30 cm of the soil profile and improve plant growth.

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REFERENCES

Butterly, C. R.; Baldock, J. A. and Tang, C. 2013. The contribution of crop residues to changes in soil pH under field conditions. *Plant and Soil,* **366**: 185-198

Cassiolato, M. E.; Meda, A. R.; Pavan, M. A.; Miyazawa, M. and Oliveira, J. C. d. 2000. Evaluation of oat extracts on the efficiency of lime in soil. *Brazilian Archives of Biology and Technology,* **43**: 533-536

C**assiolato, M. E.; Miyazawa, M.; Meda, A. R. and Pavan, M. A. 2002.** A laboratory method to estimate the efficiency of plant extract to neutralize soil acidity. *Brazilian Archives of Biology and Technology,* **45**: 183-187

Miyazawa, M.; Pavan, M. A. and Franchini, J. C. 2002. Evaluation of plant residues on the mobility of surface applied lime. *Brazilian Archives of Biology and Technology,* **45**: 251-256

W**ang, L.; Butterly, C. R.; Chen, Q.; Mu, Z.; Wang, X.; Xi, Y.; Zhang, J. and Xiao, X. 2016.** Surface Amendments Can Ameliorate Subsoil Acidity in Tea Garden Soils of High-Rainfall Environments. *Pedosphere,* **26**: 180-191

O-12

Reducing Zinc Availability in Tropical Acid Soil by using Humic Acids, Crude Fulvic Acids and Humin

***Maliana, W.A.*[1], *S. Kasim*[1], *and E.M. Shuib*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
43400 UPM Serdang, Selangor, Malaysia
; susilawati@upm.edu.my

INTRODUCTION

Zinc contamination is widespread and in many regions still increasing due to the increase of contamination from various sources of pollution that enhanced Zn availability in soils. The concentration of Zn in natural soil is usually higher than other types of heavy metal and most of them are distributed between organically complexed form and in inorganic form, as Zn^{2+} in the soil solution (Ko et al., 2007). The availability of Zn in soils is controlled by many factors such as pH, clay, organic matter and nutrient contents (Kiekens, 1995; Azura et al., 2012). Soil pH between 4 to 8 might possibly promotes chelation process to takes place (Tan, 2003). In sandy and acid soils, Zn retention capacity is increased as the amount of organic matter increased (McBride and Blasiak, 1979), however, types and levels of humic fractions are classified as the important factors for metal chelation within organic molecules structure (Stevenson, 1994). The effect of humic fractions especially from tropical region in removing Zn contamination is not well studied so far, thus this study was conducted to investigate the effect of tropical humic fractions on Zn availability in acid soils.

MATERIALS AND METHODS

The mineral soil used in this study was Nyalau Series (*Typic Paleudults*). These mineral soils were obtained from a disturbed area at Universiti Putra Malaysia Bintulu Sarawak Campus Malaysia. Soil samples were taken at a depth of 0 to 25 cm. Whilst, deep peat soil were sampled at Block 6, 2°49' 39.9" N 111°55' 36.2"E, Sungai Talau Research Station, CRAUN Research in Mukah, Sarawak at a depth of 0 to 25 cm. Both soil samples were air dried and sieved to pass through a 2 mm sieve before further use in the experiment. Humic and fulvic acids were obtained from the air dry deep peat by alkaline extraction at room temperature (Susilawati *et al*., 2007). An alkaline solution is used to extract humic fractions because it is needed to breakdown the electrostatic bonds by exchanging it with ions as well as helping in solubilizing the organic molecules by ionizing the acid and phenolic functional groups (Stevenson, 1994). The residue called humin were air dried until a constant weight achieved. The three types of humic fractions were then weighed/measured according to Table 1 before they were used in treating 150 g of air-dried mineral soil in the plastic container. The treatments were applied at 75% field capacity. Prior to treatments, the soil had been treated with 1000 mg kg^{-1} Zn in the form of $ZnSO_4$. Then, all treated soil were left at room temperature for different incubation period (0, 3, 6, 9, 12, and 15 days) before they were destructively sampled and analyzed using standard laboratory protocols. The experiment was conducted in completely randomized design with three replications and the data was analyzed using SAS version 9.3 (SAS, 2001).

RESULTS AND DISCUSSION

Nyalau series (*Typic paleudults*) soil, that was used in this study has a CEC of 24.2 $cmol^{(+)}$ kg^{-1}. Its texture was sandy loam with the pH value of 4.3 and 3.9 in water and 1M KCl, respectively. Application of different humic fractions has caused the soil pH to be different significantly ($p \leq 0.05$) (Table 2). Soil treated with either HA + crude FA (T4) or humin (T5) had higher pH value than that of other treatments (Table 2). Alkalinity (expected pH for 0.1M KOH) offered by extraction solution of the humic fractions has indirectly caused the pH to increase. The pH of treated soil fluctuated within 15 days of incubation period for all treatments. Crude FA and humin were more effective in reducing Zn^{2+} availability in acidic soil. As shown in Figure 1, the amount of Zn^{2+} in the soil solution was reduced significantly (day 0 and 3) when they were treated with either crude FA or humin. In case of HA, contrasting results were recorded. Application of HA was not able to reduce Zn^{2+} in acidic soil in short incubation period (<6 days). Solubility of the humic fractions and alkalinity could be the main reason for these findings, besides the presence of high K^+ in crude FA and humin. Generally, HA requires high pH to be dissolved and the acidity has made the HA to be less effective. At the end of incubation period, combination of the three fractions known as HA, crude FA and humin was able to reduce Zn^{2+} in acidic soil. This was because, humic substances naturally exert strong binding strength with metals, which may be attributed to the interaction of metal ions with two important acidic binding sites of carboxylic and phenolic hydroxyl groups of humic substances (Cameron and Sohn, 1992; Ashley, 1996).

Table 1: Treatments used in zinc chelation study

Trt	Zinc (mg/kg)	Humic Acid (g)	Crude Fulvic Acid (mL)	Crude Humin (g)
T0	-	-	-	-
T1	1000	-	-	-
T2	1000	0.5	-	-
T3	1000	-	5	-
T4	1000	-	-	3
T5	1000	0.5	5	-
T6	1000	0.5	5	3

Table 2: pH_{KCl} of treated soil at different incubation period

Trt/day	0	3	6	9	12	15
T0	3.81a	3.94b	3.97b	3.95b	3.93ab	3.89b
T1	3.67b	3.79c	3.87c	3.84c	3.78c	3.71c
T2	3.67b	3.75d	3.85cd	3.80c	3.73cd	3.71c
T3	3.54c	3.64f	3.77d	3.69d	3.66d	3.60e
T4	3.85a	3.99a	4.08a	4.05a	4.00a	3.94a
T5	3.81a	3.91b	4.01ab	3.81c	3.93ab	3.90ab
T6	3.56c	3.70e	3.81cd	3.91b	3.87b	3.65d

*Means with different letters within column are significantly different by using Tukey Test at $p \leq 0.05$

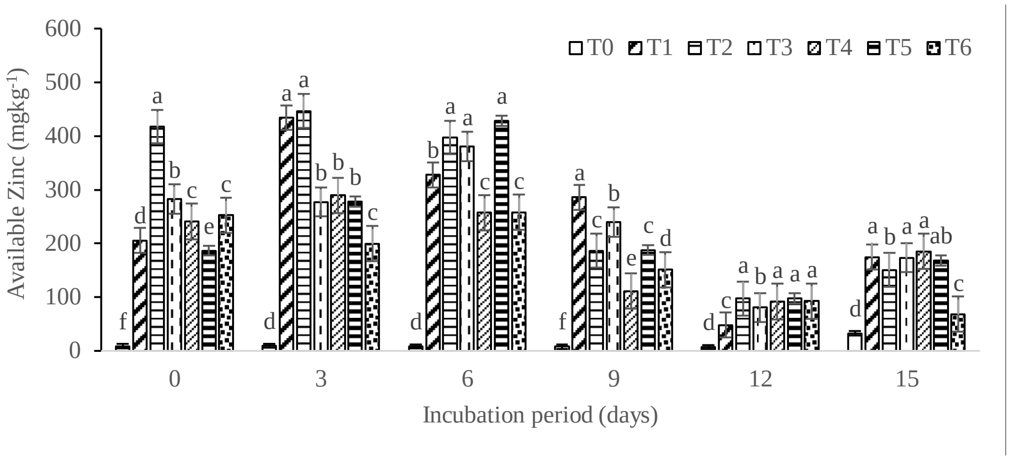

Figure 1: Amount of available zinc in soil after treated with different types of humic fractions

*Means with different letters within each incubation day are significantly different by using Tukey Test at p≤ 0.05

CONCLUSION

Humic fractions were able to reduce Zn^{2+} availability in acid soil, however different incubation period is needed for each of them to be effective. Crude FA and humin are known to be essential sources to control Zn contamination in acid soil in a short period of time. However, it is recommended to further clarify the effect of different humic fraction concentration levels on Zn chelation to extend the understanding and application, since this aspect has not been covered in the current study.

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REFERENCES

Aini Azura, A., Fauziah, C.I., and Samsuri, A.W. 2012. Cadmium and zinc concentrations in soils and oil palm tissues as affected by long-term application of phosphate rocks fertilizers. Soil and Sediment Contamination: An International Journal, **21**(5): 586-603. https://doi.org/10.1080/15320383.2012.672485

Ashley, J.T.F. 1996. Adsorption of Cu(II) and Zn(II) by estuarine, riverine and terrestrial humic acids. Chemosphere, **33:** 2175-2187.

Cameron, D.F. and M.L. Sohn. 1992. Functional group content of soil and sedimentary humic acids determined by CP?MAS 13C NMR related to conditional Zn(II) and Cd(II) formation constants. Science Total Environment, **113:**121-132.

Kiekens, L. 1995. Zinc. In B.J. Alloyway (ed.), Heavy Metals in Soils, 2nd ed., Blackie Academic & Professional, Glasgow, pp. 284-305.

Ko, T.H., M.H. Shih, and H.T. Hsueh. 2007. Spectroscopic study on the zinc-contaminated soils for the determination of zinc speciation. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, **66**(2): 442-447.

McBride, M.B. and J.J. Blasiak. 1979. Zinc and copper solubility as a function of pH in an acid soil. Soil Science Society of American Journal,

SAS, (2001). SAS/STAT Software. SAS Institute, Cary, NC.

Stevenson, F.J., 1994. Humus chemistry: genesis, composition, reactions, 2nd edition. Wiley, New York, pp: 378 - 486.

Susilawati, K., O.H. Ahmed, A.B. Nik Muhamad, and M. Y. Khanif, 2007. Effects of extraction and fractionation period on the yield of a tropical peat soil (Hemists) humic acids. American Journal of Agricultural and Biological Sciences, **2**(3): 202-205.

Tan, K.H., 2003. Humic matter in soil and the environment: Principles and controversies. Marcel Dekker, Inc., New York, pp:34-71.

O-13

Management of acid soils by liming for diversified cropping systems in Bangladesh

***M. Jahiruddin*[1], *M. Bodruzzaman*[2], *G. K. M. M. Rahman*[3], *and M. M. Rahmanl*[1]**
[1]Dept. of Soil Science, Bangladesh Agricultural University, Mymensingh, Bangladesh
[2]Bangladesh Agricultural Research Institute, Gazipur, Bangladesh
[3]Dept. of Soil Science, Bangabandhu Sheikh Mujibur Rahman Agril. Univ, Gazipur, Bangladesh
; m_jahiruddin@yahoo.com

INTRODUCTION

In Bangladesh more than 50% people (total 160 million population) live in acid soil areas and 45% people are engaged in agriculture. The adverse effects of soil acidity on crop productivity have significant impact on food security as well as people's livelihoods. Thus, management of acid soils is a great challenge in Bangladesh agriculture. Of 14.7 million ha lands, 0.108 million ha lands across the country are very strongly acidic (pH <4.5), 3.383 million ha lands strongly acidic (pH 4.5-5.5), and 1.114 million ha lands are moderately acidic (pH 5.6-6.5) in reaction. Soil acidity is increasing with time mainly because of leaching out of basic cations (Ca^{2+}, Mg^{2+}) for heavy monsoon rains and abundant use of urea fertilizer. Between 1998 and 2010, the very strongly acid plus strongly acid soils area has increased by 13% (SRDI, 1998, 2010). Acid soils may constraint crop production in more than 30% of lands in this country. Liming is a good practice for amelioration of soil acidity and improvement of crop yield.

MATERIALS AND METHODS

Lime requirement as well as its effect on diversified crops was studied in acid soils. Lime requirement was assessed by three methods: (i) soil-lime incubation, (ii) Shoemaker, McLean and Pratt single buffer method (Shoemaker et al., 1961) and (iii) decision-support system, NuMaSS (Smyth et al., 2007) over a wide range of acid soils and land types. Results of lime requirement from the three methods were verified through field experiments and an assessment was done in terms of crop productivity and profitability in the two cropping systems: wheat-rice and maize-rice in two consecutive years.

In order to evaluate the effects of lime application on soil chemical properties, soil samples were collected from the fields of above two cropping system experiments after two years. Analysis was done for soil pH and extractable K, P, Ca, Mg, Zn, Mn, Cu and Fe contents.

A number of single crop trials were done in acid soils with and without liming (1 t ha^{-1}). This was carried out at farmer's fields with 40 crops in liaison with the Department of Agriculture Extension (DAE) of the Govt.

Another investigation was done to determine the optimum time for sowing/planting after lime application. It was tested on a wide range of crops – wheat, maize, mustard, carrot, radish, tomato, potato, brinjal, sweet gourd, chili, cauliflower, amaranth, lentil, chickpea and garlic. Biomass was recorded after 30 days of sowing/planting from lime treated/ untreated plots.

RESULTS AND DISCUSSION

The dolomite lime ($CaCO_3.MgCO_3$) estimate for the incubation method was 0.04-4.7 t ha^{-1} with a mean of 1.86 t ha^{-1}, the SMP buffer method 0-4.2 t ha^{-1} with a mean of 1.22 t ha^{-1} and in contrast for the NuMaSS methods were 0-1.25 t ha^{-1} and 0-3.5 t ha^{-1} having mean values of 0.18 and 0.67 t ha^{-1} for 15% and 30% aluminium saturation, respectively.

The crop experiments revealed that dolomite application consistently increased the grain yields for all crops at all sites over the years. However, lime response was higher for dry land crops (maize, wheat) than for wetland rice. The yield increment depending on the sites and years ranged from 14-51% for wheat, 19-93% for maize and only 9-17% for rice. The yield declined at higher (4 - 6 t ha^{-1}) lime application where zinc deficiency appeared. The optimum lime rate was found as 1-2 t ha^{-1}. A set of 41 non-replicated validation trials was done in farmers' fields across eight agroecological zones (AEZs) which are strongly acid soil zones. The yield response curve showed higher crop response at 2 t ha^{-1} lime applied fields, which however was not significant over 1 t ha^{-1} lime rate. Some of the validation trials were continued to examine the direct and residual effects of dolomite over three years in the two cropping systems, wheat-jute-rice in 6 sites and potato-maize-rice in 2 sites. These trials showed positive yield response to the added dolomite at 1 or 2 t ha^{-1}.

Analysis of soil samples collected after two years from the two cropping pattern experiments exhibited that 90% of the soil reaction was complete within 3 months of lime application for 1 or 2 t ha^{-1} lime application, whereas the reaction continued and attained equilibrium after 12 months. The increased pH after lime effect was persistent up to at least three years. Extractable P increased by 34%, 53%, 57% and 33% due to 1, 2, 4 and 6 t ha^{-1} lime, respectively. Exchangeable K remained unaffected when lime was added at 1 or 2 t ha^{-1} while it declined when lime was added at higher rate (4 or 6 t ha^{-1}). The exchangeable Ca and Mg obviously increased linearly with the rates of lime application. Concerning micronutrients, the availability tended to decrease with the rates of lime application. Nevertheless, such effect was the minimum when lime was added at 1 or 2 t ha^{-1}.

The farmers' field (single crops) results showed that all the crops positively responded to the lime application. Rice among the crops tested responded the minimum and ginger, turmeric and wax gourd responded the maximum (Table 1).
The sowing/planting time experiments displayed that the biomass reached its maximum when lime was applied at least seven days before crop was sown or planted.

CONCLUSION

Application of dolomite at 1 t ha^{-1} can sustain higher crop yield in acid soils for at least three years. Crop should be sown or planted after at least one week of lime application. Rice crop responded less because of its cultivation in wetland condition which favors the soil to attain pH neutrality.

REFERENCES

Bodruzzaman, M, Rahman, G. K. M. M., Islam, M. S., Roy, M. C., Duxbury, J. M. and Lauren, J.G. 2014. Lime Technology for the Acid Soil Regions of Bangladesh.

Shoemaker, H. E., McLean, E. O. and Pratt, P. F. 1961. Buffer methods for determining the lime requirement of soils with appropriate amounts of extractable aluminium. Soil Sci. Soc. Am. Proc. **25:** 274-277.

Smyth, T.J. Osmond, D. L., Yost, R. S., Hoag, D. L., Reid, W. s., Branch, W., Wang, X. and Li, H. 2007. Nutrient Management Support Sustem (NuMaSS), v. 2.2 Soil Management Collaborative Research Support Program, NCSU, Raleigh, USA.

SRDI, 1998, 2010. Soil reaction map. Soil Resource Development Institute (SRDI), Dhaka.

Table 1. Yield benefits of different crops for dolomite application in acid soils (Bodruzzaman et al., 2014)

Crop	Yield benefits		Crop	Yield benefits	
	%	kg ha^{-1}		%	kg ha^{-1}
Amaranth	12-40	1500-7910	**Mukhi Kachu**	11-42	2710-6690
Bitter gourd	16-50	2800-10020	**Mungbean**	20-37	150-294
Black gram	33-47	250-330	**Mustard**	11-26	100-250
Bottle gourd	17-41	2590-5000	**Onion**	17-60	1310-4350
Brinjal	23-42	3934-4710	**Point gourd**	13-75	1730-7500
Cancone	25-50	1630-15000	**Potato**	23-30	4400-4880
Carrot	23-31	1650-2470	**radish**	28-51	2200-6990
Chili	25-58	1730-3500	**Rice**	11-15	320-555
Cole crops	12-44	5500-9880	**Ridge gourd**	12-41	2220-4490
Cucumber	27-50	2000-4950	**Snake gourd**	33	3670
French bean	10-30	250-3500	**Spinach**	17-45	1730-20250
Garden pea	20	2450	**Sponge gourd**	29	9880
Garlic	19-67	980-3600	**Sigarcane**	26-32	20650-21640
Ginger	10-606	900-2990	**Sweet gourd**	14-39	4400-5440
Groundnut	28-40	550-990	**Tomato**	27-69	3620-10020
Jute	20-29	400-550	**Turmeric**	12-128	2100-8890
Khira	22-28	3210-9000	**Waermelon**	22	7000
Lady's finger	15-53	2400-3460	**Wax gourd**	43-121	7410-10130
Lentil	31-51	250-700	**Wheat**	17-43	500-1020
Maize	20-50	1300-3000	**Yard long bean**	20-43	2000-3610

O-14

Transporters involved in perferential distribution of boron in rice

***Ji Feng Shao*[1,2], *Naoki Yamaji*[1], *Ren Fang Shen*[2] *and Jian Feng Ma*[1]**
[1]Institute of Plant Science and Resources, Okayama University, Chuo 2-20-1, Kurashiki, 710-0046, Japan
[2]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China
; maj@rib.okayama-u.ac.jp

INTRODUCTION

Boron (B) deficiency is one of the limiting factor of crop production on acid soil. Since B is highly required for the growth of meristem and reproductive organs for their active growth, preferential distribution of limited B to these tissues is very important for improving internal B-use efficiency in plants. Developing tissues have low transpiration, therefore, the contribution of transpiration-dependent distribution is very low and a system for the preferential distribution of B to the developing tissues is required.

Recently, nodes in gramineous plants play an important role in distribution of mineral elements (Yamaji and Ma, 2014). Many different transporters in the nodes are supposed to be required for the inter-vascular transfer of mineral elements. Our global expression analysis of rice node I revealed that a member of NIP (nodulin 26-like intrinsic protein) family, *OsNIP3;1* and *OsBOR1* showed higher expression. OsNIP3;1 was previously reported to be a plasma membrane–localized transporter for B (Hanaoka et al. 2014; Liu et al. 2015), while OsBOR1 functions as a B uptake transporter in the root (Nakagawa et al. 2007). However, their exact role in rice nodes is not clear. Here, we functionally characterized these two genes in terms of expression pattern and cellular localization of the nodes, response to different B concentrations and phenotypic analysis of knockout lines. We found that two transporters work cooperatively to preferentially deliver B to the developing tissues.

MATERIALS AND METHODS

Two T-DNA insertion mutant lines of *OsNIP3;1* (4A-01956 and 2A-00764) and their wild-type rice (WT1; cv. Dongjin, WT2; cv. Hwayoung) were used for phenotypic analysis and labelling experiment with ^{10}B in this study. For expression pattern experiments, RNA from different organs was extracted by using the RNeasy Mini Kit (Qiagen), and the expression was determined with Thunderbird SYBR qPCR mix on Master cycler real-time RT–PCR. For tissue specificity localization of OsNIP3;1 and OsBOR1, an immunohistological analysis was performed. The concentration ^{10}B was determined by ICP-MS with isotope mode after the samples were digested by HNO_3 (60%) in a plastic tube.

RESULTS AND DISCUSSION

Expression pattern of *OsNIP3;1* and *OsBOR1*

Both *OsNIP3;1* and *OsBOR1* showed high expression in the nodes at both the vegetative and reproductive growth stage although their expression was also found in other organs (Fig.1). The expression of *OsNIP3;1* in the nodes was up-regulated by B deficiency, but down-regulated by high B in 2 h (Shao et al., 2018). By contrast, the expression of *OsBOR1* was unaffected by excess B concentration

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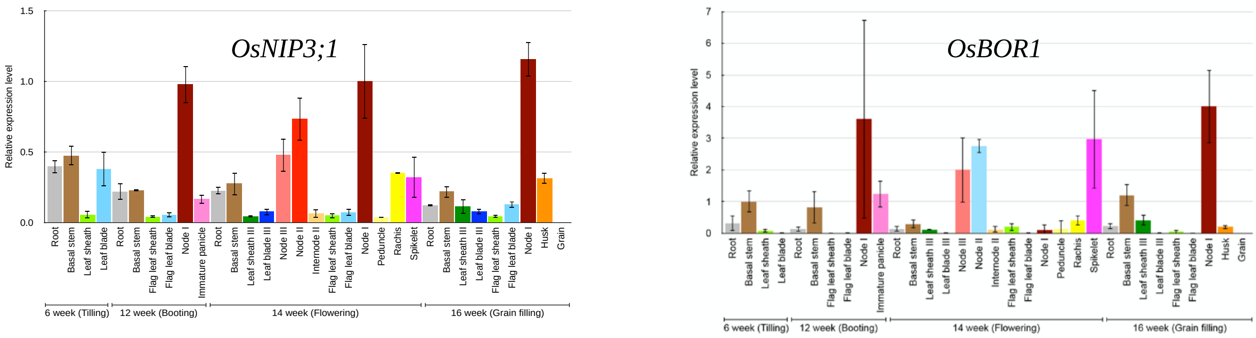
Fig. 1. Expression paterrn of *OsNIP3;1* and *OsBOR1* in different organs at different growth stage of rice grown in a field

Tissue specificity of localization of OsNIP3;1 and OsBOR1 protein

The tissue specificity localization of OsNIP3;1 and OsBOR1 were investigated



by immunohistological analysis. OsNIP3;1 was localized at the xylem parenchyma cells of enlarged vascular bundles of node I, while OsBOR1 was localized at the cell layers next to OsNIP3;1. Both proteins were rapidly degraded within a few hours in response to high B.

Role of *OsNIP3;1* in B distribution

To investigate the distribution of B, a ^{10}B-labeling experiment was conducted. Knockout of *OsNIP3;1* hardly affected B uptake, but altered B distribution; the mutant showed less distribution of B to the new leaves compared with WT, resulting in B-deficiency symptom in the new leaves under B-deficiency. Investigation of B distribution pattern in *OsBOR1* mutants is being undertaken.

CONCLUSION

These results indicate that OsNIP3;1 and OsBOR1 are co-operatively involved in preferential distribution of B to developing tissues in nodes of rice.

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REFERENCES

Hanaoka, H., Uraguchi, S., Takano, J., Tanaka, M. and Fujiwara, T. (2014). OsNIP3;1, a rice boric acid channel, regulates boron distribution and is essential for growth under boron-deficient conditions. Plant J. **78:** 890-902.

Liu, K., et al. (2015). Dwarf and tiller-enhancing 1 regulates growth and development by influencing boron uptake in boron limited conditions in rice. Plant Sci. **236:** 18-28.

Nakagawa, Y., Hanaoka, H., Kobayashi, M., Miyoshi, K., Miwa, K. and Fujiwara, T. (2007). Cell-type specificity of the expression of *OsBOR1*, a rice efflux boron transporter gene, is regulated in response to boron availability for efficient boron uptake and xylem loading. Plant Cell **19:** 2624-2635.

Shao J. F., Yamaji, N., Liu. X. W., Yokosho, K., Shen, R.F. and Ma, J. F. (2018). Preferential distribution of boron to developing tissues is mediated by intrinsic protein OsNIP3;1. Plant Physiol. **176:**1739-1750.

Yamaji, N. and Ma, J. F. (2014). The node, a hub for nutrient distribution in gramineous plants. Trends Plant Sci. **19:** 556-563.

O-15

Plant water status and root system response of NILs lines under combined aluminium toxicity and drought stress

Siecińska J.[1], and Nosalewicz A.[1]
[1]Institute of Agrophysics, Polish Academy of Science
Doświadczalna 4, 20-290 Lublin, Poland
; j.wrobel@ipan.lublin.pl

INTRODUCTION

Drought stress and aluminium toxicity are among the most common constraints on crop production worldwide. Although the primary interaction between the plants and these stresses takes place at the soil-root interface it also known, that the primary symptom of aluminium toxicity is the inhibition of root growth, while drought stress mainly affects shoot growth (Yang et al. 2013). Longer roots help drought exposed plants to uptake water from deeper soil. Aluminium inhibition of the root system may increase the susceptibility of agricultural plants to water shortages during long periods without rain.

The negative effects of soil acidity on plants may be alleviated by liming. One possible effective alternative to liming may be the cultivation of Al resistant plants. It is well known that liming can be used to improve plant yields, however, it is not known in which cases liming is cost effective because of the cost of agriculture practices, the degree of soil acidification or the species of cultivated plants. Despite the fact that excessive soil acidity and drought stress may act interactively on plant growth, most research to date has focused on the separate effects that each of the stressors has on both root and shoot growth and crop yield. It should be taken into account that two factors: the time of exposure to and the intensity of the simultaneously acting stressors both have important effects on plant response.

In the natural environment, plants growing in acid soil suffer from aluminium toxicity during the whole growth period. On the other hand, drought episodes of different durations may also affect plants several times over this period. Our previous study concerning aluminium concentration in soil solutions extracted from soil at various soil water potentials showed that a decrease in the soil water potential (increasing drought intensity) was accompanied by increasing aluminium concentration in the soil solution thereby enhancing the threat of aluminium toxicity to plants (Siecińska et al. 2016). The decrease in water uptake induced by the drought was accompanied by an increase in the aluminium concentration of the soil solution which may hinder the precise prediction of the impact of Al on plants in field conditions with variable soil water availability and an increasing frequency of drought events.

The aim of our study was to evaluate the effect of a combination of soil drought and aluminium toxicity on root system morphology and the resulting alterations in the water relations of near isogenic lines (NIL) of wheat.

MATERIALS AND METHODS

Two wheat genotypes (*Triticum aestivum* L.), Aluminium-tolerant ET8 and Aluminium-sensitive ES8 (Delhaize et al. 1993) were used as plant material. The soil material was a loamy sand textured soil with a native pH of 4.2, and after liming the soil pH rose to 6.5. Plants were grown in 10 cm in diameter and 40 cm in height soil columns filled with moderately compacted soil in controlled laboratory environment conditions. The unique system for maintaining and controlling soil moisture based on TDR probes and a precise flow meter (E-test, Poland) was used (Wilczek et al., 2013).

Soil moisture was maintained at an optimum level for the control treatment during the whole growth period.

RESULTS AND DISCUSSION

Generally there were no significant differences in the root biomass of the NILs grown in acid soil irrespective of water availability. However, in acid soil the roots of the ES8 line were thicker and shorter than the roots of the ET8 line. The stimulatory effect of drought stress on root length, but not root biomass was only noted in the ES8 line grown in limed soil.

Data obtained from the measurements of leaf transpiration at different soil water potentials were used to investigate the rate at which transpiration decreases with increasing drought intensity. Both of the NILs grown in limed soil were characterised by the similar rate at which leaf transpiration decreased with decreasing soil moisture. However, a large differentiation was observed in soil of pH 4.2, which could be caused by an increase in the aluminium concentration with decreasing levels of soil moisture (Siecińska et al., 2016). This hypothesis can be supported by the observation that a reduction in water uptake in drought treatments was accompanied by increased aluminium concentration in both NILs roots. This effect was more pronounced in ES8 than ET8. Total water uptake by plants exposed to combined drought stress and aluminium toxicity remains at a similar value between the two wheat lines. However, in this growth condition, ET8 absorbed water more effectively as observed similar water uptake by both lines was noted at much lower water potential under ET8 than ES8

CONCLUSION

Our findings provide insights into the plant water relations of wheat with different Al resistance to combined Al toxicity and drought stress and demonstrate that low soil moisture (soil drought) in an acid aluminium-toxic soil aggravates Al toxicity which decidedly restricts the plant root system and thereby has a negative effect on water and nutrient uptake by plants.

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REFERENCES

Delhaize E., Ryan PR., Randall P.J. 1993. Aluminium tolerance in wheat (Triticum aestivum L.). Plant Physiol **103:**695-702.

Siecińska J., Wiącek D., Nosalewicz A. 2016. The influence of soil acidity on aluminium and mineral nutrient concentrations in solutions from soil at different water potentials. Acta Agrophys **23(1):**97-104.

Wilczek A., Szypłowska A., Nosalewicz A., Skierucha W., Wilczek W. 2013 Project of an automatic system for soil moisture regulation using TDR technique. VI International Symposium "Farm Machinery and Processes Management in Sustainable Agriculture", Lublin, 20-22.11.2013 r. 197-200.

Yang Z.B., Rao I.M., Horst W.J. 2013. Interaction of aluminium and drought stress on root growth and crop yield on acid soils. Plant Soil 372Ł3-25.

O-16

Growth inhibition of rice (*Oryza sativa* L.) and wheat (*Triticum aestivum* L.) seedlings in Ga- and In-contaminated acidic soils is caused by Al toxicity

Syu, C.H.[1], J.Y. Su[2], L.Y. Chen[2], and D.Y. Lee[2]
[1]Division of Agricultural Chemistry, Taiwan Agricultural Research Institute, No.189, Zhongzheng Rd., Wufeng Dist., Taichung City 41362, Taiwan
[2]Department of Agricultural Chemistry, National Taiwan University, No. 1, Sec. 4, Roosevelt Rd., Taipei 10617, Taiwan
; dylee@ntu.edu.tw

INTRODUCTION

Gallium (Ga) and indium (In) compounds are being extensively utilized in semiconductor manufacturing and the electro-optical industry. Once industrial effluents containing Ga and In are discharged into irrigating systems, which may affect the growth and productivity of crops. Limited information exists on the effects of emerging contaminants Ga and In on crop growth. Therefore, this study investigated the effects on growth and uptake of Ga and In by rice and wheat plants grown in Ga and In-contaminated soils with different soil properties.

MATERIALS AND METHODS

Pot experiment was conducted and the rice (*Oryza sativa* L., cv Taikeng 9) and wheat (*Triticum aestivum* L., cv Taichung Sel. 2) seedlings were grown in two soils of different pH spiked with various Ga and In concentrations (50, 100, 200 and 400 mg kg^{-1}) for 50 days. The growth index and the concentrations of Ga, In and Al in roots and shoots of rice and wheat seedlings were measured after harvesting. The concentration of Ga and In in the plant tissues was determined by ICP-MS, and that of Al by ICP-OES.

RESULTS AND DISCUSSION

Table 1 shows the growth indices (root and shoot biomass) of rice and wheat seedlings grown in the tested soils under different Ga and In treatments. For acidic soils (Pc soils), the biomass of root and shoot were all decreased with Ga/In concentrations in soils, but there was no significant correlation between either Ga or In concentration and growth indices of rice and wheat seedlings grown in alkaline soils (Cf and Tk soils). Due to the high activity of soluble Al species (Al^{3+}) in acidic soils, it is possible that Al could be released into soil and then caused the Al toxicity to rice and wheat plants via the addition of Ga and In to acidic soils, which could be proved by the results of available-Al in acidic soils with different Ga/In treatments (Table 2). In addition, plant tissue analysis showed that the concentrations of Ga and In in roots were about one order of magnitude higher than in shoots (Fig. 1). This suggests that roots are a major sink of Ga and In accumulation in rice and wheat plants.

Table 1. The biomass of rice and wheat seedlings grown in tested soils with different Ga/In treatments

Ga/In treatment		Ga-CK	Ga-50	Ga-100	Ga-200	Ga-400	In-CK	In-50	In-100	In-200	In-400
Tested soils						mg plant^{-1}					
Rice seedling											
Pc soil	Root	281^{a}	164^{b}	94.2^{c}	42.8^{d}	30.0^{d}	281^{A}	298^{A}	183^{B}	74.2^{C}	33.8^{C}
	Shoot	636^{a}	426^{b}	240^{c}	101^{d}	78.3^{d}	636^{B}	770^{A}	558^{C}	222^{D}	92.5^{A}
Cf soil	Root	228^{ab}	223^{b}	248^{ab}	249^{ab}	276^{a}	228^{AB}	183^{B}	184^{B}	192^{B}	258^{A}
	Shoot	418^{a}	435^{a}	430^{a}	449^{a}	465^{a}	418^{A}	403^{A}	436^{A}	426^{A}	460^{A}
Wheat seedling											
Pc soil	Root	38.5^{b}	43.3^{a}	43.5^{a}	39.8^{b}	32.0^{c}	38.5^{B}	49.7^{A}	42.5^{AB}	36.4^{B}	27.4^{C}
	Shoot	151^{b}	171^{ab}	190^{a}	174^{ab}	102^{c}	151^{B}	166^{A}	151^{AB}	143^{B}	111^{C}
Tk soil	Root	306^{a}	264^{a}	217^{a}	251^{a}	268^{a}	306^{A}	226^{B}	243^{B}	237^{B}	246^{B}
	Shoot	1308^{a}	1369^{a}	1213^{a}	1188^{a}	1200^{a}	1308^{A}	1202^{A}	1231^{A}	1307^{A}	1227^{A}

The pH of Pc, Cf and Tk soils are 4.1, 7.4 and 7.6.

Different letters indicated the differences in the value among the Ga/In treatments based on the LSD test.

Table 2. The concentrations of $CaCl_2$ extractable-Al in tested soils with different Ga/In treatments

Ga/In treatment	Ga-CK	Ga-50	Ga-100	Ga-200	Ga-400	In-CK	In-50	In-100	In-200	In-400
Tested soils					mg kg^{-1}					
Rice-pot experiment										
Pc soil	50.8^{e}	74.9^{d}	81.6^{c}	94.8^{b}	133^{a}	50.8^{E}	70.6^{D}	74.9^{C}	80.9^{B}	96.7^{A}
Cf soil	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Wheat-pot experiment										
Pc soil	51.0^{e}	52.9^{d}	55.2^{c}	61.1^{b}	110.3^{a}	51.0^{D}	50.5^{D}	55.0^{C}	60.5^{B}	80.1^{A}
Tk soil	0.02^{a}	0.02^{a}	0.02^{a}	0.01^{a}	0.02^{a}	0.02^{A}	0.02^{A}	0.02^{A}	0.02^{A}	0.02^{A}

n.d.: not detection.

Different letters indicated the differences in the value among the Ga/In treatments based on the LSD test.

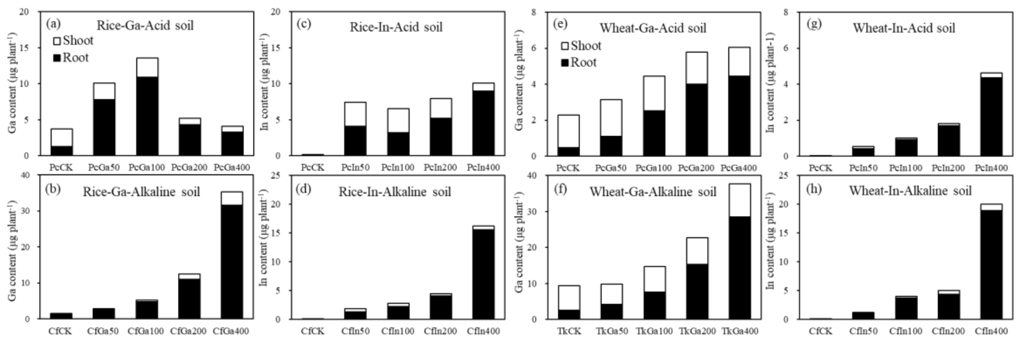

Fig. 1. The contents and distribution of Ga/In in rice and wheat seedling grown in tested soils with different Ga/In treatments

CONCLUSION

The growth inhibition of rice seedlings in Ga/In-spiked acidic soils is mainly due to Al toxicity resulting from enhanced Al release through competitive adsorption of Ga/In, rather than from Ga/In toxicity, while no such effect was found in alkaline soils due to the low availability of Ga, In and Al.

REFERENCES

Su, J.Y., C.H. Syu, and D.Y. Lee. 2018. Growth inhibition of rice (*Oryza sativa* L.) seedlings in Ga- and In-contaminated acidic soils is respectively caused by Al and Al + In toxicity. Journal of Hazardous Materials **344:**274-282.

O-17

Enzymatic responses of *Phaseolus vulgaris* l. Genotypes to Al-stress

***Tóth, B.*[1], *Z. Győri*[1], *P. Sipos*[1] *and M.A. Grusak*[2]**
[1]Institute of Nutrition, University of Debrecen, 1 Egyetem Square, 4032 Debrecen, Hungary
[2]Red River Valley Agricultural Research Center, 1605 Albrecht Boulevard N, Fargo, ND 58102-2765 USA
Present address: Institute of Plant Sciences, Faculty of Natural and Agricultural Sciences, University of the Free State, PO Box 339, Bloemfontein 9300, Republic of South Africa
; both@agr.unideb.hu

INTRODUCTION

Aluminum (Al) toxicity negatively affects the agriculture, the agronomy and the environment. Al limits the efficiency of crop production. Consequently, the financial income of the farmers decrease significantly. The World's population is growing fast, nowadays. Although human population has increased significantly the allotted areas for agriculture are decreasing. To produce a sufficient amount and quality of plant protein, is a huge challenge for growing populations, especially in developing countries. The common bean (*Phaseolus vulgaris* L.) is one of the most important legumes in Central America and in Africa.

The main objective of this study was to prove that there exists a strong genotype effect of Al toxicity and sensitivity in the examined five common bean genotypes from Pinto line. The aim was to determine which genotypes are more tolerant to Al stress. Overall, the research findings could contribute to creation of more stable agriculture production where acidic soils are implicated.

MATERIALS AND METHODS

Seeds of common bean genotypes AC Island, Croissant, Poncho, Santa Fe and Topaz from Pinto line were used. Plants were grown and treated with 0 or 20 μM $AlCl_3$ according to Rangel et al. (2005). Root length, the dry weight of shoots and roots were measured. The activity of superoxide dismutase was determined according to Misra and Fridovich (1972). Assay of peroxidise as proposed by Reddy et al. (1995) was used. Lipid peroxidation (LP) was evaluated according to the amount of MDA (Heath and Packer, 1968). To determine the amount of total ROS (reactive oxygen species), 2,7-dichlofluorescein diacetate was used. Protein was measured by the method of Bradford (1976).

RESULTS AND DISCUSSION

The main symptom of Al toxicity is rapid inhibition of root growth. At the end of the experiment, the dry weight of the shoot and roots were measured (results are not shown). Al stress does not have any negative effects on shoot growth at the early growth stage. However, the dry weight of root was significantly lower at all genotypes. The highest decrease was observed at Croissant genotype (59.62%). The effect of Al on root-elongation rate is best described as Al-induced inhibition of root-elongation (results are not shown). The genotypes were arbitrarily ranked for Al resistance in four categories based on the percentage of Al-induced inhibition of root elongation. Poncho genotype was classified as Al-hipersensitive (inhibition > 90 %). Croissant, Santa Fe and Topaz genotypes were classified as Al-sensitive (inhibition between 50 % and 90 %), and AC Island as intermediate (inhibition 30 – 50 %). This classification is based on 20 μ M Al treatment for up to three days, which is suggested by Rangel et al. (2005) with modification of the authors.

It is proved that exposure to Al could negative affect production of reactive oxygen species (ROS) in plants as Al stress causes peroxidation of lipids in the plasma membrane. This effect could be due to ROS and Al induces the expression of several genes encoding antioxidative enzymes such as superoxide dismutase (SOD). Although, the activity of SOD was significantly higher in all genotype. There was no connection between SOD and POX activity when exposed to Al stress. However, the POX activity significantly lower in Croissant and Topaz (Table 1). LP is displayed less sensitive to Al than the root elongation. Cakmak and Horst (1991) suggest that LP is the consequence rather than the primary cause of Al injury to plant roots.

Table 1: Effect of Al on enzymatic activity (SOD, POX, LP) in the roots of five common bean genotypes (AC Island, Croissant, Poncho, Santa Fe and Topaz) grown in a solution containing 0.5 mM $CaCl_2$, 0.5 mM KCl and 8 µM H_3BO_3 for 72 h at 20 µM Al, pH 4.5 (n=4) Significant difference compared to the 0 µM Al treatment: *p<0.05, **p<0.01, ***p<0.001

Genotypes	SOD($Ug^{-1}Fw$)		POX($\Delta A436\ g^{-1}\ FW\ min^{-1}$)		LP(nmol MDA g^{-1}FW)	
	0 µm Al	20 µm Al	0 µm Al	20 µm Al	0 µm Al	20 µm Al
ACIsland	0.14±0.01	0.16±0.01*	4.33±0.60	4.85±0.91	8.13±0.40	17.73±0.40
Croissant	0.15±0.01	0.17±0.00*	2.42±0.31	3.16±0.23**	23.75±10.37	14.86±5.67
Poncho	0.13±0.04	0.19±0.02*	6.23±1.16	6.52±1.81	5.31±1.03	4.14±0.52
Santa Fe	0.13±0.01	0.17±0.01*	4.98±0.34	5.63±0.25	9.94±3.17	8.67±2.57
Topaz	0.17±0.01	0.22±0.02*	2.80±0.36	3.56±0.28*	18.99±5.31	39.56±5.78***

(SOD = superoxide dismutase, POX = peroxidase, LP = lipid peroxidation, FW = fresh weight, MDA = malondialdehyde)

CONCLUSION

According to the measured parameters, a strong genotype effect was found regarding the bean plant's response to Al stress. In comparison with the four genotypes (Croissant, Poncho, Santa Fe and Topaz) AC Island genotype was less affected by the Al-toxicity. To explain these genotypic differences targeted molecular biology examinations are needed. These research findings could contribute to address the issue of food scarcity due to abiotic stress and, thus, provide food security to the malnourished population in developing countries.

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REFERENCES

Bradford, M. M. 1976. A rapid and sensitive method for the quantitation of microgram quantities to protein utilizing the principles of protein-dye binding. Anal Biochem **72**:248-254.

Cakmak I., and W. J. Horst. 1991. Effect of aluminum on lipid peroxidation, superoxide dismutase, catalase, and peroxidase activities in root tips of soybean (*Glycin max*). Physiologia Plantarum **83**: 463-468.

Heath, R. L., and L. Packer. 1968. Photoperoxidation in isolated chloroplasts. I. Kinetics and stoichiometry of fatty acid peroxidation – Arch Biochem Biophys **125**:189-198.

Misra, H. P., and I. Fridovich. 1972. The role of superoxide anion in the antioxidation of epinephrine and a simple assay for superoxide dismutase. J. Biol Chem **247**: 3170-3175.

Rangel, A. F., M. Mobin, I. M. Rao, and W. J. Horst. 2005. Proton toxicity interferes with the screening of common bean (*Phaseolus vulgaris* L.) genotypes for aluminum resistance in nutrient solution. J. Plant Nutr. Soil Sci **168**: 607-616.

Reddy, K. P., S. M. Subhani, P. A. Khan, and K. B. Kumar. 1995. Effect of light and benzyl adenine and dark-treated graving rice (*Oryza sativa*) leaves-changes in peroxidases activity. Plant Cell Physiol **26**: 987-994.

O-18

Managing and achieving high oil palm yields on low pH soils of acid sulfate and peat in Malaysia

U.T. Pupathy[1] ***and Andrew Cheng M. F[1]***
[1]Sime Darby Plantation Sdn Bhd, Regional Office-Sarawak Zone, KM 50, Bintulu-Miri Road, Rajawali Complex, P.O.Box 673, 97000, Bintulu, Sarawak, Malaysia
; pupathy.uthrapathy@simedarbyplantation.com
; andrew.cheng.muifah@simedarbyplantation.com

INTRODUCTION

Generally soils of acid sulfate and tropical peat are considered as 'problem soils', attributed to their low-pH values. An updated data revealed that about 0.5 million ha of acid sulfate soils occur in Malaysia (Rosilawati *et al.,* 2012). These acid sulfate soils have conditions that are unfavorable for agriculture as acid sulfate soils are severely affected by the presence of excess sulfates (Poon and Bloomfield, 1977). Meanwhile, there are also more than 2 million hectares classified as lowland peat in Malaysia, mostly in Sarawak State. The agricultural potential of peat soil is usually restricted by unfavorable characteristics of peat such as low bulk density, low pH value and lack of micronutrient availability (Pupathy, *et al.*, 2017). In addition to these, depth of organic layer, extent of potential subsidence, degree of humification and mineralisation are limiting factors for any agricultural cultivation on peat. As such being low pH soils, acid sulfate soils and peat possesses adverse effects on the agricultural activities, especially for oil palm *(Elaeis guineensis)* cultivation. However through best agro-management practices, oil palm generally can grow well on low pH soils with sustainable yields and vegetative growth. In this review paper, the important factors of soil, plant and water management and their interactions are discussed to ensure the sustainability productivity of oil palm on acid sulfate as well as on peat soils.

METHODOLOGY

Fresh fruit bunch (FFB) yields that were recorded in both research and commercial scales on various acid sulfate and peat soils were gathered for comparison. Solutions learnt through research and empirical knowledge to elevate oil palm yields were highlighted.

RESULTS AND DISCUSSION

FFB yield realised in Sime Darby estates on deep peat in Sarawak was slightly high/comparable (mean of 17 year-yield:16.62 mt/ha/yr) to yields from UPB (mean of 12 year-yield:11.78 mt/ha/yr, Gurmit, 1999), MPOB (mean of 15 year-yield:11.93 mt/ha/yr, Mohd Tayeb *et al.*, 2002), and Department of Agriculture, Sarawak (mean of 11year-yield:15.16 mt/ha/yr, (Jaman and Kueh, 1996)). Maximum yields recorded for acid sulfate soils were generally higher as compared to those obtained generally on peat (**Table 1**). This is due to more compounded factors involved for oil palm cultivation on peat such as low bulk density, palm leaning and termite attacks. Minimising leaching of nutrients, especially K fertilizers, maintaining water level at 50 to 75 cm from peat surface, detecting and treating termite are among the key aspects to achieve high yield in peat planting (Gurmit, 1999; Pupathy and Chang, 2003;Pupathy, *et a.l,* 2017). Al toxicity and excess sulfates are the major constraints to FFB production in oil palm. By maintaining water-table at 45 to 60 cm, accelerated pyrite-oxidation was avoided to obtain maximum oil palm yields (Hew and Khoo, 1970; Pupathy and Paramananthan, 2014).

Table 1: Summary on Maximum Fresh Fruit Bunch (FFB) for Acid Sulfate and Peat Soils

Soil Type/Series	Maximum FFB Yield (mt/ha/ year)	Year of harvesting at	Remarks
Acid Sulfate soils Jawa	33.57	10th	-High yield by controlling water table at 45 -60 cm below soil surface. Hew and Khoo (1970) found that liming was ineffective to control acidity in acid sulfate soils. Source: Pupathy U.T. & Paramananthan S, 2014. Agro-management for Oil Palms Planted on Acid Sulfate Soils, In: Selected Papers on Soil Science: Volume 1: Problem Soils, Agricultural Crop Trust.
Linau	32.11	11th	
Jawa/Sedu/Tongkang	31.29	10th	
Sedu/Briah	38.53	7th	
Peat (Peninsular) Shallow (< 1m)	23.7	10th	-Water management and micronutrient inputs are key factors for maximising yields. Source: Mohd Tayeb D, 2002. Oil Palm Planting on Peat-Progress and Future Direction, MPOB, In:R&D for competitive edge in the Malaysian oil palm industry.
Moderate (1-2m)	20.2	9th	
Deep (> 2m)	17.3	4th	
Deep	17.1	3rd	-Source: Gurmit Singh, 1999, Agronomic Management of Peat Soil for Sustainable oil Palm Production, United Plantations' Experience, ISP Seminar on Peat, Kingtown,Sibu, Sarawak.
Peat (Sarawak) Peat	32.21	6th	-Source: Sharifah Sharul R.S.A., Sathashinikisan K, Tan Choon C., Noor H.H and Tan J.S, Oil Palm Planting Material for Peat: Performance and Challenges Factors Impacting the Competitiveness of the Palm Oil Industry, Planter,Vol.93, No 1092, March 2017.
Anderson (>2m)	19.63	10th	-Source: Jaman. O. and Kueh H.S. 1996. Oil Palm Research and Development on Peat Soil in Sarawak, Agricultural Department, Sarawak, Sibu, Sarawak.
Moderate Peat (1-2m)	29.03	7th	-Source: Musa B, Ahmad Fairuz Z, Mohaimi M and Harikrishna K, 2016, Planting Materials: Performance in Sarawak- Sime Darby's Experience, Planter, Vol.92, No. 1089, December 2016.
Shallow-Deep (>0.5m)	21.35	10th	-Commercial scale data; no liming was carried out. Source: Pupathy U.T. *et al*, 2017.

CONCLUSION

With proper water and fertilizer management, FFB yields above 30-35 mt/ha are achievable on low-pH soils of acid sulfate and peat and these yields are comparable to those FFB on some best soil types.

REFERENCE

Poon, Y.C. and Bloomfield, C. 1977. The amelioration of acid sulfate soil with respect to oil palm.Trop.Agric. (Trinidad), **54 (4):** 289-305.

Pupathy U.T., Andrew Cheng M. F, Kumaran R and Abdul Razak Ijang.2017. Planters' Experience on Managing Challenges and Limiting Factors for High Oil Palm Yield on Peat. International Soil Science Conference, MSSS-UPM, Bintulu, Sarawak, Malaysia.

Rosilawati A.K., Shamsuddin J. and Fauziah C.I. 2012. Effects of applying various lime sources on pH and exchangeable Al of an acid sulfate soil under submerged condition. Soil Science Conference of Malaysia, Soils 2012, Kota Bharu, Kelantan, Malaysia, pg 468-474.

O-19

Using liquid lime to improve growth of oil palm seedlings on a tropical peat soil

Ahmed, O.H. [1,2,3], Ali, M.[1], and Audrey, A.[1]
[1]Faculty of Agriculture and Food Sciences, UPM Bintulu Campus,
[2]Borneo Eco-Science Research Center, Faculty of Agriculture and Food Sciences, UPM Bintulu Sarawak Campus, 97008 Bintulu, Sarawak, Malaysia
[3]Institute of Tropical Agriculture and Food Security, UPM, 43400 Serdang, Selangor, Malaysia
; osumanuharuna@gmail.com

INTRODUCTION

Tropical peats have low pH and base saturation (Chan, 2003). To cultivate such soils with oil palm seedlings requires liming. Calcium carbonate is widely used as a liming material in oil palm cultivation although the amount of lime needed for oil palm cultivation is usually high. Moreover, $CaCO_3$ lime reaction with acid soils is very slow and because of this, it takes long time to increase soil pH (Osman, 2013). Liquid lime is a high-quality liquid flowable and white liming material that reacts faster than ordinary lime powder without burning plant roots. The amount of liquid lime needed is lower but reacts faster to increase soil pH than any other powdered lime. Liquid lime reduces soil pH, boosts soil microbial activities, and nutrients availability (Hart et al. 2013). Additionally, it protects crops root system from damage due to increase in temperature during heavy liming. Moreover, it reacts very fast with soils thereby making more suitable and cost-effective liming material, especially for tropical peat soils. However, there is limited information on the use of liquid lime on tropical peat soils. The objectives of this study were to determine the effects of: (i) Liquid lime on selected chemical properties and nutrient uptake of oil palm seedlings on a tropical peat soil and (ii) Liquid lime on growth of oil palm seedlings on a tropical peat soil.

MATERIALS METHODE

A peat soil classified as Saprist peat soil was used in incubation and greenhouse experiments for 90 days, after which stand procedures were used to determine selected soil chemical properties. Details about the treatments are presented in Table 1. Analysis of variance was used to test treatment effects whereas treatments means were compared using Tukey's Test.

Table 1: Treatments evaluated in the pot study to determine their effectiveness on oil palm seedlings

Treatments	Peat soil	Calcium carbonate	Liquid lime		N	P_2O_5	K_2O	MgO
			Application on 1st and 15th days after transplanting		17.9	17.9	32.1	7.8
			First	Second	Urea	TSP	muriate of potash	Kieserate
	……….. $g\ pot^{-1}$………		..……. $mL\ pot^{-1}$..……..		……………..….. $g\ pot^{-1}$…………..…..			
T1	5	-	-	-	38.9	38.9	53.5	28.89
T2	5	10	-	-	38.9	38.9	53.5	28.89
T3	5	-	13	0	19.46	19.46	26.75	14.44
T4	5	-	26	0	19.46	19.46	26.75	14.44
T5	5	-	13	13	19.46	19.46	26.75	14.44

Note: The fertilizer rate is those recommended for oil palm seedling from 3 months to 6 months (Rankine and Fairhurst, 1998) and that only half of those fertilizers were applied to peat soils treated with reactive lime.

RESULTS AND DISCUSSION

In the incubation study, the Reaktif-TE significantly improved soil pH due to its high Ca and K. pH of T3, T4, and T5 (liquid lime) over 20 weeks were significantly high than that of T2 ($CaCO_3$) because of the rapid reaction of the Ca and K of the liquid lime with the peat soil. Further evidence of the effectiveness of the liquid lime is the significant improvement soil Ca, Mg, and K (Table 2) at the end of growing the oil palm seedlings. The other ripple effect of the liquid lime following increase in soil pH is the significant increase in soil nitrate, ammonium, and available P (Table 3) partly due to mineralization as the soil pH became conducive for the soil microbes to thrive. Another evidence of the effectiveness of the liquid lime is the significant increase in the N, P, K, Ca, and Mg concentrations of the oil palm seedlings' tissues (Table 3).

Table 2. Lime treatments on the chemical properties of a tropical peat soil at twenty weeks after planting oil palm seedlings

Treatment	T1	T2	T3	T4	T5
pH_{KCL}	2.85c ± 0.04	3.15b ± 0.05	3.27b ± 0.07	3.55a ± 0.04	3.54a ± 0.04
pH_W	3.50de ± 0.04	3.68dc ± 0.05	3.78bc ± 0.06	4.05a ± 0.01	3.89ba ± 0.02
	……… mg kg^{-1} ………				
Available NO^{3-}	11.58d ± 0.58	15.38c ± 0.43	21.93a ± 0.34	18.31b ± 0.56	23.69a ± 0.52
Exch. NH_4^+	100.64a ± 3.91	82.62b ± 1.68	107.07a ± 2.22	93.18ba ± 3.02	65.81c ± 4.45
Available P	38.13a ± 0.48	18.67d ± 0.87	29.73b ± 0.82	28.44cb ± 0.89	24.64c ± 0.86
	……… cmol kg^{-1} ………				
Available K^+	4.12a ± 0.08	1.34e ± 0.04	1.95d ± 0.03	2.40c ± 0.09	3.52b ± 0.14
Available Ca^{2+}	6.53c ± 0.09	4.85d ± 0.05	13.40a ± 0.40	10.65b ± 0.41	13.66a ± 0.42
Available Mg^{2+}	1.34a ± 0.04	0.55e ± 0.04	0.91dc ± 0.04	0.74d ± 0.03	1.03bc ± 0.04

Note: different letters within a row indicate significant difference between means of 3 replicates ± standard error using Tukey's test at $P \leq 0.05$.

Table 3. Lime treatments on fresh weight and leaf nutrients concentration of oil palm seedlings at twenty weeks after planting

Treatment	T1	T2	T3	T4	T5
	……… g $plant^{-1}$ ………				
Total dry weight	102.32c ± 1.42	126.85a ± 1.09	116.01b ± 1.12	117.98b ± 1.22	116.97b ± 0.75
Total N (%)	4.50d ± 0.27	5.55c ± 0.04	6.243b ± 0.08	6.80a ± 0.09	5.74c ± 0.05
Total P (mg kg^{-1})	1158d ± 41.38	1461c ± 34.28	1745b ± 60.23	1927a ± 43.97	2042a ± 42.81
	……… mg kg^{-1} ………				
Total K^+	8.12e ± 0.13	8.88d ± 0.10	12.27c ± 0.12	13.04b ± 0.12	13.65a ± 0.07
Total Ca^{2+}	5.12e ± 0.08	6.76d ± 0.13	7.69c ± 0.13	9.33b ± 0.10	10.51a ± 0.09
Total Mg^{2+}	1.66e ± 0.05	1.92d ± 0.03	2.23c ± 0.04	2.54b ± 0.04	2.77a ± 0.03

Note: different letters within a row indicate significant difference between means of 3 replicates ± standard error using Tukey's test at $P \leq 0.05$.

CONCLUSION

The chemical properties of reactive lime is significantly higher than that of $CaCO_3$. Additionally, the reactivity and reduction of peat soil acidity by the liquid lime is higher and faster as compared with $CaCO_3$. The liquid lime furthermore increased the availability of soil nutrients as compared with $CaCO_3$ thereby enhancing reduction in the use of chemical fertilizer by 50% as compared with $CaCO_3$. Moreover, it improved oil palm seedlings' nutrient uptake and growth. The suitable rate of the liquid lime on peat soil in growing oil palm seedling is 26 mL per seedling.

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REFERENCES

Chan, W.H. 2003. Characteristics of Peat and Its Nutritional Requirements for Oil Palm Cultivation by *Arif Sugandi*.

Fairhurst, T.H., and Mutert, E. 1999. "Introduction to Oil Palm Production," *Better Crops InternationalVol 13, No. 1.*

Hart, J. M., Sullivan, D. M., Christensen, N. W., Horneck, D. A., & Pirelli, G. J. 2013. Applying Lime to Raise Soil pH for Crop Production (Western Oregon). *Oregon State University, Extension Service*.

Osman, K.T. 2013. Chemical Properties of Soil. In Soils (Pp. 97-111). Problem Soils and Their Management. *In Soils* (Pp. 161-174). Springer Netherlands.

Peech, H.M. 1965. Hydrogen-ion Activity. In *Method of Soil Analysis, Part 2,* ed. C. A. Black, D. D. Evan, L. E. Ensminger, J. L. White, F.E. Clark, and R.C. Dinaue, pp 914 – 926. Ithaca: American society of Agronomy, Madison, Wisconsin.

O-20

Effect of soil pH on basal stem rot disease incidence in *Ganoderma* inoculated oil palm seedlings

Abdulla Rahman Khunaw[1], Radziah Othman[1,3], Nusaibah Syed Ali[2], Mohamed Hanafi Musa[1] ,
[1]Department of Land Management,
[2]Department of Plant Protection, Faculty of Agriculture,
[3]Institute of tropical Agriculture and Food Security, Universiti Putra Malaysia, 43400 Serdang, Selangor
; radziah@upm.edu.my

INTRODUCTION

The incidence of Basal Stem Rot (BSR) disease caused by *Ganoderma. boninense* in oil palm seedlings has been an issue that continues to devastate the oil palm industry in the tropics. Soil pH has been implicated in the development of BSR in oil palm. Most soils in Malaysia have low pH ranges from 3 to 5 and previous studies have shown that higher BSR incidence was observed in low pH soil of Selangor doil series and peat, while slightly lower BSR incidence in high pH of Kranji and Briah soil series (Parthiban et al., 2016). In a different situation, low pH has been observed to suppress the incidence of BSR disease in oil palm. Soil pH is known to influence plant nutrients availability, microbial activity in the soil, and plant health (Sewards, 2014). Lack of adequate nutrients increases the plant's susceptibility to disease, which can lead to higher disease incidence, increased disease severity and eventually plant death. Basal Stem Rot incidence and severity in oil palm have been shown to be associated with availability of plant nutrients in soil (Roslan and Idris, 2012).

MATERIALS AND METHODS

Soil samples were collected from BSR-affected oil palm area in Macap, Malacca of the United Malacca Berhad. The *G. boninense* UPM13 cultures were maintained on potato dextrose agar (PDA) and incubated at room temperature (27 ± 1 °C) for seven to ten days. A total of 30 rubber wood blocks ($6.0 \times 6.0 \times 12.0$ cm^3) were prepared according to Khairuddin (1991). Inoculum preparations were made by placing ten plugs sized 6 mm from eight-day-old mycelium cultures grown on PDA using a core-borer onto each surface of the autoclaved rubber wood block (RWB). The block were then incubated for 10-12 weeks. Soil pH was adjusted to pH 5, 6 and 7 using calcium carbonate at the rate of 1, 2.5 and 5 t/ha. *Ganoderma* inoculation was done by placing three month old oil palm seedlings on top of the fully *Ganoderma* colonized RWB and all seedlings were grown for four months. Plant top and root biomass yield, nutrient updates, disease incidence and severity were determined.

RESULTS AND DISCUSSION

The root and shoot biomass of plants were significantly affected by *Ganoderma* infection. Lower root and shoot growth was observed in plants infected with *Ganoderma* compared to plants without *Ganoderma* infection. Adjusting the soil pH with calcium carbonate improved plant growth. Soil pH more than 5 improved root and shoot growthof plants compared to that in original soil pH 4.5. In *Ganoderma* infected plants, roots and shoot improved at pH 5 and above. However, shoots declined at pH 6 and 7.

Disease incidence (DI) and disease severity (DS) of *Ganoderma* infected plants were significantly affected by soil pH. Higher disease incidence was observed in plant grown at pH 4.5 compared to soil adjusted to higher pH. Lower DI was observed in plant grown at pH 5.0. Disease severity at original soil pH at 12 weeks of growth was higher than at higher pH, but after 16 weeks of growth the DS increased at higher soil pH. It seems that increasing soil pH did not help to reduce the disease severity. The reduced root development due to *Ganoderma* infection disrupted the nutrient adsorption resulting in unhealthy plants which are more susceptible to disease infection. The results agree with the previous studies on a fungus (Gurmit, 1991) that disease severity or development might be ascribed to environmental conditions, especially pH levels.

The uptake of nutrients (N, P, K, Ca and Mg) was also significantly affected by *Ganoderma* infection. In general nutrient uptake was higher in non-infected compared to infected plants. At pH 6, higher N, P and Ca uptake was observed in non-infected plants compared to *Ganoderma* infected plants. Infected plants had lower uptake of nutrients due to poor root growth. Generally healthy plants have better nutrient uptake than plants under stress condition (Rosenani et al., 2016).

CONCLUSION

The results showed that oil palm growth, disease incidence and disease severity were significantly affected by *Ganoderma* infection. Adjusting soil pH to higher than 5 improved growth of *Ganoderma* infected plants. Basal Stem Rot disease incidence could be reduced by modifying the soil pH with application of calcium carbonate.

REFERENCES

Gurmit, S. (1991). *Ganoderma*-the scourge of oil palm in the coastal area. In *Proceedings of Ganoderma workshop, Bangi, Selangor, Malaysia, 11 September 1990.* (pp. 7-35). Palm Oil Research Institute of Malaysia.

Khairuddin, H. (1991). Pathogenicity of three *Ganoderma* species on oil palm seedlings. Journal Perak Planter's Association, 43-49.

Parthiban, K., Vanitah, R., Jusoff, K., Nordiana, A.A., Anuar, A.R., Wahid, O., & Hamdan, A.B. (2016). GIS Mapping of Basal Stem Rot Disease in Relation to Soil Series Among Oil Palm Smallholders. American Journal of Agricultural and Biological Sciences, **11 (1),** 2.12.

Rosenani, A. B., Rovica, R., Cheah, P. M., & Lim, C. T. (2016). Growth Performance and Nutrient Uptake of Oil Palm Seedling in Prenursery Stage as Influenced by Oil Palm Waste Compost in Growing Media. International Journal of Agronomy, 2016.

Roslan, A., & Idris, A. S. (2012). Economic impact of *Ganoderma* incidence on Malaysian oil palm plantation–a case study in Johor. Oil Palm Industry Economic Journal, 12(1), 24-30.

Sewards, J. (2014). Soil pH and its Effect on nutrient availability, soil microbial activity and plant health. UF/IFAS Extension, Volusia County, Florida.

O-21

An Al-inducible transcription factor, ART2 is involved in Al tolerance in rice

***Jing Che*[1], *Tomokazu Tsutsui*[1], *Kengo Yokosho*[1], *Naoki Yamaji*[1] *and Jian Feng Ma*[1]**

[1]Institute of Plant Science and Resources, Okayama University, Kurashiki 710-0046, Japan

; wonderful1122@gmail.com

INTRODUCTION

Rice (*Oryza sativa*) is more tolerant to Al toxicity compared with other cereal crops such as maize, wheat, barley, and sorghum, although there is also a genotypic difference between japonica and indica cultivars. Through mutant approach, a transcription factor, ART1 (Al resistance transcription factor 1) was found to be involved in high Al resistance in rice. ART1 is a Cys2His2-type zinc finger transcript factor, regulating more than 30 genes (Yamaji *et al.*, 2009). In rice genome, there are five homologs of ART1. However, these homologs have not been functionally characterized. In the present study, we characterized one of them, ART2 (Os04g0165200) in terms of tissue and spatial expression, subcellular localization, transcriptional activation activity, phenotypic analysis of the knockout lines.

MATERIALS AND METHODS

To investigate the subcellular localization of ART2, a translational ART2-GFP fusion was transiently introduced into tobacco (*Nicotiana tabacum* cv. Petit Havana SR1) protoplasts using the polyethylene glycol (PEG) method. The yeast one-hybrid assay was performed using MATCHMAKER GAL4 Two-Hybrid System 3 (Clontech) and MATCHMAKER One-Hybrid Library Construction and Screening Kit (Clontech) to examine the transcriptional activation potential of ART2 (Yamaji *et al.*, 2009). The expression of *ART2* in response to different Al concentrations (0-100 μM) for different times (0-72 h) or to other metals including 30 μM Cd and 10 μM La or to different pH (4.5 and 5.6) was investigated by RT-PCR. Knockout lines of *ART2* were generated by CRISP/Cas9 technique. Al tolerance was compared between wild-type (WT), *art1* mutant (Nipponbare background), and *art2* mutant (T3) by measuring the root elongation inhibition during 24 h at different Al concentrations, including 0, 10, 30, and 50 μM Al. The effect of pH on root elongation was investigated at pH ranging from 3.5 to 6.0. RNA-sequencing was performed to identify the candidate genes regulated by ART2.

RESULTS AND DISCUSSION

The full length of *ART2* is composed of 1126 bp, encoding a protein with 371 amino acids. ART2 shares 35%, 33%, and 39% identity, respectively, with ART1 in rice, STOP1, and STOP2 in Arabidopsis. Similar to these proteins, ART2 also contained the conserved C2H2 zinc finger domain. Subcellular localization observation showed that the GFP signal from protoplast expressing ART2-GFP was localized to the nucleus (Fig. 1a-c).

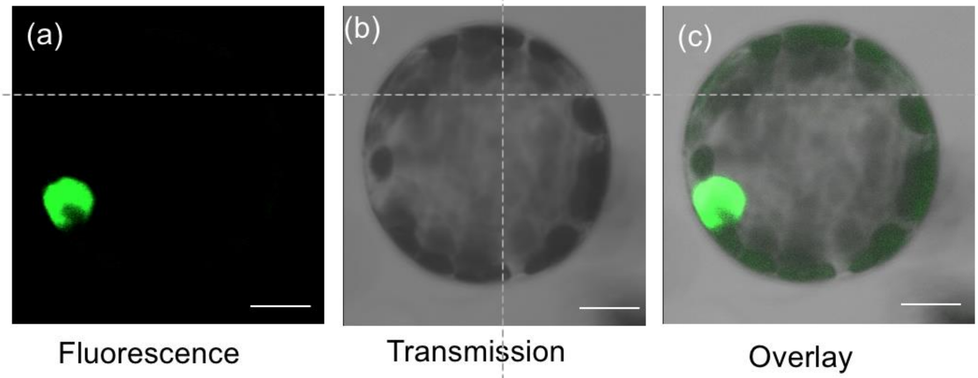

Fig. 1 Subcellular localization of ART2. ART2 fused with green fluorescent protein (GFP) at the C-terminus was transiently expressed in the tobacco protoplast. GFP signal (a), bright field image (b) and a merged image (c) are shown. Scale bars=10 μm.

Yeast one-hybrid analysis showed that that ART2 has a transcriptional activation potential in yeast, indicating that ART2 functions as a transcription factor. The Al-induced expression of *ART2* was not observed in the *art1* mutant, indicating that the expression of *ART2* was regulated by ART1. Compared with *ART1*, the expression of *ART2* in the roots was much lower. A time-dependent analysis showed that the expression of *ART2* was induced by Al after exposing to Al for 6 h. In the roots, the expression of *ART2* was affected neither by low pH nor by other metals including Cd and La. Therefore, different from constitutive expression of *ART1* (Yamaji *et al.*, 2009), the expression of *ART2* in the roots was specifically induced by Al.

The Al tolerance was compared between the wild-type rice, *art1* mutant, and *art2* mutants. In the absence of Al, the root elongation in the mutants was similar to the wild-type rice. However, the root elongation was inhibited by Al more in the *art2* mutants than in the wild-type. Knockout of *ART1* increased Al sensitivity more than that of *ART2*. The root elongation was similar between *art2* mutants and WT at each pH treatment, including pH 4.0, 4.5, 5.0, 5.5, and 6.0. These results indicate that knockout of *ART2* resulted in increased sensitivity to Al toxicity, but did not alter sensitivity to different pHs. Since identification of transcription factor for Al tolerance in Arabidopsis (STOP1) and in rice (ART1) (Iuchi *et al.*, 2007; Yamaji *et al.*, 2009), a number of their homologs (ART1/STOP1-like proteins) have been reported in different plant species. However, they play different roles in Al and proton tolerance. For example, STOP1 in Arabidopsis and NtSTOP1 in tobacco regulates both Al tolerance and proton-toxicity tolerance (Ohyama *et al.*, 2013), while VnSTOP1 in rice bean and STOP2 in Arabidopsis only regulate proton tolerance (Kobayashi *et al.*, 2014; Fan *et al.*, 2015).

To identify the candidate genes regulated by ART2, we extracted genes upregulated by Al by more than three folds in the wild-type rice, but less than two folds in the *art2* mutant. As a result, a total of 5 genes were selected as candidate genes regulated by ART2. They are supposed to be involved in cell wall maintenance, membrane protein, detoxification and stress response, and metabolism.

CONCLUSION

In conclusion, ART2 is an Al-inducible transcription factor localized in the nucleus in rice. It is involved in Al tolerance but not proton tolerance although its contribution to Al tolerance is smaller than ART1. ART2 regulates five genes implicated in Al tolerance, which may play a supplementary role in Al tolerance in rice.

ACKNOWLEDGMENTS

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REFERENCES

Fan W, Lou HQ, Gong YL, Liu MY, Cao MJ, Liu Y, Yang JL, Zheng SJ. 2015. Characterization of an inducible C2H2-type zinc finger transcription factor VuSTOP1 in rice bean (*Vigna umbellata*) reveals differential regulation between low pH and aluminum tolerance mechanisms. *New Phytologist* **208**: 456-468.

Iuchi S, Koyama H, Iuchi A, Kobayashi Y, Kitabayashi S, Kobayashi Y, Ikka T, Hirayama T, Shinozaki K, Kobayashi M. 2007. Zinc finger protein STOP1 is critical for proton tolerance in Arabidopsis and coregulates a key gene in aluminum tolerance. *Proceedings of the National Academy of Sciences, USA* **104**: 9900-9905.

Kobayashi Y, Ohyama Y, Kobayashi Y, Ito H, Iuchi S, Fujita M, Zhao CR, Tanveer T, Ganesan M, Kobayashi M, et al. 2014. STOP2 activates transcription of several genes for Al- and low pH-tolerance that are regulated by STOP1 in *Arabidopsis*. *Molecular Plant* **7**: 311-322.

Ohyama Y, Ito H, Kobayashi Y, Ikka T, Morita A, Kobayashi M, Imaizumi R, Aoki T, Komatsu K, Sakata Y, et al. 2013. Characterization of AtSTOP1 orthologous genes in tobacco and other plant species. *Plant Physiology* **162**: 1937-1946**.**

Yamaji N, Huang CF, Nagao S, Yano M, Sato Y, Nagamura Y, Ma JF. 2009. A zinc finger transcription factor ART1 regulates multiple genes implicated in aluminum tolerance in rice. *Plant Cell* **21**: 3339-3349.

O-22

Mechanisms of B in alleviating al toxicity in promoting apoplast alkalinization in root transition zone

***Xuewen Li*[1], *Yalin Li*[1], *Jingwen Mai*[1], *Mei Qu*[1], *Jiayou Liu*[1], *Yingmng Feng*[1], *Hongdong Xiao*[1], *Min Yu*[1]**
[1]Department of Horticulture, Foshan University, Foshan 528000 Guangdong, China
; yumin@fosu.edu.cn

INTRODUCTION

Boron (B) supply alleviates aluminum (Al) toxicity, but the mechanisms remain elusive.It is well-known that the activity of Al^{3+} is directly related to the apoplastic pH. Our research disclose the machanims in apoplast alkalization (PAT) in root transiton zone regualted by PM-H+-ATPase.

MATERIALS AND METHODS

Pea (*Pisum sativum* L. Cv Zhongwan no. 5) seeds were germinated and cultured in 1/4 modified Hoagland solution accoriding to Yu et al. (2006, 2009). Boron and Al treatments were applied at 0, 15 or 30μM $AlCl_3$ and 0 or 25 μM H_3BO_3 (0.5 mM $CaCl_2$, pH 4.0) for 1, 3, 12, or 24 h depending on the experimental design. Root length was determined using WinRhizo-Pro software. Lateral roots of the seedlings were stained by Haematoxylin and morin and observed under (fluorescent) microscope directly. Aluminium concentration was determined by ICP-AES. Non-invasive Micro-Test technique system was adopted to measure rhizosphere pH and IAA flux. BCECF was used to detect apoplast pH under CLSM.

RESULTS AND DISCUSSION

1. Aluminum accumulation in the root transition zone is decreased by B supply

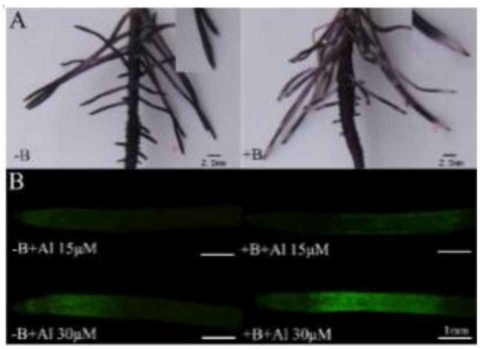

Fig. 1. (A) Haematoxylin staining,(B) Morin staining

2. B is essential for maintaining the alkalization of root surface in transition zone

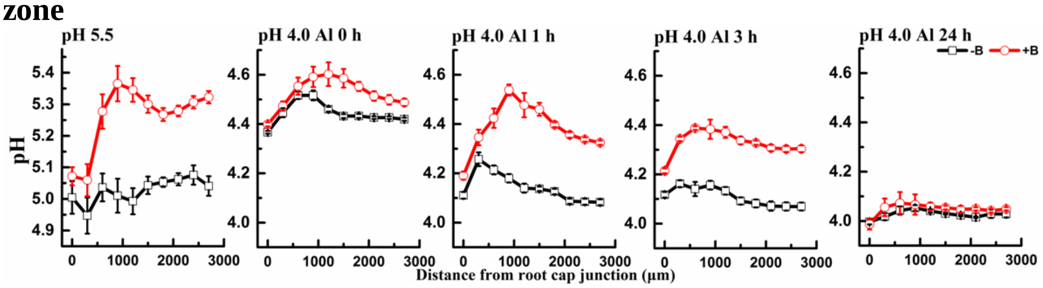

Fig. 2 Surface pH profile along pea root tips.

3. B is essential for maintaining apoplast alkalization in root transition zone

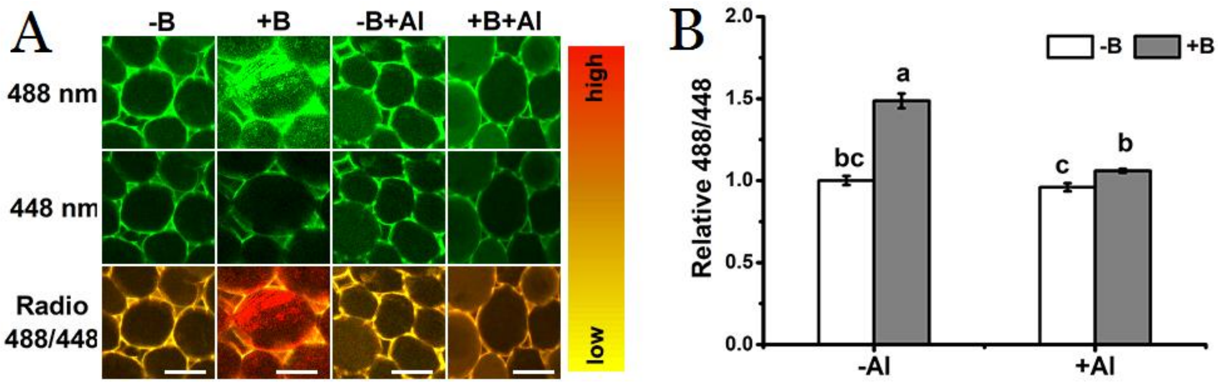

Fig. 3 Apoplast pH in root transition zone of pea.

4. Apoplast alkalization in root transition zone is regulated by PM-H+-ATPase

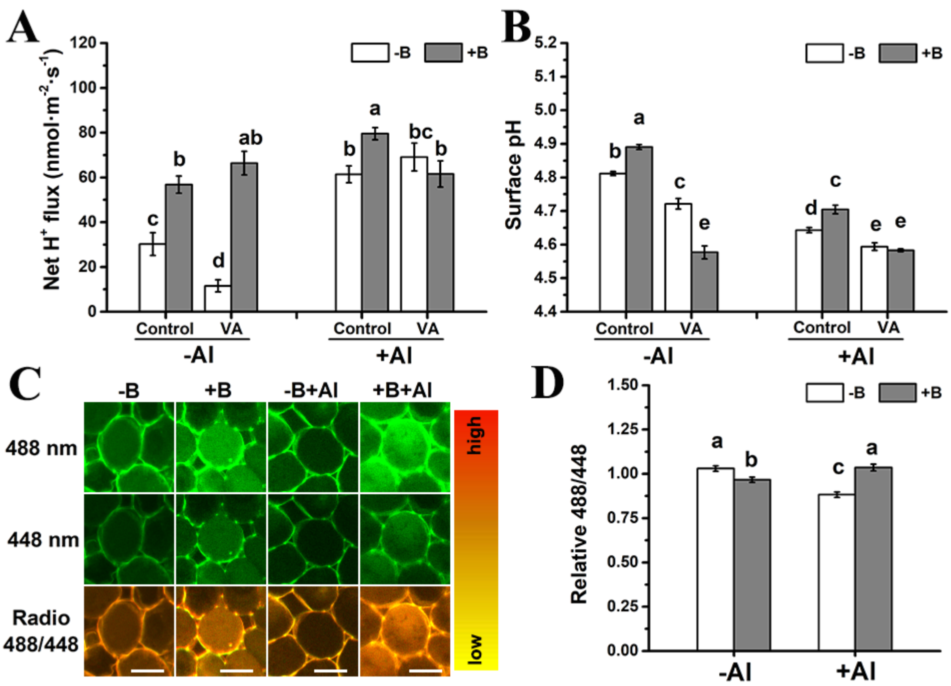

Fig. 4 Apoplast pH in root transition zone of pea with the application of Vanadate (VA).

CONCLUSION

Boron supply promotes apoplast alkalization and alleviates Al toxicity to roots of pea.

ACKNOWLEDGEMENTS

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REFERENCES

Staal M., K. Vissenberg. 2011. Apoplastic alkalinization is instrumental for the inhibition of cell elongation in the Arabidopsis root by the ethylene precursor 1-aminocyclopropane-1-carboxylic acid. Plant Physiol. **155:** 2049-2055

O-23

STOP1 transcription factor regulates the early aluminum-inducible expression of its primary target genes *via* the unfolded protein response-like pathway

Mutsutomo Tokizawa[1, 2]*, *Takuo Enomoto[1]*, *Hiroki Ito[1]*, *Yoshiharu Y. Yamamoto[1]*, *Yuriko Kobayashi[1]*, *Leon V. Kochian[2]*, and *Hiroyuki Koyama[1]
[1]Applied Biological Sciences, Gifu University, Japan,
[2]Global Institute for Food Security, University of Saskatchewan, Canada.
; mutsutomo.tokizawa@gifs.ca

Aluminum ions (Al^{3+}) are most rhizotoxic among the solubilized ions in acid soils and Al toxicity is a primary factor that decreases crop yields on acid soils. A decade ago, a zinc finger transcription factor STOP1 (Sensitive TO Proton Rhizotoxicity 1) was identified as ab essential gene involved in acid soil tolerance, including both Al and low pH stress, in *Arabidopsis* (Iuchi et al., 2007). STOP1 regulates multiple Al tolerance genes including *ALMT1* (ALuminum-activated Malate Transporter 1), *AtMATE* (a citrate transporter) and *ALS3* (ALuminum Sensitive 3) (Sawaki et al., 2009). Recently, we showed that STOP1 directly binds to *ALMT1* promoter and the binding is an essential step for the Al-induced expression (Tokizawa et al., 2015). Although several studies show that the expression of *ALMT1* is immediately (within 1h) and strongly (~100 fold) induced in response to Al, it is unclear how STOP1 regulates this induction. In addition, STOP1's primary Al-inducible target genes other than *ALMT1* are poorly understood. To understand mechanism of STOP1-mediated transcriptional activation for the primary target genes, we first searched the primary target genes that have STOP1 binding sites in the promoter in suppressed genes in the *stop1* mutant by using a combination analysis of bioinformatics-based cis-element prediction, in vitro protein-DNA binding, and *in planta* promoter assays, which were the same methods used to find the STOP1-binding site in the *AtALMT1* promoter. In this analysis, we found that STOP1 directly binds to the *STOP2* and *GDH2* (*GLUTAMATE DEHYDROGENASE 2*) promoters and regulates the Al-induced expression of these genes. However, *ALS3* and *AtMATE*, that are well-known STOP1-regulated and Al-inducible genes, both are indirectly regulated by STOP1.

To compare the expression profiles between the primary target genes (*i.e., ALMT1*, *GDH2* and *STOP2*) and the secondary target genes (*i.e., AtMATE* and *ALS3*), we quantified the expression level of these genes under Al conditions at different time points of Al exposure. The expression of primary target genes was induced within 1.5h by Al, whereas that of secondary target genes was initiated 3h-after the Al treatment. This result strongly suggests that binding of STOP1 to the promoter play critical role in the early Al response in the primary target genes.

Recently, Balzergue *et al.* (2017) reported that STOP1 accumulation in nuclei was observed in P-deficiency condition. Because it is unclear whether this accumulation can be observed under Al stress conditions, we prepared transgenic plants carrying the *STOP1* promoter:STOP1-GFP and examined the localization of STOP1. The nuclear accumulation can be observed in response to Al within 1.5h. In addition, this accumulation and the induction of the primary target genes were inhibited by a chemical chaperon that is a known inhibitor for the unfolded protein response (UPR).

UPR inducers (*e.g.*, Tunicamycine) induce STOP1 nuclear accumulation and the expression of the primary target genes. Additionally, typical UPR inducible genes (*e.g.*, *BIP3*) were induced by Al treatment. These results suggest that Al-induced STOP1 nuclear accumulation and the following transcriptional activation process in the primary target genes are activated *via* the UPR pathway.

REFERENCES

Balzergue C et al., (2017) Low phosphate activates STOP1-ALMT1 to rapidly inhibit root cell elongation. Nat Commun **8**: 15300

Iuchi S et al., (2007) Zinc finger protein STOP1 is critical for proton tolerance in Arabidopsis and coregulates a key gene in aluminum tolerance. Proc Natl Acad Sci U S A **104**: 9900-9905

Sawaki Y et al., (2009) STOP1 regulates multiple genes that protect Arabidopsis from proton and aluminum toxicities. Plant Physiol **150**: 281-294

Tokizawa M et al., (2015) STOP1, CAMTA2, and other transcription factors are involved in *ALMT1* expression. Plant Physiol **167**: 991-1003

O-24

Detecting land cover changes with satellite images after prolonged eruptions of Mt. Sinabung in north Sumatra Indonesia

Frisa Irawan Ginting[1]*, *Ahmad Yasir[1]*, *Ahmad Fanani[1]*, *Gusnidar[1]*, *Yulnafatmawita[1]*, *Malik Nelson[2]*, *Dian Fiantis[1]*, *Budiman Minasny[3]
[1]Department of Soil Science, Faculty of Agriculture, Andalas University, Kampus Limau Manis, Padang25163, Indonesia.
[2]Department of Estate Crops, Payakumbuh Agriculture Polytechnic Institute, Kampus Politani Tanjung Pati, 50 Kota, Sumbar,
Indonesia
[3]Sydney Institute of Agriculture, The University of Sydney, 1 Central Avenue, Australian Technology Park, Eveleigh, NSW 2015,Australia.
; dianfiantis@yahoo.com

ABSTRACT

Volcanic eruptions affect natural and land resources in a wide area. The present study quantifies the impact of volcanic eruption on the environment by using satellite images of Landsat and Spot. Here details on land cover changes are derived from a study of volcanic ash deposits in the vicinity of Mt. Sinabung (North Sumatra, Indonesia). By investigating the volcanic eruptions from 2010 to 2016, we document that eruptions can have very different dynamics to alter the soil surface and agriculture activities. The spectral analysis shows that air-fall volcanic deposits are characterized by low reflectance values, dark to light grey color, variations in grain size, coarser at proximal to fine materials for distal deposit of a distance between 7 to 10 km or more. The Landsat satellite recorded that 2,165 Ha of volcanic soils were covered by airfall ash deposit, after the first phase of eruption on August to September 2010. After that Mt. Sinabung ceased down for 3 years to August 2013, only about 200 Ha of bare surface land was detected and it is close to eruptive center. Then on September 2013 the volcano erupted again to present time (March 2018) alter the surrounding areas up to 30,320 Ha with the deposit of volcanic materials. The productive agriculture land become unproductive after the repetitive eruptions. Monitoring the impact of volcanic eruptions with satellite images are found to be very useful and represent a powerful tool with it spatial and temporal scale of observation.

Keywords— tephra deposits; spatial and temporal resolution; Landsat; Spot

INTRODUCTION

Volcanic ash fallout is a recurrent environmental hazards in volcanic regions. On August 27, 2010 Mt. Sinabung in North Sumatera Indonesia erupted after rested for twelve hundreds of years. The phreatic eruption ended 10 days later and commenced again on September 17, 2013 to present (March 2018). The eruptions of Mt. Sinabung produce explosive columns, disperse tephra, crater-limited lava domes, flank descending lava flows, vent-derived pyroclastic density currents (PDCs), and lava flow margin collapse-generated PDCs (1). It was estimated the prolonged eruptions of Mt. Sinabung resulted in major impacts on land surface in the vicinity of Mt. Sinabung as volcanic materials deposited above grounds. Then the lava flows can travel downward slope and led to the destruction of agriculture crops and settlement areas.

As Sinabung continues to erupt which endanger humans and their livelihoods, destructing the agriculture crops and burying their farms, the Indonesian authority set and map the volcanic hazard zonation and prohibited areas endanger of a hot ashfall hazards with 4 categories. They are zone 1 of 0-3 km from summit, zone 2 cover 3-5 km, 5-7 km consider zone 3 and 7-10 km of zone 4. Zone 1 to 2 are consider the red zone and prohibited, no one can enter and carry out any activities. Areas beyond the red zone (>5 km) are allowed to be cultivated.

The area devastated by the eruption of Mt. Sinabung offers an opportunity to observe the land cover changes after natural disaster which can be detected by satellite. Satellite images have been providing measurements of global volcanic hazards by recording the disruptions occur at spatial, spectral or temporal scales. The satellite recorded the electromagnetic spectrum of volcanic materials at the optical region of 350–2500 nm [2].

The objective of this study was to provide a detail spatial distribution and temporal changes on the extent of volcanic ash deposits and land-used after repeated eruptions of Mt. Sinabung by using Landsat and Spot 6 data from 2010 to 2016.

O-25

Chemical changes in acid sulphate soil of Sungai Raya, Negeri Sembilan

Som, A.[1], Yahya, A.A.[1], Bachmann, R.T.[1], and Karananidi, P.[1]

[1]Universiti Kuala Lumpur, Malaysian Institute of Chemical and Bioengineering Technology, Bandar Vendor Taboh Naning, Alor Gajah 78000, Melaka, Malaysia
; padmini.karananidi@s.unikl.edu.my

ABSTRACT

In Malaysia, acid sulphate soil can be found mainly in coastal regions where waterlogged soil was drained for agriculture purposes. The soil has a very low pH value and high dissolved aluminium concentration which are both detrimental for plant growth. Acid sulphate soil obtained from Kampung Sungai Raya (Negeri Sembilan) were characterized and incubated with three oil palm frond biochar application rates (20, 40 and 60 g/kg). The changes on acid sulphate soil pH and electrical conductivity (EC) value were monitored for 40 days. Oil palm frond biochar application rates had a significant effect ($P \leq 0.05$) on acid sulphate soil pH and EC. While incubation period had no significant effect ($P \geq 0.05$) on EC. Application rates of 40 and 60 g/kg was significantly greater ($P<0.05$) than control and 20 g/kg.

INTRODUCTION

Acid sulphate soil is characterised by low pH (<4) and high amount of soluble aluminum which prevents elongation and division of root cells resulting in a stunted and deformed root system and forms insoluble compounds such as $AlPO_4$ which reduces the availability of phosphate for plant intake [2]. Application of lime ($Ca(OH)_2$, CaO or $CaCO_3$) is the conventional rehabilitation method, however it can be an economic burden for most farmers. The use of biochar, a carbonaceous porous material, generally has a good adsorption capacity and its ability to adsorb aluminum to reduce toxicity to crops. Malaysia being the second largest producer of palm oil in the world, produces 82.5 kg of fronds / (palm year) and this has generated masses of agricultural waste. In this study we produced biochar from oil palm fronds, and evaluated the effect of biochar application rate and incubation time on acidic sulphate soil in terms of pH, electrical conductivity and sulphate content in laboratory scale experiments.

MATERIALS AND METHOD

Fresh oil palm fronds were collected from an oil palm plantation at Kampong Cherana Putih, Alor Gajah, Melaka and were cut into 3cm x 3cm dimension. The pyrolysis process parameters were 345°C for 45 min, which were chosen based on previous studies to produce biochar with nearest to neutral pH value and minimum hydrophobicity [4, 5].

An amount of 14 kg of soil from Kampung Sungai Raya, 71150 Linggi, Negeri Sembilan, Malaysia (2° 25' 48.0" N and 101° 57' 29.5" E) was air dried and sieved to eliminate pebbles and other foreign objects. The soil was weighed and 1000 g transferred into each container. Oil palm frond biochar was added to each of the container with varying dosage (0, 20 g/kg, 40 g/kg and 60 g/kg) and mixed thoroughly.

The 40 days room temperature incubated samples were collected every 5 days for pH and conductivity determination; in-house method base on BS1377: Part 3:1990.

Particle size distribution was determined using the Malvern Mastersizer 2000 particle size analyser. The analyser was set at obscuration of 5%, 1700 rpm stir rates and stability was reached at 3 minutes.
Water soluble sulphate was determined by using BS 1377: Part 3:1990. A 2:1 water extract was obtained from soil samples dried at 75°C overnight. The sulphate in the extract was precipitated by using $BaCl_2$.
The significant effect of application rate and incubation time was analysed by using Analysis of Variance (ANOVA) in Minitab 17.

RESULTS AND DISCUSSION

1.1. Characterisation of Soil and oil palm frond Biochar

The acid sulphate soil was collected from 20 cm top layer and has an initial pH of 3.7 and soil water-soluble sulphate content of 0.07%. According to our particle size analysis, the soil is classified as silt loam under soil textural triangle [10]. The 10% amount of clay further indicate the presence of sulphuric acid from oxidation of pyrite that caused disintegration of clay minerals [11].

1.2. Effect on Soil pH

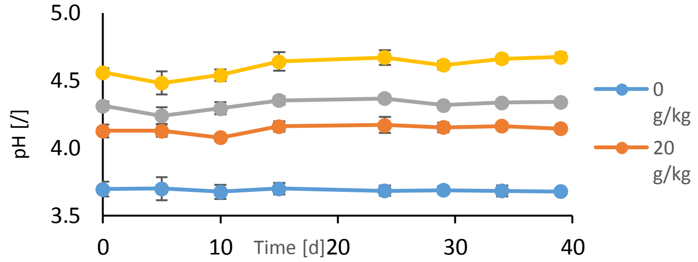

Fig. 1. Changes in pH value upon application of 20, 40 and 60 g/kg of oil palm frond biochar and changes during incubation. 0 g/kg are control samples. Standard deviation of means are marked as error bars (n = 3).

The oil palm frond biochar used in this study has a pH value of 7.6. The soil pH value increased upon incremental addition of 20, 40 and 60 g/kg oil palm frond biochar to 4.13, 4.31 and 4.56, respectively (Fig. 1). It was proposed that cations (K^+, Na^+, Mg^{2+} and Si^{2+}) presence in the feedstock such as oil palm fronds will form carbonates during pyrolysis process and become the main alkaline components in the biochar [12]. It has been identified that surface organic functional groups on the biochar contribute to long-term soil CEC and pH buffering capacity [13]. Negatively charge phenolic, carboxyl and hydroxyl group can bind with H^+ ions from the soil solution [14].

1.3. Effect on Soil Electrical Conductivity

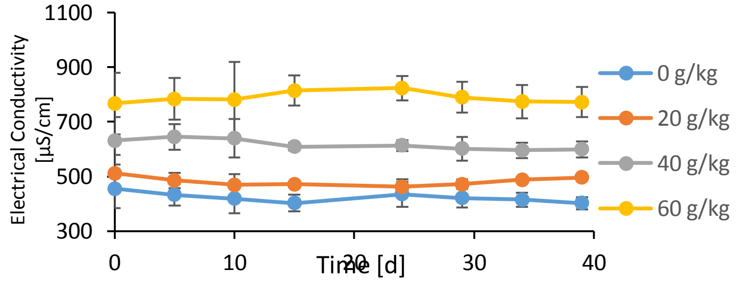

Fig. 2. EC value upon application of 20, 40 and 60 g/kg of oil palm frond biochar and changes during incubation. 0 g/kg is control sample. Standard deviation of means are marked as error bars (n=3).

The original soil tested had an EC of 450 µS/cm and indicates sodicity of soil [9]. The area where the soil was collected is sporadically flooded with seawater during high tide that may have caused the soil sodicity to occur. The EC values increased by nearly 100 units with every 10 g increment of oil palm frond biochar application rates (Fig. 2). Oil palm frond biochar has been reported to contain high amount of base cations K^{+}, Ca^{2+}, Mg^{2+} and Na^{+} [4] which upon release to soil water contributed to EC.

CONCLUSION

Oil palm frond biochar was able to increase acid sulphate soil pH value from 3.7 to a maximum of 4.7 by application of 20, 40 and 60 g/kg and EC value with significant difference ($p \leq 0.05$). Incubation period shows no significant difference ($p > 0.05$) in the EC value for each application rate. Future study must focus on integration effect of oil palm frond biochar, acid sulphate soil, plant growth, fertilizer and other additives.

REFERENCES

D**. Fanning, M. Rabenhorst, and R. Fitzpatrick,** "Historical developement in understanding of acid sulfate soils," *Geoderma*, **1972.**

I. H. Rorison, "The effect of soil acidity on the nutrient uptake and physiology of plants," in *Proceedings of the International Symposium on Acid Sulfate Soils*, **1973**, p. 293.

J. Shamshuddin, S. Muhrizal, I. Fauziah, and E. Van Ranst, "A Laboratory Study of Pyrite Oxidation in Acid Sulfate Soils," *Commun. Soil Sci. Plant Anal.*, vol. 35, no. 1–2, pp. 117–129, **2004.**

A. M. Som, Z. Wang, and A. Al-Tabbaa, "Palm frond biochar production and characterisation," *Earth Environ. Sci. Trans. R. Soc. Edinburgh*, vol. 103, no. 1, pp. 39–50, **2012.**

F. Nizam, "Comparison on the basic characteristic of oil palm frond biochars and the potential of biochar as source of potassium for oil palm cultivation in peat soil," Universiti Kuala Lumpur, **2016.**

J. H. Yuan and R. K. Xu, "The amelioration effects of low temperature biochar generated from nine crop residues on an acidic Ultisol," *Soil Use Manag.*, vol. 27, no. 1, pp. 110–115, **2011**.

J**.-D. Mao, R. L. Johnson, J. Lehmann, D. C. Olk, E. G. Neves, M. L. Thompson, and K. Schmidt-Rohr,** "Abundant and stable char residues in soils: implications for soil fertility and carbon sequestration," *Enviironmental Sci. Technol.*, vol. 46, p. 9571, **2012.**

R. Chintala, J. Mollinedo, T. E. Schumacher, D. D. Malo, and J. L. Julson, "Effect of biochar on chemical properties of acidic soil," *Arch. Agron. Soil Sci.*, vol. 60, no. 3, pp. 393–404, Mar. **2014.**

FAO, "Soil salinity assessment," *Book*, p. 165, **1999.**

C. Cruz, "Soil quality considerations in the selection of sites for aquaculture," Fishpond engineering: a technical manual for small-and medium-scale coastal fish farms in southeast asia, **1983.** [Online]. Available: http://www.fao.org/docrep/field/003/AC172E/AC172E04.htm.

C. Cruz, "Soil quality considerations in the selection of sites for aquaculture," Fishpond engineering: a technical manual for small-and medium-scale coastal fish farms in southeast asia, **1983.** [Online]. Available: http://www.fao.org/docrep/field/003/AC172E/AC172E04.htm.

C. E. Brewer, Y.-Y. Hu, T. E. Schmidt-Rohr, K., Loynachan, D. A. Laird, and R. C. Brown, "Extent of pyrolysis impacts on fast pyrolysis biochar properties," J. Environ. Qual., vol. 41, p. 1115–1122., **2012.**

J**.-D. Mao, R. L. Johnson, J. Lehmann, D. C. Olk, E. G. Neves, M. L. Thompson, and K. Schmidt-Rohr**, "Abundant and stable char residues in soils: implications for soil fertility and carbon sequestration," Enviironmental Sci. Technol., vol. 46, p. 9571, **2012.**

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O-26

Organic fertilizer tithonia plus to control iron toxicity and increasing rice yield on new paddy field in Ultisol

***Nurhajati, H.*[1], *N.Rozen.*[1], *and Y.Mala*[1]**
[1]Faculty of Agriculture, Andalas University, Kampus Limau Manis Padang, Indonesia 25163 ; nurhajatihakim44@gmail.com

INTRODUCTION

Ferrous toxicity is one of the problems that often found in lowland rice system in tropical and sub –tropical especially in new paddy field (NPF) on Ultisols. Flooding during rice growth creates a chemically reduced state in soils causing reduction of insoluble (Fe^{3+}) to soluble (Fe^{2+}) form even at excessive level as much as 381 mg.kg^{-1} in NPF on Ultisols. Excessive Fe^{2+} uptake is eventually the main cause of iron toxicity of rice (Hakim *et al*, 2012). From a series of research using Tithonia (*Tithonia doversifolia*) as organic fertilizers to improve acid soil fertility, it was found that Tithonia was able to improve soil fertility by reducing acid content, Al saturation of soil, increased soil pH, soil organic mater content, soil nutrients of N, P, K, Ca, Mg levels, and can substitute N and K commercial fertilizer (CF) application as much as 25 to 75 % for maize on Ultisols (Hakim and Agustian, 2012).

Inoculation of phosphate-solubilizing-bacteria (PSB) into Tithonia rhizosphere can produce 11.3 t dry matter, 215 kg N, 30kg P, and 253 kg K / 2000m^2.ha^{-1}.year^{-1}(Hakim *et al*, 2014). Hakim *et. al.* 2012 processed the biomass of Tithonia into organic fertilizer Tithonia plus (OFTP). The OFTP is an organic fertilizer made from pruned Tithonia, using stardec and Trichoderma as decomposers + lime as grinding CaCO3 + paddy straw or + cow manure, and + commercial fertilizers (CF). The result showed that OFTP formulation as 2t Tithonia + 5t paddy straw + 50% commercial fertilizer; and 2t Tithonia + 75% commercial fertilizers are the optimum formula to reduce Fe^{2+}solubility and enhanced levels of nutrients availability such as N, P, K, and gave higher than 4 t.ha^{-1} rice grain yield at NPF in Ultisol at Sitiung Koto Baru Sub-District, in Dharmasraya District. Therefore, further research should be continued in larger area, with an objective to find a more appropriate formulation of OFTP to alleviate iron toxicity, reduced commercial fertilizer and simultaneously increased the production of rice at NPF (3 year age) in Koto Salak sub-district in Dharmasraya District, West Sumatra province, Indonesia.

MATERIALS AND METHODS

Field experiment was conducted in NPF (3 year age) on Ultisols at Koto Salak sub-District, in Dharmasraya District, West Sumatra Province, Indonesia. The randomized block design (RBD) with four treatments and four block were implemented. There were three treatments of OFTP formulation and one treatment without OFTP+100% commercial fertilizers (CF). Formulation OFTP for 1 ha land as follow; A = 2t Tithonia + 5t paddy straw (PS) + 50% CF; B = 2t Tithonia + 5t cow manure (CM) + 50% CF; C = 2t Tithonia + 5t PS + 2t CM + 25% CF; and D = without OFTP + 100% commercial fertilizers (250kg Urea, 200kg TSP, 250kg KCl, and 100 kg Kiserite). Liming with 500 kg $CaCO_3$ ha^{-1} were applied for each treatments. The pruned Tithonia and paddy straw (size 3-5cm) was obtained by chopping in chopper machine. The mix amount of Tithonia and paddy straw or and cow manure as material of OFTP was composted for four weeks by adding *stardec* and *Trichoderma* as bio-decomposer. The size of the block was 24m x 6m, while the plot was 6m x 6m. The OFTP were broadcasted on the soil surface, evenly plowing and incubated for three weeks. Then, two seedlings of IR_{66} variety with two weeks age were planted into each plot in plant spacing 25 cm x 25 cm.

RESULS AND DISCUSSION

Results of soil chemical properties included Fe^{2+}, total-N, P-Bray-2, and exchangeable-K and rice grain yield effected by OFTP formulation at NPF at Koto Salak was presented in Table 1.

Table 1. Limited soil chemical properties after 6 weeks was incubated by OFTP, and rice grain yield effected by OFTP formulation at NPF at Koto Salak sub-District, Dharmasraya District, in West Sumatra Province, Indonesia.

Treatments formulation of OFTP		Fe^{2+}	Total-N	P-Bray-2	Exch.K	Grain yield
		$mg.kg^{-1}$	%	$mg.kg^{-1}$	$cmol.kg^{-1}$	$t.ha^{-1}$
A	2 t Tithonia+5 t paddy straw (PS)+50% CF	95.79	0.33	24.45	0.33	4.80 a
B	2 t Tithonia+5 t cow manure(CM)+50% CF	52.29	0.34	38.17	0.38	5.40 a
C	2 t Tithonia+5 t PS+2 t CM+25% CF	95.86	0.30	11.02	0.20	4.10 a
D	Without OFTP + 100% CF*	153.99	0.32	15.07	0.30	4.40 a
	Original soil	254.71	0.16	3.11	0.12	

*100%CF =250kg Urea, 200kg TSP, 250kg KCl, and 100 kg Kiserite).

Table 1 showed that NPF is rich in Fe^{2+}, but has low N, P, dan K. The addition of OFTP decrease a large amount of Fe^{2+} solubility and increase N, P, and K. The decrease of Fe^{2+} solubility is due to the reaction of $Fe2^{+}$ chelation by organic acid, mainly from Tithonia. Gusnidar (2007 *cit* Hakim and Agustian, 2012) claimed that Tithonia besides producing N, P, dan K, it also produced organic acids such as acetic, propionate, salycilic, citric, succinic, and tartaric acids. In addition, the increase of N, P, and K in soil can be because the high nutrients content in Tithonia, and can replace 50-75% N, P, K from commercial fertilizer. The decrease of iron solubility and the increase of N, P, and K have resulted a good growth of paddy plant, as it can produce rice grain yield of 4.10 to 5.40 $ton.ha^{-1}$. There is no significant differences in rice grain yield production between the four treatments, which means that the implementation of OFTP can reduce the application of commercial fertilizer (CF) of 50-75%. Further, it was found that treatment A and B in Kota Salak were better compare to treatment C and D. The result of this study strengthened the result of Hakim *et al* 2012 in Sitiung Koto Baru.

CONCLUSION

The formulation of OFTP that more appropriate to control the iron toxicity and reduced the application of commercial fertilizers till 50% to get higher rice grain yield at new paddy field (NPF) at Dharmasraya District, West Sumatra Province, Indonesia were two options (1) 2t Tithonia + 5t cow manure + 500kg CaCO3 + 50% commercial fertilizer, and (2) 2t Tithonia + 5t paddy straw + 500kg CaCO3 + 50% commercial fertilizers with rice grain yield production of 5.40 t and 4.80 t ha^{-1} respectively.

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REFERENCES

Hakim, N; N. Rozen; and Y. Mala. 2012. Application of Organic Fertilizer Tithonia Plus to Control Iron Toxicity and Reduce Commercial Fertilizer Application on New Paddy Field. J Trop Soils **17**(2): 135-142.

Hakim, N., dan Agustian.2012. Titonia untuk Pertanian Berkelanjutan. Andalas University Press.352 halaman.

Hakim, N., R.Alfina., Agustian., Hermansah., and Yulnafatmawita. 2014. Bacterial Inoculants to Increace the Biomass and Nutrients Uptake of Tithonia Cultivated as Hedgerow Plants in Ultisols. Malaysian J.Soil Sci.vol **18**:115-123.

O-27

Agronomic biofortification of upland rice with Zinc

Moraes, M.F.[1]***, S. J. F. Alves***[2]***, L. V. F. V. Alves***[1]***, M. E. P. Batista***[1]***, J. A. L. Pascoalino***[3] ***and J. L. V. Carvalho***[4]
[1]Graduate Program of Tropical Agriculture, Faculty of Agronomy, Federal University of Mato Grosso. Avenida Valdon Varjão, 6390, CEP 78600-000, Barra do Garças, MT, Brazil.
[2]Federal University of Parana (UFPR)
[3]Brazilian Soybean Strategic Committee (CESB)
[4]Brazilian Agricultural Research Corporation (EMBRAPA).
; moraesmf@ufmt.br

INTRODUCTION

Due to its food importance, rice is among the most produced cereals in the world. Among cereals, rice plays a strategic role in both economic and social aspects, being the focus of studies on biofortification programs. It is an extremely versatile crop that adapts to diverse soil and climate conditions, being considered the species with the greatest potential to combat hunger in the world (EMBRAPA, 2005). In Brazil, there is a lack of information on fertilization methods for agronomic biofortification. Thus, the objective of this study was to evaluate the method of zinc application and its relationship with the nutritional quality of grains and agronomic aspects of upland rice cultivars.

MATERIALS AND METHODS

The study was carried out in two locations: Rio Verde, GO and Palotina, PR, Brazil. At each site, the experiments were composed of four treatments: (i) without Zn application (control); (ii) soil Zn application (Zn-S); foliar Zn application (Zn-F) and (iv) soil plus foliar Zn application (Zn-S+F). The experimental design was in randomized blocks, with 4 replications, totaling 32 plots. Application of Zn in the soil was carried out at the planting, with 250 kg ha^{-1} of NPK 8-30-20 formulation, which presented 4% Zn (10 kg ha^{-1} of Zn). In the control and foliar Zn treatments, the formulation did not contain Zn. For foliar Zn application, a solution with 2% of zinc sulfate ($ZnSO_4.5H_2O$) was used, in application rate of 200 L ha^{-1} (equivalent to 910 g ha^{-1} of Zn) and carried out at the beginning of the filling of grains. The dose used for both Zn fertilization in the soil and foliar followed the protocol suggested by the international research projects on agronomic biofortification of the HarvestZinc Program (Cakmak et al., 2010; Phattarakul et al., 2012). Two upland rice genotypes: BRS Sertaneja and Zebu Ligeiro were cultivated, being the first one chosen to be a high yield commercial cultivar and the second to present potential for biofortification (higher Zn concentration in the grains). The sample units (plot) were 4 rows of 5 m, with spacing between rows of 0.30 m and 60 seeds per linear meter, corresponding to an area of 6 m^2 per plot. The useful area of each parcel consisted of 2 central rows, discounting 0.50 m of the border, totaling 2.40 m^2 of useful area. The rice plants were cultivated until the physiological maturity of the grains. Throughout the development of the crop, foliar analysis was performed and the variables yield and nutritional quality of the grains were determined.

RESULTS AND DISCUSSION

In general, Zn concentration in leaves and grains increased with application of this micronutrient, even in soil with Zn concentration above the critical level (Table 1). There was no increase in productivity due to the application of zinc, however, there was an inverse relationship between yield and Zn concentrations in upland rice grains. The cultivar that presented the highest yield in both environments was the BRS Sertaneja, with average productivity of 2134 kg ha^{-1} and 2301 kg ha^{-1}, respectively, in Palotina and Rio Verde, Brazil. The accumulation of Zn in the grains was determined by the yield differences of the cultivars, being higher in the commercial cultivars due to the high yield.

Table 1: Grain zinc concentration of two upland rice cultivars in response of zinc management.

Place: Rio Verde city	BRS Setaneja mg kg^{-1}	Zebu Ligeiro mg kg^{-1}	Place: Palotina city	BRS Setaneja mg kg^{-1}	Zebu Ligeiro mg kg^{-1}
Control*	18.54	22.30	Control	32.80	37.19
Zn-S	19.84	22.95	Zn-S	33.87	38.04
Zn-F	21.51	24.54	Zn-F	34.27	43.50
Zn-S+F	19.27	23.25	Zn-S+F	39.16	45.28

*Control: no Zn application; Zn-S: soil Zn application; Zn-F: foliar Zn application; Zn-S+F: soil plus foliar application

Zn leaf concentration considered suitable for upland rice are between 25 and 35 mg kg^{-1}. In Palotina, Zn leaf concentration were close to this range, with levels between 25.22 to 33.47 mg kg^{-1} for BRS Sertaneja and 32.59 to 39.72 mg kg^{-1} for Zebu Ligeiro. In Rio Verde, it was observed that Zn leaf contents were below that considered adequate for upland rice, presenting levels between 16.05 to 16.48 mg kg^{-1} for the cultivar BRS Sertaneja and 16.77 to 17.34 mg kg^{-1} to grow Zebu Ligero. The results of Zn analysis on upland rice grains, in response to Zn sources, show the importance of zinc fertilization to increase the quality of the agricultural product and, consequently, benefit human health by reducing the lack of Zn that afflicts 20% of the world population (Hotz & Brown, 2004).

CONCLUSION

The genotype-environment interaction determines the yield potential and, consequently, also the nutritional quality of the grains. Thus, under the conditions of the present study, due to the low yield of the cultivar with potential for agronomic biofortification, the commercial cultivars with high yield and the application of zinc (in the soil plus foliar) are more feasible, raising the concentration and accumulation of the same in the grains. It is necessary, in the programs of genetic improvement directed towards biofortification, to seek the increase of yield for these cultivars.

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REFERENCES

Cakmak, I.; Pfeiffer, W. H.; Mcclafferty, B. 2010. Biofortification of Durum Wheat with Zinc and Iron. cereal chemistry, 87(1):10-20

Embrapa - Embrapa clima temperado. Cultivo de arroz irrigado no Brasil. Sistemas de produção, 3 ISSN 1806-9207 versão electronica. Novembro **2005**.

Hotz, C.; Brown, K. H. 2004. Assessment of the risk of zinc deficiency in populations. Food and Nutrition Bulletin **25**:130-162.

Phattarakul, N.; Rerkasem, B.; Li, L. J.; Wu, L. H.; Zou, C. Q.; Ram, H.; Sohu; V. S.; Kang, B. S.; Surek, H; Kalayci, Yazici, M. A.; Zhang, F. S.; Cakmak, I. 2012. Biofortification of rice grain with zinc through zinc fertilization in different countries. Plant and Soil **361**: 131-141.

O-28

Monitoring a Soil Adjusted Vegetation Index (SAVI) from 2010 to 2017 in areas affected by Mt. Sinabung eruptions

Ahmad Yasir[1], Frisa Irawan Ginting[1], Gusnidar[1], Yulnafatmawita[1], Malik Nelson[2], Dian Fiantis[1], Budiman Minasny[3]
[1]Department of Soil Science, Faculty of Agriculture, Andalas University, Kampus Limau Manis, Padang25163, Indonesia.
[2]Department of Estate Crops, Payakumbuh Agriculture Polytechnic Institute, Kampus Politani Tanjung Pati, 50 Kota, Sumbar, Indonesia
[3]Sydney Institute of Agriculture, The University of Sydney, 1 Central Avenue, Australian Technology Park, Eveleigh, NSW 2015, Australia.
; dianfiantis@yahoo.com

INTRODUCTION

Prior to 2010, Mt. Sinabung in North Sumatra, Indonesia was considered as dormant volcano, then a series of eruptions with volcanic ash column up to 5 km height occurred on August 27 to September 7, 2010. After that the volcano rested and became active again from September 2013 to February 17, 2018. The volcanic eruption caused volcanic ash particles not just blanketed the earth surface but they also absorbed and reflected electromagnetic (EM) wave radiation from the sun. These EM reflections are detected and recorded by time series of satellite images. Using a time series of Landsat data, it is possible to monitor the changes of the soil and vegetation surfaces before and after the catastrophic events.

The reflectance features of the Landsat images were used to estimate various vegetation indices. Spectral vegetation indices have been used extensively to predict vegetation cover, above-ground biomass, and leaf-area index. The set of vegetation indices used in this study comprised of the normalized difference vegetation index (NDVI) and soil adjusted vegetation index (SAVI). NDVI is commonly used to monitor the phenology, quantity, and activity of vegetation and SAVI considered soil background condition which influence partial canopy spectra and calculated vegetation indices.

Monitoring the soil adjusted vegetation index (SAVI) and normalized difference vegetation index (NDVI) is important in agricultural land especially in area affected by recent volcanic eruption of Mt. Sinabung. These vegetation indices can be used to delineate the distribution of vegetation and soil based on the characteristic reflectance patterns of green vegetation. This study assessed the changes in the SAVI and NDVI values after eruption of Mt. Sinabung from 2010 to 2017. It also tried to link surface soil organic matter with the vegetation indices.

MATERIALS AND METHODS

Soil and volcanic ash materials were collected within hot ashfall hazard zones of Mt. Sinabung at a distance range of 3-5 km, 5-7 km and 7 – 10 km from volcanic plume or eruption center. The study sites included four districts: Naman Teran in the North, Payung in the South, Tiganderkat in the West and Simpang Empat in the East. Both soil and volcanic ash samples were sampled for chemical analysis. Time series of the Landsat 5 and Landsat 8 images were downloaded before (2010) and after eruption (2011 to 2017). Equation used to calculate vegetation indices SAVI is according to Huete (1988) and NDVI is based on Monsef and Smith (2017).

RESULTS AND DISCUSSION

Field observation showed that the thickness of volcanic ash deposit in 2016 were varied from 1 cm of volcanic ash layer in North to about 29 cm in South. The total area affected by direct deposition of volcanic ash from eruption of Mt. Sinabung was estimated at 31,297 Ha.

The results of regression analysis demonstrated a significant positive relationship ($p < .05$) between SAVI and soil organic matter. The relationship decline from East, North, South and West (Table 1). It appears that volcanic ash deposition altered the spatial and temporal resolution of the vegetation indices (Table 2). The impact of volcanic eruption reduced the vegetated land to bare land. Spatial distribution of SAVI in the danger zone of Mt. Sinabung showed that 9% without any vegetation to low vegetation, medium vegetation 17% (Figure 1).

Table 1. Relationship observed between organic matter content and SAVI

Direction	OM (%)	SAVI	correlation
	10.78	0.3188	
East	6.71	0.2579	0.91
	7.99	0.2486	
	5.32	0.4817	
South	4.77	0.4209	0.79
	5.29	0.4409	
	17.59	0.4835	
West	3.02	0.3057	0.75
	4.44	0.4412	
	11,23	0. 2994	
North	14.67	0.4368	0.89
	29.03	0.5213	

Table 2. Changes of SAVI in Mt. Sinabung from 2010-2017

Year	SAVI
2010	1.065
2011	0.621
2012	0.655
2013	0.778
2014	0.703
2015	0.747
2016	0.738
2017	0.384

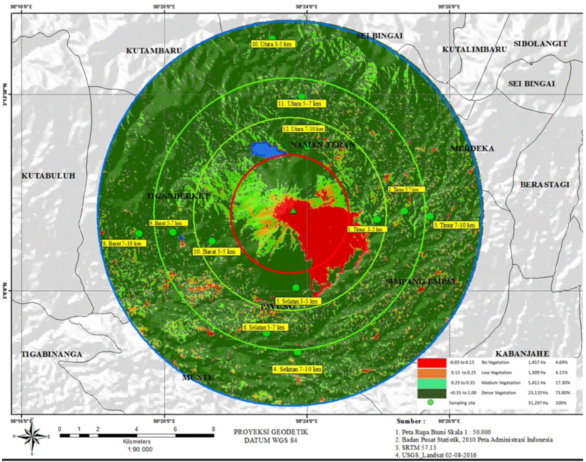

Figure 1. Spatial distribution of SAVI in hot ashfall danger zones of Mt. Sinabung

CONCLUSIONS

This study clearly shows that the satellite-derived vegetation indices are able to accurately detect the changes in the vegetation indices at hot ashfall danger zone of Mt. Sinabung before and during prolonged eruption to date. The SAVI values in 2011 declined 40% after first year of volcanic eruption, remained constant for the following 2 years, slightly increased in 2013, decreased again in 2014 and increased in 2015 and 2016 but fall again in 2017. Vegetation indices were found to be highly correlated to soil organic matter.

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REFERENCES

Huete, A. R. 1988. *A Soil Adjusted Vegetation Index (SAVI)*. Remote Sens. Of Environ. **13:**187-208.

Monsef, H. A. and S. E. Smith. 2017. A new approach for estimating mangrove canopy cover using Landsat 8 imagery. Computers and Electronic in Agriculture **135:** 183-194.

O-29

Spatial distribution of soil and root carbon storage in the Pasoh 50 ha forest dynamics plot

Jeyanny, V.[1], Turner, B.L.[2], Davies, S.J.[3], Mohamad Fakhri, I.[1], Yao, T.L.[1], Suhaimi, W.C.[†], and Fletcher, C.S.[1]

[1] Forest Research Institute Malaysia (FRIM), 52109, Kepong, Selangor

[2] Smithsonian Tropical Research Institute, Apartado Postal 0843-03092, Balboa, Ancon, Panama

[3] SIGEO–CTFS, Smithsonian Institution, Department of Botany, MRC-166, PO Box 37012, Washington, DC 20013, USA

[†] deceased

; jeyanny@frim.gov.my

INTRODUCTION

The atmospheric concentration of carbon dioxide (CO_2), the primary greenhouse gas, has increased by about 30 % from the start of the industrial revolution to 1992 due to fossil fuel combustion and land use change (Mark and Thomas, 2001). There are two alternatives to reduce CO_2: decreasing carbon sources and increasing carbon sinks. Tropical forests play an important role as a potential carbon sink, because forest carbon stocks in tropical regions can range between 164 and 250 Mg C ha^{-1} (Gibbs et al., 2007) and in Malaysia reports have shown that the range of carbon stocks of 164–196 Mg C ha^{-1} (Saatchi et al., 2011) in the above ground biomass. Besides aboveground biomass, tropical forest soils can contain more carbon than temperate forest soils due to factors related to litter inputs, organic matter decomposition and temperature (Lal, 1998). The nature and properties of tropical soils such as clay content, soil texture and mineralogy have been cited to influence soil carbon stabilization (Mutuo et al., 2006). Soil carbon stocks may also vary due to topographic positions such as elevation and temperature. In this study, we investigated whether soil carbon, fine (< 2 mm) and coarse roots (> 2 mm) differ with respect to habitat variability in the Pasoh 50 ha plot in Negeri Sembilan, Malaysia, part of the ForestGEO global network of forest dynamics plots.

MATERIALS AND METHODS

The 1000 × 500 m fifty ha ForestGEO plot is located in the 24 km^2 Pasoh Forest Reserve (2° 58' N, 102° 20' E) in the Jelebu district of the state of Negeri Sembilan, West Malaysia. The forest reserve lies in an undulating inter-montane basin between low mountains (Okuda *et al.* 2003a). Annual rainfall averages about 1800 mm, ranging from under 1200 to over 2400 mm (Noguchi et al., 2003). The 50 ha plot was established in 1988 and the tree community comprises 81 families, 295 genera and 818 species (Davies et al., 2003). The Euphorbiaceae is the most species-rich family in Pasoh with 85 species. *Shorea* was the most important genus in terms of trees and basal area (Davies et al., 2003), followed by *Syzygium*, *Diospyros* and *Aglaia* in terms of species richness. The soils in the 50 ha plot are derived from granitic alluvium in valleys and shale on ridges. Soils were grouped into different clusters and characteristics as follows in Table 1.

Table 1: Soil groups and characteristics in Pasoh 50 ha CTFS plot

Group	Soil series	Soil texture	Soil characteristics
Ridge	Gajah Mati, Terap	clay loam to clay	shale-derived, lateritic gravel, well drained
Slope	Bungor	loam to clay	shale-derived, well drained
Alluvial	Tebuk, Tawar, Tawar (pale)	sandy clay loam to clay	granitic alluvial, moderate to imperfectly drained
Swamps	Kampong Pusu, Alma, Awang	sandy clay loam	granitic alluvial, poorly to imperfectly drained

The distribution of soil types in the 50 ha plot is shown in Figure 1. Ridgetop soils contain ironstone gravel subsoils (Gajah Mati). The slopes surrounding the knolls had soils derived from shale material (Bungor). The northwest region was the floodplains with sub-recent alluvial terrain. The southwest and the southeast counterparts had alluvial soils with better drainage compared to the swamps.

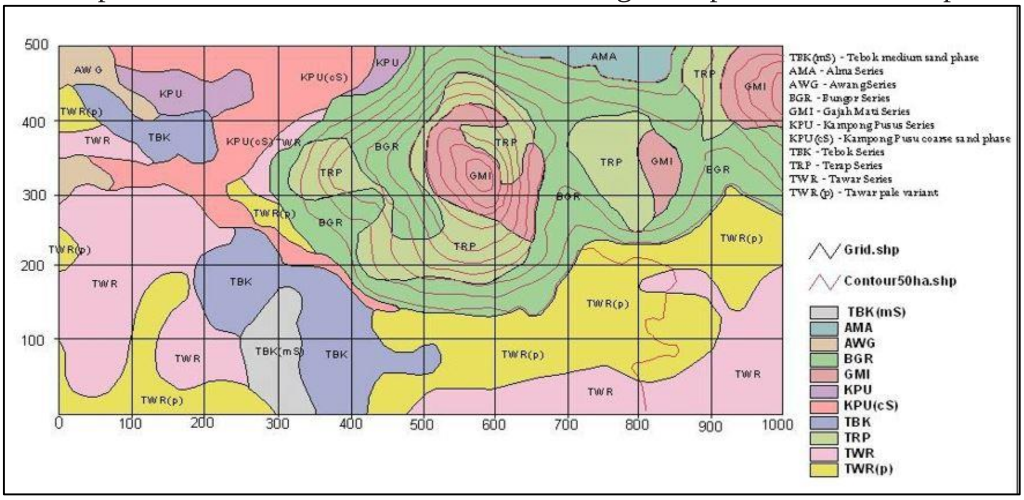

Figure 1: Distribution of soil types in 50 ha CTFS plot in Pasoh, Negeri Sembilan.

Soil pits of major soil types (Gajah Mati, Bungor, Tawar, and Kampong Pusu) were dug outside the perimeter of the CTFS plot. Each pit was described according to the Soil Taxonomy system of soil classification (Soil Survey Staff, 1999). Samples were taken from genetic horizons for the determination of bulk density, moisture content, soil pH, total C, N, P and soil texture analysis (Table 2).

Within the plot, soil samples were collected from nine locations around the center of each hectare. Surface samples (0 – 10 cm and 10 – 20 cm) were taken with a constant volume soil corer (approximately 2.5" diameter) to allow calculation of bulk density. At five of the locations, additional samples were taken using a Jarett auger at 20-50 cm, and then in 50 cm increments to 300 cm, bedrock, or the water table. For each hectare, the samples of equivalent depth were combined for analysis. All soils were air dried (approximately 7-10 days at 25°C), hand-picked to remove roots and stones, and sieved < 2 mm. Root fragments were separated into < 2 mm and > 2 mm diameter, air dried and weighed. Soil samples were oven dried at 35°C, milled and the carbon concentration determined by combustion and gas chromatography (Flash 1112NC Soil Analyzer, CE Elantech, NJ) in the STRI Soils Laboratory, Panama.

The carbon concentration in each sample was calculated on a volume basis from the carbon concentration (mass basis) combined with the bulk density and reported in per hectare basis. Roots were oven-dried at 60°C, weighed to determine biomass, and carbon concentration determined on a pooled sample per hectare and converted to total root carbon on an aerial basis. The data was analysed for significant differences using Analysis of Variance (ANOVA) at $p < 0.05$. We found that there were no significant differences for all variables tested thus the descriptive statistics (median, range and standard error) were reported in Table 3.

Table 2: Median values of selected soil properties from the dominant soil pits outside the 50 ha CTFS plot up to 1 m depth

Profile	**Bulk Density (g cm^3)**	**pH water**	**Total C -%-**	**Total N -%-**	**Total P mg kg^{-1}**
Gajah Mati	1.13	4.35	0.57	0.05	69.00
Bungor	1.45	4.60	0.55	0.04	31.09
Tawar	1.35	4.60	1.01	0.07	38.90
Kampong Pusu	1.39	5.50	0.49	0.05	48.60

RESULTS AND DISCUSSION

The soil C stocks in descending order were ridge<slope<swamps<alluvial. However, there were no significant differences among topographies for all variables tested. Mean carbon stocks up to 1m were greater for soils from the ridge and slope than from the alluvial soils (Table 3). Carbon stocks were most variable on the ridge (65.0-138.0 Mg C ha^{-1}). Fine root biomass (< 2 mm) was greatest on the slope and alluvial regions compared to the ridge and swamps (Table 3). Coarse roots were greater in the swampy area where the values were 44% higher compared to the alluvial habitat.

Table 3: Soil, fine roots and coarse roots C stocks in 50 ha CTFS plot up to 1 m depth

Topography		Soil C stocks (Mg ha^{-1})		Fine root < 2mm C stocks		Coarse roots > 2mm C stocks	
	n	Mean	Range	Mean	Range	Mean	Range
RIDGE	9	90.3	65.0-138.0	2.35	1.5-3.0	4.21	2.50-7.80
		(7.9)		(0.19)		(0.59)	
SLOPE	13	90.0	74.5-116.3	2.54	1.6-4.0	3.90	1.9-6.5
		(3.7)		(0.18)		(0.42)	
ALLUVIAL	21	87.7	71.8-115.8	2.66	1.7-4.2	3.87	1.1-7.0
		(2.5)		(0.13)		(0.38)	
SWAMP	7	81.0	58.1-95.4	2.35	1.6-3.4	5.57	2.90-8.5
		(5.7)		(2.40)		(0.75)	

Note: Values in parenthesis represents standard error

Soils in the ridge area were mainly well-drained lateritic soils with relatively high clay contents (Table 1). Tawar and Kampong Pusu soils (Table 1) on alluvial plains and swampy areas may have the tendency to retain lesser soil C stocks due to high porosity and reduced decomposition due to anoxic conditions. The Tawar soil series predominates in the south and the eastern part of the plot and is coarse grained (Adzmi et al., 2010). Soils of Kampong Pusu are imperfectly drained as the upper subsoil is known to be wet most of the year. Although soil moisture values were lesser for Tawar and Kampong Pusu (Table 2) in samples taken outside of the plots, these soils were reported to be wet most of the time within the plot.

Fine root biomass was greater on slopes and alluvial soils, might reflect aboveground phenology, soil moisture, or nutrient availability (Cheng and Bledsoe, 2002). The coarse roots C stocks in swamps were the highest, and only a small fraction were accounted for in this study. Further investigations with larger coarse roots need to be investigated to detect C trends in roots. In general, Davies et al., (2003) reported that total basal area and stem density differed significantly across the 50 ha plot, with the highest basal area on ridges, but higher stem density in alluvial areas and swamps. Thus, the C stocks in soils, as well as fine and coarse root biomass, may have been heavily influenced by the aboveground biomass distribution. The soil C stocks in Pasoh (up to 1 m depth) were slightly higher compared to that of Ngo et al., (2013) whom reported values of 77.5 Mg C ha^{-1} in primary forest plot in Bukit Timah, Singapore. However, values of fine roots for Pasoh were relatively lower compared to Ngo et al., (2013).

CONCLUSION

Soil C stocks were greater on ridges and slopes compared to the alluvial areas and swamps in dipterocarp forest at Pasoh, Malaysia. In contrast, fine root C stocks were greater on the slopes and alluvial soils. Future work will quantify above-ground C stocks to form a more complete picture of C stocks in this forest.

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REFERENCES

Bationo, A., Kihara, J., Vanlauwe, B., Waswa, B., and Kimetu, J. 2007. Soil organic carbon dynamics, functions and management in West African agro-ecosystems. *Agricultural Systems* **94**(1): 13-25

Cheng, X.M. and Bledsoe, C. S.2002. Contrasting seasonal patterns of fine root production for blue oaks (*Quercus douglasii*) and annual grasses in California oak woodland. *Plant Soil.* **240**: 263–274

Lal, R., Kimble, Follett, R.F., and Stewart, B.A. 1998. *Soil processes and the carbon cycle*. Boca Raton:CRC Press

Mark, T.H. and Thomas, M.S. 2001. Carbon emissions and economic development: future trajectories based on historical experience. – Environment and Development Economics **6**: 63-83

Mutuo, P.K., Shepherd, K.D., Albrecht, A. and Cadisch, G.,2006. Prediction of carbon mineralization rates from different soil physical fractions using diffuse reflectance spectroscopy. *Soil Biology and Biochemistry* **38**(7): 1658-1664.

Ngo, K.M., Turner, B.L., Muller-Landau, H.C., Davies, S.J., Larjavaara, M., Nik Hassan, N.F., and Lum, S. 2013. Carbon stocks in primary and secondary tropical forests in Singapore. *Forest Ecology and Management*, **296**: 81-89.

Noguchi S, Nik AR and Tani M. 2003. Rainfall characteristics of tropical rainforest at Pasoh Forest Reserve, Negeri Sembilan, Peninsular Malaysia. Pp. 51–58 in Okuda T *et al.* (Eds.) *Pasoh. Ecology of a Lowland Rain Forest in Southeast Asia.* Springer, Tokyo

Okuda T, Manokaran N, Matsumoto Y, Niiyama K, Thomas SC and Ashton PS(*Eds.*). 2003. *Pasoh. Ecology of a Lowland Rain Forest in Southeast Asia.* Springer, Tokyo

Saatchi, S.S., Harris, N.L., Brown, S., Lefsky, M., Mitchard, E.T., Salas, W., Zutta, B.R., Buermann, W., Lewis, S.L., Hagen, S. and Petrova, S., 2011. Benchmark map of forest carbon stocks in tropical regions across three continents. *Proceedings of the National Academy of Sciences*, ***108***(24), pp.9899-9904.

Soil Survey Staff. 1999. Soil Taxonomy: A Basic System of Soil Classification for Making and Interpreting Soil Surveys. United States Department of Agriculture–Natural Resources Conservation Service, Lincoln, NE.

O-30

Stabilization of natural stable Cs in each organ of blueberry bushes grown in three types of soils through acidification and/or fertilization

***Matsuoka, K[1,3].,* N. Moritsuka[2], S. Kusaba[1], and K. Hiraoka[1,4]**
[1] Institute of Fruit Tree and Tea Science, NARO, Tsukuba 305-8605, Japan
[2] Graduate School of Agriculture, Kyoto University, Kyoto 606-8502, Japan
[3] Department of Radioecology, Institute for Environmental Sciences, Kamikita-gun 039-3212, Japan
[4] Western Region Agricultural Research Center, NARO, Zentsuji 765-0053, Japan
; kaori.matsuoka@hotmail.co.jp

INTRODUCTION

Since the Fukushima Daiichi Nuclear Power Plant accident in March 2011, Japan, there have been several reports on blueberry fruit whose radiocesium level exceeded the maximum allowable limit for radiological safety (100 Bq kg^{-1}). Blueberry (*Vaccinium*) species have shallow root systems, which exposes them to radiocesium accumulated in the surface soil. For instance, blueberry fruit in the Czech Republic contained high levels of ^{137}Cs, and the ^{137}Cs concentrations were significantly correlated (r = 0.93) with the soil ^{137}Cs concentration, even 28 years after the 1986 Chernobyl nuclear power plant accident (Cervinkova and Poschl, 2014). The suitable soil management for reducing the radiocesium in blueberry fruit is important to decrease a long-term human health risk by the intake.

In blueberry culture, sulfur and NH_4^+ and K fertilizers are applied to soils for acidification (pH of 4 to 5) and fertilization by which radiocesium in soils may be released into the soil solution. In our previous pot experiment using three types of soils subjected to acidification and/or fertilization (Matsuoka et al., 2017), the average concentration of stable Cs in the soil solution was not significantly correlated with the content of Cs in the blueberry bushes. Previous reports also showed that the uptake of ^{137}Cs by blueberries and the concentration in blueberry fruit decreased by K fertilizer application or with a soil containing high exchangeable K (Iwabuchi, 2014; Mandro et al., 2014). These findings suggest that the uptake of stable or radioactive Cs by blueberries and its translocation to fruit are not solely determined by the Cs concentration in the soil solution.

In this study, therefore, we reanalyze the data from our previous experiment (Matsuoka et al., 2017) to evaluate the soil factors affecting the concentration and content of Cs in blueberries and its translocation to fruit.

MATERIALS AND METHODS

A two-year pot experiment was carried out by using three soil types (0-20 cm surface soil; an Andosol, a Cambisol, and a Fluvisol) combined with four treatments (a control with no treatment, acidification (15 g sulfur powder/ 3 kg soil per pot), combined NH_4^+ and K fertilization (0.6 g NH_4Cl-N and 0.6 g KCl-K/ 3 kg soil per pot), or a combination of acidification with fertilization). Each pot was planted with a single 4-year-old rabbiteye blueberry bush (*Vaccinium virgatum* Aiton cv. 'Onslow'), and each treatment was triplicated. The soil solution was sampled by the suction method eight times during the experimental period (March 2014 to September 2015). Within 24 h after watering, we collected the soil solution from each pot, and then the concentrations of 13 elements were determined; Na, Mg, Al, P, K, Ca, Mn, Fe, Cu, Zn, Rb, and Cs by ICP-MS; N (NH_4^+ and NO_3^-) by the colorimetric methods. The soil solution pH was also measured. Blueberry fruit was sampled during harvesting in 2014 and 2015. After the second harvest, the blueberry bush was divided into leaves, branches-plus-stem, and roots. The plant samples were digested in concentrated HNO_3, and the concentration of Cs was measured by ICP-MS.

RESULTS AND DISCUSSION

The Cs concentration in the root-zone soil solution increased (though non-significantly) by the acidification, fertilization, and combination within a given soil. However, the whole-bush Cs concentration did not change significantly by any soil treatments. It is partly due to the lower requirement of Cs by the bushes than their supply via the soil solution. Comparing among each blueberry organ in the second year, the fruit Cs concentration in the Andosol and Fluvisol decreased significantly by the acidification, fertilization, and combination. On the other hand, the fruit Cs concentration in the Cambisol decreased greatly (though non-significantly) by the fertilization but increased (though non-significantly) by the acidification. The Cs concentration in the branches-plus-stem was similar to that in the fruit. The leaf and root concentrations were not influenced significantly by any soil treatments within a given soil.

We analyzed these data by the correlation analysis to find the key cations in the soil solution that affected the Cs concentration in the blueberry organs. The Cs concentration in the fruit, leaves, branches-plus-stems was significantly negatively correlated with the Na, Mg, K, and Ca concentrations in the soil solution. On the other hand, the Cs concentration in any of the organs was not significantly correlated with the Cs concentration in the soil solution. This reconfirmed that the Cs concentration in soil solution was not main factor affecting the Cs concentration in the blueberry. The concentrations of Na, Mg, K, and Ca in the soil solution of the Cambisol were lowest among the three soils, and the K concentration in the soil solution significantly increased by the fertilization but not by the acidification, which probably influenced the fruit Cs concentration. Unlike the aboveground parts, the concentrations of the root Cs and the whole-bush Cs were not correlated significantly with the Na, Mg, K, and Ca concentrations in the soil solution. It was, therefore, suggested that the increase in the concentrations of the basic cations in the soil solution, especially K, is important to reduce the concentration of Cs in fruit, but not in the root and the whole bush.

Further analysis was carried out to reveal how the soil treatments influenced the Cs uptake by the bushes and its translocation from the roots to the fruit. The concentration of Cs in each blueberry organ was multiplied with the dry matter weight to obtain the Cs content in each organ. As a result, the whole-bush Cs content did not differ significantly in any soil treatments within a soil type. Then, we compared the distribution of the Cs in an organ as the percentage of the total Cs in the whole bush. The distribution percentage of Cs in the fruit decreased (though non-significantly for the Andosol, but significantly for the Fluvisol) by the acidification, fertilization, and combination (from 20% to 5-13% for the Andosol, from 25% to 3-13% for the Fluvisol), whereas the percentage in the root increased (significantly for the combination treatment in the Andosol, and significantly for the acidification and combination treatments in the Fluvisol) (from 49% to 64-84% for the Andosol, from 44% to 45-90% for the Fluvisol). In contrast to these soils, the percentage of Cs in the fruit in the Cambisol increased (though non-significantly) by the acidification, fertilization, and combination (from 5% to 8%), but decreased (though non-significantly) in the root (from 87% to 77-82%). These results indicated that the soil treatments significantly influenced the distribution of Cs in the bushes, but not the Cs uptake by the bush.

CONCLUSION

Our results reconfirmed that the uptake of stable Cs by the blueberries and its translocation to the fruit are not solely determined by the Cs concentration in the soil solution. Apparently, the Cs concentration in the fruit was negatively correlated with the concentrations of Na, Mg, K, and Ca in the soil solution. In our case, the effect of soil management on the fruit Cs concentration varied with the soil type; the fertilization treatment was effective to decrease the Cs concentration in all the soils, and the acidification treatment was effective for only two of the three soils. Further research is needed whether these findings can be applied to the radiocesium with a focus on the original soil properties.

REFERENCES

Cervinkova, A. and M. Poschl. 2014. Radiocaesium in forest blueberries in selected location of Jeseníky protected landscape area. Mendel Net.: 224–228. https://mnet.mendelu.cz/mendelnet2014/articles/52_cervinkova_1062.pdf.

Iwabuchi, K. 2014. Inhibition of radiocesium absorption in a blueberry. Repot on Fukushima Agricultural Technology Center, pp 82–85. http://www.pref.fukushima.lg.jp/w4/nougyou-entre/kenkyu_houkoku/kenkyu_houkoku_top.htm. (in Japanese).

Mandro, Y. N., K. Rosen and M. M. Vinichuk. 2014. Effect of wood ash and K-fertilization on ^{137}Cs uptake by selected forest plants. International Conference on Radioecology and Environmental Radioactivity. https://intranet.pacifico-meetings.com/amsysweb/faces/publicacionOnline.xhtml?id=146.

Matsuoka, K., N. Moritsuka, S. Kusaba and K. Hiraoka. 2017. Effects of soil type and soil treatment on solubilization of 13 elements in the root zone and their absorption by blueberry bushes. Hort. J. DOI: 10.2503/hortj.OKD-100. https://www.jstage.jst.go.jp/article/hortj/advpub/0/advpub_OKD-100/_pdf.

O-31

Effect of fertilizer on greenhouse gas emission in oil palm plantation in acid soils

***Nino Sakata*[1], *Hiroshi Aoki*[2], *Atsushi Sakamoto*[2], *Lulie Melling*[3] and *Kazuyuki Inubushi*[1]**
[1]Graduate School of Horticulture, Chiba University, 648 Matsudo, Matsudo-shi, Chiba, Japan
[2]JCAM AGRI. Co. LTD., Tokyo, Japan
[3]Peat Research Laboratory Unit, Sarawak, Malaysia
; inubushi@faculty.chiba-u.jp

INTRODUCTION

Oil palm (*Elaeis guineensis* Jacq.) production in Malaysia and Indonesia is currently the focus of concern due to its potential impact on the environment via greenhouse gas emissions. Oil palm plantations have been reported to release large quantities of nitrous oxide (N_2O) into the atmosphere, which is most likely linked to nitrogen (N) fertilizer use. However, there are still limited studies comparing effects of the type of soil and N fertilizer on N_2O and carbon dioxide (CO_2) emissions. This study aimed to evaluate the effects of soil types and N fertilizer on N_2O and CO_2 emissions in oil palm plantations. Interactions of soil properties and topography influencing soil N_2O and dissolved N_2O will also be reported in oil palm plantation in Malaysia and Indonesia.

MATERIALS AND METHODS

Study sites were located in oil palm plantation areas on tropical land, with one site in Indonesia and two sites in Malaysia. The first site was located in Tunggal Plantation, Riau Province, Indonesia on sandy loam soil classified as Ultisols. The Tunggal Plantation site has a sloping topography with an annual rainfall of 1387 mm. The second site was located in Simunjan Plantation, Sarawak, Malaysia on sandy soil, which was also classified as Ultisols. The Simunjan Plantation site is characterized by sloping topography with an annual rainfall of 4095 mm. The third site was located in Tatau Plantation, Sarawak, Malaysia on peat soil, classified as Histosols. The Tatau Plantation site is characterized with a flat topography, located along the coast, with an annual rainfall of 2225 mm. Three replications of the following four experimental treatments were conducted: Treatment B: no nitrogen fertilizer and no tillage. Treatment M: coated fertilizer in granular form was applied by the deep placement method: namely, after digging soil to 0‑15 cm depth at four different spots, approximately 140 cm away from palm trees, fertilizer was incorporated and covered with soil. Treatment C: conventional fertilizer (non-coated) surface application on four spots approximately 140 cm away from palm trees, with no tillage. Treatment B2: no nitrogen fertilizer, with tillage only in the soil (0‑15 cm) in a similar way as in treatment M. Except for B and B2 treatments, the annual rates (kg N ha^{-1}) of application for the conventional fertilizer were 151 as NPK (nitrogen-phosphorous-postassium) (16-4-25), 107 in the first year and 121 in the second year as NK1 (1:1 mixture of ammonium sulphate and MOP (Muriate of Potash)), and 69 as urea in Tunggal, Simunjan and Tatau, respectively. The rate of conventional fertilizer application followed each plantation's guidelines. It was considered that the coated fertilizers are more efficient due to a lower loss rate of N (Shoji and Kanno 1994). Hence, the rates of application for the coated fertilizer were about half the rate of the conventional fertilizers, namely 76, 62 and 46 kg N ha^{-1} in Tunggal, Simunjan and Tatau, respectively.
N_2O and CO_2 emissions were measured by closed chamber method for 15‑16 months in Tunggal sandy loam soil, Indonesia, and in Simunjan sandy soil and Tatau peat soil,

Malaysia. Within each site, treatments with coated fertilizer and conventional fertilizer, and unfertilized with and without tillage, were established. Also N_2O and CO_2 emissions were measured in upper, middle, and lower slope positions in Tunggal plantation. Dissolved N_2O concentration in water in puddle, drain, and well was measured by headspace method.

RESULTS AND DISCUSSION

N_2O and CO_2 fluxes showed high variabilities with seasons, types of soil and fertilizer treatments. Dissolved N_2O concentrations varied also by water sources and sampling time. The mean values of the N_2O fluxes from each treatment in the Simunjan sandy soil was the lowest among the three soils. The mean of the N2O fluxes from each treatment in the Tunggal sandy loam soil. The mean of the N_2O fluxes was found to be the highest among the three soils in each treatment of the Tatau peat soil. The N application rate of coated fertilizer was about half that of conventional fertilizer and was applied as deep placement. In the Tungal soil, coated fertilizer reduced N_2O emissions by 31 and 48% in wet and dry seasons, respectively, compared to the conventional fertilizer, and was similar to unfertilized treatment. However, N_2O emissions increased in Simunjan and Tatau soils during dry seasons. There was no significant difference between treatments. Dissolved N_2O concentrations in water sources were supersaturated as leading to possibility to be source of indirect emissions.

CONCLUSION

These results show that N_2O and CO_2 fluxes in the tropical oil palm plantations were significantly affected by the type of soil, but not always by fertilizer treatments. Agricultural landscape plays an important role in its relation to hydrological process.

REFERENCES

Shoji, S., and Kanno, H., 1994. Use of polyolefin-coated fertilizers for increasing fertilizer efficiency and reducing nitrate leaching and nitrous oxide emissions. Fertil. Res., **39**, 147‑152. doi:10.1007/BF00750913

Sakata, R., Shimada S, Arai, H, Yoshioka N, Yoshioka R, Aoki H, Kimoto N, Sakamoto A, Melling L, Inubushi K, 2015. Effect of soil types and nitrogen fertilizer on nitrous oxide and carbon dioxide emissions in oil palm plantations, Soil Sci. Plant Nutr. **61**: 48-60.

Sakata, R., Shimada S, Arai, H, Yoshioka N, Yoshioka R, Aoki H, Kimoto N, Sakamoto A, Inubushi K, 2016. Effect of topography on N_2O and CO_2 emissions and dissolved N_2O in oil palm plantation in Riau, Indonesia, Trop. Agr. Develop. **60**(4): 226-235.

O-32

Effect of slag fertilizer on methane emission and rice growth from paddy soil

Qian Li[1], ZanYang Wang[1], Takashi Nagasawa[1], Kazuyuki Inubushi[1]
[1]Graduate School of Horticulture, Chiba University, Matsudo 648, Matsudo-shi 271-8510, japan
; inubushi@faculty.chiba-u.jp

INTRODUCTION

When paddy field is submerged, the soil organic matter is finally decomposed to methane by methanogens (Asami and Takai 1970). Methane is one of the main component of greenhouse gases. A pot experiment and a field experiment were conducted to evaluate the effects of application of slag fertilizer (Minekaru) and organic fertilizer (bokashi) on methane emission, soil chemical property (pH) and rice growth, yields and quality.

MATERIALS AND METHODS

Pot experiment: The pot experiment was conducted from April 30 to September 8, 2017. Three kg sandy soil was placed into plastic pot (3L), five treatments were set up as allows: (1) slag fertilizer (200 kg/10 a); (2) bokashi fertilizer (80 kg/10 a); (3) slag (200 kg/10 a) ＋bokashi (80 kg/10 a) fertilizer; (4) chemical fertilizer (NPK fertilizer 39 kg/10 a); (5) soil only, all with 3 replications. (1) - (4) with three rice seedlings and (5) without rice seedling, all of the pots received basal dressing of NPK fertilizer (1 g) and top dressing twice with urea (0.125 g).

Field experiment: The field experiment was conducted from April to September, 2017 and designed with 3 treatments as chemical fertilizer (39kg/10a), bokashi fertilizer (80 kg/10 a), and slag (200 kg/10 a) ＋bokashi (80 kg/10 a) fertilizer, all with duplicates.

Methane flux was measured by closed chamber method, soil pH was measured with pH meter, and rice quality was measured by taste analyzer.

RESULTS AND DISCUSSION

Pot experiment: Total CH_4 emission of bokashi fertilizer treatment increased about 20% compared with chemical fertilizer treatment, but slag＋bokashi fertilizer treatment decreased about 16% and slag fertilizer decreased about 42% compared with chemical fertilizer treatments, which were much more decrease than previously reported (Furukawa and Inubushi 2004). pH of the percolating water from the slag fertilizer treatment pots were slightly higher than that without slag fertilizer. This was an expected result because there are a lot of alkalinities in slag fertilizer and the pH of slag fertilizer was 12. Among the treatments, slag＋bokashi fertilizer treatment showed a higher score about rice quality. This could be due to the slag fertilizer contains Si, Fe, Mg, P, Ca and some trace elements (Singla and Inubushi 2015).

Field experiment: Total CH_4 emission of bokashi fertilizer treatments increased about 20% compared with chemical fertilizer treatments, but slag fertilizer＋bokashi fertilizer treatments decreased about 10% compared with chemical fertilizer treatments. The result shows that pH of the soil from the slag fertilizer＋bokashi fertilizer treatment was higher than that from other treatments. The score about eating quality was higher with the application of slag fertilizer.

Application of slag fertilizer with the organic fertilizer increased the rice growth and yields compared with that just apply bokashi fertilizer or slag fertilizer, but it was also lower than chemical fertilizer treatments in pot experiment, and the field experiment did not show a big difference among the different treatments.

CONCLUSIONS

Application of slag fertilizer (Minekaru) together with organic fertilizer (bokashi) decreased methane emission from paddy soil, but soil chemical property (pH), rice growth and yields were not significantly influenced, while rice quality was improved.

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REFERENCES

Asami T and Takai Y 1970: Behavior of free-iron oxide in paddy soil (Part 4) Reduction of free-iron and metabolisms of various gases in paddy soil. Jpn. J. Soil Sci. Plant Nutr., 41, **48**, 48-55.

Furukawa Y and Inubushi K 2004: Evaluation of Slag Application to Decrease Methane Emission from Paddy Soil and Fate of Iron. Soil Sci. Plant Nutr., **50** (7), 1029-1036.

Singla A and Inubushi K 2015: Effect of slag -type fertilizers on N_2O flux from komatsuna vegetated soil and CH_4 flux from paddy vegetated soil. Paddy Water Environ., **13**, 43-50.

O-33

Potential of Cyanobacteria Isolated From Different Fresh Water Bodies of Sri Lanka as a Food Supplement

M. S. Malika[2]***, T. Bowanage***[1]***, A. Sumanasinghe***[2]***, R. R. Ratnayake***[1]
[1] National Institute of Fundamental Studies, Kandy, Sri Lanka
[2]Post Graduate Institute of Agriculture, University of Peradeniya, Kandy, Sri Lanka
; shyamamalika@gmail.com

INTRODUCTION

The projected rise of global population from the current 7.2 billion to over 9.6 billion within the next several decades and food requirement also will be gradually increased with population. This onerous target puts enormous pressure on agriculture sector to achieve the food security (Lum et al., 2013). But such a big gap in food production can be achieved by either bringing more and more land under cultivation or enhancing the production of cultivated lands available (Singh *et al.*, 2016). Today food production practices are strongly dependent on intensive tillage, irrigation, application of chemicals (fertilizers and pesticides) which in turn create environmental problem such as deforestation, soil acidity, soil erosion, concentration of heavy metals, irrigation problems and climate change. The potential of cyanobacteria photosynthesis for the production of variable compounds or for energetic use is widely recognized due to their more efficient utilization of energy as compared to higher plants (Priyadarshani and Rath, 2012). Cyanobacteria produce a wealth of high-value bio products and have been mass cultivated for centuries as a nutritional supplement (Möllers *et al.*, 2014). The first use of microalgae by human dates back 2000 years to the Chinese, who used *Nostoc* to survive during famine (Priyadarshani *et al.*, 2012). Today there are numerous commercial applications of cyanobacteria as they can be used to enhance the nutritional value of food and animal feed owing to their chemical composition. Therefore the overall objective of this study was to investigate the potential use of some selected fresh water cyanobacteria isolated from fresh water bodies of Sri Lanka as a food supplement.

MATERIALS AND METHODS

Cyanobacteria contain water samples were collected representing three climatic zones (wet, dry and intermediate zone) of Sri Lanka and cultivated them using BG 11 and GO media. Repeated subculturing in liquid and solid media were used to purifying monocultures. Semi mass culturing of monocultures were carried out by 10L size aspiratory bottles. Cyanobacteria biomass was harvested 40 days after semi mass culturing. Collected algal pellets were transferred into petri plates and placed for oven-drying overnight at 50 ^{o}C. The nutritional composition of cyanobacteria including total carbohydrate, total protein, total lipid, individual sugars, and vitamin C and essential macro- and microelements were quantified.

RESULTS AND DISCUSSION

Eleven mono cultures(*Phormidium*sp., *Cephalothrixkomarekian*, *Chrooccales* sp., *Planktolyngbia* sp., *Cephlothrix* sp., *Microcoleus* sp., *Oscillatoria* sp., *Mycrocystis* sp., *Synechococcus* sp., *Pseudoanabaena* sp., and *Dermocarpa* sp.) were obtained by repeated sub- culturing in both, solid and liquid media. The percentage of total carbohydrate in cyanobacteria dry biomass varies between 5.67%-51.94%. *Oscillatoria* sp. and *Pseudoanabaena* sp. had significantly higher values of 51.94% and 51.12%, respectively. A study carried out by (Rajeshwari and Rajashekar 2011) reported that carbohydrate content of cyanobacteria sp. S. bohneri and O. *foreaui* ranged between 8.0% and 28.4%. In nitrogen fixing cyanobacteria 16-38% of carbohydrate content was reported (Vargas *et al.*, 1998). Our study showed a very high amount of carbohydrate compared to the previous reports. Total protein content ranged between 2.45% to 69.47%. *Microcoleus* sp. show significantly higher value compared to other cyanobacteria strains.Total lipid content of eleven strains ranged between 3.85%-34.53%. *Phormidium*sp. had significantly high lipid content (Rajeshwari and Rajashekhar, 2011). Different aquatic habitats of cyanobacteria had the total lipid content range from 10 – 20% (Rajeshwari and Rajashekhar, 2011). Our study showed higher content of lipids than the previous studies. The study showed that cyanobacteria contain individual sugars such as galactose, rhamnose, glucose, arabinose and fructose.Cyanobacterialbiomasses contained a significantly high amount of galactose and glucose. Range of vitamin C was 0.0015 mg/100g – 3.3761 mg/100g in fresh biomass of cyanobacteria. However the content found in oven dried biomass was low. The selected strains were rich in macro- and microelements such as Ca, Mg, K, Zn, Mn, Co and Fe.

CONCLUSION

Out of this eleven cyanobacteria strains, *Oscillatoria* sp., *Cynechococcus* sp.and *Pseudoanabaena* sp. were rich in carbohydrates. *Planktolyngbia* sp., *Cephlothrix* sp. and *Microcoleus* sp. were rich in protein. Both *Cynechococcus* sp. and *Pseudoanabaena* sp. were rich as a lipid source. Considering macroelements *Cynechococcus* sp. and *Pseudoanabaena* sp. rich as a Ca source. Microcoleus sp. and Oscillatoria sp. suitable as Mg and K source. Microelements (Zn, Mn, Co, Cu and Fe) concentrations were high in all eleven cyanobacteria strains.

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REFERENCES

Lum, K. K., Kim, J. and Lei, X. G. (2013) 'Dual potential of microalgae as a sustainable biofuel feedstock and animal feed', *Journal of Animal Science and Biotechnology*. doi: 10.1186/2049-1891-4-53.

Möllers, K. B. *et al.* (2014) 'Cyanobacterial biomass as carbohydrate and nutrient feedstock for bioethanol production by yeast fermentation', *Biotechnology for Biofuels*, 7(1). doi: 10.1186/1754-6834-7-64.

Pr**iyadarshani, I. and Rath, B. (2012)** 'Commercial and industrial applications of micro algae – A review', *Journal of Algal Biomass Utilization*, 3(4), pp. 89–100.

Rajeshwari, K. R. and Rajashekhar, M. (2011) 'Biochemical Composition of Seven Species of Cyanobacteria Isolated from Different Aquatic Habitats of Western Ghats ', *Brazilian Archives of biology and Technology,* 54(October), pp. 849–857. doi: 10.1590/S1516-89132011000500001.

S**ingh, J. S. *et al.* (2016)** 'Cyanobacteria: A precious bio-resource in agriculture, ecosystem, and environmental sustainability', *Frontiers in Microbiology*. doi: 10.3389/fmicb.2016.00529.

V**argas, M. A. *et al.* (1998)** 'Biochemical composition and fatty acid content of filamentous nitrogen-fixing cyanobacteria', *Journal of Phycology*, 34(5), pp. 812–817. doi: 10.1046/j.1529-8817.1998.340812.x.

Poster Presentation

P1- Physical and chemical properties of low pH soil.

P1-1

Higher cation exchange capacity determined lower critical soil pH and higher Al concentration for soybean

Baquy M. A. A[1]. and R. K. Xu [1]
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Science, Nanjing 210008, China
; rkxu@issas.ac.cn

INTRODUCTION

Crop production is a big challenge in acid soils to meet the food demand of increased population in the world as well as China. Soil pH and exchangeable Al are the important soil parameters for acid soil management. Critical soil pH and Al concentration will help judicious application of liming materials in acid soils for better crop growth. The sensitivity of crop to acidity varies with plant species, cultivar within plant species and soil types (Kariuki et al. 2007). Thus, the critical soil pH and Al concentration must be investigated to better understand which acid soils need to be ameliorated or not. Our assumption was that there is different critical soil pH and Al content for different acid soils derived from different parent materials for soybean crop. Therefore, to justify our hypothesis, greenhouse soybean pot culture was conducted for the soils derived from different parent materials.

MATERIALS AND METHODS

Four sets of pot experiment with four different soils (Table 1) derived from different parent materials were established in greenhouse for soybean culture.

Table.1 Some basic characteristics of four soils. Soil properties are characterized by organic matter (OM), cation exchange capacity (CEC), exchangeable Al^{3+} (Ex. Al^{3+}), and exchangeable base cations.

Soil	Location	Parent material	Soil pH	OM	CEC	Ex. Al^{3+}	Exchangeable base cations				
							Ca^{2+}	Mg^{2+}	Na^{+}	K^{+}	Total
				g kg^{-1}	------------------------- cmol$_c$/kg --------------------------						
Alfisol	Nanjing	Loess deposit	4.71	23.10	18.2	4.51	2.27	1.44	0.009	1.01	4.72
Ultisol	Langxi	Quaternary red earth	3.97	18.17	12.5	5.24	0.60	0.32	0.01	0.50	1.43
Ultisol	Jing	Granite	4.69	24.00	9.53	2.04	1.03	0.43	0.07	0.03	1.56
Ultisol	Yingtan	Tertiary red sandstone	4.98	6.42	4.03	1.24	0.33	0.22	0.04	0.08	0.67

A pre-existing soil pH gradient was considered as different treatments for this study. The experiment was established in a complete randomized design with three replications for each soil. The 3.5 kg soils were used in each pot. Locally grown soybean variety Xudou-14 was used as test crop. Eight 3-day germinated seeds were sown in each pot. After 1 week, the seedlings were reduced to 5 in each pot to make the uniformity. Regular and sufficient distilled water was given at 70% field water holding capacity in each pot. After 30 days of growing, the crop was harvested. Plant height, chlorophyll content, shoot and root dry matter yield were measured. Soil samples were collected after crop harvest from each pot separately, dried at ambient temperature, and ground to pass through a 0.3-mm sieve for determining soil pH, exchangeable acidity and bases.

RESULTS AND DISCUSSION

An inverse exponential significant relationship occurred between soil pH and Al concentration as well as Al saturation at all sites. There was strong correlation existed between soil pH and Al concentration with r^2 of 0.92, 0.94, 0.96, and 0.92 for the soils from Nanjing, Langxi, Jing, and Yingtan, respectively. The correlation between soil pH and Al saturation was quite strong with r^2 of 0.92, 0.95, 0.83, and 0.88 at Nanjing, Langxi, Jing, and Yingtan, respectively. At given soil pH, the Al concentration and Al saturation varied at all sites.

The results indicated that plant growth parameters were affected adversely due to Al toxicity at low soil pH level in all soils. The critical soil pH varied with soil type and parent materials. They were 4.38, 4.63, 4.74, and 4.95 in the Alfisol derived from loss deposit, and the Ultisols derived from Quaternary red earth, granite and Tertiary red sandstone, respectively. The critical soil exchangeable Al was 2.42, 1.82, 1.55, and 1.44 $cmol_c\ kg^{-1}$ for the corresponding soils. At 90% yield level, the critical Al saturation was 6.94, 10.36, 17.79, and 22.75% for the corresponding soils (Fig. 1).

The variation in critical soil pH and exchangeable Al among soybean plant growth parameters was noticed for a particular soil as well as among the soils. The lower critical soil pH and higher Al concentration were detected in the Alfisol among all soils. However, higher critical soil pH and lower Al concentration were found in Tertiary red sandstone derived Ultisol among all soils. The critical values varied among the soils were due to different soil types and their CEC. The Alfisol has the highest CEC, followed by the Ultisols from Quaternary red earth and granite, while the Ultisol from Tertiary red sandstone has the lowest CEC (Table 1), which was consistent with the critical soil pH and exchangeable Al values. Thus, the higher soil CEC led to the lower critical soil pH and the higher critical soil exchangeable Al. Similar phenomenon is observed for Al saturation (Fig 1). It is well documented that soil base cations can reduce Al toxicity to crop plants (Merino-Gergichevich et al. 2010). Higher base cation saturation and greater exchangeable base cations in the Alfisol can reduce Al toxicity to soybean roots compared with three Ultisols. Therefore, the above reasons were responsible for lower critical soil pH and higher Al concentration in the Alfisol than these of three Ultisols for soybean.

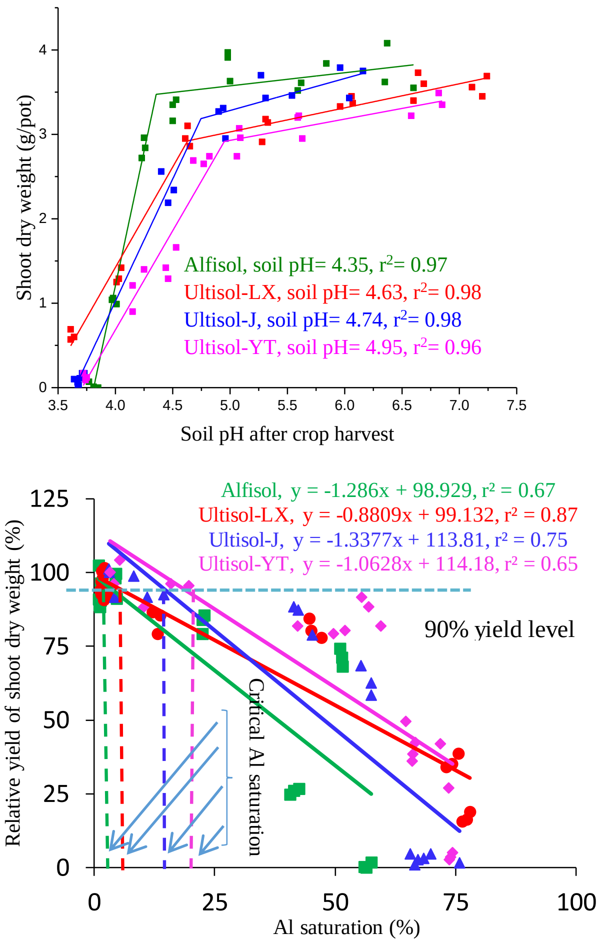

Fig. 1 Shoot dry weight and relative yield of soybean as function of soil pH and Al saturation. The fitted equations were significant at $P < 0.01$.

CONCLUSION

Greater soil CEC and exchangeable base cations led to the lower critical soil pH and Al saturation, and higher soil exchangeable Al. Therefore, we recommended that critical soil pH, soil exchangeable Al and Al saturation should be considered during judicious liming approach for soybean production.

ACKNOWLEDGEMENTS

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REFERENCES

Kariuki S. K., H. Zhang, J. L. Schroder, J. Edwards, M. Payton, B. F. Carver, W. R. Raun, E. G. Krenzer. 2007. Hard red winter wheat cultivar responses to a pH and aluminum concentration gradient. Agronomy Journal 99: 88–98.

Merino-Gergichevich C., M. Alberdi, A. G. Ivanov, M. Reyes-Diaz. 2010. Al^{3+}-Ca^{2+} interaction in plants growing in acid soils: al-phytotoxicity response to calcareous amendments. Journal of Soil Science and Plant Nutrition **10**:217–243.

P1-2

Does the rising pH of growth medium increase the tolerance or sensitivity of organisms to aluminum?

***Xue Qiang Zhao*[1], *Xiao Ying Dong*[1], *and Ren Fang Shen*[1]**
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China
; rfshen@issas.ac.cn

INTRODUCTION

The solubilization of Al^{3+} from soil minerals is linearly increased with the decreased soil pH lower than 5.0. The increased toxic Al in soil solutions inhibits the growth of organisms in soils, thereby threatening agricultural productivity and ecological stability (Zhao et al. 2014). Undoubtedly, Al toxicity mainly occurs in acid soils. Therefore, most of the investigations on Al toxicity are conducted under acid condition. Nevertheless, this does not imply that the rising pH of growth medium certainly decreases Al toxicity to organisms.

In an early report, Al tolerance in the Arabidopsis mutant is attributed to an Al-induced increase in the rhizosphere pH (Degenhardt et al. 1998). Mainly based on this finding, the increased rhizosphere pH is summarized as a mechanism of Al tolerance in plants in several review papers such as Matsumoto (2000). By contrast, an increasing body of research demonstrates the H^+ alleviation of Al toxicity in plants and bacteria (summarized by Zhao et al. 2017). Does the rising pH of growth medium increase the tolerance or sensitivity of organisms to Al?

Most organisms are sensitive to both Al and acid. The effect of acid stress on organisms can confuse that of Al toxicity. Based on this consideration, we used Al- and acid-tolerant plants and red yeast to research the effects of pH on Al toxicity.

RESULTS AND DISCUSSION

Effects of rising pH of growth medium on the Al toxicity to plants

NH_4^+ alleviates Al toxicity and reduces Al accumulation in rice and *Lespedeza* compared with NO_3^- (Zhao et al. 2009; Chen et al. 2010). NO_3^- uptake by plants increases the pH of growth medium, while NH_4^+ uptake decreases that. The rising pH from 3.5 to 5.0 enhances Al accumulation in rice tips (Zhao et al., 2009). NH_4^+-reduced Al accumulation in roots is attributable to the altered cell wall properties triggered by the decreased pH because of NH_4^+ uptake (Wang et al. 2015).

Effects of rising pH of growth medium on the Al toxicity to microbes

Bacteria are generally sensitive to acid stress, but fungi are adapted to a wider pH range. We chose one yeast strain *Rhodotorula taiwanensis* RS1 that is acid- and Al-tolerant as materials. The increased pH from 3.1 to 4.2 hugely aggravates Al toxicity and accumulation in this strain (Zhao et al. 2017), which is in agreement with the previous report in bacteria (Kinraide and Sweeney 2003).

Possible mechanisms for the increased Al toxicity by rising pH

Under low pH condition, much more Al ions are desorbed from soils into solutions, but these ions cannot be fully adsorbed by cells because H^+ occupies some of Al-binding sites at cell surface and/or increases the positive potential of cell surface (Figure 1).

Under increased pH condition, much fewer Al ions were desorbed from soils into solutions, but much more Al ions are adsorbed by cells because fewer H^+ occupies the Al-binding sites at cell surface and the negative potential of cell surface is increased (Figure 1).

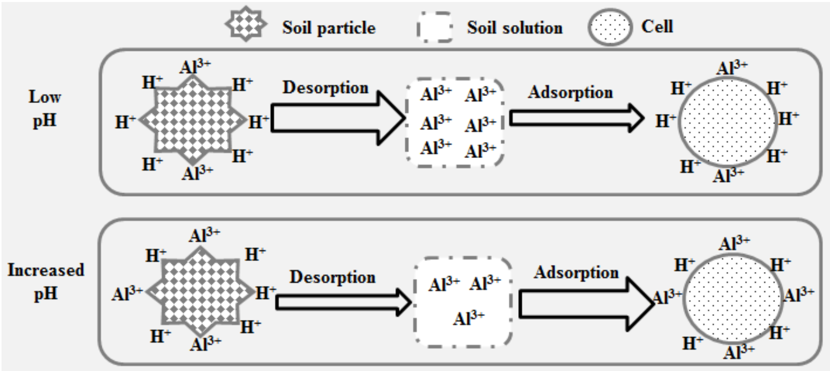

Figure 1 Schematic diagram of possible effects of increased pH on the desorption and adsorption of Al from soil to cell within acid range (lower than pH 5.0). We used Al^{3+} to represent various toxic Al forms and did not indicate the changes of Al forms.

CONCLUSION

The rising pH of growth medium within acid range (lower than pH 5.0) increases Al toxicity to plants and microbes. Although lower pH is the reason for the occurrence of Al toxicity in acid soils, it can also play a certain role in alleviating Al toxicity.

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This study was supported by the National Natural Science Foundation of China (Nos. 31672229, 41230855, and 41271257).

REFERENCES

Chen, Z. C., X. Q. Zhao, and R. F. Shen. 2010. The alleviating effect of ammonium on aluminum toxicity in *Lespedeza bicolor* results in decreased aluminum-induced malate secretion from roots compared with nitrate. Plant and Soil, **337**:389-398.

Degenhardt, J., P. B. Larsen, S. H. Howell, and L. V. Kochian. 1998. Aluminum resistance in the Arabidopsis mutant *alr-104* is caused by an aluminum-induced increase in rhizosphere pH. Plant Physiology, **117**:19-27.

Kinraide, T. B., and B. K. Sweeney. 2003. Proton alleviation of growth inhibition by toxic metals (Al, La, Cu) in rhizobia. Soil Biology and Biochemistry, **35**:199-205.

Matsumoto, H. 2000. Cell biology of aluminum toxicity and tolerance in higher plants. International Review of Cytology, **200**:1-46.

Wang, W., X. Q. Zhao, R. F. Chen, X. Y. Dong, P. Lan, J. F. Ma, and R. F. Shen. 2015. Altered cell wall properties are responsible for ammonium-reduced aluminium accumulation in rice roots. Plant, Cell and Environment, **38**:1382-1390.

Zhao, X. Q., R. F. Chen, and R. F. Shen. 2014. Co-adaptation of plants to multiple stresses in acidic soils. Soil Science, **179**:503-513.

Zhao, X. Q., R. F. Shen, and Q. B. Sun. 2009. Ammonium under solution culture alleviates aluminum toxicity in rice and reduces aluminum accumulation in roots compared with nitrate. Plant and Soil, **315**:107-121.

Zhao, X.Q, X. M. Bao, C. Wang, Z. Y. Xiao, Z. M. Hu, C. L. Zheng, and R. F. Shen. 2017. Hydroxy-Al and cell-surface negativity are responsible for the enhanced sensitivity of *Rhodotorula taiwanensis* to aluminum by increased medium pH. Archives of Microbiology, **199**:1185-1194.

P1-3

Mechanisms for increasing soil resistance to acidification by long-term manure application

***Shi, R. Y*[1,2]. *and R. K. Xu*[1]**
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China
[2]University of the Chinese Academy of Sciences, Beijing 100049, China
; rkxu@issas.ac.cn

INTRODUCTION

Soil acidification has been greatly accelerated in recent decades due to acid deposition and the excessive application of NH_4^+-base fertilizers. It is important to slow down soil acidification for sustainable agriculture in tropical and subtropical regions where the soils pH buffering capacity (pHBC) is low. The long-term application of organic manure can increase soil organic matter and CEC, and thus should increase soil pHBC and inhibit soil acidification. However, the effects and mechanisms of manure application on the soil pHBC have not been reported yet. The objectives of this study were to investigate: (i) the effect of manure application on pHBC and the resistance of soils to acidification, and (ii) the mechanisms by which manure application resist soil acidification.

MATERIALS AND METHODS

Samples of an Alfisol derived from Triassic limestone and sandy shale were collected from a long-term experimental maize field in Guiyang, Guizhou Province (106°07′ E, 26°11′ N). The long-term Alfisol experiments were established in 1995. The Alfisol samples were collected from five long-term fertilization plots namely: (1) non-fertilization (control), (2) N, P, and K (NPK) chemical fertilizers, (3) cattle manure only (M), (4) 1/2 NPK plus 1/2 M (1/2 NPKM) (50% N from manure, and 50% N from chemical N fertilizer), and (5) total NPK plus total M (NPKM). All soil samples were taken from the surface layer (0-20 cm). After being air-dried, the soil samples were ground to pass through a 0.25 mm sieve for measurement of pHBC and soil basic properties. Their basic properties were shown in Table 1. Soil pHBC was determined by acid-base titration technique (Xu et al. 2012). Simulated acidification experiment was conducted by the addition of various amounts of HNO_3 to examine the effect of manure on soil resistance to acidification (Shi et al., 2017).

RESULTS AND DISCUSSION

The application of manure increased pHBC and the resistance of soils to acidification in the Alfisol. The pHBC of the Alfisol in the M and NPKM treatments was increased by 81 and 60% compared with the control, respectively. The extent of protons consumption by the Alfisol followed the order: M > NPKM > 1/2NPKM > NPK ≈ control during stimulated acidification, which was consistent with their pHBC. These results suggest that manure application increased the resistance of the Alfisol to acidification by increasing soil pHBC.

Application of manure increased soil organic matter content and CEC significantly compared with control and thus increased soil pHBC (Table 1), which was consistent with previous reports (Aitken, 1992; Nelson and Su, 2010). The protonation of organic anions from the dissociation of weakly acidic functional groups on soil organic matter was the main mechanism responsible for the increase in pHBC and soil resistance to acidification induced by manure application. This mechanism was confirmed by the experimental observations: the release of base cations from soils increased, while soil exchangeable base cations and effective CEC decreased with as soil pH decreased. The anions of weak acid groups combined with H^+ to form neutral molecular, which led to the decrease in negative charges on soil surface.

Table 1 Properties of the soil samples from different long-term fertilization field treatments in the Alfisol. Values in the Table are presented as means ± standard deviation (n=3)

Treatments	**pH**	**OM %**	**CEC cmol kg^{-1}**	**pHBC mmol kg^{-1} pH^{-1}**
Control	6.53±0.08d	3.66±0.20bcd	19.40±1.57b	31.30±1.99c
NPK	6.25±0.08e	3.56±0.52cd	19.38±0.73b	33.04±1.88c
1/2 NPKM	7.09±0.02c	4.84±0.53abc	21.72±1.22ab	45.92±0.17b
NPKM	7.27±0.03b	5.01±0.49ab	23.30±0.59a	49.95±2.48b
M	7.44±0.03a	5.90±0.56a	23.82±1.36a	56.61±0,18a

OM: organic matter; CEC: cation exchange capacity; pHBC: the pH buffering capacity. Different letters in the same column show significant difference among the treatments ($P \leq 0.05$).

CONCLUSIONS

The application of manure can effectively increase soil pHBC due to the increase in soil organic matter content and CEC, and thus inhibited soil acidification. The protonation of organic anions from the dissociation of acidic functional groups on soil organic matter was the predominant mechanism responsible for the increase in soil resistance to acidification. The decrease in soil effective negative charge with decreasing soil pH confirmed this mechanism.

ACKNOWLEDGEMENTS

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REFERENCES

Aitken, R.L. 1992. Relationships between extractable Al, selected soil properties, pH buffer capacity and lime requirement in some acidic Queensland soils. Australian Journal of Soil Research **30**: 119-130.

Nelson, P.N., N. Su. 2010. Soil pH buffering capacity: a descriptive function and its application to some acidic tropical soils. Australian Journal of Soil Research **48**: 210-207.

Shi, R.Y., Z.N. Hong, J.Y. Li, J. Jiang, M.A. Baquy, R.K. Xu, W. Qian. 2017. Mechanisms for increasing the pH buffering capacity of an acidic Ultisol by crop straw derived biochars. Journal of Agricultural and Food Chemistry **65**: 8111-8119.

Xu, R.K., A.Z. Zhao, J.H. Yuan, J. Jiang. 2012. pH buffering capacity of acid soils from tropical and subtropical regions of China as influenced by incorporation of crop straw biochars. Journal of Soils and Sediments **12**: 494-502.

P1-4

Assessing soil properties of different landuse in Bintulu, Sarawak using soil fertility indices

Aiza Shaliha Jamaluddin[1]*, Arifin Abdu[1], Daljit Singh Karam[2], Shamshuddin Jusop[1] and Aizul Azfar Zulkeefli[2]
[1]Department of Forest Management, Faculty of Forestry,
[2]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 4300 Serdang, Selangor, Malaysia
; aiza.jamaluddin@gmail.com

INTRODUCTION

Soil fertility indices can assist in determining the impact of agriculture practices and forest management on soil properties. The evaluation of soil quality is complicated as there are many aspects of soil that can affect its fertility; soil properties ranging from physical, chemical and biological factors, all have its own role in ensuring soil fertility (Doran and Parkin, 1994). The objectives of this study were to examine the effect of soil compaction/hardness on soil properties, the soil fertility status based on soil indices and to assess the nutrient status of soil under different management practices. Therefore, the findings of this study could help in the development of optimal land management methods to ensure sustainable land resources management, thereby improving the livelihood of agrarian communities in the study area and other similar agro-ecological zones.

METHODOLOGY

Study site and soil sampling

This study was conducted at a rehabilitated forest (RF), secondary forest (SF), oil palm plantation (OP) and rubber plantation (RP) of Universiti Putra Malaysia Bintulu Campus in Sarawak, Malaysia. Soils were sampled in plots of 20 m x 20 m for each site. Each plot was divided into two subplots. Soil samples were collected at two depths, surface (0-15 cm) and subsurface (15-30 cm). Soil fertility index (SFI) (Moran *et al.* 2000) and soil evaluation factor (SEF) (Lu *et al.*, 2002) were used to evaluate the soil fertility of these four different land uses. The SFI and SEF indices were calculated to quantify the intensity of land degradation in the study area as stated:

SFI = pH + Organic matter (% dry soil basis) + Available P (mg kg^{-1} dry soil) + Exchg. K ($cmol_c kg^{-1}$) + Exchg.Ca ($cmol_c kg^{-1}$) + Exchg. Mg ($cmol_c kg^{-1}$) – Exchg. Al ($cmol_c kg^{-1}$) ……………………………..Equation 1

SEF = [Exchg. K ($cmol_c kg^{-1}$) + Exchg.Ca ($cmol_c kg^{-1}$) + Exchg Mg ($cmol_c kg^{-1}$) – log (1 + exchg. Al ($cmol_c kg^{-1}$)] x Organic matter (% dry soil basis) + 5 ………………………..…………………Equation 2

RESULTS AND DISCUSSION

Assessing soil fertility status using soil indices

The SFI and SEF values for surface and subsurface soils are shown in Figure 1 and 2 for all sites. Figure 1 and 2 shows that the value of SFI was higher than the SEF value at both depths. The highest SFI value was at oil palm plantation (11.97) followed by rubber plantation (11.44), rehabilitated forest (9.30) and secondary forest (7.77) for surface soil. While for subsurface soil, the SFI value was highest at rubber plantation (11.03) followed by oil palm plantation (10.66), rehabilitated forest (7.90) and secondary forest (7.18). However, the SEF values were lower compared to SFI values for all sites and depths. The highest SEF value was at rehabilitated forest (5.70) followed by oil palm (5.29), rubber plantation (4.31) and secondary forest (3.21), respectively, for surface soil. While for subsurface soil, the highest value was at rehabilitated forest (4.56) followed by secondary forest (3.89), oil palm plantation (3.73) and rubber plantation (2.99). The SEF values for subsurface soil for all sites were lower compared to surface soil, except for secondary forests.

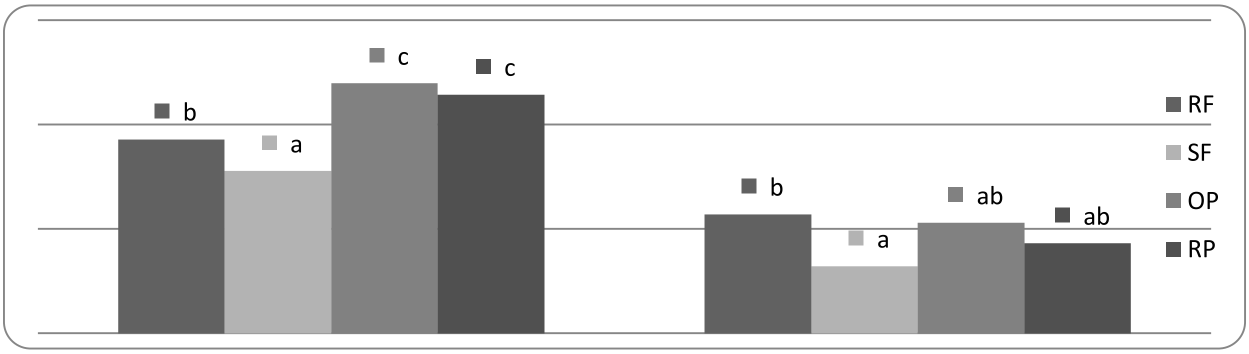

Fig.1. The Soil Fertility Index (SFI) and Soil Evaluation Factor (SEF) comparison for rehabilitated forest (RF), secondary forest (SF), oil palm plantation (OP) and rubber plantations (RP) at surface soils (0-15 cm). Different letter(s) indicate significant differences among sites at $p \leq 0.05$ using Tukey's HSD test

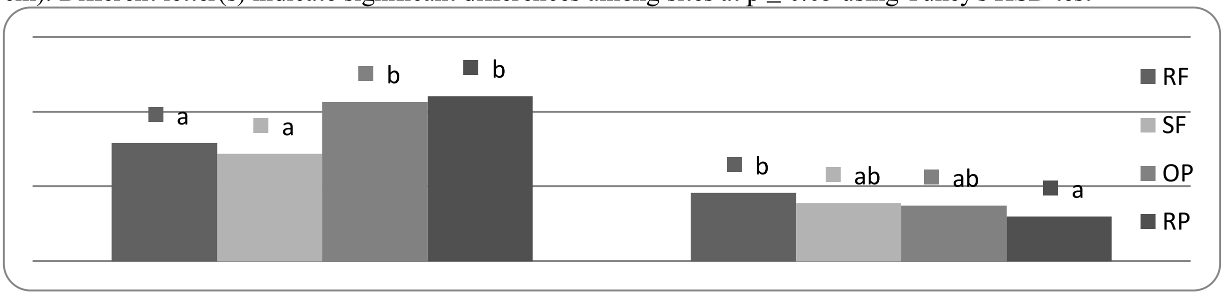

Fig.2. The Soil Fertility Index (SFI) and Soil Evaluation Factor (SEF) comparison for rehabilitated forest (RF), secondary forest (SF), oil palm plantation (OP) and rubber plantations (RP) at subsurface soils (15-30 cm). Different letter(s) indicate significant differences among sites at $p \leq 0.05$ using Tukey's HSD test.

REFERENCES

Doran J.W. and T.B., Parkin, 1994. Defining and assessing soil quality. In Defining Soil Quality for a Sustainable Environment, ed JW Doran, DC Coleman, DF Bezdicekans BA Stewart. SSSA Special Publication.No. 35.pp 3-21.

Lu, D., E., Moran and P., Mausel, 2002.Linking Amazonian secondary succession forest growth to soil properties.LandDeg. Dev., **13:** 331-343.

Moran, E.F., E.S., Brondizio,J.M., Tucker,M.C., Da, Silva-Forsberg and S.D., McCracken et al., 2000. Effects of soil fertility and land use on forest succession in Amazonia.For.Ecol, Manage. **139:** 93-108.

P1-5

Variations in chemical structure of tropical peat soil organic matter and its sensitivity to biodegradation

Faustina Sangok[1]*, *Yuki Sugiura[2]*, *Nagamitsu Maie[3]*, *Lulie Melling[1]*, *Akira Watanabe[2]
[1]Sarawak Tropical Peat Research Institute, Kota Samarahan, Sarawak, Malaysia
[2]Graduate School of Bioagricultural Sciences, Nagoya University, Nagoya, Japan
[3]School of Veterinary Medicine, Kitasato University, Aomori, Japan
; faustina270187@gmail.com

INTRODUCTION

Tropical peatland is important in global carbon (C) cycle and has been acting as C sink since millennia. In Malaysia, the peatland area is 2.7 Mha (Mutalib et al., 1992), and 64% is found in Sarawak (Tie and Kueh, 1979). Since 1990s, peatland has been actively reclaimed, and in Malaysia, 660,000 ha of peatland area has been converted for oil palm plantation (Wahid et al., 2010). The chemical structure and stability of soil organic matter (SOM) in tropical peatland differ depending on vegetation and topo-hydrological characteristics. Land use change from peat forest to agricultural land cause the near bare land to expose to a higher temperature, and soil pH is changed because of fertilizer application and liming. However, the sensitivity of peat SOM to changes in soil pH and increase in temperature is not well understood. The objective of this study is to evaluate the sensitivity of the SOM in the tropical peat soils to biodegradation under different landuse.

MATERIALS AND METHODS

Peat soil samples (0–25 cm depth) were collected from Mixed Peat Swamp forest under different land use, i.e., primary forest (PF), secondary logged-over forest (SF), oil palm plantation (OPP), and sago palm garden (SG) in Sarawak, Malaysia. ^{13}C CP/PASS NMR spectra were obtained and C composition were estimated for all the soil samples. The soil samples were incubated under 3 treatments, i.e., Control (25°C, native pH ca. 3), Neutralization (25°C, increase to pH 7), and High Temperature (35°C, native pH ca. 3) for 91 days in 100 mL Erlenmeyer flask. The concentration of CO_2 gas was measured using GC-TCD every 7 or 14 days. The significant differences between different land use were determined using one-way ANOVA.

RESULTS AND DISCUSSION

Carbon composition as estimated from the ^{13}C CP/PASS NMR spectra in Table 1 showed % alkyl C was the largest in PF likely due to waxy lipids and lignin compounds from the woody materials in peat soils (Nierop et al., 2001). % *O*-alkyl C was larger in SF, OPP, and SG soils, suggesting the increase of labile organic matter content derived from polysaccharides after landuse change. % aromatic C did not show conspicuous change under different land use. Alkyl C/O-alkyl C ratio decreased in the order of PF>SG>SF>OPP.

Table 1: Composition of soil samples used

Land use‡	% Alkyl C (0–45 ppm)	% *O*-alkyl C (45–110 ppm)	% Aromatic C (110–160 ppm)	% Carboxyl C (160–190 ppm)	% Ketone C (190–220 ppm)
PF	35.2	26.5	23.7	11.0	3.7
SF	25.9	32.2	24.8	13.1	3.9
OPP	24.7	31.4	25.5	14.4	4.0
SG	29.2	30.9	24.3	11.3	4.3

‡PF = primary forest, SF = secondary logged-over forest, OPP = oil palm plantation, and SG = sago palm garden

Mean CO_2 flux from PF, SF, OPP, and SG were in the range of 12.4–26.0, 14.8–30.7, 7.3–14.3, and 19.2–62.0 mg C kg^{-1} day^{-1}, respectively (Table 2). The rate of C decomposition increased significantly after neutralization and high temperature treatments in the SG soils, while in the PF soils, Neutralization decreased the rate of C decomposition by 2 times. High temperature showed no effect on the rate of C decomposition in PF soils, while in SF, OPP, and SG soils, the rate of C decomposition increased by 2 times. This finding suggested that the differences in the environmental conditions and management practices at the study sites contributed to the variations in the peat SOM and their sensitivity to biodegradation process.

Table 2: Mean CO_2 flux during 91-day period of biodegradation (mg C kg^{-1} day^{-1})

Land use	Control	Neutralization	High Temperature
PF	26.0 ± 7.5 bcd†	12.4 ± 10.4 cd	25.0 ± 8.7 bcd
SF	14.8 ± 1.3 cd	30.7 ± 2.4 bc	25.1 ± 2.8 bcd
OPP	7.3 ± 0.3 d	14.3 ± 2.0 cd	12.6 ± 2.9 cd
SG	19.2 ± 6.9 cd	62.0 ± 13.3 a	40.6 ± 4.6 b

†Values followed by different letter differ at $p < 0.05$.

CONCLUSION

The present study showed the variations in the sensitivity of peat SOM towards biodegradation under different landuse. Further analysis of peat soils from different forest types and their microbial biota is important to enhance the understanding of biodegradation process in tropical peat soils.

REFERENCES

Mutalib, A.A., Lim, J.S., Wong, M.H., and Koonvai, L. 1992. Characterization, distribution and utilization of peat in Malaysia, in: Tan, S.L. et al. (Eds.) Tropical Peat. Proceedings of the International Symposium on Tropical Peatland, 6–10 May 1991, Kuching, Malaysia, pp.7–16.

Nierop, K. G., van Lagen, B., and Buurman, P. 2001. Composition of plant tissues and soil organic matter in the first stages of a vegetation succession. Geoderma 100, 1–24.

Tie, Y. L., and Kueh, H. S., 1979. A review of Lowland Organic Soils of Sarawak, Technical Paper No. 4, Department of Agriculture, Sarawak, Malaysia, pp. 1–34.

Wahid, O., Aziz, N. A., Mohammed A. T., Harun, M. H., and Din, A. K., 2010. Mapping of Oil Palm Cultivation on Peatland in Malaysia. MPOB Information Series No. 473, ISSN 1511–7871.

P1-6

The effect of biochars application on agricultural soil properties in Cameron Highlands

***Khasifah, M*[1].**
[1]Malaysian Agricultural Research and Development Institute (MARDI), Persiaran MARDI-UPM, 43400 UPM Serdang, Selangor, Malaysia
; khasifah@mardi.gov.my

INTRODUCTION

Soils of the tropics are dominated by Oxisols and Ultisols (Ishak and Jusop, 2010). These highly weathered soils have a low pH, and have a mineralogy dominated by quartz, kaolinite clays and higher oxides contents (Wambeke, 1992; Ishak and Jusop, 2010; Zhao *et al.*, 2014). These characteristics result in low nutrient availability, in particular the fixation and deficiency of phosphorus (P) (Cross and Schlesinger, 1995). Tropical soils often have a low cation exchange capacity (CEC) and in some cases an anion exchange capacity, (Yamato *et al.*, 2006; Masulili *et al.*, 2010), which may result in the loss of N through leaching. Nitrogen fertilizer applications tend to be higher and the efficiency of the fertilizer is lower, because of the leaching process (Cahn *et al.*, 1993; Thomsen *et al.*, 1993). In addition, the Al and Fe concentrations are higher in the soil solution as a result of the weathering process (Cornelissen *et al.*, 2013; Alling *et al.*, 2014; Kloss *et al.*, 2014), and this can cause Al toxicity to the plants.

Furthermore, the long-term intensive farming system on the arable land have decreased the carbon content of the soils in the tropics (Lal, 2004; Lal, 2006; Lal, 2009) decreasing in the fertility of tropical agricultural soil. The decline in tropical soil fertility is due to soil erosion along with the loss of organic matter (Mekuria and Noble, 2013). According to Lal (2004) and Cerri *et al.* (2007), soils in the tropics have lost approximately 20 to 80t C ha^{-1}, and most of the carbon is released into the atmosphere.

Building soil carbon content is important for a number of reasons, including promoting the structural stability of the soil, increasing water retention and infiltration, increasing carbon fixation and reducing soil erosion (Victoria *et al.*, 2012). Ultisols and Oxisols soils are known to have a low percentage of organic matter (1% to 2%) (Ishak and Jusop, 2010), and this is not sufficient for crop growth (Ishak and Jusop, 2010). Consequently, an organic amendment is needed to enhance the soil quality and productivity (Ishak and Jusop, 2010). There are a number of organic materials (carbon sources) available to farmers such as manure and compost.

Biochar is one of these organic amendment sources and has been suggested as one way to improve the quality of soils. Biochar has attracted significant research interest into the effects of biochar application on soil in recent years, especially in the temperate region that has demonstrated that the addition of biochar to soils can have positive or negative effects on the soil and crops. However, the literature on tropical soils following biochar addition is scarce. Therefore, the aim of the study was to investigate the effects of biochar addition on soil properties and nutrient leaching on three tropical soils with different gradients of degradation (forest, non-intensive farming and intensive farming soils).

MATERIALS AND METHODS

To investigate the effect of biochar on the soil characteristics, the soils were amended with and without 2% of coconut shell (CS) and rice husk (RH) biochars by weight and incubated for up to 360 d. To assess the impact of biochar on soil leaching, 27 unplanted soil columns were amended with and without 2% CS and RH biochars by weight. Five leaching events were conducted by leaching 100 mLof deionised water through each of the soil columns. The physical and chemical properties of the soils are demonstrated in Table 1.

Table 1: Physical and chemical properties of soils and biochars

Soils and biochars	Forest Soil	Intensive Farming Soil	Non – Intensive Farming Soil	CS Biochar	RH Biochar
% Clay	28.63	35.06	33.89	-	-
% Silt	11.01	18.09	18.08	-	-
% Sand	60.36	46.86	48.03	-	-
Texture	Sandy Clay Loam	Sandy Clay	Sandy Clay	-	-
% Carbon	3.42	1.64	1.08	72.95	38.64
% Nitrogen	0.18	0.22	0.07	0.53	0.53
C/N Ratio	19.05	7.38	14.99	139.71	72.59
CEC (meq 100 g^{-1})	9.7	9.4	5.8	31.17	43.28
pH	4.62	5.03	5.52	8.33	8.46
Inorganic P (mg g^{-1})	0.06	1.85	0.2	0.39	1.75

RESULTS AND DISCUSSION

The biochar addition significantly increased ($P<0.05$) the soil pH and total carbon, but had marginal effects on the CEC and had a limited effect on the water retention. The biochar application had no effect on total nitrogen, phosphate and soil aggregation. Adding biochar to soil reduced ammonium leaching in the forest soil, but had no clear effect on the other two soils. The nitrate and phosphate concentrations in the soil leachate exhibited an inconsistent pattern after the addition of the biochars.

CONCLUSION

Results showed that biochar application at a lower rate has some effects on soil process, but these effects are not strong enough to recommend biochar as a soil conditioner for the soils studied. Therefore, further research is needed to determine whether higher application rates might make a real impact on tropical soil.

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REFERENCES

Alling, V., Hale, S. E., Martinsen, V., Mulder, J., Smebye, A., Breedveld, G. D. & Cornelissen, G. 2014. The role of biochar in retaining nutrients in amended tropical soils. *Journal of Plant Nutrition and Soil Science* **177**(5): 671-680.

Cahn, M. D., Bouldin, D., Cravo, M. & Bowen, W. 1993. Cation and nitrate leaching in an Oxisol of the Brazilian Amazon. *Agronomy Journal* **85**(2): 334-340

Cerri, C. E. P., Sparovek, G., Bernoux, M., Easterling, W. E., Melillo, J. M. & Cerri, C. C. 2007. Tropical agriculture and global warming: impacts and mitigation options. *Scientia Agricola* 64(1): 83-99.

Cornelissen, G., Martinsen, V., Shitumbanuma, V., Alling, V., Breedveld, G. D., Rutherford, D. W., Sparrevik, M., Hale, S. E., Obia, A. & Mulder, J. 2013. Biochar effect on maize yield and soil characteristics in five conservation farming sites in Zambia. *Agronomy* **3**(2): 256-274.

Cross, A. F. & Schlesinger, W. H. 1995. A literature review and evaluation of the. Hedley fractionation: Applications to the biogeochemical cycle of soil phosphorus in natural ecosystems. *Geoderma* **64**(3): 197-214.

Ishak, C. F. & Jusop, S. 2010. *Weathered tropical soils: the ultisols and oxisols.* Serdang: Universiti Putra Malaysia Press.

Kloss, S., Zehetner, F., Wimmer, B., Buecker, J., Rempt, F. & Soja, G. 2014. Biochar application to temperate soils: Effects on soil fertility and crop growth under greenhouse conditions. *Plant Nutrient Soil Science* **177**: 3-15.

Lal, R. 2004. Soil carbon sequestration impacts on global climate change and food security. *Science* **304**(5677): 1623-1627.

Lal, R. 2006. Enhancing crop yields in the developing countries through restoration of the soil organic carbon pool in agricultural lands. *Land Degradation & Development* **17**(2): 197-209.

Lal, R. 2009. Challenges and opportunities in soil organic matter research. *European Journal of Soil Science* **60**(2): 158-169.

Masulili, A., Utomo, W. H. & Syechfani, M. 2010. Rice husk biochar for rice based cropping system in acid soil 1. The characteristics of rice husk biochar and its influence on the properties of acid sulfate soils and rice growth in West Kalimantan, Indonesia. *Journal of Agricultural Science* **2**(1): p39.

Mekuria, W. & Noble, A. 2013. The role of biochar in ameliorating disturbed soils and sequestering soil carbon in tropical agricultural production systems. *Applied and Environmental Soil Science* 2013.

Thomsen, I. K., Hansen, J. F., Kjellerup, V. & Christensen, B. T. 1993. Effects of cropping system and rates of nitrogen in animal slurry and mineral fertilizer on nitrate leaching from a sandy loam. *Soil Use and Management* 9(2): 53-57.

Victoria, R., Banwart, S., Black, H., Ingram, J., Joosten, H., Milne, E., Noellemeyer, E. & Baskin, Y. 2012. The benefits of soil carbon. *Emerging issues in our global environment. UNEP Yearbook*.

Wambeke, A. V. 1992. *Soils of the tropics: properties and appraisal.* McGraw Hill.

Yamato, M., Okimori, Y., Wibowo, I. F., Anshori, S. & Ogawa, M. 2006. Effects of the application of charred bark of Acacia mangium on the yield of maize, cowpea and peanut, and soil chemical properties in South Sumatra, Indonesia. *Soil Science and Plant Nutrition* **52**(4): 489-495.

Zhao, X., Wang, J., Xu, H., Zhou, C., Wang, S. & Xing, G. 2014. Effects of crop-straw biochar on crop growth and soil fertility over a wheat-millet rotation in soils of China. *Soil Use and Management* **30**(3): 311-319.

P1-7

Influence of afforestation on soil pH in an ex-tin mine

Ho, W.M*[1]*., Sik, H.S*[1]*., Mohd Ghazali, H*[1]*. and Ang L.H*[1]*.
[1]Forest Research Institute Malaysia (FRIM), 52109 Kepong, Selangor, Malaysia
; howaimun@frim.gov.my

INTRODUCTION

Rehabilitation of degraded areas through afforestation has gained much attention as an environmentally sustainable and economically viable means in recent decades. In addition, afforestation for establishment of forest plantation is more practical in marginal areas with poor soil quality compared to agriculture. This avenue is especially useful to restore degraded areas like previously mines sites or landfills that are contaminated with harmful toxic elements with infertile soils that deter further usage of these areas.

The benefits of afforestation for the purpose of plantation establishment include improvement of aesthetic appearance, reduction of erosion and carbon sequestration with additional benefit of revenue generation. However, the establishment of plantations, particularly with fast-growing trees, may trigger other environmental effects such as requirement of higher soil nutrients and thus changes in soil properties. Berthrong *et al.* (2009) reported that mean pH decreased 0.3 units with afforestation. This study aims to investigate the effects of afforestation using timber species on selected soil properties.

MATERIALS AND METHODS

The study site is located in FRIM Research Station, Bidor, state of Perak about 120km from Kuala Lumpur. Planting and research activities in the station by Forest Research Institute Malaysia has started since year 1999 with the initial objectives to green the area and to become demonstration plots for different forest plantation species on ex-tin mine. Tree stands selected for this study are 10-year-old *Acacia mangium* and 7-year-old *Hopea odorata* on sand tailings.

Soil samples were collected from tree stands of *A. mangium* and *H. odorata* at three different depths using soil core sampler: 0–5cm, >5–10cm and >10–15cm. Three sampling points were selected randomly each from open (unplanted) areas and under tree stands (away from tree stems). Samples collected were kept in sealed bags and transported to laboratories for analysis of pH, carbon (C) and nitrogen (N). C and N were determined with elemental analyser by combustion method and thus results reported are total C and N (in %).

RESULTS AND DISCUSSION

Soil pH under tree stands of *A. mangium* and *H. odorata* as well as in open areas is presented in Table 1. We found that soil under tree stands have recorded average values below pH 5 compared to unplanted areas. Soil pH values in the open and under tree stands are found to be significantly different ($P<0.05$) by soil depth. This finding is in congruence with few others which concluded that forest soil tend to be more acidic soil than grassland or abandoned agriculture fields (Schlesinger 1997; Chapin *et al*. 2002; Falkengren-Grerup *et al*. 2006).

Table 1: Soil pH under tree stands and in open area according to soil depth

Soil depth (cm)	*Acacia mangium*		*Hopea odorata*	
	Open	Stand	Open	Stand
0–5	4.62ᵃ ± 0.19	4.31ᵃ ± 0.08	5.75ᵃ ± 0.07	4.87ᵃ ± 0.06
>5–10	5.32ᵇ ± 0.10	4.59ᵇ ± 0.03	5.75ᵃ ± 0.05	4.89ᵃ ± 0.04
>10–15	5.90ᶜ ± 0.09	4.88ᶜ ± 0.05	5.74ᵃ ± 0.05	5.19ᵇ ± 0.04
Average	5.28 ± 0.64	4.60 ± 0.28	5.75 ± 0.01	4.98 ± 0.18

Note: Different letters indicate significant differences between soil depths at P < 0.05.

Results from Table 1 also indicate a general increasing trend in soil pH whereby it tends to increase with depth. The increase in soil pH across soil depth was found to be significant (P<0.05) for tree stand and open area of *A. mangium*. Soil pH increased with increasing depth probably attributed to leaching by rainfall, and by dissolved carbonic acid and organic acids which remove metal cations, like calcium (Ca^{2+}), potassium (K^{+}) and magnesium (Mg^{2+}), and replace them with H^{+} ions. There are some concerns with regards to soil pH lower than 5.0 including potential associated problems like aluminium and manganese toxicity, calcium deficient, low CEC and low microbial activity (Craswell & Pushparajah 1989).

CONCLUSION

Our results conclude that the two young tree stands of *A. mangium* and *H. odorata* have more acidic soil than unplanted areas with a general trend of increasing pH with soil depth. Continuous monitoring of soil pH in planted areas should be carried out to further understand the development of soil properties including nutrient levels as influenced by tree stand development with age. Caution must also be taken to discourage planting of agriculture crops in similar soil with pH below 5.0 in which aluminium and manganese may reach toxic concentrations. This is because concentrations of most metallic ions increase with soil acidity due to hydrolysis of insoluble oxides in which they usually occur.

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REFERENCES

Berthrong, S.T., Jobba´ Gy, E.G. and Jackson, R.B. 2009. A global meta-analysis of soil exchangeable cations, pH, carbon, and nitrogen with afforestation. Ecological Applications. **19**(8): 2228–2241.

Chapin, F. S., Mattson, P. and Mooney, H. 2002. Principles of terrestrial ecosystem ecology. Springer, New York.

Craswell, E.T. and Pushparajah, E. 1989. Management of acid soils in the humid tropics of Asia. ACIAR Monograph No.13 (mSRAM Monograph No.1).

Falkengren-Grerup, U., Ten Brink, D. and Brunet, J. 2006. Land use effects on soil N, P, C and pH persist over 40–80 years of forest growth on agricultural soils. Forest Ecology Management. **225**: 74–81.

Schlesinger, W.H. 1997. Biogeochemistry: an analysis of global change. Second edition. Academic Press, Boston, Massachusetts.

P1-8

Effect of soil compaction on soil CO_2 flux from tropical peatland

Nur Azima Busman*[1,2]*, Che Fauziah Ishak*[2]*, Muhammad Firdaus Sulaiman*[2]*,
***Nagamitsu Maie*[3] *and Lulie Melling*[1]**
[1]Sarawak Tropical Peat Research Institute, 94300 Kota Samarahan, Sarawak, Malaysia
[2]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia 43400 UPM Serdang, Selangor, Malaysia
[3]School of Veterinary Medicine, Kitasato University, Aomori, Japan
; zima_b67@yahoo.com

INTRODUCTION

Tropical peatland stores a large amount of carbon (C) and is an important C sink. In Malaysia, 25% of the peatland area has been converted for oil palm plantation which requires drainage, compaction and groundwater table (GWT) control. Numbers of publication have addressed the contribution of this conversion to a release of C in the form of CO_2 flux, mainly focused on the effect of drainage. However the information on the effects of peat soil compaction towards those emissions is insufficient. Therefore, the objective of this study is to examine the effect of peat soil compaction through changes in soil bulk density (BD) on soil CO_2 flux.

MATERIALS AND METHODS

Soil samples (50–70cm depth) collected from Mixed Peat Swamp (MPS) forest in Maludam National Park, Betong Division, Sarawak, Malaysia were packed and compacted in a polyvinyl chloride (PVC) pipe (with inner diameter 8.3cm, length 20 cm) to three different BD; 0.14 g cm^{-3}, 0.18 g cm^{-3} and 0.22 g cm^{-}. These soil columns were incubated for three months under a laboratory controlled condition. Redox potential (E_h) probe were installed horizontally at depth of 10 cm, 30 cm and 50 cm from soil surface. Artificial rain event were simulated for each soil column by spraying approximately 150 mL of filtered rain water twice a week to maintain soil moisture. The amount of rain water added was based on the annual mean precipitation recorded at the sampling site. Soil column were pre-incubated for one week to allow the recovery of microbial communities after the sampling disturbances. Soil CO_2 fluxes and E_h were measured on a weekly basis for three months.

RESULTS AND DISCUSSION

Total C and N of the soil sample used in this study are 59% and 1.2%, respectively, while soil pH was 3.3 and ash content was 2.7%. The highest means of soil CO_2 flux rate (mg C m^{-2} h^{-1}) was from soil compacted to BD 0.22 g cm^{-3} (52.81 ± 8.54) followed by 0.18 g cm^{-3} (48.27 ± 5.30) and 0.14 g cm^{-3} (38.31 ± 4.08) .

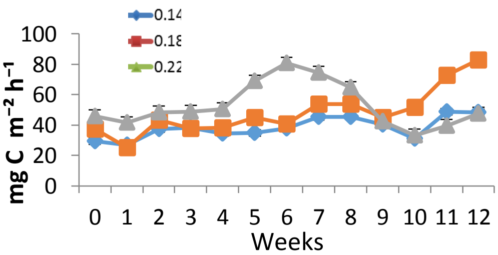

Figure 1: Weekly soil CO_2 fluxes

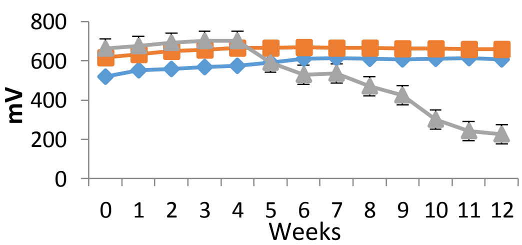

Figure 2: Weekly E_h value at 10 cm depth

Weekly soil CO_2 flux and E_h of the 10 cm depth from different soil BD were shown in Figure 1 and 2, respectively. Soil CO_2 flux from BD 0.22, increased when E_h at 10 cm depth started to decrease at week 5. Soil compaction was known to increase soil BD and reduces the total soil porosity (Silva et al., 2008), which resulted in slower water infiltration rate. In this study, gas sample were taken three days after the artificial rain event and due to the slower water infiltration rate at soil BD 0.22, the percolating rain water fill in and replaces CO_2 in the soil pore space prior to gas sampling, possibly enhance the release of soil CO_2 into the atmosphere.

Soil CO_2 flux remained high until week 8 and started to decrease at week 9 until 12. With continuous application of rain water every week and lower soil porosity, the water remain in the soil pore space longer, resulted in the further lowering of E_h in soil BD 0.22 (Figure 2). When air space in the soil were filled with water, soil O_2 status shifted from aerobic to anaerobic condition, which resulted in low CO_2 flux (Yu and Patric 2004). In this study, E_h value recorded at week 9 to 12 from BD 0.22 ranging from +424 to +226 mV indicates low O_2 level (Pezeshkiez and DeLaune 2012).

In contrast with soil BD 0.22, the water infiltrates uncompacted (BD 0.14) and less compacted (BD 0.18) soil at a faster rate. E_h and soil CO_2 flux from BD 0.14 showed consistent pattern along the incubation period. Meanwhile for soil BD 0.18, soil CO_2 flux showed no significant difference with soil BD 0.14 from week 0 until 9. However sudden increased at week 10 till 12 was possibly due to the water infiltration rate started to be slower compared to the earlier incubation stage.

CONCLUSION

Soil compaction modified soil physical properties such as soil BD, porosity and water infiltration rate which also influence the production of soil CO_2. However, longer period of incubation is needed for better understanding on the effect of compaction towards emission of CO_2.

REFERENCES

Silva S, Barros N, Costa L, Leite F (2008) Soil compaction and eucalyptus growth in response to forwarder traffic intensity and load. Rev Bras Cienc Solo **32:**921–93.

S. R. Pezeshki and R. D. DeLaune (2012) Soil Oxidation-Reduction in Wetlands and Its Impact on Plant Functioning.

P1-9

Soil chemical properties of a high quality-tea production area in Thai Nguyen province, Vietnam

***Chien, H.H[1].*, M. Tokuda[2], D.V. Minh[3], Y. Kang[2], K. Iwasaki[2], and S. Tanaka[1]**
[1]Kuroshio Science Program, Graduate School of Integrated Arts & Sciences, Kochi University,
[2]Faculty of Agriculture, Kochi University, and [3]Faculty of Recourses Management, Thai Nguyen University of Agriculture and Forestry, B200 Monobe, Nankoku, Japan 783-8502 ; chienbk87@gmail.com

INTRODUCTION

Tea (*Camellia sinensis*) is widely planted and its infusion is a popular beverage throughout the world. Because tea quality such as theanine and catechin contents can be improved with increasing amounts of ammonium in soils and because of the nature of tea plants to be Al-tolerant, or Al-phile (Konishi et al. 1985), excessive N fertilizers tend to be applied to tea gardens. As the results of absorption of ammonium by the plants and nitrification in soils, many studies reported severe acidification and rising levels of active Al of soils and the concomitant nutritional imbalances and leaching loss of nutrients (Yokota et al. 2005; Yang et al. 2018).

With tea-drink culture, Vietnam is one of the famous tea producers, as ranked 6th and 11th for export in quantity and value, respectively (FAOSTAT). Thai Nguyen Province is the center for special tea production with high quality. However, the information are still quite lacking on soils of tea gardens in this country; although only Dang (2002) conducted the detailed study, his study sites were placed in Song Cau commune, located in the mountainous region and a peripheral area in terms of tea production in the province. As such, we have launched an international study to find out important soil factors to determine tea quality. This paper will report general physico'chemical properties of tea garden soils in Tan Cuong commune, the core area for tea production in the province with special reference to terrain condition and planting ages.

MATERIALS AND METHODS

Tan Cuong commune was placed in the flood plain on the left side of Cong River. The terrain of tea gardens varied from the flat land to the gently-sloped dikes and river terraces with increasing distance from the river. According to preliminary interview to the owners, tea leaf with higher quality in terms of the farm-gate price is produced at the gardens close to the river. Therefore, three transect lines (named as A, B and C) were established perpendicularly to the river. Four gardens on each line were selected as study sites. Transect A was composed of the current natural dikes (A1), old dikes (A2, A3), and river terrace (A4). Transect B and C were flat lands (B1, C1) and old dikes (B2 to B4, C2 to C4). While A4 and B3 were terraced, B4 was severely disturbed by excavation for constructing material, resulting in rock abundant feature even at the ground surface. The planting ages ranged from 4 years to more than 50 years. Soils were classified as Entisols at flat lands and Ultisols at dikes and river terraces.

Soil samples were collected from the depths of 0-10 cm and 20-30 cm at the middle point between hedges in triplicates. For the comparison, soil samples were also collected from three sites in Song Cau commune. The samples were air-dried and passed through a 2 mm sieve in diameter. Soil pH, EC, Total C and Total N, CEC, exchangeable Ca, Mg, K, Na, Al and H, NH_4^+-N and NO_3^--N, available P and soil texture were analyzed.

RESULTS AND DISCUSSION

Soil texture class ranged from sandy loam to light clay in Tan Cuong and mostly light clay in Song Cau. The 0-10 cm soils were more acidified in Tan Cuong with pH(H_2O) of 3.2 to 4.4. No difference in pH at 20-30 cm was found between two communes. The levels of exchangeable bases were lower and that of exchangeable Al was higher in Tan Cuong than Song Cau.

In Tan Cuong, as a whole, soil texture was finer with distance from the river on the transect lines except for A. In terms of terrain, the soils at the new dike (A1) showed finer texture than those at flat lands (B1, C1). While the soil texture at old dikes (A2, A3, B2, C2 to C4) was coarser than that at the new dike (A1), the disturbed sites by terracing or excavation (A4, B3, and B4) showed higher clay contents than the others, probably due to mixing or exposure of deeper soils with high clay contents. The levels of exchangeable Al were correlated with clay contents (r=0.45 and 0.51 for 0-10 cm and 20-30 cm, respectively) and tended to increase along the transect line. Meanwhile, T-C and T-N were correlated with garden age in spite of disturbance in some gardens (r=0.78 and 0.67 for T-C in 0-10 cm and 20-30 cm, respectively), suggesting build-up of soil organic matter pools through leaving pruned plant materials in the field. In spite of N application as ammonium sulfate or urea, NO_3^--N contents were higher than NH_4^+-N, indicating the occurrence of nitrification under highly acidic condition as reported by other researchers. Significant levels of exchangeable Ca, Mg, and K and mineral N were found in both layers. The contents of available P was high occasionally exceeding 1000 and 500 mg P kg^{-1} in 0-10 cm and 20-30 cm, respectively. These results of exchangeable bases and available P indicate that appreciable amounts of fertilizer nutrients even including P would move down to deeper soil layers.

The gate prices of tea leaf obtained from the interview tended to be lower with increasing clay contents (r=-0.50 and -0.57 for 0-10 cm and 20-30 cm, respectively, not significant). This relation would not mean that the tea quality is directly determined by soil texture but imply that soil texture-related properties such as physical properties, mineralogical properties and the contents of trace elements might be the candidates as the determinant of the tea quality. In general, while the physical properties largely depend on soil texture, mineralogical properties and the contents of trace elements are affected by soil parent material and/or weathering status. In the study area, the latter might reflect the difference in terrains of study sites with the distance from the river. Further analysis should be done for such properties of soils.

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REFERENCES

Dang, M.V. 2002. Effects of tea cultivation on soil quality in the northern mountainous zone, Vietnam. 175pp. Dissertation thesis, University of Saskatchewan.

Konishi, S., S. Miyamoto, and T. Taki 1985. Stimulatory effects of aluminum on tea plants grown under low and high phosphorus supply. Soil Science and Plant Nutrition, **31:** 361-368.

Yang, X., K. Ni, Y. Shia, X. Yi, Q. Zhang, L. Fang, L. Ma, and J. Ruana. 2018. Effects of long-term nitrogen application on soil acidification and solution chemistry of a tea plantation in China. Agriculture, Ecosystems and Environment, **252:** 74-82.

Yokota, H., A. Morita, and F. Ghanati. 2005. Growth characteristics of tea plants and tea fields in Japan. Soil Science and Plant Nutrition, **51:** 625-627.

P1-10

Innovative mudflat corer system for bulk density sampling in mangrove mudflats

***Jeyanny, V*[1]*., Fakhri, M.I*[1]*., Norhisyam, I*[1]*., and Wan Rasidah Kadir*[1]**
[1]Forest Research Institute Malaysia (FRIM), 52109, Kepong, Selangor
; jeyanny@frim.gov.my

INTRODUCTION

Currently, collection of bulk density samples in mangrove mudflats is laborious and sometimes unsuccessful using common stainless corer rings due to structureless soil and water suspension which cannot be retrieved mechanically. This invention facilitates the collection of core/undisturbed samples in an innovative way which prevents contamination, assists bulk density determinations which can be used to compliment soil profile descriptions in mangrove ecosystems. Profile descriptions are important for soil structure determination, vegetation structure analysis and predicting soil movements in mudflats. Besides that, bulk density samples are required to calculate soil carbon stocks in mangrove forest ecosystems. The mudflat corer system is a combination of existing technology with the mud profile sampler (FRIM ID 02-2012) to collect bulk density and undisturbed soil samples from mangrove mudflats. It can collect massive and unstructured soils under inundated waters at required depths.

MATERIALS & METHODS

The mudflat corer is an object made out of polyvinylchloride material which is non-corrosive with a length of 100 cm and a diameter of 5 cm. This corer needs to be used in combination with mud profile sampler. It also has a cap for retrieval of samples. This system is made out of durable polyvinyl chloride material with 5 mm thickness which can prevent damage on the outer layer and can be used in oven drying at high temperature. It uses vacuum principle, which creates a suction effect to retain samples and is cost effective. The steps involved in sampling and the pictorial methods are given in Figures 1.

Fig.1a: Insertion of mudflat sampler

Fig. 1b: Insertion of mudflat corer into sampler with closed cap

Fig. 1c: Retrieval of mudflat corer

Fig. 1d: Mudflat corer cut to 5 cm

RESULTS & DISCUSSION

The mudflat corer system was useful to collect bulk density samples at required depths. Samples were then weighed using a laboratory weighing scale and dried in the oven at 105°C. Determination of bulk density was done using standard methods (Burt, 2011). Values derived from mudflat corer system for bulk density and soil moisture content were comparable to samples collected in the same area using stainless steel core rings in a previous study up to 10 cm soil depth (Jeyanny et al., 2009). This tool potentially can be utilized in slurry soils such as in an ex-tin tailings area.

CONCLUSION

The mudflat corer system is an alternative method to using stainless steel core rings in mudflats to retrieve information on soil bulk density and water content. This invention disclosure of this tool is registered under the Forest Research Institute Malaysia (No: ID4/2017).

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REFERENCES

Burt, R. 2011. Soil Survey Laboratory Information Manual: Soil Survey Investigations Report No. 45. *USDA NRCS, Nebraska*.506p

Jeyanny, V., Suhaimi, W.C., Wan Rasidah, K., Adi, F. & Azian, M. 2009. Preliminary analysis of soil properties of an eroding mangrove shore in Selangor, Malaysia.*GLOMIS* **7**(3):5-6 Available online at http://www.glomis.com/ej/pdf/EJ_7-3.pdf

P2- Physiological, molecular mechanism and plant adaptation to acid soil condition

P2-1

Properties of mineral accumulation of solfatara plants in western Japan

Wasaki, J.[1]***, A. Yamamoto***[1]***, T. Saito***[1]***, H. Tsubota***[2]***, T. Watanabe***[3]***, and T. Nakatsubo***[1]
[1]Graduate School of Biosphere Science, Hiroshima University, Kagamiyama 1-7-1, Higashi-Hiroshima 739-8521, Japan
[2]Miyajima Natural Botanical Garden, Graduate School of Science, Hiroshima University, 1156-2, Mitsumaruko-yama, Miyajima-cho, Hatsukaichi 730-0543, Japan
[3] Research Faculty of Agriculture, Hokkaido University, N9W9, Kita-ku, Sapporo 060-8589, Japan
; junw@hiroshima-u.ac.jp

INTRODUCTION

Many active volcanoes are present in Japan including Kyushu district, located in Western Japan. Strong acid soils (less than pH 4) are frequently found around the region close to fumaroles (volcanic vents). The acid soil around fumaroles, so-called solfatara, limits the plant growth by the low pH itself and Al toxicity. Some specific plants such as *Miscanthus sinensis* (Poaceae) and *Fimbristylis dichotoma* subsp. *podocarpa* (Cyperaceae) were found in solfatara fields (Yoshii 1937, Negoro 1943). However, the properties of mineral accumulation and the tolerance to Al stress of plants species dominated in solfatara fields is not well known. Thus, this study aimed to investigate the properties of mineral accumulation by comparing same species found in solfatara and normal (non-solfatara) fields in Western Japan.

MATERIALS AND METHODS

Study Sites

Study sites are Oita, Kagoshima, and Hiroshima, in Western Japan. 'Oita' located in Eastern Kyushu district and included 3 sites as follows: O1, Myoban Hot Spring (Beppu City, 33°19'N, 131°26'E, about 390 m a.s.l.); O2, Tsukahara Plateau (Yufu City, 33°18'N, 131°25'E, about 930 m a.s.l.); and O3, Yutsubo (Kuju Town, 33°06'N, 131°11'E, about 1,070 m a.s.l.). 'Kagoshima' located in Southern Kyushu district and included 3 sites as follows: K1, Hokonagi Hot Spring (Kirishima City, 31°54'N, 130°48'E, about 720 m a.s.l.); K2, Iwodani Hot Spring (Kirishima City, 31°53'N, 130°50'E, about 690 m a.s.l.); and K3, Kurinodake Hot Spring (Yusui Town, 31°57'N, 130°46'E, about 700 m a.s.l.). 'Hiroshima' located in Western Honshu district and included 4 sites as follows: H1, Hiroshima Univ. (Higashi-Hiroshima City, 34°24'N, 132°42'E, about 220 m a.s.l.); H2, Hachihonmatsu (Higashi-Hiroshima City, 34°25'N, 132°40'E, about 270 m a.s.l.); H3, Toyohira (Kita-Hiroshima Town, 34°38'N, 132°26'E, about 400 m a.s.l.); H4, Kurahashijima Island (Kure City, 34°05'N, 132°27'E, about 10 m a.s.l.); and H5, Miyajima Island (Hatsukaichi City, 34°17'N, 132°17'E, about 20 m a.s.l.).

Plant Materials and Analyses

Soil pH was analyzed (soil: water = 1:2.5). Shoots (*Fimbristylis dichotoma* and *Lycopodium cernuum*) or mature leaves (fully expanded leaves, *Vaccinium bracteatum*) were harvested and air-dried in an oven at 70˚C for 3 days. An aliquot of powdered plant sample was digested by HNO_3-H_2O_2 for mineral analysis. ICP-MS (inductively coupled plasma mass spectrometry; ELAN, DRC-e; Perkin Elmer, Waltham, MA, USA) was used for analysis of 26 mineral elements in digested solution (Chen et al. 2009).

RESULTS AND DISCUSSION

Distribution of Solfatara Plants

We focused on three species as solfatara plants: *Fimbristylis dichotoma* subsp. *podocarpa* (Cyperaceae), *Lycopodium cernuum* (Lycopodiaceae), and *Vaccinium bracteatum* (Ericaceae), because they were frequently found not only in normal environments (excepting *F. dichotoma* subsp. *podocarpa*) but also in many solfatara fields in our study sites. *F. dichotoma* subsp. *podocarpa* was found at O1, O3, K1, K2, and K3 sites. Another subspecies/varieties of *F. dichotoma* var. *tentsuki* was harvested at normal fields at H1 and close to K1 and K3 sites (very far from solfatara). *L. cernuum* was found at O2, O3, K1, K2, and K3 sites in solfatara, and H2, H3, and H4 sites in normal sites. *V. bracteatum* was found at K1 and K3 sites in solfatara, and H1 and H5 sites in normal sites. The average of soil pH was 5.29 ± 0.30, 3.57 ± 0.63, and 3.48 ± 0.33 at Hiroshima, Oita, and Kagoshima sites, respectively.

Mineral Concentration of Solfatara Plants

Table 1 showed the concentration of mineral elements of solfatara plants. S was extremely high in all plants grown solfatara fields, suggesting that the high input of S at fumaroles is the source. P accumulation was also higher in solfatara fields, although it was not significant in *F. dichotoma*. Solubility of inorganic P could be improved at solfatara environments by very low pH. K concentration of *L. cernuum* was high in solfatara, whereas Na was low. It may imply the selective K uptake strategy of *L. cernuum*.

Interestingly, Al concentration was similar level between solfatara and normal fields in each plant. *L. cernuum* highly accumulated Al (around 10 mg/gDW) in their shoots like known Al-accumulators, such as buckwheat (Ma et al. 2001), *Melastoma* (Watanabe et al. 1997). This suggests that *L. cernuum* is an Al accumulator, although the uptake may not be stimulated by high Al^{3+} ions solubilized under low pH. Al concentration of *F. dichotoma* and *V. bracteatum* was low level, suggesting that they had any strategy to avoid Al accumulation into root tissues. Ca and Mn concentration of *F. dochotoma* and *L. cernuum* was significantly low in solfatara than in normal environments. On the other hand, Mg accumulation was increased in all plants in solfatara. It is considered that differences of solubility of these elements reflect to the concentration in plants grown different pH.

Table 1: Concentration of mineral elements in shoots or mature leaves (mg/gDW).

Plant Name	Variety	Field Type	n	Al		Ca		Mg		Mn	
Fimbristylis dichotoma	subsp. *podocarpa*	Solfatara	15	0.549 ± 0.092	n.s.	1.483 ± 0.210	**	1.555 ± 0.168	*	0.098 ± 0.014	**
	var. *tentsuki*	Normal	9	0.387 ± 0.208		6.265 ± 0.393		0.970 ± 0.128		0.231 ± 0.036	
Lycopodium cernuum		Solfatara	18	11.16 ± 1.20	n.s.	1.369 ± 0.135	**	1.915 ± 0.239	n.s.	0.062 ± 0.006	**
		Normal	12	9.75 ± 1.32		2.324 ± 0.172		1.648 ± 0.143		0.210 ± 0.041	
Vaccinium bracteatum		Solfatara	9	0.516 ± 0.058	n.s.	11.41 ± 1.137	n.s.	2.979 ± 0.224	*	0.299 ± 0.065	n.s.
		Normal	4	0.604 ± 0.048		11.6 ± 0.816		1.991 ± 0.145		0.530 ± 0.095	

Plant Name	Variety	Field Type	n	K		Na		P		S	
Fimbristylis dichotoma	subsp. *podocarpa*	Solfatara	15	23.98 ± 1.841	n.s.	0.263 ± 0.125	*	1.974 ± 0.206	n.s.	6.142 ± 0.789	**
	var. *tentsuki*	Normal	9	21.69 ± 4.562		0.035 ± 0.020		1.646 ± 0.280		1.684 ± 0.306	
Lycopodium cernuum		Solfatara	18	13.82 ± 1.19	**	0.326 ± 0.069	*	1.225 ± 0.134	**	3.290 ± 0.269	**
		Normal	12	7.34 ± 0.81		0.896 ± 0.265		0.490 ± 0.075		1.004 ± 0.119	
Vaccinium bracteatum		Solfatara	9	8.330 ± 0.576	n.s.	0.035 ± 0.015	n.s.	0.936 ± 0.052	*	6.363 ± 0.539	**
		Normal	4	7.392 ± 1.203		0.101 ± 0.087		0.682 ± 0.045		1.904 ± 0.101	

CONCLUSION

Fimbristylis dichotoma subsp. *podocarpa* (Cyperaceae), *Lycopodium cernuum* (Lycopodiaceae), and *Vaccinium bracteatum* (Ericaceae) were found in solfatara fields. It is concluded that their strategies to adopt the low pH is varied, especially in Al tolerance. It is expected to clarify the detail strategies to adopt low pH in physiological and molecular aspects in the future.

ACKNOWLEDGEMENTS

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REFERENCES

Chen, Z., T. Watanabe, T. Shinano, T. Ezawa, J. Wasaki, K. Kimura, M. Osaki, and Y.G. Zhu 2009. Element interconnections in *Lotus japonicus*: A systematic study of the effects of element additions on different natural variants. Soil Science and Plant Nutrition **55**:91-101.

Ma, J., P. Ryan, and E. Delhaize 2001. Aluminium tolerance in plants and the complexing role of organic acids. Trends in Plant Science **6**: 273–278.

Negoro, K. 1943. Study on solfatara plants. In: Progress in Biology 1. pp 151-193 Kyoritsu Shuppan, Tokyo.

Yoshii, Y. 1937. Aluminum requirement of solfatara plants. Botanical Magazine Tokyo **51**: 262-270.

Watanabe, T., M. Osaki, and T. Tadano 1997. Aluminum-induced growth stimulation in relation to calcium, magnesium, and silicate nutrition in *Melastoma malabathricum* L. Soil Science and Plant Nutrition **43**: 827–837.

P2-2

Dissecting the genetic architectures of Aluminum tolerance in Arabidopsis thaliana accessions

Yuki Nakano[1], Kazutaka Kusunoki[1], Satoshi Iuchi[2], Masatomo Kobayashi[2], Hiroyuki Koyama[1,3] and Yuriko Kobayashi[1,3]
[1]The United Graduate School of Agricultural Science, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan
[2]Experimental Plant Division, RIKEN BioResource Center, 3-1-1 Koyadai, Tsukuba, Ibaraki 305-0074, Japan
[3]Faculty of Applied Biological Sciences, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan
; k_yuriko@gifu-u.ac.jp

INTRODUCTION

Aluminum (Al) rhizotoxicity is one of the major stress factors of acid soil syndrome. Plants have adapted to acid soil environment by evolving several Al tolerance mechanisms such as organic acid (OA) excretion to the rhizosphere, activation of ROS-scavenging enzymes, and others [1, 2]. Investigation of molecular mechanisms determining the physiological and genetic polymorphisms among natural variation would be one approach for understanding complex Al tolerance. In fact, the polymorphism of important Al tolerance genes with large contribution in Al tolerance variation, such as ALMT1 (Al-activated malate transporter) and MATE (multidrug and toxic compound extrusion), have been identified in several species [1, 3, 4]. However, it remains unclear a series of tolerance genes which explain large part of Al tolerance variation among accessions. In this study, we performed genome wide association study (GWAS) of Al tolerance using approximately 200 accessions of Arabidopsis thaliana, which have adapted to various environments due to large genetic diversity. Our GWAS study identified multiple novel candidates genes of Al tolerance, which were validated by comparison of gene expression between contrasting accessions (i.e. expression level polymorphism: ELP), and by comprehensive reverse genetics analysis of candidate genes.

MATERIALS AND METHODS

Al tolerance of 196 A.thaliana accessions were estimated by relative root length (RRL; root length in stress condition/root length in control condition) of five days old seedlings grown hydroponically with and without 5 μM $AlCl_3$. The GWAS was conducted with approximately 200K genome-wide SNPs using "Q+K" method in TASSEL 3.0 [5]. The population structure used in the GWAS was constructed by STRUCTURE 2.3.4 software [6]. GP analysis was conducted using ridge regression by "glmnet" R package [7]. The cumulated effects were estimated by 5-fold cross-validation using coefficient of determination (R^2) and root mean square error (RMSE) using an index. Linkage disequilibrium (LD) region of GWAS detected SNPs ($r^2 \geq 0.8$) were calculated in 10-kb window by PLINK 1.07 [8]. The AtALMT1promoter::GUS transgenic plants were generated using the AtALMT1 promoters of -2235bp from start codon isolated from Col-0 and Bil-7 accessions. The GUS expression levels were measured by real-time PCR using gene-specific primer pairs. T-DNA insertion mutants were obtained from Nottingham Arabidopsis Stock Centre.

RESULTS AND DISCUSSION

The RRL of 196 A.thaliana accessions showed normal distribution ranged from 6.6% to 70.8% with high broad sense heritability (H_b^2 = 0.98), suggesting the Al tolerance variation was controlled by polygenes. The GWAS detected a number of SNPs associated with Al tolerance across the genome. In order to determine the threshold for the GWAS, we calculated the cumulative effects of associated SNPs for the Al tolerance variation by Genomic prediction (GP). This analysis estimated that more than 70% of the Al tolerance variation was explained by top-140 associated SNPs in the GWAS, and 168 candidate genes located within LD region of the SNPs were identified. These results suggested that our GWAS identified a series of genes which control large part of Al tolerance variation among A.thaliana accessions. The candidate genes contained previously reported and a priori Al tolerance genes. A major Al tolerance gene AtALMT1 was located near the 14th and 15th associated SNPs, and the accessions with minor alleles showed significantly higher gene expression level than sensitive accessions. The sequence analysis of the AtALMT1 promoter identified several variants including 497- ________ 1 bp transposable element (TE) insertion linked with the major and minor allele. Promoter activity analysis using AtALMT1promoter::GUS transgenic plants demonstrated that the ELP of AtALMT1 was determined by promoter polymorphism among accessions. Most Al-tolerant accessions with high- expression type of AtALMT1 promoter were genetically closed and inhabited in Western Europe area where acid soil is dominant. This result suggested that the several accessions adapted to acid soil area of Western Europe through enhancing AtALMT1 expression level.

We performed comprehensive reverse genetics analysis of the candidate genes to identify novel Al tolerance gene. We evaluated Al tolerance of 73 knock out (KO) mutant plants of the candidate genes, and identified novel four KO lines which showed less than 30% of the Al sensitivity of WT. Expression analysis of the novel tolerance genes identified the ELP correlated with Al tolerance. These results indicated that the GWAS identified multiple novel tolerance genes which explain a part of Al tolerance variation among A.thaliana accessions.

CONCLUSION

The natural variation of A.thalaina is a powerful resource for investigation of adaptive strategies of plants to acid soil environment. Our GWAS for Al tolerance identified 168 candidate genes which could explain more than 70% of the Al tolerance variation. A major Al tolerance gene AtALMT1 was contained in the candidate genes. Sequence analysis and expression analysis revealed that the ELP of AtALMT1 caused by promoter polymorphism were associated with Al tolerance among accessions. In addition to AtALMT1, comprehensive reverse genetics analysis of the candidate genes identified multiple novel Al tolerance genes. Our results revealed a series of Al tolerance genes with causal polymorphism which explain large part of Al tolerance variation among A. thaliana accessions.

REFERENCES

Hoekenga OA, Maron LG, Piñeros MA, Cançado GMA, Shaff J, Kobayashi Y, et al. AtALMT1, which encodes a malate transporter, is identified as one of several genes critical for aluminum tolerance in Arabidopsis. Proc Natl Acad Sci U S A. **2006;103**: 9738–9743.

Ezaki B, Gardner RC, Ezaki Y, Matsumoto H. Expression of aluminum-induced genes in transgenic arabidopsis plants can ameliorate aluminum stress and/or oxidative stress. Plant Physiol. **2000;122**: 657–665.

Magalhaes J V, Liu J, Guimarães CT, Lana UGP, Alves VMC, Wang Y-H, et al. A gene in the multidrug and toxic compound extrusion (MATE) family confers aluminum tolerance in sorghum. Nat Genet. **2007;39**: 1156–61.

Sasaki T, Ryan PR, Delhaize E, Hebb DM, Ogihara Y, Kawaura K, et al. Sequence upstream of the wheat (Triticum aestivum L.) ALMT1 gene and its relationship to aluminum resistance. [Internet]. Plant & cell physiology**. 2006**. pp. 1343–54.

Bradbury PJ, Zhang Z, Kroon DE, Casstevens TM, Ramdoss Y, Buckler ES. TASSEL: software for association mapping of complex traits in diverse samples. Bioinformatics. **2007;23**: 2633–2635.

Pritchard JK, Stephens M, Donnelly P. Inference of population structure using multilocus genotype data. Genetics. **2000;155**: 945–959.

Friedman J, Hastie T, Tibshirani R. Regularization Paths for Generalized Linear Models via Coordinate Descent. J Stat Softw. **2010;33**: 1–22.

Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MAR, Bender D, et al. PLINK: A Tool Set for Whole-Genome Association and Population-Based Linkage Analyses. Am J Hum Genet. **2007;81**: 559–575.

P2-3

The distribution pattern of pectin in transition zone of different al-resistance pea (*Pisum sativum*) cultivars

***Xiao Hongdong*[1]*, Li Xuewen*[1]*, Yu Min*[1]**
[1]Department of Horticulture, Foshan University, Foshan, 528000, Guangdong, CHINA
; 1041909160@qq.com; yumin@fosu.edu.cn

INTRODUCTION

Aluminum (Al) toxicity is the primary constraint of agriculture production on acid soils. Therefore, it is of great significance to carry out research on Al tolerance in plants to improve the yield and quality of crops, which is the major problem for food production in acidic soils. Previous studies have demonstrated that the transition zone is most sensitive and plays an important role in plant Al tolerance. Pectin is a major component of cell wall and is responsible for the sensitivity of root transition zone. In this study, Al-tolerant and Al-sensitive cultivars of pea (*Pisum sativum*) were employed to investigate the distribution pattern of pectin in transition zone.

MATERIALS AND METHODS

Pea (*Pisum sativum* L) seeds were germinated and cultured, and treated with 30μM $AlCl_3$ solution (25 μM H_3BO_3, 0.5 mM $CaCl_2$, pH 4.5) for 24 h. Root length determined using WinRhizo-Pro software. Selected lateral roots of the seedlings to stain with morin, JIM5 and JIM7. Roots were observed under fluorescent microscope directly, free-hand sections were observed under a Laser-Scanning Confocal Microscope. The measurement of Al concentration by ICP-AES. The pectin content and PME activity was measured by colorimetric method, modified from Yang (2008).

RESULTS AND DISCUSSION

1 Different Al resistance in cultivars of pea

Kobayashi et al. (2004) have shown that cv Alaska is an Al-tolerant cultivar and cv Hyogo is an Al-sensitive cultivar. Our results showed that root elongation was inhibited by Al toxicity in both cv Alaska and cv Hyogo, but relative root elongation in cv Alaska was higher than that in cv Hyogo (Fig. 1A). Meanwhile there was significantly less Al accumulation in the transition zone of cv Alaska compared to cv Hyogo (Fig. 1B). The green fluorescence indicated by Morin were stronger in Al-sensitive cv Hyogo than that in Al-tolerant cv Alaska, especially in the transition zone (Fig. 2C and 2D). And the distribution of Al in Al-sensitive cv Hyogo was wider than in Al-tolerant cv Alaska (Fig. 2E and 2F).

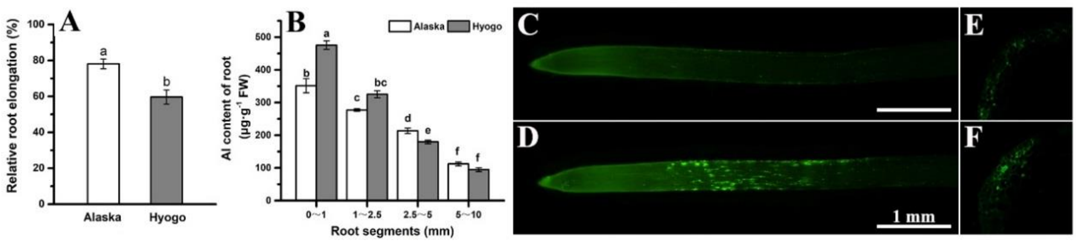

*Fig. 1 Effect of Al application on root elongation (A) and Al content (B) in different cultivars of pea

2 The property of pectin in transition zone

The pectin content and PME activity of transition zone in Al-sensitive cv Hyogo was higher than that of Al-tolerant cv Alaska (Fig. 2A and 2C). The higher PME activity of cv Hyogo resulted in lower DM (Fig. 2B). Our results showed that, under Al toxicity, there were higher pectin content and lower DM in the transition zone of Al-sensitive cultivar than the Al-tolerant cultivars.

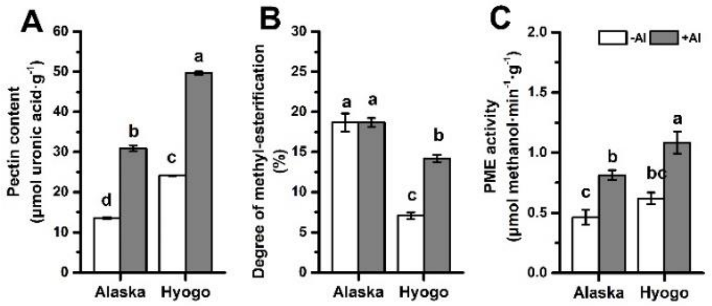

*Fig. 2 Cell wall pectin properties. A. Cell wall pectin content. B. the degree of pectin methyl-esterification. C. The demethylesterified pectin content. D. PME activity.

3. The distribution pattern of pectin in transition zone

Monoclonal antibodies (JIM5 and JIM7) were used for immunofluorescence localization of cell wall pectins. Under Al stress, the difference in JIM5 and JIM7 between the two cultivars was mainly in the outer layer (Fig. 3). In the outer layer, the fluorescence of JIM5 was brighter in cv Hyogo (Fig. 3D), while JIM7 was brighter in Alaska (Fig. 3F).

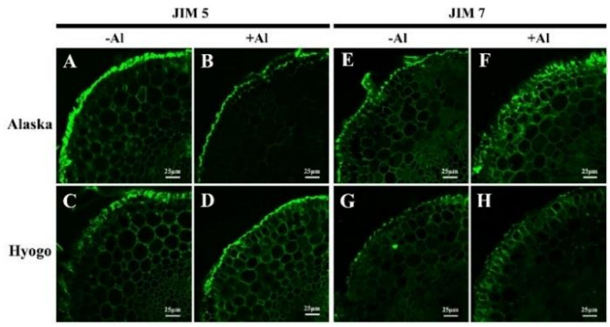

*Fig. 3 Immunofluorescence localization of JIM5 and JIM7 in the transition zone of different peas (*Pisum sativum*). Roots cross-section was collected at 1500 μm in Alaska (A, B, E, F) and Hyogo (C, D, G, H) with or without Al treatment. The cross sections were stained with JIM5 (A, B, C, D) and JIM7 (E, F, G, H) observed under CLSM.

CONCLUSION

The transition zone is the most Al-sensitive apical root zone. Morin stain show that Al-sensitive cv Hyogo have accumulated more Al in the transition zone, especially its outer layer, meanwhile the fluorescence of JIM5 in the outer layer was brighter. Therefore, the accumulation of Al in the transition zone of Al-sensitive cultivars may be related to the low methyl esterification of pectin in the outer layer.

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REFERENCES

Kobayashi, Y., Yoko Y., and Hideaki M. (2004). Studies on the mechanism of aluminum tolerance in pea (*Pisum sativum* L.) using aluminum-tolerant cultivar ‘Alaska’ and aluminum-sensitive cultivar ‘Hyogo’. *Soil Science and Plant Nutrition* **50.2:** 197-204

Yang, J., Li, Y., Zhang, Y., and Zhang, S. (2008). Cell wall polysaccharides are specifically involved in the exclusion of aluminum from the rice root apex. *Plant Physiology* **146,** 602-611

P2-4

Impact of aluminum induced malate excretion on primary metabolism of Arabidopsis root

***Takuo Enomoto[1]*, Akira Oikawa[2], Yuriko Kobayashi[1,3], Hiroyuki Koyama[1,3,]**

[1]United Graduate School of Agricultural Science, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan;

[2] Faculty of Agriculture, Yamagata University, 1-23 Wakaba-macho, Tsuruoka-shi, Yamagata 997-8555, Japan

[3] Applied Biological Sciences, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan

; koyama@gifu-u.ac.jp

INTRODUCTION

Aluminum (Al) ions solubilized in the rhizosphere disturb root growth of plants grown on acid soils. Plants have evolved various resistance strategies to alleviate Al^{3+} rhizotoxicity such as organic acids (OAs) excretion from the roots. *Arabidopsis thaliana* excretes malate from the roots to protect root tip from Al toxicity. The malate release is mediated by the malate transporter Aluminum-Activated Malate Transporter 1 (AtALMT1), and dysfunction of *AtALMT1* completely suppresed root growth by sub micromolar of Al^{3+} (Hoekenga et al., 2006, Kobayashi et al., 2007). However, this strategy by OA excretion may require to maintain balance between internal C-metabolism for energy and metabolite production, and for detoxification of Al rhizooxicity. By contrast, a little is known about the interaction between malate release and metabolism. In this study, we investigated the change of OA metabolism in the transgenic Arabidopsis overexpressing *AtALMT1* to evaluate effect of malate release on the C-metabolism under Al condition.

MATERIALS AND METHODS

Arabidopsis plants of wild type (WT, Col-0) and *AtALMT1*-overexpressin (OX) were used for the experiments. The seedlings were pre-grown for 10 days under sterilized conditions in 2 % MGRL medium with 1 % sucrose at pH 5.5 and then their roots were immersed in 2 % MGRL (removed Pi) medium with or without 10 µM Al at pH 5.0 in the presence of 1% sucrose for 24 hours. Roots were sampled from each plant and used for experiments. Metabolome analysis were used by CE-MS. Gene expression analysis were performed by microarray.

RESULTS AND DISCUSSION

Metabolome analysis identified 120 metabolites in all plant samples. Metabolic changes under Al conditions were analyzed in the wild type (WT) and *AtALMT1*-OX, which excreted 5-10 times greater amount of malate than WT. Metabolites related to sugar and tricarboxylic acid cycle (TCA) such as G6P;F6P;M6P, citrate and fumarate were significantly decreased in the roots of WT under Al condition. TCA metabolites were generally lower in the *AtALMT1*-OX, suggesting that enhanced malate excretion associates with reduction of other TCA metabolites. By contrast, several amino acids were more accumulated in the *AtALMT1*-OX (e.g. Gln and Glu), which was concomitant with upregulation of genes encoding N-metabolic pathway such as *nitrate transporter 1.1* (*NRT1.1)* and *nitrate reductase* 1 (*NIA1)*. These results suggest that the malate excretion to Al solution decrease in the TCA metabolites but enhanced Glu/Gln production associated with enhanced N-assimilation. On the other hand, Al treatment increased several metabolites of the WT, which play roles to reduce cellular damage due to Al-induced ROS damage in the WT such as agmatine and g-Glu-Cys. These metabolites tended to be higher in WT than *AtALMT1*-OX. This result suggested that accumulation of these metabolites was not exaggerated by the amount of malate excretion and detoxification of Al^{3+} in the rhizosphere.

CONCLUSION

Aluminum induced malate excretion altered C-metabolism in Arabidopsis roots. TCA metabolites tended to decrease by Al treatment, which was more evident in the *AtALMT1*-OX that excreted greater level of malate released under Al condition. On the other hand, *AtALMT1*-OX accumulated greater level of Glu/Gln with the upregulation of genes for N-assimilation. These results suggested coupling of Al-activated malate excretion and N-assimilation.

REFERENCES

Hoekenga O. A., Maron L. G., Pineros M.A., Cancado G. M. A., Shaff J., Kobayashi Y., Ryan P. R., Dong B., Delhaize E., Sasaki T., et al (2006) AtALMT1, which encodes a malate transporter, is identified as one of several genes critical for aluminum tolerance in Arabidopsis. Proc Natl Acad Sci **103**: 9738–9743

Kobayashi Y., Hoekenga O.A., Itoh H., Nakashima M., Saito S., Shaff J.E., Maron L.G., Piñeros M., Kochian L. V., Koyama H (2007) Characterization of AtALMT1 expression in aluminum-inducible malate release and its role for rhizotoxic stress tolerance in Arabidopsis. Plant Physiol **145**: 843–852

P2-5

Functional analysis of a magnesium transporter gene OsMGT2 in rice

***L. D. Zhang*[1], *J. Li*[1] *and Z. C. Chen*[1]**
[1]Root Biology Center, Fujian Agriculture and Forestry University, Fuzhou, 350002, China
; zcchen@fafu.edu.cn

INTRODUCTION

The growth of crops in acidic soils is constrained by many limiting factors, such as aluminum and manganese toxicity, as well as the lack of phosphorus and magnesium (Mg). The deficiency of Mg in plants causes leaf interveinal chlorosis and growth retardation (Chen et al., 2018). It has been known that *MGT* (magnesium transporter) family genes are responsible for Mg transport in rice (Chen et al., 2012; Saito et al., 2013). However, the physiological role of *MGTs* is not well investigated. In this study, we functionally characterized an *MGT* family gene, *OsMGT2*, which encodes a putative Mg transporter in rice.

MATERIALS AND METHODS

Two-week-old rice seedlings were hydroponically grown in 1/2 kimura nutrient solution with or without Mg supply for 11 days. The shoot and root samples were separately harvested. Mg concentration was measured by ICP-MS. The chlorophyll content (SPAD value) was measured using a chlorophyll meter (SPAD-502 Plus). *Tos-osmgt2* and *Cas9-osmgt2* mutants were constructed by Transposon insertion and CRISPR-Cas9 technique, respectively.

RESULTS AND DISCUSSION

The biomass and SPAD value of the two mutants was significantly lower than that of WT under Mg deficiency (Fig. 1a, b), indicating that *OsMGT2* is required for rice growth under low Mg stress. The mutants accumulated much higher Mg in the roots than WT, resulting in a much lower root-to-shoot translocation of Mg in mutants (Fig. 1c). A complementation assay in the yeast CM66 strain showed that OsMGT2 has Mg^{2+} transport activity. Spatial expression analysis revealed that *OsMGT2* was expressed mainly in vascular tissue of both roots and shoots, suggesting its important role in rice Mg translocation.

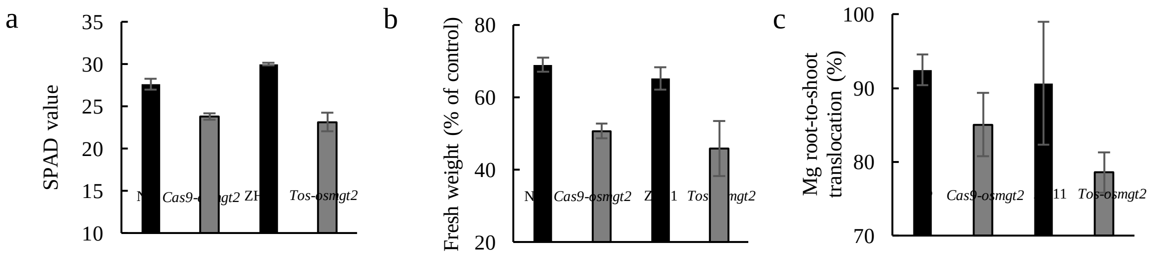

Figure 1. Comparison of SPAD value (a), fresh weight (b), and Mg translocation rate (c) in wild-type and *osmgt2* mutants under low Mg stress.

CONCLUSION

Taken together, we conclude that OsMGT2 functions as a Mg transporter in rice vascular tissues, which is required for root-to-shoot translocation of Mg. It plays an important role in rice growth under low Mg condition.

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REFERENCES

Chen, Z.C., Peng, W.T., Li, J. and Liao, H. 2018. Functional dissection and transport mechanism of magnesium in plants. Seminars in Cell & Developmental Biology **74**: 142-152.

Chen, Z.C., Yamaji, N., Motoyama, R., Nagamura, Y. and Ma, J.F. 2012. Up-regulation of a magnesium transporter gene *OsMGT1* is required for conferring aluminum tolerance in rice. Plant Physiology **159**: 1624-1633.

Saito, T., Kobayashi, N.I., Tanoi, K., Iwata, N., Suzuki, H., Iwata, R. and Nakanishi, T.M. 2013. Expression and functional analysis of the CorA-MRS2-ALR-type magnesium transporter family in rice. Plant & Cell Physiology **54**: 1673-1683.

P2-6

Genotypic variation of manganese tolerance in rice

Sheng Huang[1], Walter Johannes Horst[2], Jian Feng Ma[1]
[1]Institute of Plant Science and Resources, Okayama University, Chuo 2–20–1, Kurashiki 710–0046, Japan
[2]Institute of Plant Nutrition, Faculty of Natural Sciences, Leibniz University Hannover, Herrenhäuser Str. 2, 30419 Hannover, Germany
; maj@rib.okayama-u.ac.jp

INTRODUCTION

Manganese (Mn) toxicity is considered as a limiting factor of crop production after Al toxicity on acid soils [1]. Its toxicity symptoms are characterized by brown spots on the old leaves as a result of increased peroxidase activity [2,3]. However, the tolerance to Mn toxicity varies among plant species and cultivars within a species. Rice is one of the most Mn-tolerant crops among small-grain cereal crops. Furthermore, rice is also able to accumulate high Mn in the shoots; some rice cultivars can accumulate Mn concentration in the leaves as high as 5000 µg/g dry weight without showing any toxic symptoms [4]. However, it was also reported that there was a genotypic difference in Mn tolerance in rice. To understand the mechanism underlying the genotypic difference in Mn tolerance, as the first step, we screened 140 rice core collection accessions and physiologically characterized some of these accessions.

MATERIALS AND METHODS

A total of 140 rice accessions (including 72 japonica and 68 indica) from rice core collections was used for screening of Mn sensitivity. The seedlings (12-d-old) grown in nutrient solution were exposed to a solution containing 200 or 500 µM Mn. After exposure for 9 days, the Mn toxicity symptoms（brown spots and blade curl of the leaf）were observed and recorded. Both the root and shoot of the seedlings were then harvested for biomass measurement and the Mn concentration determination by ICP-MS.

RESULTS AND DISCUSSION

Among 140 accessions, we found 22 accessions (including 5 japonica and 17 indica) showed typical Mn toxicity symptom (Fig. 1). To physiologically characterize genotypic difference in Mn tolerance, we selected five Mn-sensitive and three Mn-tolerant accessions for further analysis. The shoot dry weight was hardly affected by high Mn concentration (200 and 500 µM) in Mn-tolerant accessions but was inhibited in Mn-sensitive accessions. The Mn-sensitive accessions accumulated higher concentration of Mn than tolerant accessions, however, there was no large difference in other mineral concentrations of the shoots between Mn-sensitive and –tolerant accessions. There was a good negative correlation between relative shoot growth and Mn concentration (Fig. 2). These results indicate that the high Mn concentration may be responsible for the increased Mn sensitivity.

Similar to shoot dry weight, the inhibition of root dry weight was more in Mn-sensitive cultivars than Mn-tolerant cultivars. This was confirmed by root elongation experiment with a short-term (24 h) exposure. There was no consistent trend in root Mn concentration between Mn-sensitive and –tolerant cultivars. Correlation analysis also showed there was no good correlation between relative root growth and root Mn concentration. In some Mn-sensitive cultivars, the root Cu concentration was higher than in Mn-tolerant cultivars. On the other hand, the root Zn concentration was greatly inhibited by high Mn concentration in some cultivars, suggesting that Mn and Zn may share the same transporters.

Comparison of the root-to-shoot translocation showed that there was no difference in the translocation of Mn, Fe, Cu and Zn between Mn-sensitive and Mn-tolerant accessions. Furthermore, compared with Mn, Fe and Zn, Cu is less translocated from the roots to the shoots.

We have developed some mapping populations between Mn-sensitive and –tolerance accessions and the QTL analysis for Mn tolerance is being undertaken.

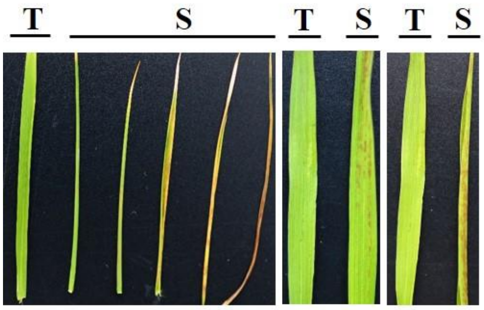

Fig.1. Mn toxicity symptoms. Brown spots appear on older leaves. T, tolerant; S, sensitive.

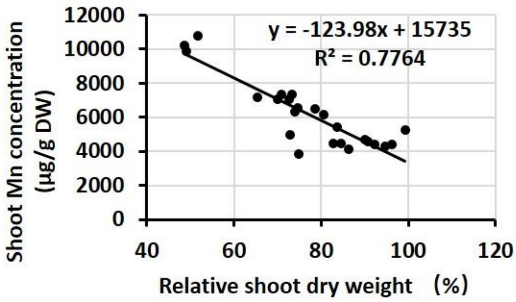

Fig.2. Correlation between relative shoot growth and Mn concentration. Seedlings grown in 0.5 µM Mn were treated with 200 µM Mn for 9 d. Shoot dry weight grown in 200 µM was normalized by dry weight grown in 0.5 µM. Mn.

CONCLUSION

There was a large genotypic variation in Mn tolerance in both japonica and indica subspecies. Mn tolerance may be associated with low Mn concentrations in the shoots in the selected rice accessions.

ACKNOWLEDGEMENTS

This work was supported by Grant-in-Aid for Specially Promoted Research (JSPS KAKENHI Grant Number 16H06296 to J.F.M.).

REFERENCES

Chen, Z., Fujii, Y., Yamaji, N., Masuda, S., Yuma, T., Kamiya, T., *et al*. (2013) Mn tolerance in rice is mediated by MTP8.1, a member of the cation diffusion facilitator family. J. Exp. Bot. **64**: 4375–4387.

Wissemeier, A.H. and Horst, W.J. (1987) Callose deposition in leaves of cowpea (*Vigna unguiculata* [L] *Walp*) as a sensitive response to high Mn supply. Plant Soil **102**, 283–286.

Führs, H. *et al*. (2009) Characterization of leaf apoplastic peroxidases and metabolites in *Vigna unguiculata* in response to toxic manganese supply and silicon. J. Exp. Bot. **60**, 1663–1678.

Sasaki, A., Yamaji, N., Xia, J., and Ma, J.F. (2011). OsYSL6 is involved in the detoxification of excess manganese in rice. Plant Physiol. **157**:1832–1840.

P2-7

An alternative splicing transcript of *FeALS1.1* is implicated in Al detoxification in buckwheat

Lei, G.J[1]., Yokosho, K[1]., Yamaji, N[1]., Fujii-Kashino, M[1]. and Ma, J.F[1].
[1]Institute of Plant Science and Resources, Okayama University, Chuo 2-20-1, Kurashiki 710-0046, Japan
; leiguijie@126.com

INTRODUCTION

Buckwheat (*Fagopyrum esculentum*) shows high tolerance to Al ion toxicity and accumulates high Al in the leaves without showing any toxicity symptoms. Some transporters involved in accumulation and detoxification of Al have been reported in buckwheat. For example, the tonoplast-localized FeIREG1, belonging to IRON REGULATED/ferroportin, is involved in the internal Al detoxification by sequestering Al into the root vacuoles (Yokosho et al., 2016). Recently, FeMATE1, a member of the multi-drug and toxic compound extrusion (MATE) family, was reported to be required for the Al-activated citrate secretion in the roots, while FeMATE2 is probably responsible for the internal detoxification of Al by transporting citrate into Golgi system in the roots and leaves (Lei et al., 2017a). Besides, two half-size ABC transporters, FeALS1.1 and FeALS1.2, are involved in the internal detoxification of Al in the roots and leaves, respectively, by sequestering Al into the vacuoles (Lei et al., 2017b). However, the molecular mechanisms underlying the high Al tolerance and accumulation are still poorly understood in buckwheat.

MATERIALS AND METHODS

RNA from roots and leaves was extracted using the RNeasy Mini Kit (Qiagen). Gene expression was determined by real time RT-PCR. Transient assay in buckwheat leaf protoplasts was performed by PEG method. Transgenic Arabidopsis was generated by the *agrobacterium tumefaciens*-mediated floral dip method.

RESULTS AND DISCUSSION

We found that the *FeALS1.1* gene had alternative splicing based on our RNA-seq data and 5'-RACE experiment, resulting in five mRNA transcripts (Fig. 1). One transcript encoded a long ORF (reported as *FeALS1.1* in Lei et al., 2017b), but other four transcripts encoded a short ORF (named as *FeALS1.1-S* here), lacking the first two predicted transmembrane domains of the long FeALS1.1. *FeALS1.1-S* was mainly expressed in the leaves, and up-regulated by Al in both the leaves and roots, but not induced by low pH or La. In the root tip region, *FeALS1.1-S* was induced by Al in both 0-1 cm and 1-2 cm segments, and showed similar expression level in the different tissues of the roots separated by laser microdissection.

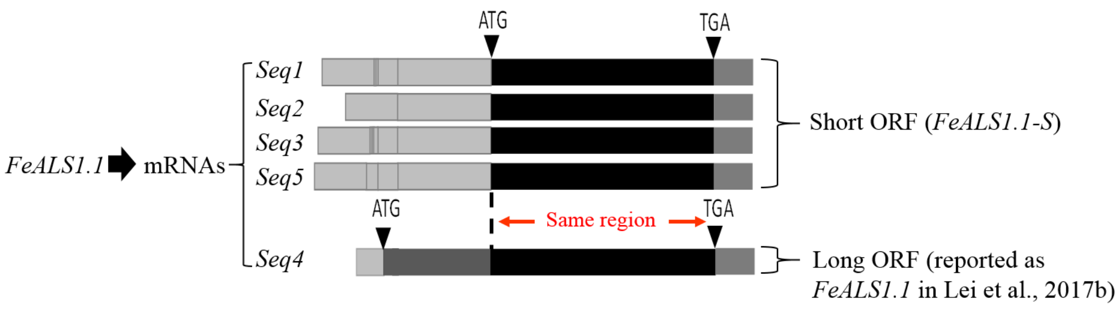

Fig. 1. Five transcripts of *FeALS1.1* due to alternative splicing.

When FeALS1.1-S-GFP fusion was transiently expressed in the buckwheat leaf protoplast, it was localized to endoplasmic reticulum and plasma membrane, but GFP-FeALS1.1-S fusion was localized to unknown vesicles in the cytosol. It looks like that GFP affected the localization of FeALS1.1-S. Therefore the subcellular localization of FeALS1.1-S needs to be further investigated.

When *FeALS1.1-S* was introduced to *atals1* mutant under the control of *AtALS1* promoter, it recovered the root elongation at 40 □M Al, but not at 60 □M Al (Fig. 2). The Al concentration in the roots and Al distribution in the root cells observed by Morin staining were not altered in the transgenic lines. These results suggest that *FeALS1.1-S* might play a different role from *FeALS1.1-L* in Al tolerance of buckwheat.

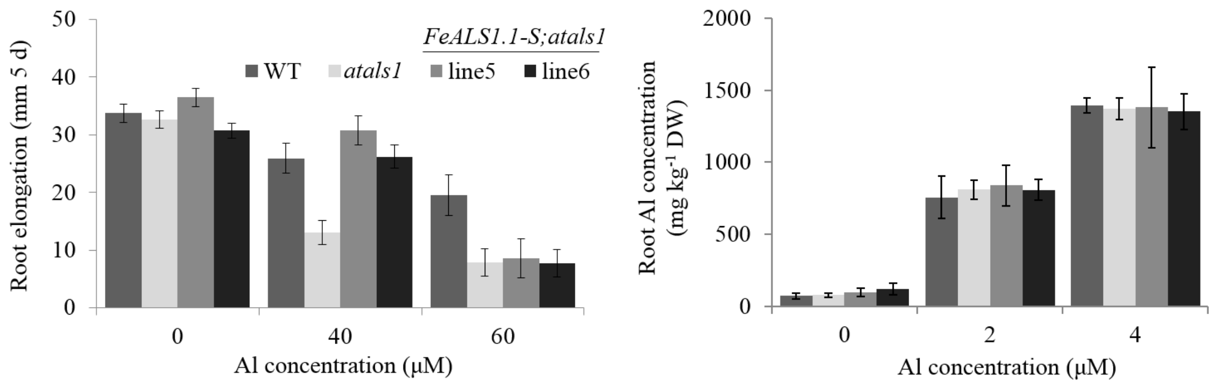

Fig. 2. Al tolerance and accumulation in the transgenic Arabidopsis lines carrying *FeALS1.1-S*. Plants were grown on an agar plate containing 0, 40 or 60 µM Al for 5 d and the root length was measured. Al concentration in the roots were determined after the plants were exposed to a solution containing 0, 2 or 4 µM Al for 1 d.

CONCLUSION

Our results suggest that *FeALS1.1-S*, a short ORF by alternative splicing of *FeALS1.1*, is probably involved in detoxification of Al mainly in the leaves of buckwheat through unknown mechanism.

ACKNOWLEDGEMENTS

This work was supported by Grant-in-Aid for Specially Promoted Research (JSPS KAKENHI Grant Number 16H06296 to J.F.M.).

REFERENCES

Lei, G.J., Yokosho, K., Yamaji, N. and Ma, J.F. 2017a. Two MATE transporters with different subcellular localization are involved in Al tolerance in buckwheat. Plant and Cell Physiology **58**:2179-2189.

Lei, G.J., Yokosho, K., Yamaji, N., Fujii-Kashino, M. and Ma, J.F. 2017b. Functional characterization of two half-size ABC transporter genes in Al-accumulating buckwheat. New Phytologist **215**: 1080-1089.

Yokosho, K., Yamaji, N., Mitani-Ueno, N., Shen, R.F. and Ma, J.F. 2016. An aluminum inducible IREG gene is required for internal detoxification of aluminum in buckwheat. Plant and Cell Physiology **57**: 1169-1178.

P2-8

Functional characterization of OsBBPI3, a putative ART1-interactive protein in rice

***Kengo Yokosho*[1], *Zhi Chang Chen*[1], *Naoki Yamaji*[1] *and Jian Feng Ma*[1]**
[1]Institute of Plant Science and Resources, Okayama University, 2-20-1 Chuo, Kurashiki, Okayama 710-0046, Japan
; maj@rib.okayama-u.ac.jp

INTRODUCTION

Rice (*Oryza sativa*) shows the highest tolerance to aluminum (Al) among the gramineous crops including maize, wheat, barley, and sorghum (Famoso *et al*., 2010; Ma, 2007). Recent identification of a transcription factor, ART1 (Al resistance transcription factor 1), revealed that high Al-tolerance in rice is achieved by multiple genes involved in the detoxification of Al (Yamaji *et al*., 2009). ART1 is a C2H2-type zinc-finger transcription factor, which is localized in the nucleus of all root cells. ART1 regulates 32 genes by binding to a cis-acting element (GGN[T/g/a/]CV[C/A/g]S[C/G]) present in the promoter (Tsutsui *et al*., 2011). The expression of all these ART1-regulated genes is specifically up-regulated by Al. However, the expression of *ART1* is not induced by Al. Therefore, activation of ART1 is required, but the underlying mechanism is unknown. To investigate the mechanism involved in ART1 activation, a yeast two-hybrid screening was carried out and several candidates were obtained. Here we report functional characterization of OsBBPIs (Bowman-birk protease inhibitor), which have potential to interact with ART1.

MATERIALS AND METHODS

To generate *bbpi3* triple mutant by CRISP/Cas9, we used the Cas9 plant expression vector (pU6gRNA) and sgRNA expression vector (pZDgRNA_Cas9ver.2_HPT) provided by Dr. Endo (National Institute of Agrobiological Sciences, Japan). The derived construct for mutation was transformed into calluses (cv. Nipponbare) by Agrobacterium-mediated transformation. To investigate Al tolerance, seedlings of wild-type rice and *bbpi3* mutant were exposed to 0, 10 and 50 μM Al for 24 h. Root length was measured at 0 and 24 h with a ruler. For expression experiment, total RNA was extracted from the roots of wild-type rice or *bbpi3* mutant exposed to Al at different treatment time. The relative transcript levels were determined by quantitative RT-PCR.

RESULTS AND DISSCUSSION

The yeast two hybrid screening showed 11 proteins interact with ART1. Among them, 5 proteins (OsBBPI2.1:Os01g0124000, OsBBPI2.2:Os01g0123900, OsBBPI3.1:Os01g0124200, OsBBPI3.2:Os01g0124100, OsBBPI3.3:Os01g0124400) belong to BBPIs family, which was reported as serine protease inhibitors in legumes (Bowman 1946). Expression analysis showed *OsBBPI3.1*, *OsBBPI3.2* and *OsBBPI3.3* were mainly expressed in root, while *OsBBPI2.1* and *OsBBPI2.2* were expressed in the shoot. The expression of *OsBBPI3.2, OsBBPI3.1* and *OsBBPI 3.3* in roots were 12, 4 and 4 times increased by Al treatment for 6 h, respectively. However, this induction was not regulated by ART1. To investigate the physiological role, we generated a triple mutant using by CRISP/Cas9. The Al tolerance was compared between the wild-type rice and *bbpi3* triple mutant by measuring root elongation inhibition during 24 h. The root elongation was inhibited more in the *bbpi3* triple mutant than in the wild-type rice at 50 μM, but not at 10 μM Al (Figure 1). The expression analysis showed that some ART1 down-stream genes were lower in the *bbpi3* triple mutant than in the wild type.

For example the expression of *OsFRDL4* was reduced 40% in the *bbpi3* triple mutant than in the wild-type rice, but that of *STAR1* was not altered (Figure 2). We propose that OsBBPI3 might be involved in maintaining ART1 function in rice roots.

CONCLUSION

We functionally characterized three *OsBBPI3* genes (*OsBBPI3.1, OsBBPI3.2* and *OsBBPI3.3*), which their encoded proteins have potential interaction with ART1 based on yeast two-hybrid screening. These genes were mainly expressed in the roots and the expression level was induced by Al, but not regulated by ART1. The triple mutants of *bbpi3* showed an increased Al sensitivity compared with the wild-type rice. The expression analysis showed that some ART1 down-stream genes were lower in the *bbpi3* triple mutants than in the wild-type in the presence of Al. We propose that OsBBPI3 might be involved in maintaining ART1 function in rice roots.

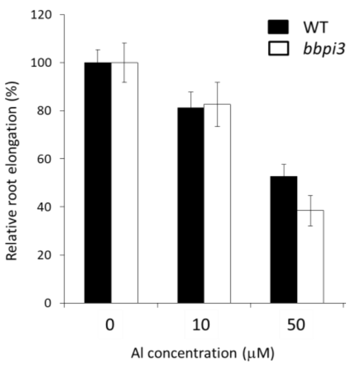

Figure 1 Al tolerance of WT and *bbpi3* mutant.

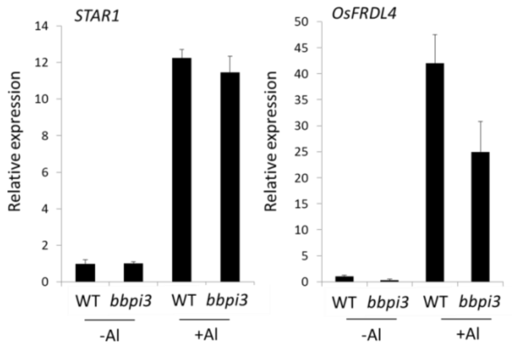

Figure 2 Expression analysis of *STAR1* and *OsFRDL4* in WT and *bbpi3* mutant.

ACKNOWLEDGEMENT

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REFERENCES

Bowman D.E. 1946. Differentiation of soybean anti-tryptic factors. Proc Soc Exp Biol Med **63**: 547-550.

Famoso A.N., Clark R.T., Shaff J.E., Craft E., McCouch S.R. and Kochian L.V., 2010. Development of a novel aluminum tolerance phenotyping platform used for comparisons of cereal aluminum tolerance and investigations into rice aluminum tolerance mechanisms. Plant Physiol **153**, 1678-1691.

Ma J.F. 2007. Syndrome of aluminum toxicity and diversity of aluminum resistance in higher plants. Int Rev Cytol **264**: 225-252.

Tsutsui T., Yamaji N. and Ma J.F. 2011. Identification of a cis-acting element of ART1, a C2H2-type zinc-finger transcription factor for aluminum tolerance in rice. Plant Physiol **156**: 925-931.

Yamaji N., Huang C.F., Nagao S., Yano M., Sato Y., Nagamura Y. and Ma J.F. 2009. A zinc finger transcription factor ART1 regulates multiple genes implicated in aluminum tolerance in rice. Plant Cell **21**: 3339-3349.

P2-9

GmINS1, a candidate gene for a nodulation QTL, is a key contributor to nodule development in soybean

***J. K. Zheng*, Y[1]. Q. Yang[1], X. X. Li[1] and H. Liao[1]**
[1]Root Biology Center, Fujian Agriculture and Forestry University, 350002, Fuzhou, China
; hliao@fafu.edu.cn

INTRODUCTION

About 50% arable lands in the world are acidic and nutrient deficiency, especially low nitrogen (N) availability is the primary factor limiting crop production on acidic soils. Biological nitrogen fixation (BNF) could provide N source for plants, and thus might reduce fertilizer application and increase nutrient efficiency. Since soybean has a superior capacity to fix N_2 via rhizobia, it has been widely used to improve soil fertility, including in acidic soils. The BNF capacity varied among soybean genotypes is a complex trait controlled by multiple quantitative trait loci (QTL) (Santos et al., 2013; Yang et al., 2017). To date, the genetic and molecular mechanisms underlying BNF remain largely unknown.

MATERIALS AND METHODS

In the present study, both field experiment and sand culture had been carried out. For the field experiment, 175 $F_{9:11}$ RILs were grown at the Dishang experimental farm (Shijiazhuang City, Hebei Province, China), nodules were harvested to analyze nodule number and individual nodule size, and to identify QTLs for nodulation associated traits. To explore effects of *GmINS1* on nodulation, the transgenic composite plants were inoculated with rhizobium BXYD3, and grown in sand culture for 30 days. Nodules were harvested to determine nodule number and individual nodule size

RESULTS AND DISCUSSION

In this study, we constructed a high resolution genetic map consisting of 27 dCAPs markers, and the gene *GmINS1* was selected as a candidate gene for explaining *qBNF-11* QTL effects on nodulation (Fig. 1a). qRT-PCR analysis indicated that *GmINS1* expression was strongly associated with the number of big nodules and individual nodule size in progeny RILs (Fig. 1b-d). The results suggest that *GmINS1* might be the candidate gene responsible for the nodulation QTL identified in the field studies.

GmINS1 was identified as a homolog of *GmEXPB2*, which played an important role in soybean nodulation. Here, we found that overexpression of *GmINS1* significantly facilitated nodule development and led to more big nodules ($D \geq 2$ mm). In contrast, suppression of *GmINS1* obviously limited nodule enlargement and reduced individual nodule sizes (Fig. 2). This suggests that *GmINS1* might play a dominant role in nodule enlargement. These results might be valuable for marker assisted selection to breed elite soybean varieties with optimized BNF capacity and yield.

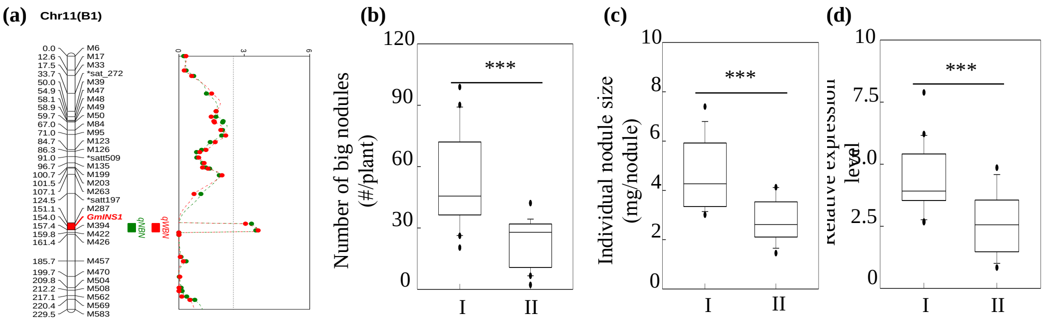

Fig. 1 Genetic localization of *GmINS1*, and relationships between *GmINS1* expression and nodule development in RILs. (a) Co-location analysis of the putative QTL for nodulation traits and *GmINS1*. (b) Number of big nodules. (c) Individual nodule size. (d) Transcription of *GmINS1*.

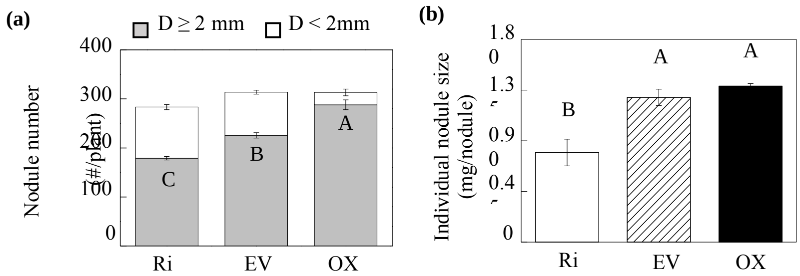

Fig. 2 Effects of RNA interference (Ri) or overexpression (OX) of *GmINS1* on soybean nodulation. (a) Nodule number. (b) Individual nodule size. Ev, Soybean transgenic plants harboring empty vector as control.

CONCLUSION

Taken together, we conclude that *GmINS1* is a candidate gene for *qBNF-11* QTL effects on soybean nodulation in the field, with the functions of this gene primarily occurring in nodule development, especially during enlargement of nodules.

ACKNOWLEDGEMENTS

This work was financially supported by China National Key Program for Research and Development (2016YFD0100700).

REFERENCES

Santos, M. A., Geraldi, I. O., Garcia, A. A. F., Bortolatto, N., Schiavon, A., Hungria, M. 2013. Mapping of QTLs associated with biological nitrogen fixation traits in soybean. Hereditas **150**: 17-25.

Yang, Y. Q., Zhao, Q. S., Li, X. X., Ai, W. Q., Liu, D., Qi, W. D., Zhang, M. C., Yang, C. Y., Liao, H. 2017. Characterization of genetic basis on synergistic interactions between root architecture and biological nitrogen fixation in soybean. Frontiers in Plant Science **8**: 1466.

P2-10

Genetic Localization of Root Hair Traits in Soybean

***H. Y. Lv*[1], *Y. Q. Yang*[1] *and H. Liao*[1]**
[1]Root Biology Center, Fujian Agriculture and Forestry University, 350002 Fuzhou, Fujian, China
; hliao@fafu.edu.cn

INTRODUCTION

Low phosphorus (P) availability is one of the major limiting factors for plants grown on acidic soils (Kochian et al., 2004). Plants have developed strategies to adapt to low P by increasing root hair elongation and density. It has been reported that hormones and environmental factors, such as auxin and nutrient deficiency, control root hair formation and development (Müller et al., 2004). Furthermore, a number of genes have been identified, which are specifically expressed in either root hairs or non-root hair cells, and are related to a certain growth stage of root hairs, such as *RHD6*, *RSL2* and *RSL4*. However, the underlying genetic mechanism remains largely unknown.

MATERIALS AND METHODS

The tested soybean population was constructed from a cross between soybean accessions YC03-3 (with normal root hairs) and RBC-RL (a mutant accession with abnormal root hairs). F1 and F2 populations, and recombined inbred lines (RILs) were generated and used for phenotypic and genotypic evaluation. Two-day soybean seedlings were observed under microscope for root hairs. Soybean leaves were collected from the parents and 150 $F_{5:7}$ RIL lines in the field.

Plant genomic DNA was extracted for whole genomic sequencing and GBS library construction for detecting SNP and Indels. GBS libraries were constructed using the two-enzyme modification following the original GBS protocol. The qualified SNP bin markers were used to construct the genetic linkage map using Join Map 4.1.

RESULTS AND DISCUSSION

In this study, we found a soybean mutant material (Laboratory accession number: RBC-RL) with few root hairs at seedling stage. In order to understand the inheritance of root hairs, we constructed a population consisting of five generations of genetic materials. The results of $\chi 2$ test showed that the segregation ratio of F_2, $F_{2:3}$ and RIL ($F_{5:7}$) fit well to 15:1, 7:8:1 and 3:1, respectively, indicating that the phenotype of few root hairs might be controlled by two independent recessive dominant genes (Table 1).

We further constructed a high resolution genetic map consisting of 8784 SNP markers using a RIL population through WGS and GBS method. QTL analysis was carried out and two QTLs with high LOD value were detected. Both QTLs were located at the end of chromosome. One of them, *qRHLa*, with a LOD score of 14.41 was mapped on the chromosome 01 at the marker Ch01.56510449, explaining 35.7% phenotypic variation. The other one, *qRHLb*, with a LOD score of 11.47 was mapped on the chromosome 11 between the markers Ch11.31494 and Ch11.554419, explaining 29.7% phenotypic variation (Fig. 2).

The results might be valuable for further map-based cloning for the target genes, and could help to better understand root hair development in soybean.

Table 1: Inheritance analysis for root hairs

	NL*	Seg. *	RL*+Med*	Total	Expected ratio	χ2	*P* value
YC03-3	20		0	20			
F_1	20		0	20			
F_2	593		135	728	15:1	0.452	0.798
$F_{2:3}$	23	31	4	58	7:8:1	0.399	0.819
RIL	105		45		3:1	1.742	0.157
RBC-RL	0		20	20			

*NL, Med and RL represented the phenotype of root hairs were normal, medium and few, respectively. *Seg: segregation.

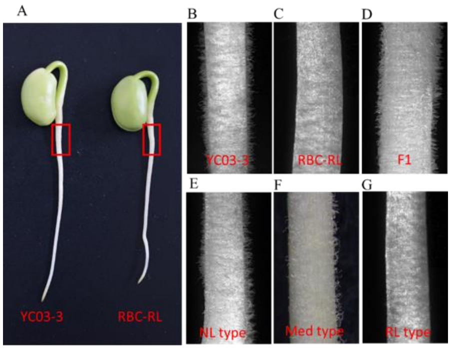

Fig. 1 The phenotype of root hairs among parents and progeny. A-D, Phenotype of root hairs in YC03-3, RBC-RL and F1 plants. Red box indicated the observation area of root hairs in A. E-G, Normal (NL), Medium (Med) and few (RL) root hairs in F_2 progeny.

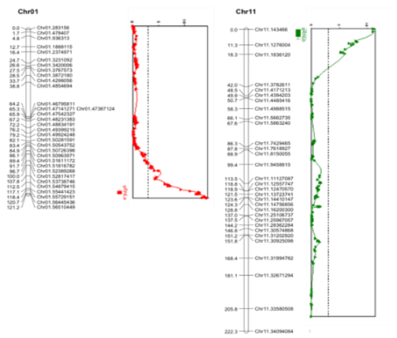

Fig. 2 Location of two quantitative trait loci (QTL) identified for root hairs in a simple genetic map in soybean.

CONCLUSION

In this study, two independent loci were identified for root hair development, which were further delimited to a 381 kb candidate region on the chromosome 01 and 580 kb candidate region on the chromosome11, respectively.

ACKNOWLEDGEMENTS

This work was financially supported by China National Key Program for Research and Development (2016YFD0100700).

REFERENCES

Kochian, L. V., O. A. Hoekenga, and M. A. Pineros. 2004. How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorous efficiency. Annual Review of Plant Biology **55:**459-493.

Müller, M. and W. Schmidt. 2004. Environmentally induced plasticity of root hair development in Arabidopsis. Plant Physiology **134:**409-41

P2-11

Adaptability of the acid soil stress in tea plants

Hiroto Yamashita[1], Akio Morita[1], Takashi Ikka[1]
[1]Faculty of Agriculture, Shizuoka University, 836 Ohya, Suruga-ku, Shizuoka, Japan
; yamashita.hiroto.16@shizuoka.ac.jp

INTRODUCTION

Acid soils significantly limit crop production worldwide because approximately 50% of the world's potentially arable soils are acidic (Kochian et al., 2004). Many crops grow poorly in acid soils owing to acid soil syndrome, which consists of multiple stress factors, including toxicities of aluminum (Al^{3+}), protons (H^{+}) and/or deficiencies of minerals such as calcium (Ca^{2+}) and phosphates (PO_4^{2-}) (Kochian et al., 2004). In contrast, tea plants (*Camellia sinensis* L.), known as one of the species adapted to acidic soil conditions, grow especially well because they can accumulate a large amount of Al in mature leaves, which does not inhibit but instead stimulates growth (Maysumoto et al., 1976; Morita et al., 2008). However, the physiological responses, genetic diversities and molecular mechanisms for multiple acid soil syndrome in tea plants is still unclear. Here, we report responses to various pH conditions with or without Al in tea plants.

MATERIALS AND METHODS

To evaluate growth, one-year-old rooted cuttings of tea cultivar 'Yabukita' were grown in a greenhouse at Shizuoka University in hydroponic solutions (Konishi et al., 1985) that were adjusted to various pH levels (2.8-7.5) with or without Al 400 μM. The solution were renewed every 2 d. After one month, plants were harvested, divided into six parts as described by Morita et al., (2008), then freeze-dried and weighed. To evaluate cell viability and gene expression, after growing under above condition which induced white new roots in control nutrient solution (pH 4.2 with Al 400μM), plants were transferred for 24 h and 48 h to the test nutrient solutions that were adjusted to various pH levels (2.8-5.8). After 24 h, root tips (0-1 cm) were excised, then immediately frozen in liquid nitrogen. After 24 h and 48 h, roots were stained with Evans blue (EB). EB staining was performed to visualize root cell viability as described by Jacyn et al. (1994). After staining, images of root tips were acquired using a digital camera. To analyze gene expression, we performed quantitative RT-PCR with SYBR Premix Ex Taq II (TaKaRa). Relative expression levels were calculated by using the comparative ΔΔCt method.

RESULTS AND DISCUSSION

The growth of whole tea plants was better at pH 3.2-4.2 with and without Al conditions. However, when compared with or without Al, the growth of white roots and new shoots (new leaves and new stems), which were those that had emerged during treatment, were vigorous with Al treatment, but were almost barely present without Al treatment. Root tips at pH 2.8 and 5.8 showed a stronger loss of cell viability than those at pH 3.2 and 4.2 by EB staining. These results suggest that tea plants adapted to low pH (3.2-4.2) conditions and more they needed to vegetatively grow the presence of Al in this pH conditions.

In gene expression, we focused on the genes involved in cellular H^{+} homeostasis, to reveal the molecular adaptation mechanism for the low pH of tea plants. We analyzed *Glutamate decarboxylase* (*GAD*) 1-3, which is a gene involved in the pH stat pathway called the GABA shunt, and plasma membrane H^{+}-ATPase (HA) genes that isolated two HA genes, *CsHA1* and *CsHA4*, from an RNA-seq database constructed in our lab. In root tips, *CsGAD1* expression was higher than *CsGAD2* and *CsGAD3*, and *CsHA1* expression was higher than *CsHA4*. In conditions without Al, *CsGAD1* expression was induced in response to low pH, but *CsGAD2* and *CsGAD3* expression was not. In contrast, in conditions with Al, all *CsGADs* expression did not change based on pH conditions. *CsHA1* and *CsHA4* also showed expression pattern in response to low pH such as *CsGAD1*. These results indicate that *CsGAD1*, *CsHA1* and *CsHA4* were involved in cellular H^+ homeostasis in tea plant roots.

REFERENCES

Jacyn Baker C, Mock NM, Baker CJ, Mock NM (1994) An improved method for monitoring cell death in cell suspension and leaf disc assays using evans blue. Plant Cell Tissue Organ Cult **39:** 7–12

Kochian L V, Hoekenga OA, Pi MA (2004) How Do Crop Plants Tolerate Acid Soils ? Mechanisms Of Aluminum Tolerance And. doi: 10.1146/annurev.arplant.55.031903.141655

Konishi S, Miyamoto S, Taki T (1985) Stimulatory effects of aluminum on tea plants grown under low and high phosphorus supply. Soil Sci Plant Nutr 31: 361–368

Matsumoto H, Hirasawa E, Seiichiro M, Takahashi E (1976) Localization of aluminium in tea leaves. Plant Cell Physiol **17:** 627–631

Morita A, Yanagisawa O, Takatsu S, Maeda S, Hiradate S (2008) Mechanism for the detoxification of aluminum in roots of tea plant (Camellia sinensis (L.) Kuntze). Phytochemistry **69:** 147–153

P2-12

Effect of Nitrogen Fertilization on Root Physiological Activity in Upland Rice

***Adibah Mohd Amin**[1]**, Zaharah A Rahman**[1]**, Mohamed Hanafi Musa**[1] **and Siti Nor Akmar Abdullah**[2]*
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia,
[2]Department of Agricultural Technology, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
; adibahamin@upm.edu.my

INTRODUCTION

Root is the most important organ for plant in nutrient uptake, both for macro- and micro nutrients (King et al., 2003; Gastal and Lemaire, 2002). The effect of N fertilization on root growth is partly understood. Studies of the relationship between root growth and N availability have shown a general tendency for plant roots to proliferate in nutrient-rich zones while root growth is suppressed in zones of low nutrient supply. Presumably, this response enables plants to compensate partially for non-uniform supplies of nutrients (Robinson, 1994). Root morphology affects the amount of water transpired by crop, the efficiency of water is used to produce dry matter and the proportion of dry matter that ends up in grain yield (Passioura, 1982). The aims of this study were to measure root physiological activity (bleeding rate) of selected upland rice varieties as influenced by nitrogen fertilization.

MATERIALS AND METHODS

The experiment layout was a factorial arrangement in a randomized complete block design (RCBD) with four replications. The treatment consisted of 5 upland rice landraces in 2 types of N fertilizers with 3 N levels. Treatments were applied as followed: (1) 0 kg/ha N of ammonium sulphate (2) 75 kg/ha N of ammonium sulphate (3) 150 kg/ha N of ammonium sulphate (4) 0 kg/ha N of potassium nitrate (5) 75 kg/ha N of potassium nitrate (6) 150 kg/ha N of potassium nitrate. All the N fertilizer were applied in 3 split applications. The root bleeding sap was collected from plants. The root bleeding rate (g h^{-1} $plant^{-1}$) was estimated from the increase in cotton weight (Fan et al., 2010). Data were analysed statistically using a one-way analysis of variance (ANOVA) from the statistical analysis software (SAS) 9.0 package. Means separation was performed using the least significant difference (LSD) at 5% level of significance.

RESULTS AND DISCUSSION

There are significant differences among landraces and fertilizer N rates on bleeding rate. Landrace III had higher bleeding rate compared to Landrace I and II (Table 1). Bleeding rate increased as the rate of N fertilizer applied increased. The results showed that N fertilizer application could promote better root growth, so the plant can take up more water and nutrients. The rate of bleeding sap also increased which suggest that the root activity was increased, and the water uptake and transfer from root to above ground part increased (Morita et al., 2000). Plant nutrient and water uptake are influenced by its root morphology, on the other hand, can also be influenced by many factors including the amount of N applied (Fan et al., 2010).

Table 1: Root bleeding of upland rice landraces as influenced by N fertilization

	root bleeding (g/hour/plant)						
	Rate of fertilizers						
	0 kg N/ha		75kg N/ha		150kg N/ha		
Landraces	**$(NH_4)_2SO_4$**	**KNO_3**	**$(NH_4)_2SO_4$**	**KNO_3**	**$(NH_4)_2SO_4$**	**KNO_3**	**means $_{landraces}$**
I	1.77	1.94	1.80	2.07	3.09	2.12	2.10^{b}
II	2.02	1.50	1.92	2.35	2.44	2.46	2.09^{b}
III	2.33	2.41	2.57	2.57	3.54	2.40	2.65^{a}
IV	2.46	2.28	2.30	3.16	2.82	3.46	2.77^{a}
V	2.49	2.28	3.16	2.83	2.15	2.67	2.58^{a}
means $_{Nrate}$	2.15^{b}		2.46^{ab}		2.72^{a}		
means $_{Ntype}$	2.46^{a}			2.43^{a}			

Values with the same letter(s) are not significantly different according to the LSD Test at $P \geq 0.05$

CONCLUSIONS

Results of the findings from this study suggested that these upland rice landraces had no preference on type of N to be taken up. The physiological activity of a whole root system in field-grown crops can be determined by bleeding rate as it is based on active water absorption caused by root pressure.

REFERENCES

Fan, J. B., Zhang, Y. L., Turner, D., Duan, Y. H., Wang, D. S. and Shen, Q. R. (2010). Root physiological and morphological characteristics of two rice cultivars with different nitrogen-use efficiency. Pedosphere. 20(4): 446–455. http://dx.doi.org/10.1016/S1002-0160(10)60034-3

Gastal, F. and Lemaire, G., 2002. N uptake and distribution in crops: an agronomical and ecophysiological perspective. J. Exp. Bot. 53, 789–799, http://dx.doi.org/10.1093/jexbot/53.370.789

King, J., Gay, A., Sylvester-Bradley, R., Bingham, I., Foulkes, J., Gregory, P., Robinson, D., 2003. Modelling cereal root systems for water and nitrogen capture: towards an economic optimum. Ann. Bot. 91, 383–390, http://dx.doi.org/10.1093/aob/mcg033.

Morita, S., Okamoto, M., Abe, J. and Yamagishi, J. 2000. Bleeding rate of field-grown maize with reference to root system development. Japanese Journal of Crop Science. Vol. 69 No. 1 pp. 80-85 DOI: 10.1626/jcs.69.80

Passioura, J. B. (1982). The role of root system characteristics in the drought resistance of crop plants. In: IRRI Drought resistance in crops with emphasis on rice. IRRI, Los Banos, Philippines, pp 71-82. http://books.irri.org/9711040786_content.pdf

Robinson, D., (1994). The responses of plants to non-uniform supplies of nutrients. New Phytol. **127,** 635–674, http://dx.doi.org/10.1111/j.1469-8137.1994.tb02969.x

P3- Soil-microbe-plant interactions at low pH.

P3-1

Plant-dependent soil bacterial responses following amendment with mineral and chemical fertilisers are driven by soil pH

Mickan, B[1]., A. Alsharmani[1], Z. M. Solaiman[1] and L.K. Abbott[1]
[1]UWA School of Agriculture and Environment (M079) and UWA Institute of Agriculture, The University of Western Australia
; lynette.abbott@uwa.edu.au

INTRODUCTION

Alternative fertilisers and biostimulants are gaining momentum as soil amendments (Abbott et al. 2018). Plant growth responses to some alternative fertilisers and microbial inoculants could increase microbial diversity and provide greater access to less soluble nutrients in soil by plants.

MATERIALS AND METHODS

The effect of fertilisers with different elemental solubilities was assessed for their effects on soil bacterial communities associated with two annual pasture species. The four fertiliser treatments were: no fertiliser (Control), mineral-based fertiliser at a rate of 15 kg ha^{-1} P (MnF), chemical fertiliser at a rate of 15 kg ha^{-1} P (CF), and a microbial inoculant (Mic). Two annual pasture plants, subterranean clover and Wimmera ryegrass, were grown separately and harvested after 10 weeks in a glasshouse experiment with application of these soil amendments. The plants were maintained with soil at 70% of field capacity. Bacterial communities in soil were assessed using high throughput sequencing analysis.

RESULTS AND DISCUSSION

Application of the chemical fertiliser and mineral fertiliser had the greatest increase in plant growth for subterranean clover and Wimmera ryegrass, but these treatments had the lowest alpha diversity of the bacterial community across most diversity indices assessed (including Fisher and Inverse Simpson indices). Additionally, these responses were plant-dependent with bacterial OTU richness being highly correlated with soil pH for subterranean clover (R^2 =0.61), but not for Wimmera ryegrass (Figure 1).

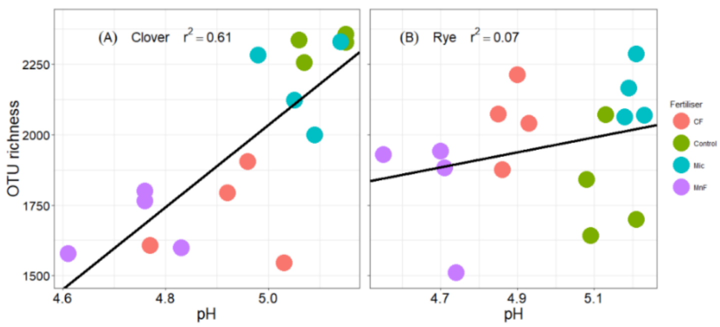

Figure 1: Bacterial community (OTU) richness compared with soil pH for soil amended with a chemical fertiliser (CF), a mineral fertiliser (MnF), a microbial inoculant (Mic) and untreated soil (Control) for (A) subterranean clover and (B) Wimmera ryegrass.

Richness was lowest following application of the chemical and mineral fertilisers when subterranean clover was grown. When Wimmera ryegrass was grown, richness was lowest for the control and mineral fertiliser. Beta diversity at the bacterial OTU level of resolution by permanova demonstrated a significant impact of soil treatments; plant species had a lesser impact on beta diversity. The dominant bacterial phylum was Actinobacteria in this soil in the presence of both subterranean clover and Wimmera ryegrass following application of all soil amendments and when the soil was untreated soil (Figure 2).

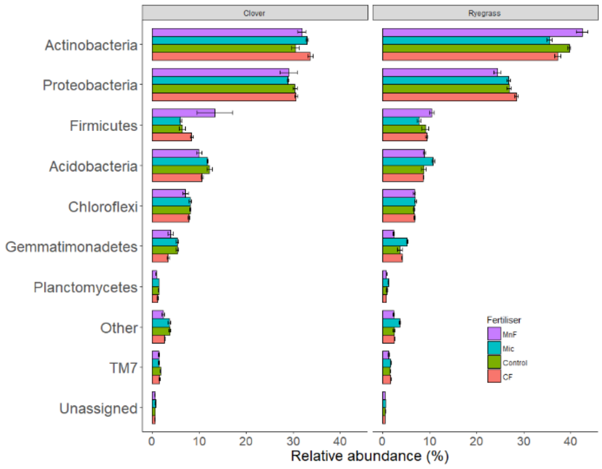

Figure 2: Relative abundance of bacterial phyla for soils amended with a chemical fertiliser (CF), a mineral fertiliser (MnF), a microbial inoculant (Mic) and for untreated soil (Control) in the presence of subterranean clover (Left) and Wimmera ryegrass (Right).

CONCLUSION

These data highlight the complexity of how soil amendments and microbial inoculants influence soil bacterial diversity and that caution is required when linking soil biodiversity to plant growth.

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REFERENCES

Abbott, L.K., Macdonald, L.M., Wong, M.T.F., Webb, M.J., Jenkins, S.N., Farrell, M. 2018. Potential roles of biological amendments for profitable grain production – A review. Agriculture, Ecosystems and Environment **256**:34-50

P3-2

Soil acidification induced by inorganic N fertilization affects soil diazotrophic population in a farmland ecosystem

Chao Wang[1], Man Man Zheng[1], Ren Fang Shen[1]
[1]State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China
; rfshen@issas.ac.cn

INTRODUCTION

To meet the food demand of an ever-increasing world population, the resulting intensive agriculture practices have included applying an overload of inorganic fertilizers (Liu et al., 2015). The increased long-term input of inorganic fertilizers, especially N-based fertilizers, in agricultural ecosystem improved soil fertility and crop yields over the past decades (Liu et al., 2015; Zeng et al., 2016) but also had various negative effects such as soil acidification, metal toxicity, lower nutrient use efficiencies, that threaten soil quality, crop growth, biodiversity, and environmental health (Guo et al., 2010; Zhong et al., 2015) and changed the biogeochemical cycles of soil nutrient elements (i.e., carbon (C), N, and P) (Geisseler and Scow, 2014). Thus, there is an increasing concern on how to safely enhance agricultural sustainability.

The N-fixing bacteria (diazotrophs) are responsible for biological N fixation, a functionally important biogeochemical N cycle (Levy-Booth et al., 2014). Because this bioprocess can supply additional N sources to the ecosystem to improve soil fertility, which is beneficial to plant productivity, it is a potentially sustainable alternative to inorganic N fertilizer use in agricultural systems (Gupta et al., 2006). However, the response patterns of diazotrophic bacteria to long-term fertilization in agricultural ecosystems are not well understood, although some short-term micro-zone experiments have been carried out (Rodríguez-Blanco et al., 2015; Simonsen et al., 2015). Detailed information on diazotrophic community composition and abundance, as well as their relationships to the soil environment, would improve our understanding of the long-term ecological effects of fertilization and of the biological functions of diazotrophic bacteria in the agroecosystem, as well as reduce inorganic N fertilizer use.

MATERIALS AND METHODS

The fertilization experiment started in 1990 at a site located in the Red Soil Experimental Station at Qiyang, Hunan Province, China (26^{o}45’ N, 111^{o}53’ E). The size of each fertilizer treatment was 20 m × 10 m and designed with two replicate plots. The cropping system was an annual rotation of summer maize (*Zea mays* L. Yedan 13) and winter wheat (*Triticum aestivum* L. Xiangmai 4). The inorganic fertilizers were applied as urea (300 kg N ha^{-1} year^{-1}), superphosphate (53 kg P ha^{-1} year^{-1}) and potassium chloride (100 kg K ha^{-1} year^{-1}). Before sowing, fertilizers were applied by banding at a depth of 10 cm. For annual input, 30% of fertilizers were applied for wheat and 70% for maize. As the plots receiving N and N+P+K (NPK) fertilizers showed severe soil acidification, these two plots were each divided into two parts in 2010. One part maintained the same fertilization as before, while the other also received 2,550 kg ha^{-1} of quicklime based on the same fertilization protocol, followed by the addition of 1,500 kg ha^{-1} of quicklime in 2014. In this trial, six different fertilizer treatments were chosen: unfertilized control (CK), inorganic N fertilizer alone (N), inorganic N fertilizer plus quicklime (NCa), inorganic P+K fertilizers (PK), inorganic NPK fertilizers, and inorganic NPK fertilizers plus quicklime (NPKCa).

Soil samples were collected on June 12, 2015 (at the maize flowering stage). The rhizosphere soils (0–15 cm depth) from five maize plants and bulk soil were collected. Shoots and roots of each plant were separately placed into envelopes and oven dried at 70°C to obtain a plant dry weight. The air-dried soils were analyzed for soil pH, total carbon (TC), total nitrogen (TN), soil organic carbon (SOC), available P (AP), and available K (AK). Fresh soil samples were used to determine soil ammonium (NH_4^+-N), nitrate (NO_3^--N), dissolved organic carbon (DOC), and organic nitrogen (DON).

The dinitrogenase reductase subunit gene *nifH* is the most used marker gene for analyzing the abundance and diversity of diazotrophic bacteria (Levy-Booth et al., 2014). Soil DNA was extracted from 0.5 g of soil (fresh weight). The *nifH* gene copy number was determined by real-time quantitative PCR (qPCR) on a LightCycler 480 real-time PCR system with SYBR®Premix Ex Taq™. The *nifH* gene-specific primers PolF/PolR (Poly et al., 2001) were used for qPCR amplification. A nested PCR approach was used to amplify the *nifH* gene fragments for pyrosequencing. The primer sets PolF/PolR and RoeschF/RoeschR (Roesch et al., 2006) were used in the first and second PCR amplification, respectively. The sequencing of the amplicon libraries was carried out on an Illumina MiSeq platform. Sequencing reads were processed with Mothur version 1.31.1, and data analysis was carried out using the vegan package of the R software (Version 3.1.2).

RESULTS AND DISCUSSION

Compared with the non-fertilized CK, the long-term N treatment significantly reduced the maize biomass, but the application of quicklime (NCa and NPKCa) markedly increased the biomass. The pH values displayed no significant differences between CK (5.38), NCa (5.20), and NPKCa (5.12), which were higher than those of N (4.04), PK (4.95), and NPK (4.00), with the lowest occurring under both N and NPK treatments. However, root effect (bulk versus rhizosphere soils) did not influence these parameters soil pH. These result suggested that the long-term input of N fertilizers is an important factor inducing the decrease of soil pH.

The *nifH* gene copy number in the rhizosphere was obviously higher than in the bulk soil under each treatment. Compared with CK, PK had a higher *nifH* gene copy number, whereas those of N and NPK were significantly lower. These inhibitory effects of N-based fertilizers were markedly alleviated by quicklime applications in NCa and NPKCa, and even the *nifH* gene copy number in NPKCa was significantly higher than in CK. Pearson's correlation analysis showed that in the bulk soil, the *nifH* gene copy number was positively correlated with soil pH, C/N, and AP and negatively correlated with NH_4^+-N and NO_3^--N. The input of N fertilizers (such as N and NPK treatments) reduced soil diazotrophic abundance, probably because of negative feedback from soil acidification or/and high N content. Such a negative response in microbial abundance to low soil pH has be well demonstrated in several long-term trials (Geisseler and Scow, 2014; Levy-Booth et al., 2014), resulting in soil pH being considered a decisive factor affecting soil microbial abundance. The significant increase in diazotrophic abundance after adding quicklime further supports this explanation.

In bulk soil, all of the fertilizer treatments significantly reduced the number of operational taxonomic units (OTUs). There were no significant differences among N, NCa, NPK, and NPKCa, suggesting that quicklime applications did not relieve the inhibitory effects of N fertilizer. These result showed that soil nutrients are as important as pH in decreasing the diazotrophic OTU richness when subjected to long-term inorganic fertilization.

The result of NMDS showed three different groups, as follows: bulk and rhizosphere samples of CK and PK; bulk and rhizosphere samples of N and NCa; and all of the samples of NPK and NPKCa. Thus, the influence of the fertilizer treatment on the community composition and structure was far more obvious than that of root effect and quicklime application. The Mantel test revealed that the diazotrophic community structures in the bulk soils were closely correlated with multiple soil variables, and the correlation coefficient followed the trend: C/N > NH_4^+-N > AK > NO_3^--N > AP > TC > SOC > DOC > pH. Redundancy analysis showed that the diazotrophic communities of both CK and PK treatment were associated with higher pH values. Soil pH has been well investigated in a number of ecosystems and is frequently considered an important determinant of bacterial community structure (Geisseler and Scow, 2014). However, such a decisive role was not observed in the current study, in which soil pH was less important compared with the soil nutrient availability. This result further confirmed that soil nutrients resulting from long-term inorganic fertilization have a greater effect than soil pH on soil diazotrophic community structure.

CONCLUSION

This study demonstrated that 25 years of inorganic fertilization resulted in strongly selective forces acting on the soil diazotrophic population. It is likely that N has a greater influence than P and K on diazotrophic bacteria. A main reason is soil acidification induced by inorganic N fertilization. And quicklime applications further confirmed this, which increased soil diazotrophic abundance. However, soil acidification showed less effect on diazotrophic community composition, compared with the soil nutrient availability. In addition, for the overall community composition, fertilizer treatments showed a greater influence than the rhizosphere effect. Thus, the different response patterns of diazotrophic abundance, community composition, and OTU richness to soil characteristics revealed a complicated mechanism behind the diazotrophic population's adaptation to long-term inorganic fertilization.

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REFERENCES

Geisseler, D., Scow, K.M., 2014. Long-term effects of mineral fertilizers on soil microorganisms-a review. Soil Biology & Biochemistry **75**, 54-63.

Gupta, V.V.S.R., Roper, M.M., Roget, D.K., 2006. Potential for nonsymbiotic N_2-fixation in different agroecological zones of southern Australia. Australian Journal of Soil Research **44**, 343-354.

Guo, J.H., Liu, X.J., Zhang, Y., Shen, J.L., Han, W.X., Zhang, W.F., Christie, P., Goulding, K.W.T., Vitousek, P.M., Zhang, F.S., 2010. Significant acidification in major Chinese croplands. Science **327**, 1008-1010.

Levy-Booth, D.J., Prescott, C.E., Grayston, S.J., 2014 Microbial functional genes involved in nitrogen fixation, nitrification and denitrification in forest ecosystems. Soil Biology & Biochemistry **75**, 11-25.

Liu, Y.B., Pan, X.B., Li, J.S., 2015. A 1961-2010 record of fertilizer use, pesticide application and cereal yields: a review. Agronomy for Sustainable Development **35**, 83-93.

Poly, F., Ranjard, L., Nazaret, S., Gourbiere, F., Monrozier, L.J., 2001. Comparison of *nifH* gene pools in soils and soil microenvironments with contrasting properties. Applied and Environmental Microbiology 67, 2255-2262.

Rodríguez-Blanco, A., Sicardi, M., Frioni, L., 2015. Plant genotype and nitrogen fertilization effects on abundance and diversity of diazotrophic bacteria associated with maize (*Zea mays* L.). Biology and Fertility of Soils **51**, 391-402.

Roesch, L.F.W., Olivares, F.L., Passaglia, L.M.P., Selbach, P.A., de Sa, E.L.S., de Camargo, F.A.O., 2006. Characterization of diazotrophic bacteria associated with maize: effect of plant genotype, ontogeny and nitrogen-supply. World Journal of Microbiology and Biotechnology **22**, 967-974.

Simonsen, A.K., Han, S., Rekret, P., Rentschler, C.S., Heath, K.D., Stinchcombe, J.R., 2015. Short-term fertilizer application alters phenotypic traits of symbiotic nitrogen fixing bacteria. Peerj 3, e1291.

Zeng, J., Liu, X.J., Song, L., Lin, X.G., Zhang, H.Y., Shen, C.C., Chu, H.Y., 2016. Nitrogen fertilization directly affects soil bacterial diversity and indirectly affects bacterial community composition. Soil Biology & Biochemistry **92**, 41-49.

Zhang, H.M., Wang, B.R., Xu, M.G., Fan, T.L., 2009. Crop yield and soil responses to long-term fertilization on a red soil in southern China. Pedosphere 1**9**, 199-207

P3-3

Rice husk biochar (RHB) influences on arbuscular mycorhizal fungi (AMF) and growth of maize

A.M. Akhir Rudin[1] and N.M. Jaafar[1]
[1]Department of Land Management, University Putra Malaysia, 43400 Serdang, Malaysia
; j_noraini@upm.edu.my

INTRODUCTION

Biochar, the pyrolysed material is currently preferred as an alternative soil amendment especially for its liming effects and its porous nature. Previous studies have reported the beneficial effects of varying biochar sources including rice husk biochar (RHB). RHB is one of the popular biochar currently applied as soil amendment in Malaysia due to its waste abundance. Biochar via several mechanisms may improve soil conditions and plant growth as well as affecting the activity of beneficial soil microorganisms such as arbuscular mycorrhizal fungi (AMF) (Warnock et al. 2007). However, many aspects are not known on the effects of RHB on AMF as well as their interactions in Malaysian highly weathered soils especially factors related to the pyrolysis condition and RHB amount applied to soil. Thus, this experiment was carried out with the objectives to determine the effects of rice husk biochar (RHB) on AMF and maize growth.

MATERIALS AND METHOD

This pot experiment was carried out with 4 different amounts of RHB (0, 1.5, 3.0 and 6.0 t/ha), with and without AMF inoculation on maize in completely randomized design with 4 replications in glasshouse Complex 11A, Faculty of Agriculture, Universiti Putra Malaysia. Three kilograms of Serdang series soil were placed per pots. The maize seeds were placed on the top of AMF inoculum. Maize plants were harvested 4 weeks after sowing. Maize plant growth, mycorrhizal root infection and spores in soil and soil nutrient concentrations were determined. Analysis of variance (ANOVA) was performed using Statistical Analysis System (SAS) software version 8.02 for Windows (SAS Institute Inc. (1992-98).

RESULTS AND DISCUSSION

Application of RHB or AMF or their interactions (AMF*RHB) had significant effects ($P \leq 0.05$) on the plant height (Table 1). Treatment 7 (T7) which is the combination of 3.0 t/ha RHB and AMF inoculation (+AMF) showed the highest in plant height with 86.25 cm as compared to control (without RHB). Maize shoot dry weight, root dry weight and root volume were not significantlly ($P \geq 0.05$) affected by either AMF or RHB application or their interactions (AMF*RHB) (data not shown). The application of varying amount of RHB noted that AMF spores were high in 1.5 t/ha RHB with 173 spores/10g soil. Root infection was not significantly affected ($P \leq 0.05$) by RHB or AMF application or their interactions (AMF*RHB). In conclusion, AMF could function well without full amount of RHB application (6.0 t/ha) (sporulated better at 1.5 t/ha RHB) while showing effects on plant height at 3.0 t/ha RHB.

Table 1: Effects of RHB on AMF and Maize Growth

Treatments	AMF Treatments	Amount of Biochar	Plant Height (cm)	AMF development	
				No. of spore / 10 g of soil	Root infection (%)
T1	-AMF	0 t/ha	62.00 b	61 b	4.15 a
T2	-AMF	1.5 t/ha	67.07 b	63 b	27.80 a
T3	-AMF	3.0 t/ha	79.50 ab	104 ab	14.20 a
T4	-AMF	6.0 t/ha	78.87 ab	126 ab	13.86 a
T5	+AMF	0 t/ha	81.35 ab	96 ab	27.08 a
T6	+AMF	1.5 t/ha	78.0 ab	**173 a**	26.13 a
T7	+AMF	3.0 t/ha	**86.25 a**	106 ab	9.75 a
T8	+AMF	6.0 t/ha	68.25 ab	68 ab	16.85 a
ANOVA PR>F VALUES					
FACTOR 1 (AMF)			0.0058*	0.0016*	0.41^{ns}
FACTOR 2 (RHB)			0.0044*	0.0027*	0.11^{ns}
INTERACTIONS AMF*RHB AMOUNTS			0.0006*	<.0001*	0.20^{ns}

** note: means with the same letter in column are not significantly different P≥0.05 according to Tukey's Standardised Range (HSD)

CONCLUSION

It was further concluded that AMF functioned well in the treatment without full amount of RHB The combined application of 1.5 t/ha to 3.0 t/ha RHB AMF with was enough to affect maize plant but higher amount of (6.0 t/ha) might be insignificant to promote AMF sporulation. At reduced amount of RHB application (1.5 t/ha), AMF might still benefit maize plant growth and soil properties. Future research should look into more trials on the effects of RHB on different AMF species at various maize growth stage.

REFERENCES

Warnock DD, Lehmann J, Kuyper TW and Rillig MC (2007). Mycorrhizal responses to biochar in soil–concepts and mechanisms. Plant and soil, **300**(1-2). pp.9-20

P3-4

Characterization of Silicate Solubilizing Bacteria from rubber plantation for growth promotion and antagonistic properties against *Rigidoporus microporus* pathogen

***Imran Shabbir*[1], *Radziah Othman*[1], *Zulkefly bin Sulaiman*[2], *Mohd Yusoff bin Abd Samad*[1], *Mui Yun Wong*[3] *and Noraini bt Md Jaafar*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
[2]Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia
[3]Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia 43400 Serdang, Selangor, Malaysia
; radziah@upm.edu.my

INTRODUCTION

Silicon (Si) enhances the growth and development in many plants and also decreases the incidence of many fungal diseases by strengthening the cell walls to increase resistance to the penetration of pathogenic fungi (Francois *et al*., 2005). Silicon benefits the plants by accelerating growth and mitigating the effects of biotic and abiotic stresses in several plants. Despite of its abundance in earth's crust, it is mostly present in insoluble forms. Silicate solubilizing bacteria (SSB) can play an efficient role here by solubilizing insoluble forms of silicates. *Rigidoporus microporus* is pathogenic fungus which causes white root disease in rubber trees and results in decreased latex productivity and can even cause death of plants. The application of SSB was reported to be one of the important mechanisms for the biological suppression of phytopathogens through polymerisation of silicate from the soil for plant uptake SSB are also advocated as a biofertilizer to solubilize silicates. However, studies on silicate solubilizing bacteria are limited. The present study was carried out to isolate and characterize silicate solubilizing bacteria and evaluation of antagonistic activity against *Rigidoporus microporus* pathogen.

MATERIALS AND METHODS

1. Collection of soil and roots samples from rubber plantation area

Rhizosphere soil and roots samples were collected from rubber plantation area in Universiti Putra Malaysia (UPM). Serial dilutions of each sample, ranging from 10^{-1} to 10^{-7} was prepared in sterilized water. For both rhizosphere soil and roots samples 1 mL aliquot of the appropriate dilution was spread on Luria Bertani (LB) agar.

2. Screening for Silicate Solubilisation: Selected bacterial isolates were purified and plated on LB agar plates, and were screened for silicate solubilisation activity.

2.1. Plate Assay: 20 µL of each bacterial isolates was placed on glucose agar (glucose 10g; agar 20g; distilled water 1000 ml; pH 7.0-7.2) plates containing silicate medium with 0.25% insoluble magnesium trisilicate (Vasanthi *et al*., 2013). After 4 days of incubation period, solubilization index (SI) was measured.

2.2. Liquid Assay: Basal medium containing calcium silicate (0.25 g) in 100 ml polyacrylate bottles sterilized & inoculated with a loopful of 24 hours fresh culture (Vasanthi *et al*., 2013). After 10 days of incubation, silica concentration was determined.

3. Phosphate (P) and Potash (K) solubilization and Nitrogen fixation activity: 20 µL of each bacterial isolates were placed on respective media. After 4 days of incubation period, solubilization index (SI) was measured for P and K solubilization and observed for change in color of the media from pale green for N fixation activity.

4. *In-vitro* Antagonistic assay: Antifungal activity of SSB isolates was checked against *Rigidoporus microporus* by dual culture test using an agar plug of the fungus and respective SSB isolates on PDA media. After 4 days of incubation period, the percentage of inhibition of radial growth (PIRG) was determined.

5. Experimental design and statistical analysis: All the *in-vitro* experiments were conducted in a complete randomized design with five replications. The data were analyzed using SAS statistical software and mean differences were compared using Least Significant Difference test at the 5% level of probability.

Table 1. Silicate, P, K solubilization, N-fixation and antagonistic effect of Silicate Solubilizing Bacteria (SSB) against *Rigidoporus microporus* pathogen

Strain	Solubilization Index (SI)			Nitrogen fixation	Silicate Solubilization (mg/L) DAI*=10	Antagonism (PIRG**)
	Silicate	Phosphate	Potassium			
SSB-4	1.26 d	1.68 b	1.25 c	++	9.05 b	41.48 b
SSB-7	4.67 a	2.52 a	2.61 a	+++	11.55 a	57.27 a
SSB-8	1.75 c	1.29 c	1.55 bc	++	9.38 b	41.70 b
SSB-9	2.51 b	1.13 c	1.62 bc	++	9.07 b	42.11 b
SSB-10	2.55 b	1.13 c	1.83 b	+++	10.14 ab	45.12 b
Control	-	-	-	-	7.36 c	-

+ indicate strength for nitrogen fixation
*DAI-Days after inoculation; **PIRG- Percent Inhibition of Radial Growth
Means within the same column followed by the same letter are not significantly different at $P > 0.05$.

RESULTS AND DISCUSSION

A total of 13 potential SSB were isolated from the healthy rubber roots and rhizosphere. Five bacterial isolates out of 13 bacterial isolates that showed silicate solubilization, were selected for further studies. The plate assay showed that the highest silicate (solubilizing index 4.67), phosphate (solubilizing index 2.52) and potassium solubilization (solubilizing index 2.61) were observed for bacterial isolate SSB-7. The liquid assay for silicate solubilization also showed that the highest solubilization (11.55 mg/L) was observed for bacterial isolate SSB-7 (Table 1). This finding was supported by Muralikannan and Anthomiraj (1998), who reported that efficient silicate solubilizing bacteria can help release other essential nutrients in soil. All SSB isolates showed the nitrogen fixation ability. SSB antagonistic activity against *Rigidoporus microporus* showed that largest percent inhibition (PIRG 57.27%) of radial growth was recorded for SSB-7 isolate followed by SSB-10 (PIRG 45.12%). These bacteria can not only directly combat phyto-pathogenic fungi but indirectly by release of Si in soil which in turn induce disease resistance in plants (Sahebi *et al*., 2015).

CONCLUSION

The antagonistic potential of these silicate solubilizing bacteria in lab condition is an important ability of practical utility in development of sustainable approach against the *Rigidoporus microporus* pathogen. These bacterial isolates will be further investigated for suppression of *Rigidoporus microporus* pathogen in rubber seedlings under glasshouse condition for a possible development of bioformulation for commercial purpose.

ACKNOWLEDGMENT

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REFERENCES

Francois, F., Wilfried, R.B., James, G.M. and Richard, R.B. (2005) Silicon and Plant Disease Resistance against Pathogenic Fungi. FEMS Microbiology Letters, **249,** 1-6.

Muralikannan, N. and Anthomiraj, S. (1998) Occurrence of Silicate Solubilizing Bacteria in Rice Ecosystem. The Madras Agricultural Journal, **85**, 47-50.

Sahebi, M., Hanafi, M., Siti, A., Akmar, N., Rafii, M.Y., Azizi, P., Tengoua, F.F., Azwa, J.N.M. and Shabanimofrad, M. (2015) Importance of Silicon and Mechanisms of Biosilica Formation in Plants. Biomed Research International, 2015, Article ID: 396010.

Vasanthi, N., Saleena, L.M. and Raj, S.A. (2018) Silica Solubilization Potential of Certain Bacterial Species in the Presence of Different Silicate Minerals

P3-5

Effect of soil pH on phosphorus and mycorrhizal availability in cocoa orchard

Izzah, A.H*[1]*., Daylon, J.H*[1]*., Wan Asrina, W.Y*[1]*. and Chubo, J.K*[1]*.
[1]Department of Crop Science, Faculty of Agriculture and Food Sciences,
Universiti Putra Malaysia Bintulu Sarawak Campus, 97008 Bintulu, Sarawak, Malaysia
; asrina@upm.edu.my

INTRODUCTION

Soil pH in tropical soil was reported to effect P solubility which later becomes unavailable to the crop. However, P availability can be increased with the presence of mycorrhizal colony in the soil surroundings. Because of this, understanding the relationship between soil pH, P and mycorrhizal availability may become a solution to overcome P depletion (Clark, 2002). Therefore, the objective of this research was to determine the effect of soil pH on P and mycorrhizal availability in three cocoa zones.

MATERIALS AND METHODS

The research was conducted in a cocoa orchard located at Universiti Putra Malaysia Bintulu Sarawak Campus (UPMKB). Three zones (i.e. A, B and C) were identified based on plant age and landform, and eventually soil samples (0 to 20 cm) were taken accordingly. A total of 115 cocoa trees were recorded with zone A, B and C having 21, 44 and 50 trees, respectively. The collected soil samples were air dried and analysed for soil pH using water (Tan, 2011), available P was extracted using double acid extraction and determine using UV/Vis Spectrophotometers at 882 nm (Tan, 2005) and mycorrhiza's spore extraction using sucrose method by Ianson and Allen (1986). All samples were analysed in triplicates and subjected to means of variance through Tukey's studentized test (ρ=0.05) using statistical analysis system SAS Ver. 9.4 and correlation study using principal component analysis (PCA) through XLSTAT Ver. 2014.

RESULTS AND DISCUSSION

Results on soil parameters are shown in Table 1 with P availability and soil pH was insignificantly different in each zone. Generally, soil pH in this orchard indicated extremely to very strongly acidic which may show P availability fixed by Al and Fe (Brady and Weil, 2017). In acidic condition (pH <5), solubility of Al and Fe were higher and attributed to decreasing P availability (Hartemink, 2005). Therefore, about 11.72, 14.41 and 14.06% P uptake were recorded in zone A, B and C, respectively.

Table 1: Soil pH and P availability in three different cocoa zones

Zone*	pH	Available P (mg kg^{-1})
A	$4.54^a \pm 0.08$	$12.98^a \pm 2.28$
B	$4.30^a \pm 0.04$	$15.96^a \pm 2.95$
C	$4.31^a \pm 0.09$	$15.57^a \pm 2.06$

* average crop age between five to ten years
** similar alphabet was insignificantly different at ρ=0.05

The correlated relationship is shown in Figure 1 with 74.59%. The F1 component indicated positive on mycorrhizal (Mycorr) and soil pH and negative on available P (P). This result has revealed P availability in all zones has closed relationship with Mycorr and pH values. Whereas the concentration of P may be controlled by Mycorr and pH. According to Clark (2002), pH has major effect on P availability while Mycorr makes P availability increased steadily. However, Mycorr and pH eventually had indicated mutual relationship where changes in pH have unaffected the Mycorr inoculation. This can be clarified through capability of Mycorr to withstands acidic condition, typically in tropical soil (Ortas *et al.*, 2007; Clark, 2002).

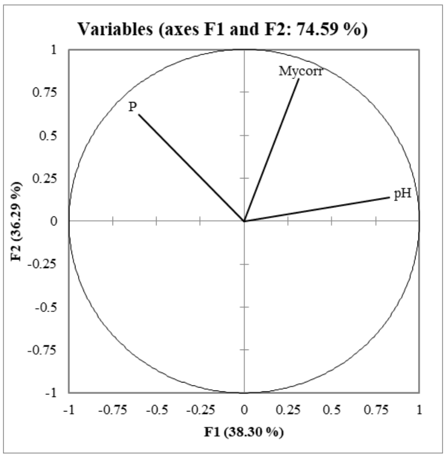

Figure 1: PCA analysis of soil pH, available P and mycorrhizal availability

CONCLUSION

Phosphorus availability and soil pH was insignificantly being affected by cocoa zone. Symbiotic relationship between soil pH and mycorrhizal inoculation was observed that lead to consistent amounts of P.

REFERENCES

Brady, N. C., & Weil, R. R. (2017). *The Nature and Properties of Soils.* Edinburgh Gate, England: Pearson Education Limited.

Clark, R. (2002). Differences among mycorrhizal fungi for mineral uptake per root length of switchgrass grown in acidic soil. *Journal of Plant Nutrition*, **25**: 1753-1772.

Hartemink, A. E. (2005). Nutrient stocks, nutrient cycling, and soil changes in cocoa ecosystems: a review. *Advances in agronomy*, **86**: 227-253.

I**anson, D. C., & Allen, M. F. (1986).** The effects of soil texture on extraction of vesicular-arbuscular mycorrhizal fungal spores from arid sites. *Mycologia*: 164-168.

O**rtas, I., Rowell, D., & Harris, P. (2007).** Effect of mycorrhizae and pH change at the root-soil interface on phosphorus uptake by sorghum using a rhizocylinder technique. *Communications in soil science and plant analysis*, **35**: 1061-1080.

Tan, K. H. (2005). *Soil Sampling, Preparation, and Analysis.* Boca Raton, FL: Taylor & Francis.

Tan, K. H. (2011). *Principles of Soil Chemistry.* Boca Raton, FL: Taylor & Francis.

P3-6

Microbial population in soil amended with various green manures

***Nur Liyana, I*[1]*., Norziana, Z.Z*[1]*., Illani Zuraihah, I*[1]*., and Zurin Aida, J*[1]**
[1]Soil Science and Water Programme (SS3), Crop and Soil Science Research Centre
Malaysian Agricultural Research and Development Institute (MARDI), MARDI Headquaters Serdang, Malaysia
; liyanais@mardi.gov.my

ABSTRACT

Green manures are an ideal method of sustaining soil fertility in the tropics and in organic farming, for both soil fertility and microbial activity. The presence of a diverse soil microbial community is crucial to the productivity since microorganism affects all levels within the ecosystem. Field studies were carried out to compare how green manures *Arachis pintoi*, *Medicago sativa*, *Gliricidia sepium* and *Moringa oleifera* affected the microbial community in the soil. Results showed that green manure had significant impacts on microbial population. These findings show that the addition of green manures improved soil biology by increasing microbial population.

INTRODUCTION

Microbial diversity in soils is considered important for maintaining sustainability of agricultural production systems. It is widely believed that the return of legume or other green manure improve the soil fertility, however it is not easy to measure improvement in soil fertility in the short-term. Soil microbe which is a small but labile component of soil can be used as an early indicator of changes occurring in soil and responds quickly to changes in soil management and is used as an indicator of soil quality (Biederbeck *et al.*, 2005).

MATERIALS AND METHODS

2.1 Site Description and Sampling

The study was carried out at soil health management research plot which was established in Integrated Organic Farm, MARDI Serdang, Selangor. The experiment was laid out in a randomized complete block design (RCBD) consisted of five (5) treatments and four (4) replications. There were total of 20 beds measuring 2 meter x 3 meter size for each treatment and replicate. The treatments were applied as follows: T1: Control; T2: *Arachis pintoi* (planted as cover crop); T3; *Medicago sativa* (planted as cover crop); T4: *Gliricidia sepium* (leaf as mulch); T5: *Moringa oleifera* (leaf as mulch). *Gliricidia sepium* and *Moringa oleifera*'s leaves were applied as mulch every 1 month for 4 months of cultivation period. Soil samples (0-20 cm) were collected before planting and after harvest from each treatment plot and ground to pass through 12 mm mesh before subjected to laboratory analysis. Analysis of variance was conducted to test for treatment effect while means of treatments were compared using Tukey's test.

2.2 Microbial Analysis

The number of soil microorganisms was determined using the dilution spread plate technique. Nutrient agar (NA) and potato dextrose agar (PDA) were used to culture bacteria and fungi, respectively

RESULTS AND DISCUSSION

Soil analysis for microbial analysis was conducted on three occasions (beginning, middle and end) of the experiment. Figure 3.1 showed soil amended with gliciridia (2.3x10^5 cfu g-1 soil) and moringa (5.6x 10^5 cfu g-1 soil) has highest microbial population compared to soil treated with medicago (2.4x10^4 cfu g-1 soil), arachis (8.0x10^4 cfu g-1 soil) and control (6.3x10^4 cfu g-1 soil). In this study, soils treated with green manures had slightly increased the C/N ratio (8.6 to 9.1) and organic carbon content (1.05 to 1.3 cfu g^{-1} soil).

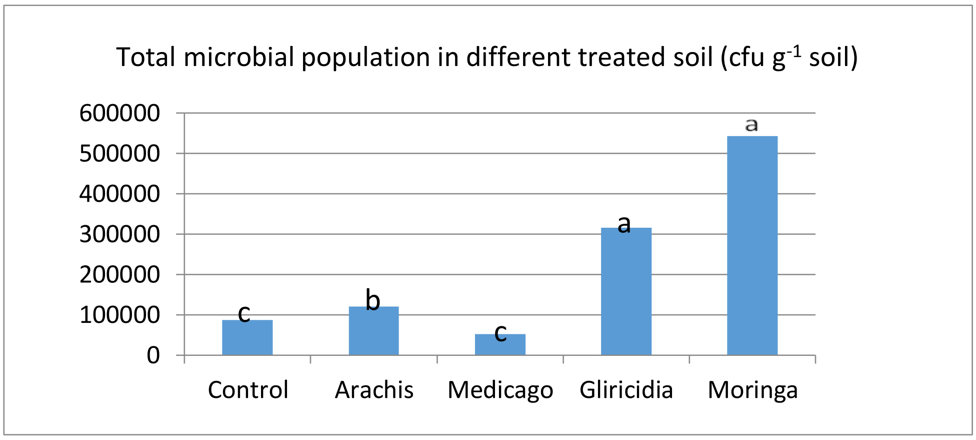

Figure 3.1. Application of moringa and gliricidia has significantly increased the microbial population in the soil

In the present study, higher colony forming unit (CFU) values for the three microb es i.e bacteria, fungi and actinomycetes were found in soil amendment with green manures and it increased from the beginning towards end of experiment (Figure 3.2). Meanwhile, numbers of bacteria and fungi in the non-treated green manure soil was significantly lower ($p \leq 0.05$). We speculated that the increased biodiversity were resulted from the increased soil nutrients. It has been reported that green manure had positive influence on microbial biomass (Cherr *et al*., 2006) which was consistent with our results.

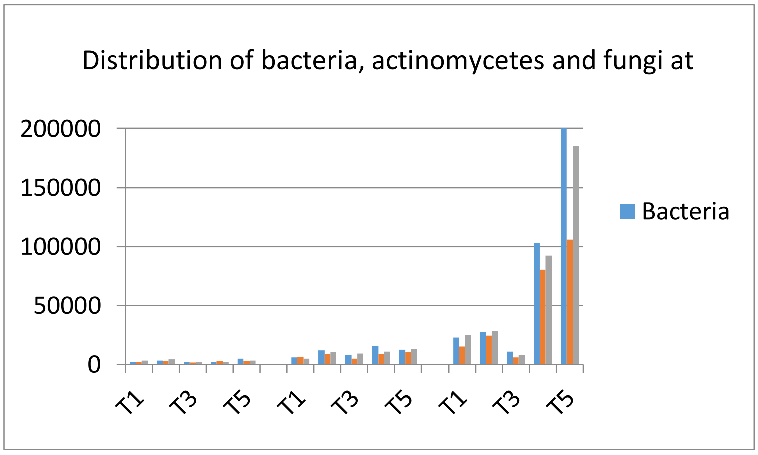

Figure 3.2: Bacteria dominated the microbial population followed by fungus and actinomycetes

Soil microbial community diversity was higher in groups treated with green manure. It is suggested that the increased biodiversity resulted from the increased soil nutrients (from the green manure). Green manure had positive influence on soil microbe population, which was consistent with the results. Increased microbial population in soil could be due to enhanced organic matter inputs from the green manure legumes (Stromberger *et al*., 2007).

CONCLUSION

As the conclusion, the application of green manure such as Moringa, Gliricidia and alfalfa increased the soil microbe population in the soil.

REFERENCES

Biederbeck, V.O., R.P. Zentner and C.A. Campbell. 2005. Soil microbial populations and activities as influenced by legume green fallow in a semiarid climate. Soil Biology and Biochemistry. **37**(10): 1775-1784.

Cherr, C., Scholberg, J., McSorley, R. (2006). Green manure approaches to crop production. Agronomy Journal. **98**(2):302-319.

N'Dayegamiye, A., and Tran, T. S. Effects of green manures on soil organic matter and wheat yields and N nutrition. Canadian Journal of Soil Science. **81** (4): 371-382

Stromberger, M., Z. Shah and D.G. Westfall. 2007. Soil microbial communities of no-till dryland agroecosystem across an evaporation gradient. Applied Soil Ecology, **35**: 94-106.

Sui, X., Feng, F., Lou, X., Zheng, J., and Han, S., 2012. Relationship between microbial community and soil properties during natural succession of forest land. African Journal of Microbiology Research, **6**(42), 7028-7034.

Zahir Shah, S., Rashidi Ahmad., and Hidayat Rahman. (2010). Soil microbial biomass and activities as influence by green manure legumes and fertilizer in rice wheat system. . Pakistan Journal of Botany. **42**(4): 2589-2598

P3-7

Effect of different media on growth of orange spike Medinilla (*Medinilla scortechinii*)

***Mohamed Hafeifi, B.*[1], *Zulhazmi, S.*[1], *Hanim, A.*[1], *Sakinah, I.*[1], *Intan Nadhirah, M.*[3], *Mohd Seth, S.*[2] *and Zulkifli, M. S.*[2]**

[1]Pusat Penyelidikan Hortikultur, IbuPejabat MARDI, 43400, Serdang, Selangor, Malaysia
[2]Pusat Penyelidikan Hortikultur, MARDI Cameron Highlands, 39000, Tanah Rata, Pahang, Malaysia
[3]Pusat Penyelidikan Sains Tanaman dan Tanah, IbuPejabat MARDI, 43400, Serdang, Selangor, Malaysia
; hafeifi@mardi.gov.my

INTRODUCTION

Orange Spike Medinilla (*Medinilla scortechinii*), from the family of Melastomataceae, to be a new native potted plant introduce in floriculture industry. Currently, research information on suitable media combination is rarely found especially when using conventional media substrate. This study was done to evaluate the effect of different combination media on growth performance of Medinilla scortechinii.

MATERIALS AND METHODS

The experiment was done at MARDI Cameron Highlands, under 50% of shaded area with the temperature between 16ºC to 26ºC. Relative humidity was at 70-90%. Rooted cuttings of the plant species (*Medinilla scortechinii*) not exceeding 5 cm long or at least with single internodes were used as planting materials. They were planted in 20 cm pots using various combinations of media mixtures as follow: *1)*100% sphagnum moss (T1), *2)*1 part perlite: 1 part peatgrow (T2), *3)*1part perlite: 1 part peatgrow: 1 part cocopeat (T3), *4)*1 part soil: 1 part sand: 1 part peatgrow (T4) (control).
Each pot was filled with 2 litres of media combination. NPK 15:15:15 were mixed in the media. The treatments consist of 5 pots/treatments were arranged in RCBD with four replications. The parameters taken were mortality of the plant, plant height, branch count, and leaf count. All data were taken weekly (week one until week five).
Analysis was performed on data using SPSS version 17.All data was analysed with Kolmogorov-Smirnov Test for the normal distribution. All abnormal data were tested using Kruskal-Wallis test. Comparison of significant values were made using Mann-Whitney U test. Differences were considered to be significant at $P<0.05$.

RESULTS AND DISCUSSION

Transplanting Shock Rate

The rooted cutting media used were 100% perlite. The rooted cuttings were transplanted into new media combination with the media attached to the root zone. The rooted cutting used were visually analysed with the presence of primary and secondary roots. Results showed that the means for T1, T2 and T3 were not significantly different (Fig 1). However, T4 give significantly different value compared to T1, T2 and T3. T4 showed the highest mortality rate compared to others. The transplanting shock effect is crucial in nursery management. Highest mortality rate in transplanting will reduce the economic value of producing new planting materials of *Medinilla scortechinii*. Hence, T1, T2 and T3 gave good interactions between media before transplanting and after transplanting.

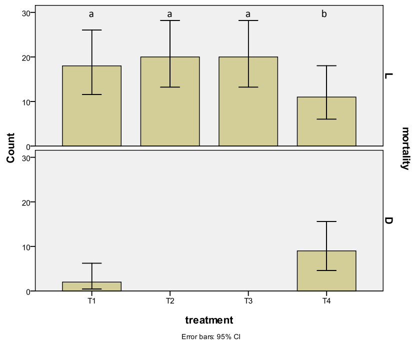

Fig 1: Mortality Rate of *Medinilla scortechinii*

Note: Means with the same letters are not significantly different at P<0.05according to Mann-Whitney U Test. L and D represent Live and Dead respectively.

Plant Height and Relative Plant Growth Rate

The highest mean of plant height was T1 (mean: 38.68 cm) compared to T2 (mean: 26.10 cm), T3 (mean: 23.89 cm) and T4 (mean: 11.89 cm), thus gave significant difference value (Fig 2). Means for T2 and T3 are not significantly different. Data from week one until week five for all treatments are not significantly different. However, relative growth rate (cm/day) showed no significant difference for all treatments (Fig 3). We can assume that the growth of *Medinilla scortechinii* on all media combination were the same, morphologically and physiologically. Hence, all media combination showed no effect on plant growth rate. Data on biomass (dry weight) were not taken due to limited samples of planting materials. Overall, all treatments showed positive growth on plant height and relative growth rate.

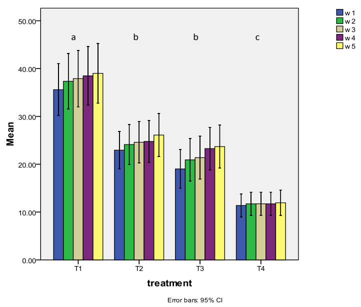

Fig 2: Mean Plant Height of *Medinilla scortechinii*

Note: Means with the same letters are not significantly different at P<0.05according to Mann-Whitney U Test. W1, W2, W3, W4 and W5 represent data taken on Week 1, Week 2, Week 3, Week 4 and Week 5, respectively.

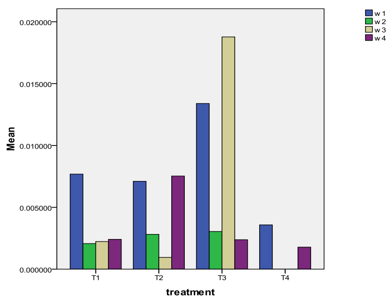

Fig 3: Relative Plant Growth Rate of *Medinilla scortechinii*

Note: Means with the same letters are not significantly different at P<0.05according to Mann-Whitney U Test. W1, W2, W3, W4 and W5 represent data taken on Week 1, Week 2, Week 3, Week 4 and Week 5, respectively.

Flowers and Fruits

Flowers were absence during this experimental observation. New technique to enhance flowering and fruiting will be discussed and developed further. However, according to Camara and Veldkamp (2011), pruning is the best and economic way to provide new shoot and new flowers. Fruits and seeds productions in Medinilla sp. were dependent on environmental conditions including suitable temperature, relative humidity and the presence of pollinators.

pH media

Medinilla scortechinii is favoured to grow in high organic matter. Some of the plants are grown on rock areas that have high level of moisture. Medinilla sp. grows well under acidic condition. pH was determined to select suitable potted media for *Medinilla scortechinii*. From the result (Fig 4), T1 (4.31) show highly significant difference amongst all media treatments. T3 (6.13) and T4 (5.98) were not significantly different to each other. T2 (6.54) gave the highest pH reading and significantly different from T1, T3 and T4. Comparing pH with other parameters, *Medinilla scortechinii* gave same growth performance in T1, T2 and T3 media. However, as T4 is conventional potting media for almost any tropical plants, *Medinilla scortechinii* show significantly low growth performance compared to other treatments. Thus, *Medinilla scortechinii* can grow well in acidic condition and organic media combination especially to grow under nursery conditions.

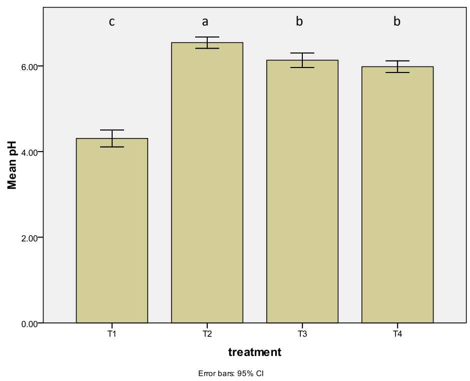

Fig 4: pH Media of *Medinilla scortechinii*

Note: Means with the same letters are not significantly different at P<0.05 according to Mann-Whitney U Test. d1, d2, d3, d4 and d5 represent data taken on Week 1, Week 2, Week 3, Week 4 and Week 5, respectively.

DISCUSSION

This study provides information on the growth performance of *Medinilla scortechinii* using rooted cuttings taken from Close Capillary Propagation System (CCPS) as planting materials. The planting materials were then transferred into four different media combinations. All the rooted cuttings were developed and grew well. Sphagnum moss planting media material was found to be the best media for the growth and development of *Medinilla scortechinii*. Meanwhile, combination of two materials, namely peatmoss and perlite can be used as potting media for the species, while combination of conventional media used in ornamental nursery were not preferred. Based on this study we can recommend the media combination also can be used for other Medinilla sp. and Melastomataceae family plants.

CONCLUSION

The plant growth data showed that T1 gave higher plant height compared to the other treatments. This appeared that the growth was probably enhanced by high organic content and high moisture content compared to the other media combination. Treatments T4 gave the lowest plant growth and higher in mortality rate, thus gave an indication that conventional method use as potted media are not suitable for the plant species (*Medinilla scortechinii*). Slightly acidic potted media are found to be favourable for the plant to grow. It is preferred that sphagnum moss can be suitable potted media for *Medinilla scortechinii*. Sphagnum moss sometime is hard to find, and expensive. Thus, using of other media (1 part perlite: 1 part peatgrow) combination is recommended.

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REFERENCES

Ab KaharSandrang, Sakinah Idris, Zabedah Mahmood. (2013). Lemon Myrtle: Tanaman Baru yang Berpotensi di Malaysia. BuletinTeknologi MARDI, Bil. **3** (2013): 41-45

Dytham, C. (2003). Choosing & using statistics. A biologists guide. York: Blackwell Publishing Company. 248 pp.

Dunnet and Hitchmough, J.D.(2004). The dynamic landscape: Design, ecology and management of naturalistic urban planting, Spon Press, London

R. Cámara-Leret and J.F. Veldkamp.(2011).A remarkable new Medinilla (Melastomataceae) from Celebes (Sulawesi), Indonesia.Gardens' Bulletin Singapore **62** (2): 213–221. 2011

Zulhazmi,S. (2015). Naturalistic planting design: A new concept in urban landscape. Scientia MARDI. Vol. 6: 3

P3-8

Effect of arbuscular mycorrhiza fungion the early establishment of *Juniperus procera* Hochst. ex Endl. In Saudi Arabia

***Amal A. M. AL-Ghamdi*[1]*, and Hasnah M. Jais*[2]**
[1]Department of Botany, Environment Program, Faculty of Biological Sciences, King Abdul Aziz University, P.O. Box 35009, Jeddah 21488, Saudi Arabia.
[2]Agrobiology Program, School of Biological Sciences, Universiti Sains Malaysia, 11800 Penang, Malaysia.
; amalalgamdi@gmail.com

INTRODUCTION

The principal problem for *J. procera* to germinate is thought to be due to seed dormancy (Teketay, 1993).In Asir, a region located in the southwest of Saudi Arabia, vast areas of productive woodlands of *J. procera* are rapidly declining (IUCN, 2010).

Seeds can be pre-treated to speed up and synchronize the germination through stratification and water soaking, which can disrupt physical dormancy in juniper seeds (El-Juhany et al., 2009).

Arbuscular mycorrhiza can influence the establishment of plants initially and help young plants to absorb nutrients from the soil. This eventually affects seedling establishment at reforestation sites (Smith et al., 2004). Therefore, this study was conducted with following objectives of 1) to evaluate artificial germination of *J. procera* seeds using different pre-treatments, and 2) to study the effects of arbuscular mycorrhiza fungi (AMF) and soil physico-chemical properties on the germination of *J. procera* seeds.

MATERIALS AND METHODS

Seed treatments

The ripe cones of *J. procera* grown in Al-Baha, southwestern region of Saudi Arabia, were collected in this experiment. The seeds of *J. procera* were extracted from the cones by manual de-pulping.

To determine seed viability, these were placed into a container of water after their extraction from the cones. Viable seeds were pre-treated before the beginning of the experiment to determine the germination percentage of the seeds by scarification, cold stratification, soaking in water and without treatment (El-Juhany *et al.* 2009). After pre-treatment, seedsm were germinated in the greenhouse was documented and the proportion of germinated seeds were computed.

Mycorrhizal inoculation

The seeds were inoculated using 50g of soil inoculums containing mycorrhizal. Mycorrhizal inoculums were not applied to uninoculated seeds. Nursery and field soil used in this experiment were sterilized. The experiments were replicated five times.

Germination of juniper seeds

Ten seeds from each pre-treatment were used in each pot. The seeds were watered daily to provide moisture. The experiment was replicated five times and monitored weekly for thirteen weeks.

Soil physico-chemical properties

Soil pH was determined according to the method by (Conklin, 2005). Soil organic matter was determined according to the method by Wilde et al., (1992) and soil moisture was determined in accordance to the methods by Yousef, (1999) and Conklin, (2005).

Soil analysis

After air-drying, the soil samples were sieved using a 2 mm mesh-size sieve. The Technicon Auto Analyzer II was used to determine the amount of N in the soil by employing the Kjeldahl technique. The Molybdenum Blue Method, which makes use of ammonium fluoride as extractant (Bray and Kurtz No. 2), was used to determine the P concentration in the samples. Using a wavelength of 660 nm, the UV Spectrophotometer 120-01 was employed to measure color formation. The amount of Ca, K and Mg in the samples were determined using the NH_4OAc, pH 7 leaching method, and then determined using atomic absorption spectrophotometer/ 5100 (Perkin Elmer).

Statistical analysis

Data on seeds that were successfully germinated were transformed into an arc-sine value before using one-way ANOVA for analysis. Means were then separated using Tukey's honest significant difference ($P < 0.05$). Paired t-test were used to determine any differences in the composition of N, P, K, Ca and Mg between nursery and field soil and also differences in soil physico-chemical properties (pH, organic matter and soil moisture) between these two soil types at $\alpha = 0.05$. All analyses were done using Statistix® Version 7.0 (Analytical Software, Tallahassee, Florida).

RESULTS AND DISCUSSION

Seed germination

Table 1 shows the proportion of germination in all pre-treatments, with and without AMF in nursery and field soils. In general, the germination percentage was higher in seeds inoculated with AMF and grown in the nursery soil (44.0 ± 8.1 to 58.0 ± 8.8 % compared to seeds grown in the field soil, of which 26.0 ± 10.3 to 36.0 ± 9.3% successfully germinated).

For seeds planted in nursery and field soil, significant differences in the percentage of germination was also observed when these were pretreated with tap water, with and without AMF. Overall, regardless of soil type, inoculation with AMF resulted in a higher percentage of germination.

Table 1: Mean (± S.E) germination percentage of *J. procera* seeds in different soils, with or without inoculation of AMF after pre-treatments.

Pretreatment	Germination (%, mean ± S.E.)			
	Nursery soil		Field soil	
	With AMF	Without AMF	With AMF	Without AMF
Sulphuric acid (98%)	48.0 ± 6.6$^{a(a)}$	28.0 ± 7.3$^{a(a)}$	30.0 ± 5.5$^{a(a)}$	4.0 ± 2.4$^{a(b)}$
Cold stratification	44.0 ± 8.1$^{a(a)}$	36.0 ± 9.3$^{a(a)}$	38.0 ± 6.6$^{a(a)}$	16.0 ± 6.8$^{a(a)}$
Tap water	58.0 ± 8.8$^{a(a)}$	26.0 ± 10.3$^{a(b)}$	42.0 ± 8.6$^{a(a)}$	8.0 ± 3.7$^{a(b)}$
Control	46.0 ± 5.1$^{a(a)}$	34.0 ± 10.3$^{a(a)}$	34.0 ± 6.0$^{a(a)}$	2.0 ± 2.0$^{a(b)}$

[a] Means in the same column followed by the same letters were not significantly different, $P > 0.05$ (Tukey's honest significant difference).
[a] Means in the same row followed by the same letters were not significantly different at $\alpha = 0.05$ (paired t-test).

In the field soil, a significant difference in the percentage of germination was observed when the seeds were treated with 98% sulphuric acid. In this study, the germination of *J. procera* seeds after a 20-minute immersion in sulphuric acid varied from 4% to 48% depending on the soil source and presence of AMF.

The proportion of pre-treated seeds that successfully germinated was highest in AMF-inoculated seeds grown in nursery soil (>40%). The least percentage of germination was observed amongst non-inoculated seeds grown in field soil (< 20%). It appears that AMF associations show benefit only during specified phases of growth (Johnson, 1997), including the seedling (Gange et al., 1993) and reproductive phase (Wilson and Hartnett, 1998).

Soil analysis

Significant differences were found between nursery soil and field soil for soil pH, content of organic matter and moisture. These factors can possibly affect the germination of *J. procera* seeds. The pH level in the nursery soil was nearly neutral whereas that of field soil was slightly alkaline.

Initially, the field soil was drier, with approximately 4.6% soil moisture compared to the nursery soil, which had around 14.9% soil moisture also the organic mater content of the field soil was lower than that of the nursery soil. Previous research (Finch-Savage et al., 1994) suggested that the success of seed germination depended mainly on the seeds' accessibility to moisture. The results of this study showed that the concentration of the nutrients Ca, Mg, N, K and P was significantly higher in nursery soil than in field soil (Table 2). However, Ca and P concentration in both nursery and field soil were not significantly different. Among the five nutrients, Mg recorded the highest amount, measuring about 1116 mg/kg and 795 mg/kg in the nursery soil and the field soil, respectively.

In general, the tested field soil had a lower nutrient content compared to nursery soil. Furthermore, in the absence of AMF, a lower germination percentage was recorded. However, in the presence of AMF, seed germination was significantly higher although the nutrient content was lower than in the nursery soil. In the control plants, the germination of *J. procera* seeds was better in nursery soil (with or without the inoculation of AMF) and field soil (with the inoculation of AMF).

Table 2: Mean (± S.E) of elements analysis in the nursery and field soil.

Elements analysis (mg/kg)		Soil	
		Nursery	Field
i)	Nitrogen	793.3 ± 17.6[a]	216.7 ± 8.8[b]
ii)	Phosphorus	76.0 ± 1.5[a]	94.0 ± 20.6[a]
iii)	Potassium	870.0 ± 63.5[a]	60.2 ± 46.0[b]
iv)	Calcium	1027.7 ± 26.6[a]	611.4 ± 259.9[a]
v)	Magnesium	1116.3 ± 52.6[a]	795.3 ± 3[b]

[a]Means in the same row followed by different letters were significantly different at $\alpha=0.05$ (Paired t-test).

CONCLUSION

Generally, pre-treatment of *J. procera* seeds in the respective soil types (nursery and field soil) were not necessary for germination. Nevertheless, the presence of AMF improved seed germination. Soil organic matter, moisture and nutrients (N, K and Mg) were relatively higher in the nursery soil than in the field soil. This factor should be considered in establishing seed germination for future preparation of seedling for replantation.

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REFERENCES

Clark, R.B. & S.K. Zeto. (2002) Arbuscular mycorrhizae: mineral nutrient and water acquisition. In: Sharma A.K. & B.N. Johri, (eds). Arbuscular mycorrhizae: Interactions in plants, rhizosphere and soils, Enfield: Science Publishers Inc. Pp.159-187.

Conklin, A.R. (2005) Introduction to Soil Chemistry. Analysis and Instrumentation, 3rd Edition, Hoboken: John Wiley and Sons. P.218.

El-Juhany, L.I., I.M. Aref & M.A. Al-Ghamdi. (2009) Effects of Different Pretreatments on Seed Germination and Early Establishment of the Seedlings of *Juniperus procera* Trees. World Applied Sciences Journal, 7, p.616-624.

Finch-Savage, W.E., K. Phelps & J.R.A. Steckel. (1994) Seedling emergence modeling and the timing of irrigation during vegetable crop establishment. In: Fourth National Symposium on stand establishment of horticultural crops. California Pp.103-108.

Gange, A.C., V.K. Brown & G.S. Sinclair. (1993) Vesicular-arbuscular mycorrhizal fungi: a determination of plant community structure in early succession. Functional Ecology, 7, p.616-622.

IUCN. (2010) International Union for Conservation of Nature Red List of Treatment Species. *Juniperus procera* (East African Cedarwood). [Online] 2010.4, [Assessed 20th February 2011], Available from World Wide Web: www.iucnredlot.org

Johnson, N.C., J.H. Graham & F.A. Smith. (1997) Functioning of mycorrhizal associations along the mutualism-parasitism continuum. New Phytologist, 135, p.575-585.

Pérez-Fernández, M.A., E. Calvo-Magro, J. Montanero-Fernández & J.A. Oyola-Velasco. (2006) Seed germination in response to chemicals: Effect of nitrogen and pH in the media. Journal of Environmental Biology, 27, p.13-20.

Smith, S.E. & D.J. Read. (2008) Mycorrhizal Symbioses. 3rd Edition. London: Academic Press. P.787.

Smith, S., F. Smith & I. Jacobsen. (2004) Functional diversity in arbuscular mycorrhizal (AM) symbioses: the contribution of the mycorrhizal P uptake pathway is not correlated with mycorrhizal responses in growth or total P uptake. New Phytologist, 162, p.511-524.

Teketay, D. (1993) Problems associated with raising trees from seeds. In: H. Lieth & M. Lohmann (eds). Restoration of tropical forest ecosystems. Dordrecht: Kluwer. Pp.91-100.

Wilde, S.A., G.K. Voigt & J.G. Iyer. (1972) Part 1: Analysis of physical properties of soils. In: Soil and Plant Analysis for Tree Culture. 4th Edition. New Delhi: Oxford and IBH Publishing Co., Pp.6-34.

Wilson, G.W.T. & D.C. Hartnett. (1998) Interspecific variation in plant responses to mycorrhizal colonization in talgrass prairie. American Journal of Botany, 85, p.1732-1738.

Yousef, A.F. (1999) Analysis methods and devices for soil and water. In Arabic: أجهزه وطرق تحليل التربه والمياه. King Saud University, Riyadh.

P4- Soil fertility and chemistry and amelioration and remediation of low pH soils.

P4-1

A Combined effect of nitrogen and phosphorus on the N uptake and yield of Bambara groundnut (*Vigna subterranea)*

***Md Mahmudul Hasan*[1], *Md Kamal Uddin*[1], *Mahmud Tengku Muda Mohamed*[1], *Ali Tan Kee Zuan*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
43400 UPM Serdang, Selangor, Malaysia
; mmhasanlimon93@gmail.com

INTRODUCTION

Bambara groundnut is an African leguminous crop which goes to genus *Vigna* and family Papillionaceae. Bambara groundnut react positively on many agronomic parameter with the application of N and P fertilizer. For completing a balanced food the ripe seeds of Bambara groundnut contain protein 16 to 21%, fat 4.5 to 6.5% and carbohydrate 50 to 60%. Limited research work is reported on the effects of N and P on Bambara Groundnut especially in Malaysia. Hence, the current study was employed to fill the gap by evaluating the impact of N and P on N content and yield of Bambara groundnut.

MATERIALS AND METHODS

A pot experiment was conducted for four month (January to April) in the Agriculture Faculty at the Ladang 15 of the Universiti Putra Malaysia, Malaysia. The size of the pot was 65.94 cm^2. Randomized Complete Block Design (RCBD) with four replications was used. Each unit was containing 10 kg of soil. 16 treatments of this experiment was the combination of Urea and Triple Super Phosphate (TSP) fertilizer along with the control. Total Nitrogen content was determined by dry combustion technique using a LECO CR-412 carbon analyzer. Shelling outturn (%) = $Ws/\ Wp$ x 100 for groundnut; Ws = weight of groundnut seed, and Wp = weight of groundnut pods. Harvest index (%) = A/ B x 100; A = pod weight, B = total plant weight with pod. The highest (A) percent (%) increase of parameters compared to control (B) treatment were measured by the this equation, Increase = A – B / B x 100. If the value of control is greater than other treatment then calculate as percent (%) decrease using same formula.

RESULTS AND DISCUSSION

Both the rates of application of N and P in the form of Urea and TSP fertilizer exerted significant influence on the number of Mature pod per plant (MPPP), 100- Seed weight (SW-100), Harvest index (HI) and Pod weight per plant (PWPP),N contentment in leaf (LN),Shoot (SN), root (RN) of the crop. This result clearly supported with those of (VAGHASIA & BHALU, 2016) who reported that increasing the rate of applied N and P fertilizer enhanced N uptake by leaf, shoot, root in all growth stages .Variation in SW-100 was also observed for the different N and P levels and varied from 60.75 to 71.38 g; the lowest being recorded for T_1 (control) and the highest was recorded in T_{16}. The highest SW-100 (g) was 1.93% compare to control (T_1). The results clearly showed that the SW-100 follow an increasing trend with the increasing rate of N and P fertilizer.

Similar observation was reported by (M. Altab hossain and A. hamid, 2007).Other researchers reported that application of N and P fertilizer combined with full rate of compost regarding its quality significantly increased legumes yields (Verde & A, 2014).

Table 1. Effect of N and P on N content and yield parameter of Bambara groundnut.

Treatments	LN	SN	RN	MPPP	PWPP (g)	SW-100 (g)	SP(%)	HI(%)
T_1	1.71i	1.05g	1.21g	30.25c	31.33e	60.75e	56.33c	33.54cd
T_2	1.78hi	1.09fg	1.33e-g	30.75c	34.45de	61.25de	64.93ac	32.72d
T_3	2.13fg	1.20e-g	1.45d-f	32.00bc	39.75b-d	67.88a-c	62.50a-c	36.43b-d
T_4	2.76bc	1.64b	1.75bc	37.75bc	41.33bc	67.75a-c	69.38a-c	35.09b-d
T_5	1.99gh	1.21d-f	1.29fg	32.00bc	35.80c-e	62.75b-e	59.25a-c	34.07b-d
T_6	2.34ef	1.23d-f	1.51de	33.00bc	38.08b-d	63.63b-e	70.75ab	34.62b-d
T_7	2.41e	1.36cd	1.65cd	32.00bc	39.25b-d	66.13a-e	69.53a-c	36.21b-d
T_8	2.90a-c	1.76b	1.82bc	32.25bc	40.83bc	68.00a-c	71.78a	36.56b-d
T_9	2.31ef	1.26c-e	1.38e-g	32.00bc	37.75b-d	62.50c-e	60.93a-c	35.02b-d
T_{10}	2.40e	1.30c-e	1.51de	33.00bc	40.65bc	66.38a-e	61.28a-c	36.81b-d
T_{11}	2.46de	1.40c	1.65cd	31.75bc	39.60b-d	64.25b-e	68.65a-c	36.06b-d
T_{12}	2.99ab	1.78b	1.95ab	39.50ab	42.98b	68.23ab	72.73a	37.84ab
T_{13}	2.24e-g	1.29c-e	1.47d-f	31.75bc	38.68b-d	63.13b-e	57.10bc	36.79b-d
T_{14}	2.48de	1.36cd	1.62cd	32.00bc	41.35bc	64.13b-e	65.50a-c	37.14bc
T_{15}	2.69cd	1.42c	1.75bc	32.50bc	41.90b	67.23a-d	66.10a-c	37.44bc
T_{16}	3.10a	2.00a	2.11a	47.25a	50.13a	71.38a	61.35a-c	41.61a
$LSD_{0.05}$	0.25***	0.16***	0.22***	8.17*	5.69***	5.92*	14.09ns	4.13*

LSD = least significant difference. Means with the same letter have no significant difference, $P \leq 0.05$ = significant (*), $P \leq 0.01$ = significant (**), $P \leq 0.001$=significant (***).

CONCLUSION

Based on the results of this study, it could be concluded that the optimum application of N and P fertilizer (N-30 and P-60 kg/ha) was the most effective approach to improve growth, uptake of N and yield of Bambara Groundnut than, the other combination as compared to the control.

REFERENCES

M. Altab Hossain And A. Hamid, (2007). Effect of nitrogen, phosphorus and zinc application on yield and uptake of nutrients by groundnut. *Bangladesh J. Agril. Res*, **32**(3), 369–374.

Verde, B. S. M. M., & A. (2014). Effects of manure, lime and phosphorus fertilizer on soil properties and soybean, Kenyatta University

Vaghasia, P. M., & Bhalu, V. B. (2016). Nutrient requirement for bold seeded confectionery groundnut (*Arachis hypogaea* L.) under irrigated condition. *International Journal of Agricultural Sciences*, **12**(1), 81–84.

P4-2

Effects of vegetation and flooding on nitrogen cycling in ozegahara mire, central Japan

***H. Shigeta*[1], K. Inubushi[1], and M. Sakamoto[2]**
[1]Graduate School of Horticulture, Chiba University, 648 Matsudo, Matsudo-shi, Chiba, Japan
[2]Scool of Environment Science, The University of Shiga Prefecture, 2500 Hassaa, Hikone, Shiga, Japan
; ahwa4256@chiba-u.jp

INTRODUCTION

Ozehgahara Mire is the largest high moor and peatland in the main island of Japan. It has a high level of biodiversity and was registered under Ramser Convention in 2005. However, recent reports indicate accelerated environmental changes in Ozegahara Mire, such as the increase in frequency of heavy rain causing more frequent flooding on the mire. The flooding could supply external nutrients to oligotrophic peat soil. Another one is the increase of the distribution area and biomass of *Myrica gale* L. var. *tomentsa* with nitrogen fixing mycorrhiza (Ohmori *et al.*, 2009), which may cause nitrogen enrichment in the peat soil. It is important to understand the effects of flooding and the increase of density of *M. gale* vegetation on nitrogen dynamics in acid peatland soil. This study aimed to investigate the relationships among soil physico-chemical properties, nitrogen dynamics in the peat soil, the density of *M. gale* vegetation, and flooding.

MATERIALS AND METHODS

Ozegahara Mire is located at 35° 55-57' N 139° 12-15' E and about 1400 m above sea level. Mean annual temperature and the highest temperature are 5.4°C and 24.5°C, respectively. Peat soil in Ozegahara Mire is made mainly of *Sphagnum* materials.

Table 1 Sampling sites and vegetation types, flooding, water content, and pH (H_2O) of soil

Site	Individual density of *M. gale*	Flooding*	Dominate plant species	Water content (%)	pH (H_2O)
K-NM-f	no	flooded	*Osmunda cinnamomea*	74.1	4.7
K-HM-f	high	flooded	*Myrica, Carex* spp.	91.1	5.7
N-NM-nf	no	non-flooded	*Carex* spp., *Moliniopsis japonica*	94.4	4.3
N-LM-f	low	flooded	*Myrica, Carex* spp.	92.2	5.3

*Whether the study sites have been flooded or not flooded were decided from distribution pattern of riverine micro sediment particles (Sakaguchi and Soma, 1999).

Soil sampling was conducted from July 31 to August 2, 2017. Four sites with different vegetation types were selected (Table 1). We measured water contents, soil pH (H_2O) with glass electrode, ammonium nitrogen and nitrate nitrogen content in soil water, acetylene reduction activity (ARA) in soil to measure nitrogen fixation activity and denitrification rate using acetylene inhibition method with the anaerobic and aerobic incubation experiments. After wet soil (5 g) was placed in bottles and pre-incubated at 15°C for 24 hours, acetylene was injected to a 10 % v/v in the headspace of bottle and incubated at 25°C for 48 hours. After incubation, gas samples were taken to be analyzed with GC-FID (C_2H_4) and GC-ECD (N_2O) to measure ARA and denitrification rate, respectively.

Table 2 Ammonium and nitrate nitrogen, acetylene reduction activity, and denitrification rate

Site	Ammonum N	Nitrate N	ARA (nmol C_2H_2/g wet soil/day)		Denitrification (µg N/kg wet soil/ day)	
	(mg N/kg wet soil)		Anaerobic	Aerobic	Anaerobic	Aerobic
K-NM-f	13.3	19.4	0.8	0.07	49.5	6.9
K-HM-f	12.5	1.5	8.9	2.19	3.8	2.0
N-NM-nf	9.4	1.7	27.9	1.24	2.7	4.8
N-LM-f	17.5	1.4	2.2	0.72	2.1	4.9

RESULTS AND DISCUSSION

Water content (Table 1) in K-NF-f on the hummock was the lowest. It is assumed that more aerobic condition on the hummock increased nitrate nitrogen content and result in increase of denitrification rate (Table 2). It has been generally regarded that denitrification activity was low in low pH environment activity (Simek and Cooper, 2002). However, any significant correlation between soil pH and denitrification rate was not clearly observed in this study. ARA was high at the site with less inorganic nitrogen (Table 2). Soil pH at the site of high and low density occurrence of *M. gale* sites were higher than at no *M. gale* sites. There was no significant difference between *M. gale* and soil nitrogen dynamics in this investigation. Inorganic nitrogen of non-flooded site was the lowest. The source of nitrogen supply was needed to be revealed.

CONCLUSION

To evaluate the effect of the increase of *Myrica gale* L. var. *tomentosa* and flooding in Ozegahara Mire, Central Japan, we measured soil physico-chemical properties and nitrogen dynamics. Density occurrence of *M. gale* tended to be higher in the area with higher soil pH. Regardless of *M. gale*, inorganic nitrogen was important factor of nitrogen fixation and denitrification in soil. Inorganic nitrogen contents in flooded sites showed higher.

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REFERENCES

M. Simek, and J.E. Cooper, 2002. The influence of soil pH on denitrification: progress towards the understanding of this interaction over the last 50 years, European Journal of Soil Science, **55**: 345-354.

T**. Ohmori, Y. Yamamura, and Y. Hori, 2009.** Distribution patterns of *Myrica gale* var. *tomentosa* and its relationship to microtopography in the Ozegahara Mire, central Japan. Vegetation Science, **26**(1): 1-8.

Y. Sakaguchi, and H. Sohma, 1998. Geological problems related to Ozegahara, in: Oshima Y (ed) Scientific researches of the Oze area in central Japan. Oze Scientific Research Group, 85–106. (In Japanese)

P4-3

Changes in red soil pH and crop yield under different fertilization regimes

***Wen, S*[1]*., B. Wang*[1]*, and Z. Cai*[1]**
[1]Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing 100081; Qiyang Agro-ecosystem of National Field Experimental Station, Hunan 426182, China
; caizejiang@caas.cn

INTRODUCTION

Approximately 6.5 % (~57 Mha) of the total arable land in China is the highly weathered acidic red soil (Ferralic Cambisol) (FAO 2006). Especially in the last 30 years, the red soils are becoming more acidic due to increased use of N fertilizers (Guo et al. 2010; Zhou et al. 2014). Soil acidification with nutrient deficiency, especially calcium (Ca) and phosphorus (P), and aluminum (Al) toxicity has become a major cause of severe yield. Animal manure has the potential to increase soil pH (Naramabuye and Haynes 2006). The objectives of this study were to investigate the effect of different fertilization regimes on soil pH, crop yield, and plant uptake of nitrogen (N), phosphorus (P) and potassium (K) as well as their relationship based on 18-year data of the field fertilization experiment (by 2008), and evaluate the best fertilization practices for sustainable agriculture production in the red soil.

MATERIALS AND METHODS

A long-term field experiment was carried out since 1990 at Qiyang Red Soil Research Station of Chinese Academy of Agricultural Sciences, located in Hunan Province. Six fertilization treatments were examined: (1) no fertilizer (control), (2) chemical N fertilizer only (N), (3) chemical N and P fertilizers (NP), (4) chemical N, P and K fertilizers (NPK), (5) manure only (M), and (6) chemical NPK plus manure (NPKM). Winter wheat and summer corn were grown in rotation every year. Urea was applied at 300 kg N ha^{-1} $year^{-1}$ for all chemical N required treatments. Superphosphate and potassium chloride were applied at 120 kg P_2O_5 ha^{-1} $year^{-1}$ and 120 kg K_2O ha^{-1} $year^{-1}$, respectively for all chemical P or K required treatments. For the NPKM treatment, 30% of the total N was applied as urea and the remaining 70% was applied as total nitrogen in the manure. The manure and urea were applied to soil at the same time, followed by sowing of each crop, and then covered with surface soil. For annual input 30% of total N was applied when wheat was planted and 70% of total N was applied when corn was planted. Due to field size limitation, each treatment was duplicated in relatively large plots (196 m^2) that allowed common field operation and management similar to farm’s field. During each harvest, crop yields were determined, and plant samples were collected. Soil samples were collected after corn harvest in the fall each year. To obtain representative samples, five cores of soils at 0–20 cm depth from each treatment plot were collected using a 5-cm inner diameter auger and thoroughly mixed. The soil samples were air dried, ground, sieved to pass through a 1.0-mm sieve for pH measurement. The selected chemical properties of a Ferralic Cambisol in 1990 was shown in Table 1.

Table 1: Selected chemical properties

	pH	Organic C	Total properties			Available properties			Exchange properties		
			N	P	K	N	P	K	Al^{3+}	H^+	CEC
		(g kg^{-1})				(mg kg^{-1})			(cmol(+) kg-1soil)		
Soil	5.70	6.06	1.07	0.52	13.7	79.0	13.9	104	0.10	0.17	8.99

RESULTS AND DISCUSSION

After 18-year fertilization, soil pH followed the order among treatments: N<NP=NPK <Control<NPKM<M. The significant effect of long-term application of urea-N on soil acidification was observed in the N, NP, and NPK treatments. The N only treatment had the greatest decrease in soil pH by 1.5 units from the initial 5.7. The control showed no change in soil pH, which was significantly higher than that in chemical fertilizers only treatments. In contrast, the M treatment showed about 1.0 unit increase and the NPKM showed no change in soil pH. Over the 18-years, wheat yield decreased significantly at the rates of 11 to 104 kg ha^{-1} $year^{-1}$ in control or chemical fertilizers only treatments. In contrast, wheat yield in manure treatments (M and NPKM) remained high with no decrease, and the highest yield (1639 kg ha^{-1}) was found from the NPKM treatment. The control (no fertilizer) and all chemical fertilizers only also markedly decreased corn yield at the rates of 24 to 210 kg ha^{-1} $year^{-1}$. Manure application (M and NPKM) resulted in no change or significant increase at 101 kg ha^{-1} $year^{-1}$ in corn yield. For N, NP and NPK treatments, wheat yields were found positively and significantly correlated with soil pH (r=0.815**, r=0.814** and 0.675**, $P<0.01$, respectively), and corn yields also showed positive and significant correlations with soil pH in the N treatment (r=0.926**, $P<0.01$) and NP treatment (r=0.720**, $P<0.01$). Thus, soil acidification from chemical N applications was one of the main factors for crop yield decrease.

CONCLUSION

Soil acidification from chemical N fertilizer is proven to be one of the main factors limiting crop yields. In contrast, continuous annual applications of manure as 100 or 70 % of total N source resulted in either an increase or unchanged soil pH during the 18-year study period. Addition of manure in combination with chemical fertilizers maintained the red soil pH and achieved high yields in the wheat and corn rotation system and thus in the long run can be the best fertilization management model for preventing red soil acidification, maintaining soil productivity, and achieving sustainable crop production. Further studies should determine the minimum amount of manure that can prevent acidification while increasing soil fertility to sustain crop yield.

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REFERENCES

Food and Agriculture Organization (FAO). 2006. World reference base for soil resources 2006—a framework for international classification, correlation and communication. World Resources Reports, Rome.

Guo, J. H., X. J. Liu, Y. Zhang, J. L. Shen, W. X. Han, W. F. Zhang, P. Christie, K. W. T. Goulding, P. M. Vitousek, and F. S. Zhang. 2010. Significant acidification in major Chinese croplands. Science **327**: 1008–1010.

Naramabuye, F. X., and R. J. Haynes. 2006. Short-term effects of three animal manures on soil pH and Al solubility. Australian Journal of Soil Research **44**: 515–521.

Zhou, J., X. Fang, X. M. Liu, Y. He, J. M. Xu, and P. C. Brookes. 2014. Effects of nitrogen fertilizer on the acidification of two typical acid soils in South China. Journal of Soils and Sediment **14**: 415–422.

P4-4

Quantification of manure required to prevent red soil acidification in an 8-year maize field experiment

***Cai Z*[1]., *B. Wang*[1], *and S. Wen*[1]**
[1]Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing 100081; Qiyang Agro-ecosystem of National Field Experimental Station, Hunan 426182, China
E-mail: caizejiang@caas.cn

INTRODUCTION

Soil acidification, along with the deficiency of nutrients particularly calcium and phosphorus, and aluminum toxicity, has become a major cause of crop failure or yield reduction in the red soil (Ferralic Cambisol) region in southern China (Ritchie 1989; Wang et al. 2008). Conventional organic fertilizers (i.e., animal manure) application could inhibit soil acidification by the return of base cations, ammonification of labile organic N in manure, and decarboxylation of organic anions (Butterly et al. 2012; Rukshana et al. 2012; Xiao et al. 2013). However, little information is available on the amount of manure needed to prevent acidification and maintain the soil productivity for high crop yield. This research determined the effects of various combinations of manure with urea on acidification process in an 8-year maize field experiment at Qiyang, Hunan Province, southern China.

MATERIALS AND METHODS

The field experiment was carried out since 2009 at Qiyang Red Soil Research Station of Chinese Academy of Agricultural Sciences. Treatments included chemical N, P and K fertilization (NPK), and NPK plus manure at 20% (NPKM1), 40% (NPKM2), and 60% (NPKM3) of total N supplied from swine manure with a total N input of 225 kg N ha^{-1} $year^{-1}$. Urea was used for all chemical N required treatments. Superphosphate and potassium chloride were applied at 32.7 kg P ha^{-1} $year^{-1}$ and 62.2 kg K ha^{-1} $year^{-1}$, respectively for all chemical P or K required treatments. The manure and chemical fertilizers were applied to soil at the same time, followed by sowing maize, and then covered with surface soil. All treatments were tested in triplicate. During harvest, crop yields were determined, and plant samples were collected. Soil samples were collected after maize harvest in the fall each year. Soil pH, exchangeable acidity, and maize yield were determined annually from 2009 through 2016 with soil exchangeable base cations measured in 2016. The initial (2009) topsoil (0–20 cm) had a total SOC of 7.16 g kg^{-1}; a total N (TN) of 0.96 g 95 kg^{-1}; a bulk density (BD) of 1.18 g cm^{-3}; a pH (1:1 w:v) of 4.81; and available N plus P and 96 potassium (K) of 61.49, 10.71, and 303.39 mg kg^{-1}, respectively.

RESULTS AND DISCUSSION

Soil acidity and maize yield were significantly affected by swine manure application rates. Soil pH decreased from an initial value of 4.81 to 4.46 and 4.62 by 2016 in NPK and NPKM1 treatments, respectively, with increased soil exchangeable acidity (dominated by Al^{3+}) by 3.24 and 1.48 $cmol^{(+)}$ kg^{-1}, decreased exchangeable base cations (to 1.92–3.53 $cmol^{(+)}$ $kg^{-1}$of exchangeable Ca and to 0.23–0.84 $cmol^{(+)}$ $kg^{-1}$of exchangeable Mg), and reduced maize yields to 2430 and 4815 kg ha^{-1}, respectively. In contrast, at manure incorporation rate of 40% or 60% of total N ($NPKM_2$ or $NPKM_3$) either no change or a significant increase in soil pH (to 5.47), increased exchangeable base cations to 4.38–5.31 $cmol^{(+)}$ $kg^{-1}$of exchangeable Ca and to 1.23–1.62 $cmol^{(+)}$ $kg^{-1}$of exchangeable Mg, and the highest yield (5193 and 5411 kg ha^{-1}, respectively) were achieved. Swine manure high rich in ash alkalinity (82.7 cmol$^{(+)}$ kg^{-1}). During manure decomposition, ammonification of labile organic N in manure and decarboxylation are the mechanisms that result in proton consumption that can increase soil pH (Ano and Ubochi 2010; Wong and Swift 2003). In addition, the manure pH was about 8.8, causing soil pH increase from the initial pH of 4.81. These values indicate that sufficient manure treatments provided more alkalinity to neutralize the acidity generated from nitrification.

CONCLUSION

Swine manure not only significantly increased soil nutrient contents such P, but also maintained maize in the red. As 40 % total N source, continuous manure application can either fully prevent red soil acidification or reduce accumulation of P and reduce the risk to environment. The application of manure is an effective nutrient and waste management tool in the red soil region.

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REFERENCES

Ano, A. O., and C. I. Ubochi. 2010 Neutralization of soil acidity by animal manures: mechanism of reaction. African Journal of Biotechnology **6**: 364–368.

Butterly, C. R., J. A. Baldock, and C. Tang. 2012. The contribution of crop residues to changes in soil pH under field conditions. Plant and Soil **248**: 51–66.

Guo, J. H., X. J. Liu, Y. Zhang, J. L. Shen, W. X. Han, W. F. Zhang, P. Christie, K. W. T. Goulding, P. M. Vitousek, and F. S. Zhang. 2010. Significant acidification in major Chinese croplands. Science **327**: 1008–1010.

Ritchie, G. S. P., and P. J. Dolling. 1985. The role of organic-matter in soil acidification. Australian Journal of Soil Research **23**: 569–576.

Rukshana, F., C. R. Butterly, J. A. Baldock, J. M. Xu, and C. Tang. 2012. Model organic compounds differ in priming effects on alkalinity release in soils through carbon and nitrogen mineralisation. Soil Biology & Biochemistry **51**: 35–43.

Wong, M. T. F., and R. S. Swift. 2003. Role of organic matter in alleviating soil acidity. In: Rengel Z (ed) Handbook of soil acidity. Marcel Dekker, New York, pp 337–358.

Xiao, K., J. Xu, C. Tang, J. Zhang, and P. C. Brookes. 2013. Differences in carbon and nitrogen mineralization in soils of differing initial pH induced by electrokinesis and receiving crop residue amendments. Soil Biology & Biochemistry **67**: 70–84.

P4-5

Use of different source of amendments for amelioration of acidic soils and rice production

***Hari Mohan Meena[1]* and H. C. Prakash[1]**
[1]Department of Soil Science & Agricultural Chemistry, UAS, GKVK, Bengaluru, India
; harimohanmeena46@gmail.com

INTRODUCTION

Soil acidity refers to presence of higher concentration of H^+ in soil solution and at exchange sites. They are characterized by low soil pH and with low base saturation. In India, 30 per cent of the cultivated land is considered acidic, where efficient fertilizer management is a problem. Out of 49 million ha of acid soils, 26 million ha have soil pH below 5.6 and 23 million ha have a pH between 5.6 and 6.5. The main causes of soil acidity in the region are intense weathering in association with humid climate and heavy precipitation (Panda, 1979). Under acidic conditions, calcium and magnesium supply is reduced and plant growth suffers. In addition to these effects, other beneficial nutrients, such as nitrogen, phosphorus and sulfur are also deficient. Ameliorating of acid soils and creates favorable conditions for better uptake of essential nutrients. Biochar and lime application are important management practice in acid soils. Biochar and lime have been recognized as an effective soil ameliorant as it raising pH and reduces Al, Fe and Mn toxicity and increases base saturation, cation-exchange capacity, P and Mo availability and improved the growth and yield of crop plants in acid soils.

MATERIALS AND METHODS

The present investigation was carried out during *kharif* season 2016 at experimental farm of Zonal Agricultural Research Station (ZARS), Mandya, University of Agricultural Sciences, Bengaluru (Karnataka). At the initiation of the experiment, the pH of the soil of the experimental field was 5.2 and EC value recorded 0.142 dS m^{-1}. The contents of organic carbon, available N, available P and available K were 4.5 g kg^{-1}, 212.95 kg ha^{-1}, 12.38 kg ha^{-1} and 139.20 kg ha^{-1}, respectively. The eight treatments were as follows: T_1 - Farmer Practices; T_2 - RDF + FYM + Zn (POP; T_3 - T_2 + lime; NPK T_4 - T_2 + bio char; T_5 - T_2 + mangla setright; T_6 - Soil test values based nutrient recommendation + FYM + Zn; T_7 - T_6 + lime; T_8 - T_6 + bio char. In this experiment, there were three replications. The experiment was conducted in randomized block design (RBD). The field experiment plot size was 10.8 m^2 (3.6m × 3m). Application of fertilizers in the treatment T_1 (Farmer Practices) was 120:35:20 kg N: P_2O_5:K_2O ha^{-1}, respectively. Recommended dose of fertilizers for rice: 100:50:50 kg N : P_2O_5 : K_2O ha^{-1}, respectively, FYM: 10 t ha^{-1} & $ZnSO_4$: 20 t ha^{-1}. The application of Biochar, lime and Mangla setright @ 2.3 t ha^{-1}, 4.1 t ha^{-1} and 150 kg ac^{-1}, respectively.

RESULTS AND DISCUSSION

The research result in Table 1 indicated that grain and straw yield of rice crop recorded significantly highest value in the treatment which received soil test value based fertilizers + Biochar (T_8) followed by soil test value based fertilizers + Lime (T_7) as compared to package of practices and farmer's practice. The chemical properties of soil such as pH, EC, OC and CEC value (Table 1) were increased from initial value of 5.20, 0.142 dS m^{-1}, 4.5 g kg^{-1} and 6.48 {c mol (p^+) kg^{-1}} to 6.76, 0.234 dS m^{-1}, 6.33 g kg^{-1} and 12.80{c mol (p^+) kg^{-1}}, respectively with the application soil test value based fertilizers + Biochar (T_8) followed by soil test value based fertilizers + Lime (T_7) as compared to package of practices. The increase of soil pH due to the application of biochar makes the soil surface more negative (Steiner et al. 2007). The application of biochar increased the CEC of acid soils may be due to the charge density per unit surface of biochar is generally high and its incorporation can increase the cation sorption of soils (Liang et al. 2006). The application of soil test values based fertilizers along with Biochar/Lime significantly increased the primary and secondary macro nutrients of the acid soil at harvest of the crop as compared to package of practices and farmer's practice (Fig. 1).

Table 1 Use of different source of amendments on chemical properties and yield of rice crop in an acidic soil

Treatments	Soil pH	EC (dS m^{-1})	OC (g kg^{-1})	CEC {c mol (p^+) kg^{-1}}	Grain Yield (q ha^{-1})	Straw Yield (q ha^{-1})
T_1 - Farmer's practice	5.53	0.155	4.69	7.07	42.39	48.59
T_2 - RDF + FYM + Zn (POP)	5.83	0.172	5.20	8.93	46.03	59.06
T_3 - T_2 + Lime	6.75	0.207	5.91	10.93	53.71	71.86
T_4 - T_2 + Biochar	6.53	0.217	6.24	12.33	54.86	74.61
T_5 - T_2 + Mangala Setright	6.11	0.200	5.82	10.60	52.78	71.42
T_6 - STV + FYM + Zn	6.03	0.177	5.33	9.73	47.29	62.76
T_7 - T_6 + Lime	6.36	0.227	6.11	11.00	55.53	79.17
T_8 - T_6 + Biochar	6.76	0.234	6.33	12.80	56.01	84.45
SE (d)	0.16	0.003	0.106	0.506	0.61	1.51
CD	0.34	0.007	0.23	1.09	1.33	3.25

STV - Soil test values based nutrient recommendation

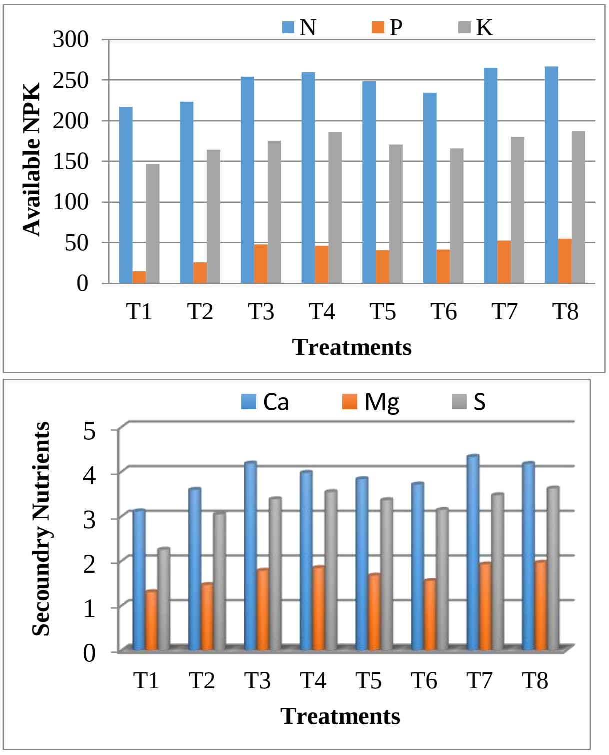

Fig. 1. Use of different source of amendments on available N, P, K, S (kg ha^{-1}) and exchangeable Ca & Mg (cmol (p^+)/ kg) in acidic soil

CONCLUSION

It is evident from the results that the application of fertilizers along with amendments (Biochar/lime) brought out a marked increase in the productivity of rice crop. The integrated use of the optimal dose of NPK along with amendments (Biochar/lime) influenced the organic carbon, CEC and available N, P and K, significantly. The use of amendments along with chemical fertilizers is absolutely essential to sustain the productivity of acid soils and to maintain the soil health.

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REFERENCES

Liang, B. L., J. Solomon, D. Kinyangi, J. Grossman, J. O'Neill, B. Skjemstad, J. O. Thies, F. J. Petersen, and J. Neves. 2006. Black carbon increases cation exchange capacity in soils. Soil Science Society of America **70**:1719-1730.

Steiner, C., W. G. Teixeira, J. Lehmann, T. Nehls, J. L. V. de Macedo, W. E. H. Blum, W. Zech. 2007. Long term effects of manure, charcoal, and mineral fertilization on crop production and fertility on a highly weathered central Amazonian upland soil. *Plant Soil*. **291**:275-290.

Panda, N. 1979. Fertilizer management in acid soils for increased efficiency, Fertilizer News **24**:75-83.

P4-6

Bioavailability of selected micronutrients in tropical peat soils

M. A. Zulhilmy[1,2]*, *M.M. Hanafi[2], I. Roslan[2], M. Hasmah[3] and M. Lulie[1]
[1]Sarawak Tropical Peat Research Institute, 94300 Kota Samarahan, Sarawak, Malaysia
[2]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia 43400 UPM Serdang, Selangor, Malaysia
[3]Universiti Teknologi MARA Sarawak, Kampus Kota Samarahan, 94300 Kota Samarahan, Sarawak, Malaysia
; zulhilma@sarawak.gov.my

INTRODUCTION

Tropical peatland is one of the important wetlands in Borneo. Tropical peatland can be identified by concentric forest zones with different forest types. It is quite distinct in Sarawak due to extensiveness of each peat basins. The forest formation in tropical peat swamp across a peat dome is influenced by the topo-hydrological characteristic of peatland affecting the fertility of peat soil. In the most mature swamps, a major forest types are Mixed Peat Swamps forest (MPS), Alan Batu forest (A.Bt), and Alan Bunga forest (A.Bg), located at the edge, middle and nearly at the center of peat dome. Substrate quality (composition/structure) and chemical properties of peat soils may differ across the peat dome because of the differences in the vegetation and environmental conditions, subsequently, influence the rate of soil organic matter (SOM) decomposition. This may affect the availability of micronutrients as peat soil is generally oligotrophic, having very low bases and micronutrients, high carbon: nitrogen (C: N) ratios, and low available N. The objective of this study is to investigate the availability of micronutrients i.e., copper (Cu), zinc (Zn) and boron (B). For this purpose, the availability of micronutrients were determined via adsorption and desorption batch experiment using soil samples collected from MPS, A.Bt and A. Bg forests.

MATERIALS AND METHODS

Peat soil samples were collected from Maludam National Park, Betong Division, Sarawak, Malaysia under three different forest types, MPS, A.Bt, and A.Bg forests at depth of 0–25 cm and 25–50 cm with five replications using peat auger (Eijkelkamp, The Netherlands). The samples were stored at 4^{o}C prior to chemical analysis. Fresh peat samples were used for determination of moisture and ash content, pH, total carbon (C) and nitrogen (N), cation exchangeable capacity (CEC), and extractable Cu, Zn and B. Substrate quality of labile SOM was determined as dissolved organic carbon (DOC) and pyrophosphate solubility index as humification level. Adsorption of Cu, Zn, and B was conducted through equilibrium batch experiment and the data was fitted into Freundlich equation [Q (mg/kg) = $K_f (C_{eq})^{1/n}$], followed by desorption experiment using different extractant to determine the readily available, exchangeable and complex form of Cu, Zn and B. Differences among the forest types were determined by analysis of variance (ANOVA) followed by means comparison of Tukey test.

RESULTS AND DISCUSSION

Physico-chemical properties of peat soil from MPS, A.Bt and A.Bg showed moisture and ash content in the range of 87- 90% and 0.4-1.4% respectively. Soil pH between 3.4-3.9 and total C and N were 50.5–57.3% and 1.4-1.8% respectively. CEC value was from 23.2-41.3 cmol/kg.The adsorption data for Cu, Zn and B fitted well to the linear Freundlich equation as shown in **Table 1**. Adsorption isotherms shape of Cu and Zn data for three different soil samples of MPS, A.Bt and A.Bg showed the L-type isotherm reflecting a relatively high affinity between the adsorbate and adsorbent, suggesting a chemisorption reaction (McBride, 2000).

Table 1: The Freundlich adsorption parameter for Cu, Zn and B from different forest types

Forest Type	Cu			Zn			B		
	r^2	K*f* L kg^{-1}	*n*	r^2	K*f* L kg^{-1}	*n*	r^2	K*f* L kg^{-1}	*n*
MPS	0.986	122.12±4.4ab	0.55	0.953	28.55±0.6ab	0.69	0.971	2.94±0.2b	1.31
A.Bt	0.983	136.53±4.8a[†]	0.54	0.956	26.99±0.7b	0.75	0.939	2.57±0.2b	1.43
A.Bg	0.974	119.95±4.4b	0.56	0.972	29.58±0.6a	0.72	0.924	3.75±0.2a	1.22

[†]Values followed by different letter differ at $P \leq 0.05$.

However, B was more favorable to S-type isotherm based on *n* value (*n*>1), suggesting cooperative adsorption (Foo and Hameed, 2010) where adsorbate-adsorbate was stronger than adsorbate-adsorbent interaction. The presence of dissolve organic matter (DOM) may also modify adsorption producing S-type isotherm and these ligands inhibit the adsorption at low metals concentration (McBride, 2000). This effect showed that K_f value for B was higher in A.Bg as DOC value was lower (A.Bg: 4.78 g kg^{-1}<A.Bt: 6.33 g kg^{-1}<MPS: 20.66 g kg^{-1}). In general, K_f value showed higher capacity of adsorption in order of Cu>Zn>B. However, Cu and Zn showed no significant difference of K_f value for MPS forests. This suggested that substrate quality i.e., humification degree and DOC contents were not a primary factor influencing the adsorption capacity of Cu and Zn on peat soils surface because humification degree and DOC level were higher in MPS compared to A.Bt and A.Bg. The efficiency of adsorption may rely on other factors such the chemical nature of the reactive surface group, level of adsorbent/adsorbate ratio, pH, ionic strength of solution, and the presence of soluble ligands (McBridge, 2000). On the other hand, the desorption experiment showed the mean readily available of Cu, Zn and B from MPS, A.Bt and A.Bg soils were in the range of 9.14–16.01 mg kg^{-1}, 42.02–57.52 mg kg^{-1} and 91.65–115.15 mg kg^{-1} respectively. Exchangeable form of Cu was 9.62–14.35 mg kg^{-1} while exchangeable Zn was 45.59–63.70 mg kg^{-1}. Complex form for Cu was in the range of 180.67–323.18 mg kg^{-1} while complex Zn was 101.0–135.04 mg kg^{-1}. Cu element was strongly bonded compared to Zn while B elements are mostly in available form and it shows B molecules interaction with the surface of soil was weak.

CONCLUSION

Adsorption capacity, K_f was greater in the order: Cu> Zn > B. Substrate quality from different forest type not a major factor to the adsorption capacity of Cu and Zn. Nevertheless, there is some effect on B adsorption in which lower DOC had greater adsorption capacity. Furthermore, desorption was greater for B followed by Zn and Cu because B are mostly in readily available form. Further investigation is necessary to understand B interaction with soil surface as anions may bond indirectly to organic groups through bridging metals ion. A right formulation and interaction of Cu and Zn with other cationic elements in peat soils also need to be well understood which may increase its availability for plant uptake.

REFERENCES

Foo, K.Y. and Hameed, B.H. (2010). Insight into the modelling of adsorption isotherm systems. Chemical engineering journal, **(156)**, 2-10

McBride,M.B. (2000). Chemisorption and precipitation. In: Sumner, M.E. ed. Handbook of soil science. CRC Press, Boca Raton, pp B-265 – B-302

P4-7

Effect of acidity on phosphorus availability in matured black pepper farms

Izzah, A.H[1]. and Wan Asrina, W.Y[1].
[1]Department of Crop Science, Faculty of Agriculture and Food Sciences, Universiti Putra Malaysia Bintulu Sarawak Campus, 97008 Bintulu, Sarawak, Malaysia
; asrina@upm.edu.my

INTRODUCTION

Soil acidity has been reported enormously to affect P availability, especially through formation of complexation ion such as Al/Fe-P. Soil pH <5.0 has effect on solubility and availability of P for plant uptake (Brady and Weil, 2017). Even though P uptake is considered minimum in black pepper (31 g/vine/year), however, excessive complexation and inability to retain the ion through exchangeable site may leach from soil colloidal surfaces (de Campos *et al.*, 2016; Ann, 2012). This problem may accelerate P deficiency and disturbed flower formation (Ravindran, 2003). Therefore, the objective of this study was to determine effect of soil acidity on Fe and P availability and examine the accumulation of P in black pepper leaves in three different black pepper farms located in Sarawak in different divisions (i.e. Kapit, Sri Aman and Kuching).

MATERIALS AND METHODS

This study is conducted in three matured (aged >24 months) black pepper farms located in Kapit, Sri Aman and Kuching, Sarawak. Surface soil sample (0 to 20 cm) was collected through non-purposive sampling techniques, while black pepper leaves were collected from active production stem in the middle and external portion of the canopy (Ann, 2012). The soil sample was air dried, pulverised and sieved through 2 mm while leaves were cleaned with distilled water, air dried to evaporate moisture, oven dried at 60°C for 3 days and ground to powder.

All the samples were analysed in triplicates for soil pH using water (Tan, 2011), total Fe on leaves through wet digestion (Hach *et al.*, 1985) and available Fe and P in soil through double acid extraction (Tan, 2011). The soil pH was measured using portable pH meter, while total and available Fe using atomic absorption spectrometer (Perkin Elmer, Model AA800) and available P (blue method) through UV/Vis spectrophotometer (PerkinElmer, Lambda 25) at 882 nm (Sarker *et al.*, 2014), respectively. All equipments in this study were calibrated accordingly using standard solution and procedure provided by the manufacturs. The means values were tested through Tukey's studentized test (p=0.05) using statistical analysis system SAS Ver. 9.4.

RESULTS AND DISCUSSION

The soils pHs, Fe and P availabilities are shown in Table 1 which indicated extremely acidic to very strongly acidic with decreasing acidity according to the order of Sri Aman >Kapit >Kuching. Acidity has shown significant different; however, Fe availability was insignificantly affected which may arise from soil mineralogy from this farm, especially on tropical soil (Agegnehu and Amede, 2017). The implication of acidity and the nature of the soil insignificantly affected the availability of P. This is because soil pH <5.0 has greater solubility of Fe which is tightly bound with P by forming ion complexation Fe-P (Brady and Weil, 2017; Vadas and Sims, 2014). A greater Fe availability in soil of Kuching (0.09 g kg^{-1}) has reduced availability of P

(0.006 g kg^{-1}). Similar trend of Fe toward P availability can be simplified through increasing Fe on soils of Kapit >Sri Aman >Kuching has decreased P accordingly.

Table 1: Soils and foliar chemical properties of different black pepper farms

Farms	Soil			Leaf		
	pH	Fe	P	Fe	P	P*
		------- g kg^{-1} -------				
Kapit	4.41 ± 0.04^b	0.05 ± 0.002^c	0.019 ± 0.0012^a	0.11 ± 0.002^a	1.31 ± 0.02^a	
Sri Aman	4.16 ± 0.01^c	0.06 ± 0.002^b	0.009 ± 0.0007^b	0.11 ± 0.006^a	1.33 ± 0.03^a	2.10
Kuching	4.91 ± 0.07^a	0.09 ± 0.004^a	0.006 ± 0.0006^b	0.11 ± 0.007^a	1.26 ± 0.02^a	

* Average accumulation of P (30 months old) in Semengok Aman
** Means with different alphabets within a column are significantly different at P=0.05

Although availability of Fe was significantly different in all farms, however, the accumulation of Fe in leaves was comparable at 0.11 g kg^{-1} (Table 1). Farm in Sri Aman (1.33 g kg^{-1}) has exhibit greater accumulation of P compared to other farms. Additionally, P availability in leaves for all farms were almost two folds lower compared to the well established farm (2.10 g kg^{-1}) (Ann, 2012). This could be caused by the amount of fertiliser applied and presence of Fe which slightly limits nutrient uptake.

CONCLUSION

Soil acidity in this study have insignificantly affected availability of Fe and P with Fe solubility manly originating from soil mineralogy. The accumulation of P in leaves were insignificantly different among farms which indicated a minimum nutrient uptake. Hence, Fe availability was the main effect on P availability in soil and black pepper uptake.

REFERENCES

Agegnehu, G., & Amede, T. (2017). Integrated soil fertility and plant nutrient management in tropical agro-ecosystems: A review. *Pedosphere,* **27:** 662-680.

A**nn, Y. C. (2012).** Determination of nutrient uptake characteristic of black pepper (*Piper nigrum* L.). *Journal of Agricultural Science and Technology,* **2:** 1091-1099.

B**rady, N. C., & Weil, R. R. (2017).** *The Nature and Properties of Soils.* Edinburgh Gate, England: Pearson Education Limited.

de Campos, M., Antonangelo, J. A., & Alleoni, L. R. F. (2016). Phosphorus sorption index in humid tropical soils. *Soil and Tillage Research,* **156:** 110-118.

Hach, C. C., Brayton, S. V., & Kopelove, A. B. (1985). A powerful Kjeldahl nitrogen method using peroxymonosulfuric acid. *Journal of Agricultural and Food Chemistry,* **33:** 1117-1123.

Ravindran, P. N. (2003). *Black Pepper: Piper nigrum.* Netherlands, UK: Taylor & Francis.

S**arker, A., Kashem, M. A., Osman, K. T., Hossain, I., & Ahmed, F. (2014).** Evaluation of available phosphorus by soil test methods in an acidic soil incubated with different levels of lime and phosphorus. *Open Journal of Soil Science,* **4:** 103.

Tan, K. H. (2011). *Principles of Soil Chemistry.* Boca Raton, FL: Taylor & Francis.

Vadas, P., & Sims, J. T. (2014). Soil Fertility: Phosphorus in Soils☆. In "*Reference Module in Earth Systems and Environmental Sciences*": Elsevier.

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Effect of biofertilizer on soil nutrient status of matured cocoa (*Theobroma cacao l.*) tree in low soil pH in Jengka, Pahang

Nurfadzilah, M[1]., Helmi, S[1]., Boney, M[1]., Nurafiza, A[1]., Rozita, O[1]. and Haya, R[1].
[1]Malaysian Cocoa Board, 5th, 6th & 7th Floor, Wisma SEDCO, Lorong Plaza Wawasan Off Coastal Highway, Locked Bag 211, 88999 Kota Kinabalu, Sabah, Malaysia
; nurfadzilah@koko.gov.my

INTRODUCTION

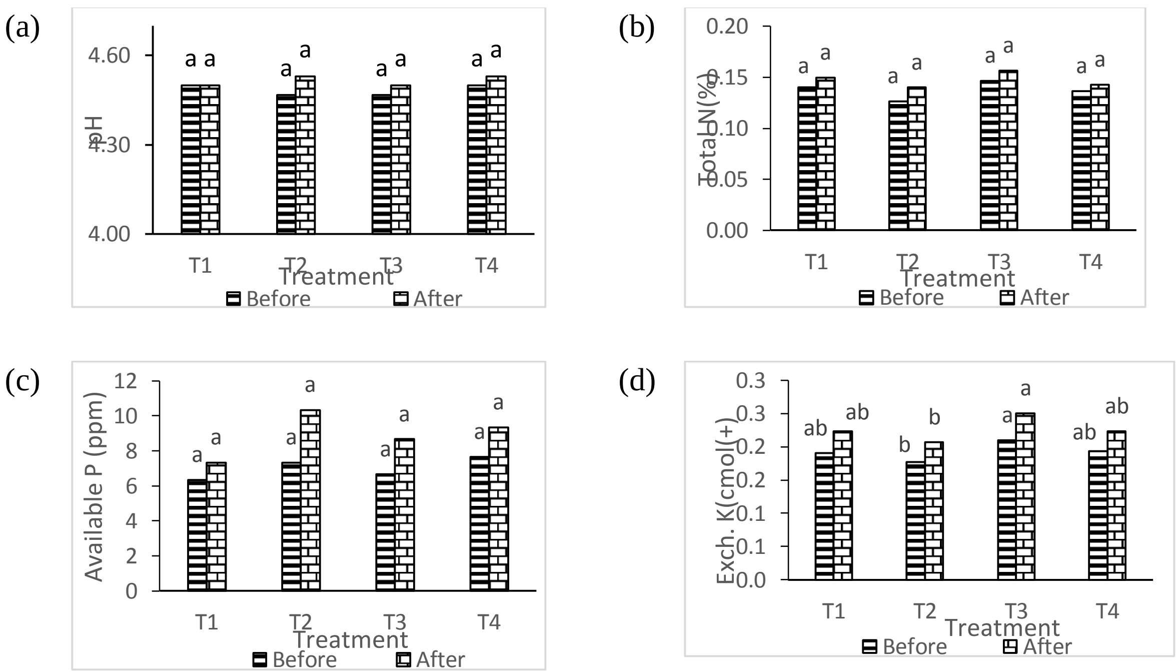
A fertilizer is a substance, chemical or natural substances which under favourable conditions when added to a soil will produce a better growth crops, increase its fertility, whether through direct or indirect action on the crop or on the properties of the soil. Inorganic fertilizer, also known as mineral or commercial, is a fertilizer mined from mineral deposit, while organic fertilizer, is a fertilizer derived from animal matter, human excreta or vegetable matter. Biofertilizer, on the other hands, is a substance containing living cells of efficient microorganisms which helps to improve plant nutrients uptake by their interactions in the rhizosphere when applied to the soil. Biofertilizer is a cost effective and renewable source of plant nutrients to supplement with chemical fertilizer. Microorganisms in the biofertilizer help in restoring the soils natural nutrient cycle and helps in builds soil organic matter. It also helps in converting complex organic material into simple compounds that can be taken up by the plant root.

MATERIALS AND METHODS

This study was carried out at Cocoa Research and Development Centre, Jengka, Pahang. The main objective of this study was to determine the effect of biofertilizer on mature cocoa tree in low soil pH. It consisted of four treatments as listed: 1) NPK Blue 12:12:17:2 with standard field practice, T2) biofertilizer using the recommended rates, T3) Compound fertilizer (NPK Blue - 12:12:17:2) plus biofertilizer with recommended rates, and 4) Control (no fertilizer added throughout the trial). All treatments (except no fertilizer application) have been applied using 16L knapsack sprayer (BioJadi fertilizer) every week for the first 7 applications, subsequence applications were done every 2 weeks until the end of the trial, whilst NPK Blue fertilizer was applied manually every four months. The trial duration has been evaluated for two years (24 month) therefore; at least 4 main fruiting seasons was observed. Four random samplings of the soil per plot by using stainless steel auger were taken with 0-20 cm depths before application, 12 months after application and shortly after the last application. The parameter studied were pH and major nutrients in the soil such as nitrogen, phosphorus and potassium. All data were recorded throughout the year and were analysed using SAS for means separation of the treatments effects.

RESULTS AND DISCUSSION

Based on the soil analysis result (Figure 1), there is no significant difference ($p>0.05$) between all treatments before and after treatments application in (a) soil pH, (b) total N and exchangeable K. However, the result of soil pHs show an improvement from the application of biofertilizer, compound fertilizer (NPK Blue - 12:12:17:2) plus biofertilizer with recommended rates and control as compared to T1, as application of lime do assisted in correcting the soil pH, whilst for conventional fertilizer usually would cause acidification to the soil. Although lime was applied, it often takes a year or more before a response can be seen. Due to the application of NPK blue for total N, slight improvement was seen in total N in T2 (Figure 1), even though nutrient for all treatments after application were still less than recommended adequate range. For available P, there are improvements on availability of P in the soil except for T1. According to Allen (2010), available P were slowly released from insoluble phosphates thus limiting the plant uptake. Therefore, there is a big gap between adequate ratios of available P (15 mg/kg) with the soil nutrient result below. Thus, it is suggested to put more lime of 700-800 g/tree per year in order to increase the mean value of soil pH for 4.5 to 6.6, which helps the availability of phosphorus in soils. For exchangeable K, there is a significant difference between treatments. For T2, T3 and T4, there a slight increase in exchangeable K after 2 years of applications. Nevertheless, when soil-potassium levels are high, generally plants will take up more potassium than needed for healthy growth (luxury consumption).



Note: Column means followed by the same letter are not significantly difference (P>0.05) according to Duncan's Multiple Range Test. Treatments are T1-NPK Blue fertilizer, T2-Biofertilizer, T3-Biofertilizer +NPK Blue Fertilizer and T4 – No fertilizer applied

Figure 1: Soil chemical charecteristics which are (a) pH, (b) Total N, (c) Available P and (d) Exchangeable K for different treatments.

CONCLUSIONS

Overall, there are improvements on pH, concentration of nitrogen and potassium for all treatments when compared to control. Phosphorus was still below the recommended suitability range, thus lime should be added to the soil in order to increase the pH and eventually produce the availability of phosphorus in soils. The effects of T3 for exchangeable K, indicated that combination between biofertilizer and NPK Blue had given some positive effect on nutrient status after treatments were applied. Therefore, this study suggested that combinated application of biofertilizer with NPK Blue should give the best effect in certain nutrients in the soil particularly, for exchangeable K, thus, will help in increasing the yield of matured cocoa tree.

ACKNOWLEDGEMENT
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REFERENCES

Allen, V.B. 2010. Science and Technology of Organic Farming. CRC Press, Taylor and Francis Group.

P4-9

Fractionation of aluminium in acidified soil-plant systems

***Matúš, P*[1]., *M. Bujdoš*[1], *M. Matulová*[1], *B. Farkas*[1]**
[1]Slovak Spectroscopic Society, member of the Association of Slovak Scientific and Technological Societies; Institute of Laboratory Research on Geomaterials, Faculty of Natural Sciences, Comenius University in Bratislava
Ilkovičova 6, 84215 Bratislava, Slovakia
; matus@fns.uniba.sk

INTRODUCTION

Aluminium occurs in various chemical species, of which free Al^{3+}, $AlOH^{2+}$, and $Al(OH)_2+$ species were found to be the most crucial to evaluate its toxicity and to predict the impact of proton inputs on soils and surface waters. The concentration of such Al species may be difficult to determine directly. For that reason these species are determined in one or more fractions. Generally, these fractions are defined operationally and according to the method of determination are termed reactive or labile aluminium.

Several single extractions, optimized BCR three-step sequential extraction procedure (SEP) and the reactive aluminium (Alr) determination were used for the fractionation of Al in acid soils. The obtained Al fractions were valuated from aspect of Al contents in plants (grass) growing on these soils. Also the calculation of the aluminium toxicity indexes (ATI) was used for assessment of Al toxicity to the plants.

MATERIALS AND METHODS

The acid samples were collected from a mining area (Šobov, Slovakia) with sulphidic deposits (ten soils 1-10, pH ~ 1.8-3.6 and five plants (grass *Festuca Rubra*) growing on soils A-E, pH~2.8-5.1).

Total Al content in samples was determined after their decomposition by $HF+HNO_3+HClO_4+ H_2O_2$ in open system at 200 °C for soil samples, and by mixture of $HF+HNO_3$ in autoclave at 160 °C for plant samples.

Single extractions were applied to the soils using 16 different agents in the following ratios: redistilled H_2O (v/w=5/1); solutions of 1 M KCl (w/v=1/10), 1 M NH_4Cl (w/v=1/10), 0.01 M $CaCl_2$ (v/w=10/1), 0.1 M $BaCl_2$ (w/v=1/10), 0.5 M $CuCl_2$ (w/v=1/10), 0.3 M $LaCl_3$ (w/v=1/10), 0.2 M $(NH_4)_2C_2O_4$ (v/w=20/1), 0.2 M $Na_2S_2O_4$ (v/w=10/1), 0.5 M NH_4F (v/w=10/1), 0.005 M NTA (v/w=10/1), 0.005 M EDTA (v/w=10/1), 0.05 M EDTA (v/w=10/1), 0.005 M DTPA (v/w=10/1), 0.1 M $Na_4P_2O_7$ (w/v=1/10) and 0.5 M HCl (v/w=20/1). The BCR SEP was adopted from Rauret et al. (1999).

Alr was determined in filtered (0.40 µm) water extracts of soils using solid phase extraction by chelating ion-exchanger Iontosorb Salicyl, see Matus and Kubova (2005).

Flame atomic absorption spectrometry (FAAS) was used for Al, Ca, Mg, Na, K determination.

RESULTS AND DISCUSSION

From Fig. 1 it is obvious that used extractants in accordance with their extractive efficiency can be divided into three groups. The first group consists of weakly efficient extractants (H_2O, $CaCl_2$, NTA, DTPA and dilute acetic acid from the first step of the BCR SEP) which release a small amounts of available Al fractions. The more efficient extraction agents as $BaCl_2$, KCl, NH_4Cl, $CuCl_2$, $LaCl_3$, EDTA and ammonium oxalate (AO) leached approximately the same amounts of Al as grass Al. The third group of extractants contains most aggressive agents as dilute HCl, hydroxylammonium hydrochloride/HNO_3 and H_2O_2/ammonium acetate from the second and third step of the BCR SEP, respectively. These extractants release the highest amounts of soil Al, approximately double as total grass Al contents. Nevertheless, most suitable agents for leaching of bioavailable Al forms are extractants from the first group.

The values of ATI (Fig. 2) calculated as the ratio of nutrient cations (Ca, Mg, K, Na) sum to Al content for soil samples are non-selective considering soil pH and Alr values but ATI values for two soil extracts and plant samples is more useful (the low ATI values < 1 mean Al toxicity risk to environment). The selective ATI values correspond well to soil pH, soil Alr and plant Al values; the highest ATI values conform to the highest soil pH and the lowest Alr values (sample A); the lowest ATI values conform to the highest content of Al in plant root and stem (sample E).

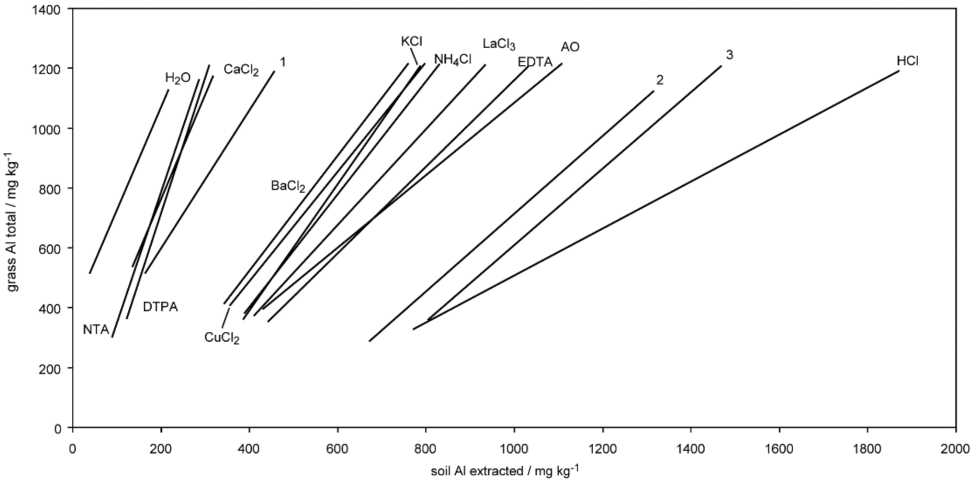

Fig. 1: The dependence of Al concentrations in individual soil extracts on total Al content in grass (1, dilute acetic acid; 2, hydroxylammonium hydrochloride/HNO_3; 3, H_2O_2/ammonium acetate)

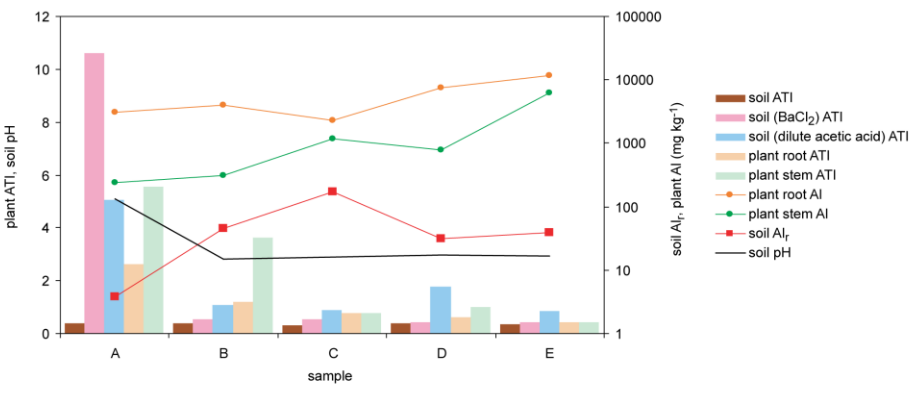

Fig. 2: The values of aluminium toxicity indexes (ATI) in soils, soil extracts and plants

CONCLUSION

It can be concluded, that acid extractable Al fraction is most Alr selective from optimized BCR three-step SEP. The dilute HCl extraction can be the alternative for routine monitoring of Al mobility instead of optimized BCR three-step SEP when the phase associations estimation is not necessary. Some simple salt extractants, mainly

$CaCl_2$ seem to be suitable for the estimation of labile and reactive Al species in acid soils and they can be a rapid tool in some applications of Al fractionation. The determination of Alr (samples 1-10, A-E) and calculation of ATI data (samples A-E) are most useful simple tools for the estimation of Al availability and toxicity to plants.

ACKNOWLEDGEMENTS

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REFERENCES

Matúš, P., J. Kubová. 2005. Complexation of labile aluminium species by chelating resins Iontosorb – a new method for Al environmental risk assessment. Journal of Inorganic Biochemistry **99**:1769-1778

Rauret, G., J.F. Lopez-Sanchez, A. Sahuquillo, R. Rubio, C.M. Davidson, A.M. Ure, P. Quevauviller.
1999. Improvement of the BCR three step sequential extraction procedure prior to the certification of new sediment and soil reference materials. Journal of Environmental Monitoring **1**:57

P4-10

Extractability utilization for soil-plant element transfer predictions in acidified lands

Hagarová, I[1].**, **P. Matúš[1], J. Medveď[1], M. Šebesta[1], F. Polák[1]
[1]Slovak Spectroscopic Society, member of the Association of Slovak Scientific and Technological Societies; Institute of Laboratory Research on Geomaterials, Faculty of Natural Sciences, Comenius University in Bratislava, Ilkovičova 6, 84215 Bratislava, Slovakia
; ingrid.hagarova@uniba.sk

INTRODUCTION

The prediction of soil metal phytoavailability using the chemical extractions is a conventional approach routinely used in soil testing. The aim of the present study was to evaluate the adequacy of optimized BCR (Community Bureau of Reference) three-step sequential extraction procedure (SEP) and single extraction with dilute HCl for the prediction of the soil pollutants effects affected by different anthropogenic sources of the acidification. These procedures were used after their validation to obtain the distribution of both the major (Al, Fe, Mn) and trace (As, Cd, Cu, Ni, Pb, Zn) risk metals in acid soils.

MATERIALS AND METHODS

The acid soil and plant samples were collected from different locations of Slovakia:
Locality A; near open quartzite mine Šobov (central Slovakia) with weathered product of pyritized quartzite as dump material (acid sulphatic earth with pH H_2O 2.99, grass *Festuca Rubra*).
Locality B; in the part of National park of the Low Fatra with podzolization process and anthropogenic acid atmospheric inputs (Histo-humic Podzol soil with pH H_2O 3.40; blueberry *Vaccinium Myrtillus* L.).
Locality C; in the region of Pezinok, Malé Karpaty Mts. with sulphide oxidation, mine waters with high Fe, SO_4^{2-}, As and Sb (Fluvi-eutric Gleysol soil with pH H_2O 3.51, blackberry *Rubus fruticosus*).

Total concentrations of studied elements in soil and plant samples were determined after their decomposition by acid mixture of $HF+HNO_3+HClO_4+H_2O_2$ in open system at 200 °C (soils) and by acid mixture of $HF+HNO_3$ in an autoclave at 160 °C (plants). For As determination the soil and plant samples were decomposed with HNO_3 in an autoclave at 160 °C.

The optimized BCR SEP was adopted from Rauret et al. (1999). Further, the soils were shaken with 0.5 M HCl (v/w ratio=20/1) for 1 h and obtained suspensions were centrifuged (2500 g) for 20 min.

Flame atomic absorption spectrometry (FAAS) was used for determination of Cd, Cu, Fe, Mn, Ni, Pb, Zn. Electrothermal atomic absorption spectrometry (ET AAS) was used for determination of Cd, Ni, Pb at low concentration levels. Inductively coupled plasma optical emission spectrometry (ICP OES) was used for Al. As was measured by hydride generation atomic absorption spectrometry (HG AAS).

RESULTS AND DISCUSSION

The soil-plant transfer coefficients in Fig. 1a were calculated from ratio of the metal total plant concentration to metal total concentration in corresponding top soil horizon where plant had grown to establish a relative sequence of analyzed metals mobility on the examined localities. The data allow to assess the plant intake intensity, phytoavailability and phytoaccumulation of studied metals for each examined site individually. The results show that Mn, Cd and Zn are the most phytoavailable from all nine studied metals in all three acidified ecosystems. These metals are significantly absorbed and accumulated by almost all given plants and their individual parts. Manganese is characterized by the highest soil-plant transfer coefficients (normalized for 100 %) and the highest phytoaccumulation (the Mn soil-plant transfer coefficients range from 25 to 388 %) from all studied metals for all examined ecosystems. In contrast to this fact Al, As, Pb and Fe are mostly inert to the plant ability to take up these metals at given pH value in spite of the finding that both As and Pb occur in the soils on sampling sites at very high concentration levels.

The extraction yield data in Fig. 1b with the first three BCR SEP steps sum and HCl extraction yields allow to assess the soil distribution and mobility of studied metals for each examined site individually. Sums of the results obtained from first three BCR SEP steps represent so-called mobilizable fractions of given metals. These data for all studied metals are related to the results from dilute HCl single extraction. It can be concluded that Cd and Cu are the most mobile metal from all nine studied metals in all three acidified ecosystems. Cadmium is characterized by the highest extraction yields of BCR SEP mobilizable fractions (normalized for 100 %) from all studied metals for all examined ecosystems. In contrast to this fact As is mostly inert to the leaching by both used extraction methods.

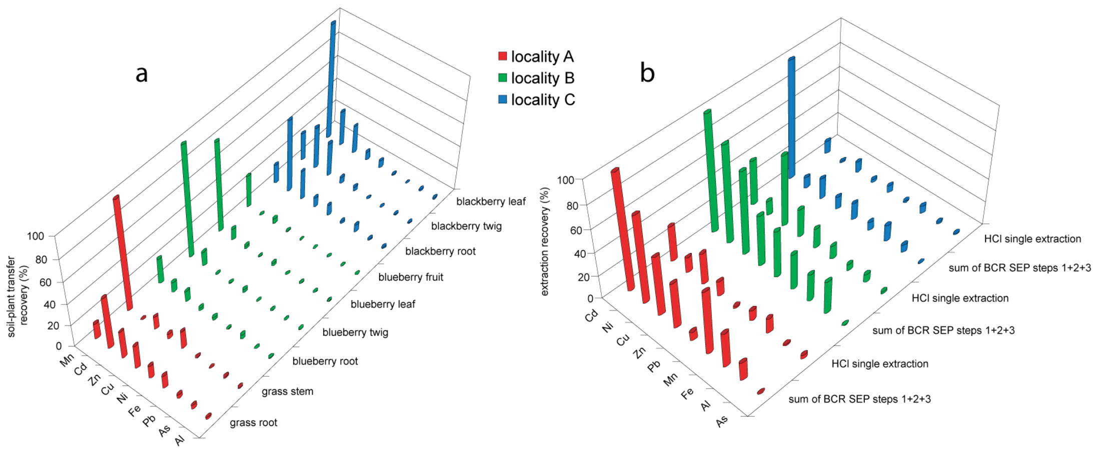

Fig. 1: Distribution of the soil-plant transfer (a) and extraction (b) recoveries normalized separately for each one from three sampling sites regardless of plant part (a), metal and extraction procedure (b)

CONCLUSION

In certain conditions the studied plants can be recommended as the bioanalytical tools for *in situ* separation of phytoavailable metals species directly in the ecosystem and the calculated soil-plant transfer coefficients represent the yields of such bioseparations. They express the ratio of partial soil metal concentration separated by the plants (represented by total plant metal concent) to total soil metal content. In this case the studied plants can be considered for the long-term extraction medium. Also the analyte phytoaccumulation can play the important role in such phytoseparations what it is reflected by the soil-plant transfer coefficients higher than 100 %. Therefore the calculated soil-plant transfer coefficients can be compared with the extraction yield data of all steps and their sums of optimized BCR SEP and single extraction by dilute HCl applied to soil samples. Based on these results, it could be classified as the relative mobility of studied metals in different soil systems. However for quantitative and more reliable estimation of the metal phytoavailability this approach requires to complete this study by other soil parameters.

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REFERENCES

Rauret, G., J.F. Lopez-Sanchez, A. Sahuquillo, R. Rubio, C.M. Davidson, A.M. Ure, P. Quevauviller.1999. Improvement of the BCR three step sequential extraction procedure prior to the certification of new sediment and soil reference materials. Journal of Environmental Monitoring **1**:57-61

P4-11

Effects of Soil Nutrient Status on main metabolites of Oolong Tea

Liu, Y*[1]*., Sun, L. L*[1]*., Liao, H*[1]*.
[1]Root Biology Center, Fujian Agriculture and Forestry University, 350002 Fuzhou, Fujian, China
; lilionsun@163.com

INTRODUCTION

It is well documented that tea plants prefer to grow on acidic soils, and therefore, most tea plantations are located on acidic soils. However, there are many nutritional problems on acidic soils, including nutrient deficiencies (e.g. P deficiency) and elemental toxicities (e.g. Al toxicity) (Ruan *et al.*, 1986). In recent years, with increasing fertilization for high yield, soils in tea plantations are becoming more acidic. However, no studies about the effects of fertilization on soil acidification and tea quality in tea plantation have been reported. As one of the best tea in China, Oolong tea is mainly produced in Fujian Province where most soils are acidic. In order to improve nutrient management in tea plantation, we sampled oolong tea leaves and their precisely located soils from the two most reprehensive production areas of Oolong tea, Wuyi Mountain and Anxi districts. After analyzed the correlations between soil nutrients and leaf metabolites using hundreds of samples, we proposed the optimal criterions of soil nutrients for oolong tea, so as to provide theoretical guidance for culturing sustainable and environmental friendly tea plantation with better tea quality.

MATERIALS AND METHODS

Three tea varieties were investigated in this study, including Tieguanyin from Anxi, and Shuixian and Rougui from Wuyi Mountain. Tea leaves and their precisely located soils in tea plantation were sampled from 22 tea gardens in Anxi and 22 tea gardens in Wuyi Mountain. In total, there were 315 leaf and 105 soil samples. Tea leaves were harvested following the local farmers' standards. The soils were sampled in 0-20 cm surface layer precisely under the sampled tea trees. Soil nutrients were analyzed according to Analysis of soil agrochemical (Bao *et al.*, 2000). Main metabolites of tea leaves were analyzed as described (Wang *et al.*, 2011). Boundary-Line analysis method was used to analyze the relationships between soil nutrients and tea metabolites.

RESULTS AND DISCUSSION

The soil pH value, alkali hydrolytic nitrogen and available phosphate in Anxi tea planation were dramatically lower than those in Wuyi Mountain, but there were no significant differences between them in the concentrations of soil exchangeable potassium and organic matter (Table 1). The tea quality from Anxi and Wuyi Mountain were different, as indicated by main metabolites (Fig.1). For example, the concentrations of total catechins, caffeine, theanine and rutin in Anxi tea were much higher than those in the tea leaves from Wuyi Mountain. The soil nutrient status in tea planation differed in different districts, which probably due to their different maternal soils or fertilization, and therefore, the effect of each soil nutrient on each tea metabolite was distinctive in two districts (Fig.2).

For example, Epigallocatechin gallate (EGCG) as the mostly accepted metabolite for tea quality, was mostly affected by the content of soil exchangeable potassium in Anxi,

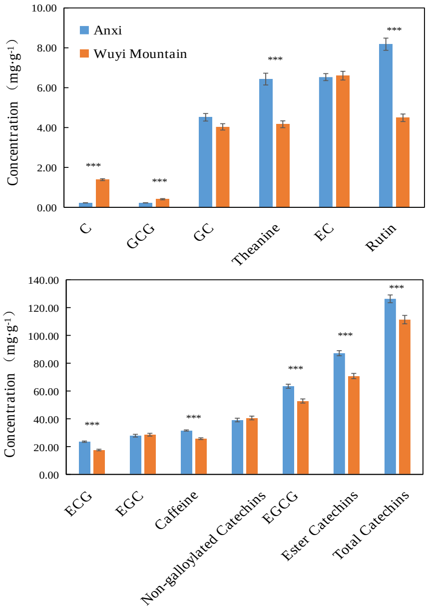

Fig.1 Concentrations of main metabolites in the tea from Anxi and Wuvi Mountain.

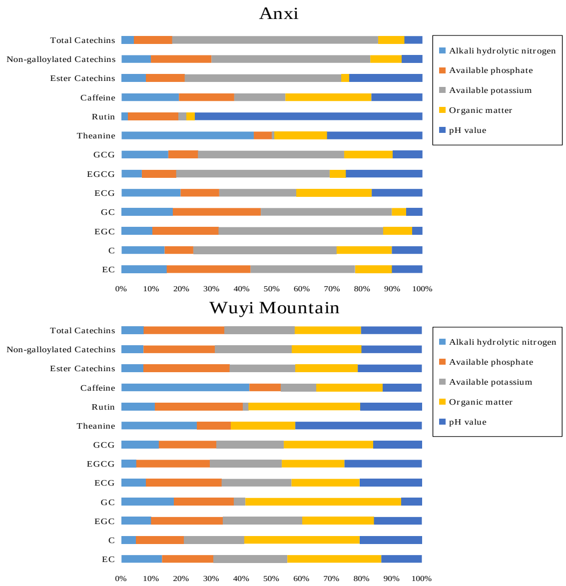

Fig.2 Dominant factors of soil nutrients limiting tea quality in Anxi and Wuyi Mountain.

but by available phosphorus in Wuyi Mountain.

Table 1 The soil properties in Anxi and Wuyi Mountain.

Parameter	Anxi （n=50）	Wuyi Mountain （n=55）
Alkali hydrolytic nitrogen （mg·kg^{-1}）	51.68 ± 1.81 b	80.32 ± 2.67 a
Available phosphate（mg·kg^{-1}）	119.36 ± 9.32 b	219.47 ± 13.77 a
Exchangeable potassium（mg·kg^{-1}）	136.71 ± 6.82 a	133.96 ± 5.47 a
Organic matter （g·kg^{-1}）	26.20 ± 0.09 a	27.60± 0.11 a
pH value	4.36 ± 0.04 b	4.63 ± 0.02 a

CONCLUSION

1. Soil nutrient status differs in different districts, which is mainly caused by material soils and fertilization.
2. Different soil nutrients have different contributions to tea metabolites.

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REFERENCES

Bao, S. D. 2000. Analysis of soil agrochemicals. China Agriculture Press.

Ruan, Y. C., and R. F. Chen. 1986. Effect of Aluminium phosphorus on growth and nutrient absorption of tea plants. China Tea **1**:3-6.

Wang, Z. M., Q. D. Guo, F. Xie, X. P. Wang, and X. G. Mei. 2011. Determination of catechins and caffeine in tea polyphenols by HPLC. Chinese Pharmaceutical Journal **46**:1674-1676.

P4-12

Effects of Tea Intercropping with Soybean on Wuyi Rock Tea

Zhi Zhou[1], and Hong Liao[1]
[1]Root Biology Center, Fujian Agriculture and Forestry University
350002 Fuzhou, Fujian, China
; hliao@fafu.edu.cn

INTRODUCTION

Acidic soils comprise up to 50% of the world's potentially arable lands. Phosphorus (P) is an essential element required for both plant growth and development, and P-deficiency is a major factor limiting plant growth in acidic soils, particularly in the highly weathered acidic soils of the tropics and sub-tropics, in which free iron and aluminium (Al) oxides bind native and fertilizer P into forms unavailable to plants. Tea plants (*Camellia sinensis*) are cultivated in humid and sub-humid tropical, sub-tropical, and temperate regions of the world, mainly in pH 4.5–5.5 acidic soils (Li et al, 2008). Tea is one of the most widely consumed beverages in the world and is one of the main economic crops in Fujian province of China. Excessive fertilization in tea gardens causes soil acidification and serious environmental pollution. Intercropping of tea plants with different crops is a cultivation mode with long history. A lot of experimental results showed that the residues from rational intercropping benefits to form soil aggregates, and therefore improves soil fertility (Ma et al, 2017). It is expected that intercropping of tea with soybean could optimize soil chemical-physical properties, such as increase the content of organic matter and nutrient in tea garden, as well as promote the growth of tea plants and improve tea quality. However, few related studies on tea plants intercropping with soybean have been reported. Wuyi rock tea is an exceptional Chinese Oolong Tea. The aim of this experiment is to study the effects of tea plants intercropping with soybean on soil fertility, yield and quality of Wuyi Rock Tea, so as to provide a practical strategy for developing sustainable and environmental friendly tea garden. .

MATERIALS AND METHODS

A nutrient efficient soybean (*Glycine max (L.) Merrill*) cultivar, Huaxia 1, was selected for intercropping with two tea varieties (*Camellia sinensis L.*), Rougui and Baiyaqilan. Before sowing, soybean seeds were inoculated with the mixture of high effective rhizobial strains. There were 3 treatments, including before sowing, after maturity and control, 5 repetitions for each treatment. Tea leaves were sampled following the local tea making standard after harvested soybean grains. The tea yield traits, including the weight of 100 buds and biomass of tea leaves were measured. Some secondary metabolites of tea, such as catcatin, theanine and caffeine were monitored. The basic soil physical and chemical properties in the two tea gardens, like soil pH, organic matter, available N, P and K were measured in the top 0~20 cm soil layer before and after soybean growth.

RESULTS AND DISCUSSION

After intercropping with soybean, the soil pH and soil fertility in the tea garden were significantly increased. As shown in (Table 1), the soil pH in the two locations was increased by 0.15 and 0.19, respectively. Furthermore, soil organic matter, available N, P and K in the two plantations were increased 0.28% and 0.07%, 25.03%

and 13.83%, 20.30% and 31.50%, 17.47% and 31.63%, respectively.

Table 1 Effects of tea-soybean intercropping on soil nutrient status

Parameter	Rougui			Baiyaqilan		
	Before sowing	After maturity	Control	Before sowing	After maturity	Control
pH	4.52±0.14b	4.97±0.08a	4.82±0.05c	4.57±0.08b	5.12±0.07a	4.93±0.10c
Organic matter (%)	3.50±0.35b	3.93±0.1a	3.65±0.10b	3.41±0.17b	3.70±0.05a	3.63±0.08c
Alkali hydrolytic nitrogen (mg·kg^{-1})	90.06±12.23b	130.20±6.58a	104.13±9.20c	117.73±17.44ab	111.90±6.67a	98.30±3.90b
Available phosphate (mg·kg^{-1})	31.65±13.89b	72.40±3.91a	60.18±5.16c	148.09±23.64b	93.11±8.30a	70.12±6.65c
Available potassium (mg·kg^{-1})	47.49±17.82b	68.80±5.08a	58.57±2.18b	94.59±24.32b	66.12±4.39a	50.23±5.19c

Data in the table are the mean of 5 replications with standard error. Same letter in same tea variety of same line after data means not significant at 0.05 level. The same as below.

Intercropping with soybean significantly increased tea yield, as indicated by 61.20% and 28.31% increase of the weight of 100 buds, and 47.39% and 28.01% increase of leaf biomass in the two gardens, respectively (Table 2).

Intercropping with soybean dramatically improved tea quality, as proved by 15.84% and 35.74% increase of catechin, 18.79% and 4.42% increase of theanine, 12.89% and 34.54% increase of caffeine, and 2.16% and 30.81% decrease of ratio of polyphends to free amino acids in the two gardens, respectively (Table 3).

Table 2 Effects of tea-soybean intercropping on growth of tea

Treatment	Weight of 100 buds(g)	Yield of Per Mu (Kg)
Rougui-Huaxia1	113.78±13.20 a	234.23±21.13a
Control	70.58±16.46b	158.91±14.84 b
Baiyaqilan-Huaxia1	46.26±15.92a	137.71±23.28a
Control	36.02±10.98b	107.58±22.70 b

Table 3 Effects of tea-soybean intercropping on tea quality

Parameter	Rougui		Baiyaqialn	
	Huaxia1	Control	Huaxia1	Control
Catechin （mg·g^{-1}）	56.00±2.67a	48.34±3.22b	54.76±6.34a	40.34±2.17b
Theanine (mg·g^{-1})	24.78±0.72a	20.86±1.09b	26.45±0.56a	25.32±0.64b
Caffeine (mg·g^{-1})	19.44±1.53a	17.22±1.43b	23.72±2.24a	17.63±1.84b
Ratio of polyphends to free amino acids	2.26±0.08a	2.31±0.13b	2.08±0.23a	1.59±0.09b

CONCLUSION

Based on the results above, we conclude that intercropping with nutrient efficient soybean variety inoculated with effective rhizobia could not only improve soil fertility, but also increase tea yield and favorite tea quality, which could be considered as a cultivation mode to build up the sustainable and ecological tea garden producing high quality of tea.

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REFERENCES

Li, J. L., P. F. Tu, N.Chen, J. C. Tang, X. R. Wang, H. Nian, H. Liao, And X. L. Yan. 2008. Effects of Tea Intercropping with Soybean. Scientia. Agricultura Sinica: 2040-2047.

Ma, Y.H., S. L. Fu, X. P. Zhang, K. Zhao, Y. Han, And H. Chen. 2017. Intercropping improves soil nutrient availability, soil enzyme activity and tea quantity and quality. Applied Soil Ecology **119** (2017): 171-178.

P4-13

Evaluation of two different compost materials as organic fertilizers on soil fertility status

***Illani Zuraihah*[1]., *Z.Z. Norziana*[1] *and M. Theeba*[1]**
[1]Crop and Soil Science Research Centre, MARDI Headquarters, Persiaran MARDI-UPM, 43400 Serdang
; illani@mardi.gov.my

INTRODUCTION

In organic vegetable cultivation, inorganic fertilizer is not allowed with the exception of naturally occurring mineral sources. The substitutes for inorganic fertilizer are mostly in organic nutrient sources that can be grouped into organic fertilizer such as from animal sources, plant sources, green manures and compost (Aini et al. 2005). In improving soil fertility, application of organic fertilizer has become an important practical measure. Organic fertilizer or compost is able to enhance soil microbial activity (Ren et al. 1996), increased microbial biomass (Suresh et al. 2004) and improved soil structure (Bin 1983; Dauda et al. 2008). Compost is widely used as a soil amendment to improve soil structure, provide plant nutrients and facilitate the re-vegetation of disturbed soils (Brady and Weil, 2002). Though high amounts of compost are required initially at the organic farm, the quantity of subsequent application may be reduced as soil physical, chemical and biological improve with time (Aini et al. 2005). Thus, the objectives of this study was to determine the effect of different source of composts on the soil chemical and biological properties.

MATERIALS AND METHODS

Field experiment and treatment used

The experiment was carried out at MARDI's Integrated Organic Farm, Serdang, Selangor. The test crop was *Lactuca sativa* grown under rain shelter with two cycles of planting and treated with two types of compost as organic fertilizers. They were plant based compost (PC), food waste compost (FC) and non-amended soil which served as CONTROL. Food waste is from restaurant which consist of cooked food and vegetables/meat leftoverb which had undergohe composting using hi-tech machine (CW500) while plant based is from leaves and grass cutting mixed with goat manure composted manually. The application rate for this study was 2 kg/m^2. The following treatments were made in a randomized complete block design (RCBD) in five replicates. Raised beds were prepared before treatment and soil samples were collected during this stage for chemical and microbial analysis.

Laboratory analyses

Compost samples from two types of composts were weighed into crucibles and placed in the sampler for C, N determination using CHNOS analyser (ELEMENTAR-Vario Macro Modules 11.44 - 5201). Total P in compost samples was determined using recommended methods for plant chemical analysis (MS677: pt I to VIII: 1980, Part IV). Soil sample were collected at depth of 0-20 cm before application of organic fertilizers and after harvesting with five replicates for chemical and biological properties. The samples were air dried and ground to pass a 2 mm sieve for analyses. Organic carbon was determined by Walkley and Black employing rapid titration method. Soil pH was measured at 1:2.5 ratios with water. Wet soil samples were used for microbial population for total colony forming unit (cfu-log$_{10}$) using total plate count on nutrient agar.For biological properties, a 10 g of wet weight of soil samples were diluted in 90 ml of physiological saline water (0.85% NaCl). Then, a series of dilution factor which consist of 100 µL of diluted samples were then spread on non-selective media (growth media), nutrient agar (Oxoid) and counted for the microbial population of total colony forming unit (CFU/mL). The number of colony forming unit (cfu) after incubation period (1-7 days) represented the population density (cfu per g wet weight of biofertilizer). Selected microbes from samples were screened on the selective media using N-free agar for free-living dinitrogen fixing bacteria (NF) and phosphate solubilizing media (PSM) for phosphate solubilizing bacteria.

RESULTS AND DISCUSSION

Chemical Content of the Compost analysis

The PC and FC were analysed for their chemical properties and the data are shown in Table 1. The FC was found to be acidic with pH 3.21 and very low CEC of 0.06 cmol/kg which support the phyto-toxicity effects on low percentage of seed germinations in early study. The FC then was subhected to heavy metals analysis assessment. However, the heavy metal content of the FC is detected lower than the maximum level of Department of Agriculture requirement of permitted limit (Table 2). Major nutrients in FC such as N, P and K are found lower than 2%. Nevertheless, the FC samples still follow the SIRIM standard of organic fertilizer to consist N value from 1.5% and above. Heavy metals in FC product are below the maximum permissible value in organic fertilizer according to MS1517:2012. For PC compost, the NPK ratio is 3:4:2 with high CEC value and this properties support the nutrients availability and uptake by the plant.

Table 1. Chemical properties for two types of compost used in the experiment

Type of compost	pH (H_2O)	CEC (cmol$^{(+)}$/kg)	Organic carbon (%)	Total N (%)	P_2O_5 (%)	K_2O (%)	Rate of application (kg/ha)
PC	7.5	18.0	54.0	2.93	4.63	2.38	20,000
FC	3.12	0.06	24.49	1.50	0.86	1.64	20,000

Table 2. Heavy metals content in FC compared to maximum permissible value according to the Department Agriculture Malaysia

Heavy metals	Maximum allowable limit (mg/kg)	FC heavy metals content (mg/kg)
Chromium (Cr)	200	72
Lead (Pb)	300	2
Nickel (Ni)	150	14
Cadmium (Cd)	5	0.1
Mercury (Hg)	2	n.d
Arsenic (As)	50	4

n.d – not detected

Changes in soil chemical and biological properties

Changes in soil microbial population from initial cropping to after harvest are as shown in Figure 1. Results showed FC contained the highest total microbial count at the initial cropping stage especially for P-solubilizing bacteria. However total microbial count after planting were identified the highest in PC treatment. N-Fixer population was the highest in control treatment. Generally, FC was found to sustain soil microbial population throughout the cropping cycle better than PC and CONTROL plot.Table 3 shows, the changes in some of selected soil chemical properties before and after cropping for all the treatments. Soil CEC, organic C and organic matter were found the higher in PC> FC> CONTROL. Soil pH value for all the treatments and control were neutral at pH 7-8.

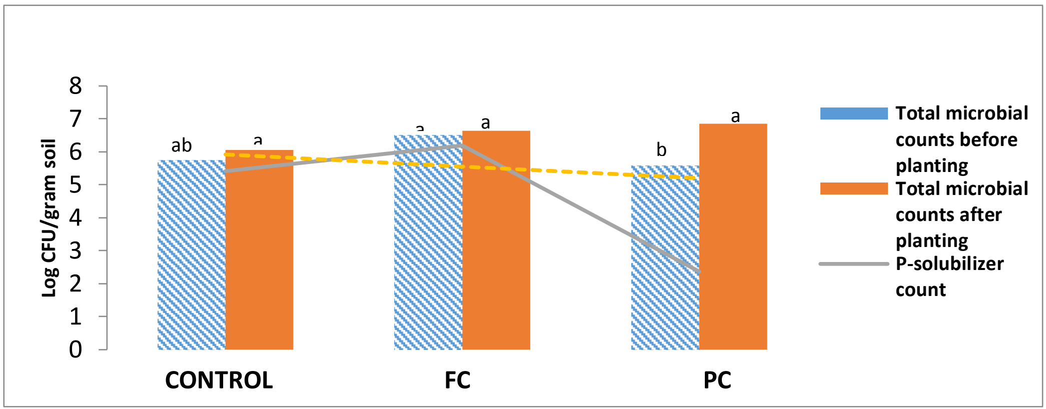

Figure 1. Microbial population trend in experimental plot

Table 3. Selected soil chemical properties at experimental plots

Parameter	Soil sampled before planting started			Soil sampled after of planting		
	CONTROL	FC	PC	Control	FC	PC
CEC (meq/100gmsoil)	5.06 ±0.75	4.42 ±0.35	5.78±0.89	4.63±0.13	9.88±3.57	11.81±2.08
pH	7.36 ±0.03	7.26 ±0.07	7.30±0.11	7.59±0.04	7.67±0.01	7.60±0.01
Conductivity (us/cm)	271.10 ±30.5	372.90 ±86.9	491.80±202	168.63±11.46	205.13±4.42	202.35±27.35
Organic C (%)	1.95 ±0.05	2.01±0.02	1.93±0.06	1.70±0.07	2.01±0.06	2.47±0.54
Organic matter (%)	3.38 ±0.09	3.47±0.03	3.35±0.10	2.92+0.13	3.48±0.11	4.26±0.93

CONCLUSION

It can be concluded that the performance of FC as fertilizer for organic crop was moderately good and can be recommended for various crops. However, the FC was found to give phyto-toxicity effects through low seed germination at first cycle planting. Thus, refinement of the fertilizer product is suggested through further decomposition or applied with effective microbes to reduce the phyto-toxic compound in the fertilizer. On the other hand, application of FC three weeks earlier before the crop seeding or transplanting is recommended for better crop performance as the phyto-toxicity effect can be minimized. The pH of FC fertilizer is very low (acidic) which is expected form the toxic compound and it needs to be increased to above 5.5 to perform as suitable organic fertilizer. FC was found not to increase the soil properties significantly where very little changes were found before and after cropping in the treatment. Meanwhile, PC compost from plant based material showed higher soil fertility improvement followed by the CONTROL.

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REFERENCES

Aini, Z., Sivapragasam, A., Vimala, P., and Mohamad Roff, M.N. (2005). Organic vegetable cultivation in Malaysia. Malaysian Agricultural Research and Development Institute. Pp 47-67.

Bin, J. (1983). Utilization of green manure for raising soil fertility in China. *Soil Sci.,* **135**:65-9.

Brady, N.C, Weil, R. R (2002). *The Nature and Properties of Soil*. Macmillian; New York.Pp. 616 - 660.

Dauda, S.N., F.Ajayi and E.E.Ndor. (2008). Growth and yield of water melon (*Citrullus lanatus)* as affected by poultry manure application. *J.Agric. Soc Sci* **4**:121-4.

Luo, H.F., Qi, H.Y and Xue,C. (2003). A preliminary application of PCR-DGGE to study microbial diversity in soil. *Acta Ecological Sinica* **23**: 1570-1575.

Mosier, A.R., J.K. Syers and J.R. Freney. (2004). Agriculture and the nitrogen cycle. Assessing the impacts of fertilizer use on food production and environment. Scope 65. *Island Press, London.*

Steiner, C., W.G. Teixeira, J. Lehmann, T. Nehls, de Macêdo, J. L. V., W.E.H. Blum and W. Zech. (2007). Long term effects of manure, charcoal and mineral fertilization on crop production and fertility on a highly weathered Central Amazonian upland soil. *Plant and Soil.* Vol. **291**:1, pp. 275-290.Steiner, C., K.C. Das,

Suresh, K.D., G. Sneh, K.K.Krishn and C.M.Mool. (2004). Microbial biomass carbon dan microbial activities of soils receiving chemical fertilizer and organic amendments. *Archives Agron. Soil Sci.,* **50**:641-7

Van Zwieten, L. (2006). Impact of agriculture inputs on soil organism. *Aust. J. Soil Res.* **3**:1-15.

MS1517:2012 Organic Fertilizer Specification (First Revision)

MS677: Pt I to VIII: (1980). Part IV: Determination of Phosphorus, Recommended Methods for Plant Chemical Analysis

MS677: Pt I to VIII: (1980). Part 11: Ashing and Preparation of Ash solution, Recommended Methods for Plant Chemical Analysis, SIRIM

P4-14

Evaluation of macroelements and physical analysis of soils at five natural populations of *Chromolaena odorata* L. for future herbal plantation programme

Farah Fazwa, M.A1*., Norhayati, S[1]., Syafiqah Nabilah, S.B[1]., Jeyanny, V[1]., Mohd Zaki, A[1]., Masitah, M.T[1]. and Mohd Asri. L[1]
[1]Forest Biotechnology Division, Forest Research Institute Malaysia, 52109, Kepong, Selangor, Malaysia
; farah@frim.gov.my

INTRODUCTION

Chromolaena odorata L. is commonly known as Siam weeds or in Malaysia is a medicinal plant which is getting high demand from herbal industry for its various purposes. Researchers from Forest Research Institute Malaysia (FRIM) has taken the initiative to conduct some studies on its agronomic requirements especially on soil types and properties at the natural conditions where the species are always found. This paper presents the investigation of macronutrients compositions and physical texture of soils at five natural populations of *C. odorata* in Peninsular Malaysia. The data presented can be used as a guidance in the establishment of *C. odorata* herbal plantation as well as a reference point for furtherculture managementof the species.

METHODOLOGY

Soil Sampling

The soils were collected from five populations; i) Mata Ayer, Perlis; ii) Jasin, Melaka iii) Maran, Pahang iv) Kota Tinggi, Johor and v) Setiu, Terengganu). Soil samples were randomly collected at depth of 0-50 cm using auger at three points of *C. odorata* mother plant and composited for each location.

Soils Analysis

Analyses were focused on soil texture (percentage of silt, clay and sand) and macroelementa composition such as nitrogen (N), available phosphorus (P) and exchangable potassium (K). Total nitrogen in soils was analyzed by micro Kjeldahl digestion followed by distillation and titration with 0.1 N HCl. Available phosphorus in soils was extracted using the Bray and Kurtz no. 2 extractant and its concentration was determined using UV-Visible spectrophotometer (Cary 60 - UV). Potassium in soil was extracted by 1 N ammonium acetate and K concentration was analyzed using Inductively Couple Plasma (ICP) - Optical Emission Spectrometer (Varian 725-ES). The soil acidity was measured using pH meter of a mixture of 1:2.5 ratios of soil and water. For physical properties, soil texture was analyzed using the pipette method. All the analyses were conducted at the Soil Chemistry Laboratory, Forest Research Institute Malaysia (FRIM).

Growth performances of Chromolaena odorata

A total of 150 mother plants were collected from the five populations and labelled with different code numbers. The topographic information such as coordinates, altitudes and dates of assessment were also recorded. The locations of origin were also tagged using Global Positioning System (GPS) for records (Table 1). Morphology characteristics of all mother plants such as height (cm), collar diameter (mm), leaf length (cm), leaf width (cm) were measured.

Table 1: Topographic information of five *Chromolaena odorata* populations

Population	Altitude (m)	Point (X)	Point (Y)	Date of survey
Mata Ayer, Perlis	53.95	250791.55	732232.78	1-3 March 2017
Jasin, Melaka	31.68	638250.52	199172.32	15-18 May 2017
Kota Tinggi, Johor	31.04	491942.81	254241.37	16-20 Feb 2017
Maran, Pahang	45.18	536110.36	400435.74	17-20 April 2017
Setiu, Terengganu	11.38	564747.03	597568.38	3-7 April 2017

RESULTS AND DISCUSSION

Table 2 presents the results of the microelements of chemical analysis at five locations. It is recorded that population Setiu, Terengganu has the highest total nitrogen (0.33%) and available phosphorus (10.26 mg/kg). Soil in Kota Tinggi, Johor has the lowest total nitrogen (0.11%) and available phosphorus (1.45 mg/kg). All soil types show pH ranging from 4.8 to 7.22. Table 3 presents the physical texture, where coarse sand content varied from 23-82%; fine sand (5-75%); silt (2-21%) and clay (11-35%).
Table 2: Macroelements of soil chemical compositions of *Chromolaea odorata* at five natural populations

Populations	pH	N (%)	Available P (mg/kg)	Exch. K (cmol/kg)
Mata Ayer, Perlis	7.22	0.16	3.67	0.10
Jasin, Melaka	5.10	0.12	1.91	3.15
Kota Tinggi, Johor	4.80	0.11	1.45	0.04
Maran, Pahang	5.20	0.13	2.52	0.11
Setiu, Terengganu	5.67	0.33	10.26	0.026

Table 3: Texture of soil planted with *Chromolaea odorata* at five natural populations

Populations	Sand (%)		Silt (%)	Clay (%)
	Coarse	Fine		
Mata Ayer, Perlis	23.35	75.75	21.41	34.8
Jasin, Melaka	32.10	20.58	19.5	30.5
Kota Tinggi, Johor	42.80	15.10	8.40	32.5
Maran, Pahang	31.95	18.35	14.1	33.75
Setiu, Terengganu	82.95	5.95	2.50	11.4

Overall results (Table 4) indicated that growth of the species is better in acidic soil than in alkaline soil. Beside Mata Ayer, Perlis (alkaline soil), plant growth in other areas (acidic soil) was better in terms of collar diameter, height, leaf width and leaf length. Most plants including flowers, vegetables, shrubs, and trees grow best where the soil pH ranges from 5.8 to 7.0. (Mason, 2017). However, growth can be inhibited under low acidic soil condition below the pH of 5.

Table 4: Morphological data of *Chromolaea odorata* mother plants at five natural populations

Population	Morphological data			
	Height (cm)	**Collar diameter (mm)**	**Leaf Length (cm)**	**Leaf Width (cm)**
Mata Ayer, Perlis	$163.73^{b}\pm5.76$	$6.20^{b}\pm 0.29$	$7.28^{b}\pm 0.26$	$3.96^{b} \pm 0.20$
Jasin, Melaka	$179.43^{b}\pm5.86$	$7.73^{a}\pm 0.31$	$9.95^{a}\pm 0.36$	$5.39^{a} \pm0.23$
Maran, Pahang	$175.80^{b}\pm5.43$	$8.24^{a}\pm 0.41$	$9.94^{a}\pm 0.16$	$5.54^{a} \pm0.11$
Kota Tinggi, Johor	$159.55^{ab}\pm6.29$	$7.67^{a}\pm 0.29$	$10.20^{a}\pm0.13$	$5.31^{a} \pm0.10$
Setiu, Terengganu	$197.37^{a}\pm8.91$	$7.44^{a}\pm 0.39$	$9.86^{a}\pm 0.30$	$5.79^{a} \pm0.23$

In actual plantation situations, materials are added to the soil to adjust the pH**.** On a field scale, this is most commonly done for acidic soils to raise the pH from 4.5 to 5.5 up to 6.5 or approaching neutrality.

CONCLUSION

Therefore, if *C. odorata* is to be planted as a plantation crop in the future, soil physico chemical amelioration and fertilization has to be undertaken to ensure that the plantation soil is under acidic condition. This is due to the fact that most plants, shrubs, and trees including *C. odorata* can grow best on the soil with pH ranges from 5.8 to 7.0.

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REFERENCES

Iwu, M.M., Duncan, A.R. and Okunji, C.O. 1999. New antimicrobials of plant origin. Perspectives on new crops and new uses. ASHS Press, Alexandria, VA., p. 457-462.

Sandra, M. 2017. Understanding Acid-Loving Plants and Soil pH **http://web.extension.illinois.edu/cfiv/homeowners/080626.html**

P4-15

Total phenolic content in *Labisia pumila* var. *Alata* (clone kfefrim01) planted at four different locations

Farah Fazwa, M.A[1].*, *Syafiqah Nabilah, S.B[1].*, *Norhayati, S[1].* and *Masitah, M.T[1].
[1]Forest Biotechnology Division, Forest Research Institute Malaysia, 52109, Kepong, Selangor, Malaysia
; farah@frim.gov.my

INTRODUCTION

Labisia pumila or commonly known as kacip fatimah, belongs to the family of Primulaceae. It possesses several medicinal properties such as pre and post-partum medication (Jamia, 2004), anti-obesity (Fazliana et al., 2009), anti-aging (Hyun-kyung et al., 2010) and anti-microbial Karimi et al., 2011; Ali and Khan, 2011. The secondary metabolites in the plants are the contributors to the biological and pharmacological activities. The antioxidants obtained from plants have greater benefit compared to synthetic ones. The basic aim of this research was to determine the total phenolic content (TPC) which is related to anti-oxidant properties of *Labisia pumila* var. *alata* (clone KFeFRIM01) after planted at four different populations. The output of this study can be used as guidance to herbal planter in determining the best soil condition of the species in order to produce high secondary metabolites content.

METHODOLOGY

Preparation of planting materials and determination of total phenolic content

One elite clone identified from the previous study (namely as KFeFRIM01) were propagated using leaf cuttings. The plants produced from the propagation were planted at four different locations in Peninsular Malaysia, i) Mata Ayer, Perlis ii) Maran, Pahang, iii) Setiu, Terengganu and iv) Kepong, Selangor. After nine months, the leaves were harvested and screened for total phenolic content (TPC) using using Folin – ciocalteu method (Singleton & Rossi, 1965).

Soil Sampling and analysis

At the early establishment of planting plots, soil samples were randomly collected at depth of 0-25 cm using auger at three points at each location. All soils samples were packed into plastic bags and transported back to FRIM. Analysis was focused on physical texture (percentage of silt, clay and sand) and macroelement composition such as nitrogen (N), available phosphorus (P) and exchangable potassium (K). All analysis was conducted at Soil Chemistry Laboratory, Forest Research Institute Malaysia (FRIM).

RESULTS AND DISCUSSION

Soil analysis at four locations

Table 1 presents the soil chemical chercteristic at four locations. It is recorded that the soil in Kepong, Selangor has the highest total nitrogen (0.20%) and exchangeable potassium (18.04 mg/kg). The soil in Setiu, Terengganu, has the lowest rate for exchangeable potassium (0.48 cmol/kg) and CEC (0.01 cmol/kg). All soil types showed pH greater than 8.0 except for soil in Setiu, Terengganu where the pH is low (1.88). The results indicated that most of the locations are categorized as alkaline soils except soil in Setiu, Terengganu which is acidic. Table 2 presents texture analysis, where coarse sand content varied from the range of 3-91%; fine sand (7-47%); silt (0.2-

33%) and clay (7-34%). It was found that location from Setiu, Terengganu comprised the highest coarse sand and it was classified as BRIS soil (beach ridges interspersed with swales).

Table 1: soil chemical properties at four planting plots of *Labisia pumila var. alata* (KFeFRIM01)

Location	**Nutrient Contents in soil**				
	N (%)	**Av. P (mg/kg)**	**Ex. K (cmol/kg)**	**CEC (cmol/kg)**	**pH**
Mata Ayer, Perlis	0.1	5.43	3.02	0.24	8.16
Maran, Pahang	0.1	4.50	2.87	0.20	9.89
Setiu, Terengganu	0.1	5.06	0.48	0.01	1.88
Kepong, Selangor	0.2	5.05	18.04	0.07	8.37

Table 2: soil texture at four planting plots of *Labisia pumila var. alata* (KFeFRIM01)

Location	**Percentage Soil texture (%)**				**Soil Texture**
	Coarse sand (%)	**Fine sand (%)**	**Silt (%)**	**Clay (%)**	
Mata Ayer	3.7	47.0	33.7	18.3	Sandy loam
Maran	28.3	40.7	9.0	27.0	Sandy clay loam
Setiu	91.0	7.0	0.23	7.0	Sand
Kepong	37.7	21.7	7.3	34.3	Clay loam

Determination of total phenolic content (TPC)

From the screening, it was found that leaves of *L. pumila* var. *alata* (KFeFRIM01) from all populations contained of total phenolic content (TPC). Table 3 indicated that plants located in Setiu, Terengganu has the highest total phenolic content (515.00 ± 132.86 mg/100 g GAE) compared to all locations. Whereas, Mata Ayer, Perlis gave the lowest value of total phenolic content at 237.50 ± 41.99 mg/100 g GAE. Overall result indicated that production of total phenolic content of the species is better in acidic soil than in alkaline soil. This finding is in line with a study conducted by Gisela et al. (2012) which indicated that the higher value of soil pH, the lower kernel yield and protein of sweet Lupine (*Lupinus angustifoliu*). Other study indicated that the highest correlation of the production of the glycoside salidroside compound in *Rhodiola sachalinensis* was affected by the soil pH. It is clearly shown that soil pH condition has an impact on the production of secondary metabolites of the plants.

Therefore, it is suggested that sites selection for growing *L. pumila* as a plantation crop in future should be in a low pH condition in order to get optimum yield of its secondary metabolites.

Table 3: Total phenolic content (TPC) from *Labisia pumila var. alata* (KFeFRIM01) leaves harvested from four different locations

Location	**Total phenolic content (mg/100 g GAE)**
Mata Ayer, Perlis	$237.50^{c} \pm 41.99$
Maran, Pahang	$262.50^{c} \pm 37.50$
Setiu, Terengganu	$515.00^{a} \pm 132.86$
Kepong, Selangor	$468.75^{b} \pm 72.54$

CONCLUSION

Results of this study indicated that certain chemical and physical properties of soils with low pH condition can positively affect the production of total phenolic content in *L. pumila* plants. There are also other environmental factors which influence the performances of the plants such as age, rainfall and temperature. Other than that, the data presented can be used as guidance in the establishment of *L. pumila* plantation crop as well as a reference point for plant breeder to produce high quality clone of *L. pumila* based on chemical compound in the future.

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REFERENCES

Ali, Z. and I. A. Khan, 2011. Alkyl phenols and evaluation of an aqueous extract of *Labisia pumila*, saponins from the roots of *Labisia pumila* a Malaysian herb, Food Chem. Toxicol, 47: 2661-5. (Kacip Fatimah), Phytochem. **72:** 2075-80.

Farah Fazwa, M.A., Aminah, H., Lokmal, N. & Mohd Zaki, A. 2012. Macroelement and physical analysis of different soil types at naturally grown *Labisia pumila* populations. Proceedings of The Soil Science Conference of Malaysia 2012. Pp 381-385

Fazliana, M., Wan Nazaimoon, W.M., Gu, H.F., & Östenson, C.G. 2009. *Labisia pumila* extract regulates body weight and adipokines in ovariectomized rats. Maturitas, **62**(1), 91–97. http://doi.org/10.1016/j.maturitas.2008.10.004.

Gisela, J., Hans-Ulrich, J., Edgar, S., and Frank, O. 2012. Effect of the soil pH on the alkaloid of *Lupinus angustifoliu. International Journal of Agrinomy*, Vol. 12, Article ID 269878.

Hyun-kyung ,C., Dong-hyun, K., Jin Wook, K.,3Sulaiman, N., Mohamad Roji, S., and Chang S.P. 2010. *Labisia pumila* extract protects skin cells from photoaging caused by UVB irradiation. Journal of Bioscience and Bioengineering, **109**(3), 291–296. www.elsevier.com/locate/jbiosc.

Jamia Azdina, J., Ibrahim, J. Khairana, H. & Juriyati, J. 2004. Perkembangan penyelidikan dan pembangunan Kacip Fatimah. *Proceedings of the Seminar on Medicinal Plants and Aromatic Plants 2004*, Pp: 13-19.

Karimi, E., Hawa, Z. E. J. and Sahida, A. 2011. Phytochemical analysis and antimicrobial activities of methanolic extracts of leaf, stem and root from different varieties of *Labisia pumila* Benth, Molecules, **16**: 4438-50.

Singeleton, V.L. & Rossi, J.A. 1965. Calorimetry of total phenolic with phoyospomolybdicphosphotungstic acid reagents. *American Journal of Enology and Viticulture*, **16:144** (158).

P4-16

Effects of low ph compost on early growth of *Labisia pumila* var. *Alata* at nursery stage

Syafiqah Nabilah, S. B[1]., Farah Fazwa, M. A[1]., Norhayati, S[1]. and Masitah, M. T[1].
[1]Plant Improvement Program, Forestry Biotechnology Division, Forest Research Institute Malaysia, 52109 Kepong, Selangor
; syafiqah@frim.gov.my

INTRODUCTION

Labisia pumila is a traditional medicinal plant which has wide therapeutic application including post-partum treatment, dysentery, dysmenorrhea and gonorrhea. In natural habitat, *L. pumila* favor slightly acidic soil condition (pH 4.0 - 4.2) as reported by Farah Fazwa et al. (2015). However, most of the field trial site as well as the plantation site of *L. pumila* nourished with alkaline soil (ph 8.0 – 9.0) (Norhayati et al., 2016). Among the symptom observed on *L. pumila* growth in alkaline soil is the yellowing of the plants which affect the plant productivity. According to Reich (n.d), the symptom indicates iron deficiency due to insufficient soil acidity to put iron into a form that a plant can absorb. Therefore, low pH compost (pH below 6) was prepared using dried leaves of *L. pumila* with the mixture of controlled-release fertilizer (CRF) to improve the soil fertility and increase the plant productivity. The compost was labelled as CompAcc which recommended to be used during the acclimatization stage, growing stage as well as at the early stage of field plantation. In this study, CompAcc was tested on plantlets of *L. pumila* var. *alata* during the second stage of acclimatization process to evaluate the plant growth performance on amended soil using low pH compost.

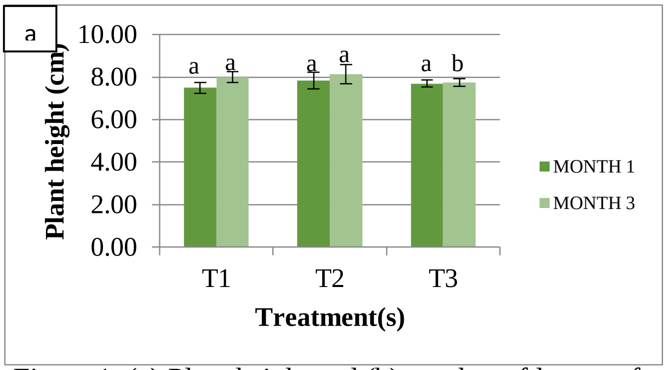
MATERIALS AND METHOD

Plantlets of *L. pumila* var. *alata* produced from Centre of Bioentrepreneur Biotehcnology FRIM lab were hardened in acclimatization chamber for a month. Then, plants with homogenize height and number of leaves were selected and transferred to the nursery for another hardening process. At this stage, different combinations of planting media consist of soil, compost and sand were prepared; Treatment 1 (T1): Soil: CompAcc: Sand - (4:1:1); Treatment 2 (T2): Soil: CompAcc: Sand - (3:1:1) and Treatment 3 (T3): Soil: COC: Sand – (3:2:1) (Control). The pH of CompAcc is in the range of 4.7 – 5.2 while the pH of commercial organic compost (COC) is 7.0 – 7.5.
The plant growth such as plant height and number of leaves was recorded during the second stage of hardening process. The experiment was conducted for three months. Data collected was subjected to analysis of variance (ANOVA) and the means value were compared by the Duncan's Multiple Range Test at $p \leq 0.05$ using the IBM SPSS Statistics version 22.

RESULTS AND DISCUSSION

The analysis of variance (ANOVA) showed there is significant difference between different treatment media with plant height and number of leaves at $p < 0.05$. Figure 1(a) showed plant height of *L. pumila* var. *alata* measured after first and third month of experiment. During the first month of experiment, plants in all treatment media have homogenized height in the range of 7.5 ± 0.26 cm to 7.8 ± 0.4 cm. After three months of observation, plants growth in treatment media containing CompAcc (T1 and T2) showed better plant height increment with 8.10 ± 0.25 cm and 8.14 ± 0.44 cm respectively, as compared to commercial organic compost (T3) with 7.75 ± 0.20

cm. Figure 1(b) showed number of leaves produced after first and three month of experiment. Similar with plant height, leaves number of the plants in all treatment were around 7.60 ± 0.27. After three months, plants in T1 and T2 produced greater leaf number with 8.11 ± 0.28 and 8.11 ± 0.31 respectively compared with T3 (7.75 ± 0.20). T1 and T2 each showed 50% increment of leaf production after three months while T3 only increased about 15%. The addition of CompAcc into *L. pumila* growing media has influenced the growth rate of the plants in terms of height and leaf production. The low pH compost significantly improved the growth of *L. pumila* var. *alata* which favor slightly acidic condition. Beside, *L. pumila*, this compost also showed good growth performance on other herbal species such as *Andrographis paniculata* (data not shown). CompAcc is one of the examples for low cost soil amendment substrate that give positive effect to the plant growth. It could reduce the maintenance cost in *L. pumila* plantation and increase the yield per hectare.



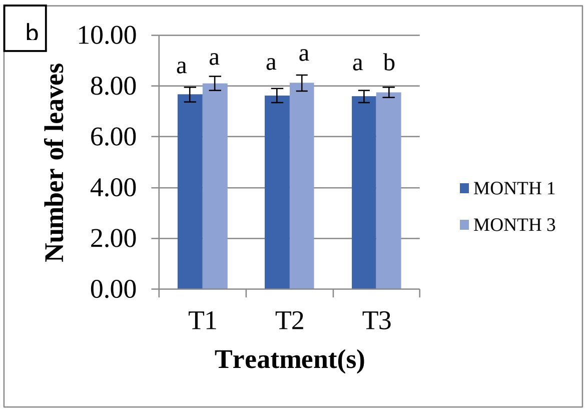


Figure 1: (a) Plant height and (b) number of leaves of *Labisia pumila var. alata* in different treatment media

CONCLUSION

The low pH compost (CompAcc) improved growth performance of *L. pumila* var. *alata* compared to the used of commercial organic compost. Therefore, it is important to study the suitable pH of growing media used for each plants as it slightly influence the nutrient absorption that give impact to plant productivity.

REFERENCES

Farah Fazwa, M. A., Maideen, H., & Mohamad, O. (2012). Selection Among Two Varieties of Labisia pumila that Yield High Phenolic Contents For Establishing Plant Stock and Further Cultivation. *Journal of Tropical Medicinal Plants*, ***13***(1).

Norhayati, S., Farah Fazwa, M.A., Mohamad, O., Syafiqah Nabilah, S.B., Shamsiah A. (2016). Evaluation of Genotype x Environment Interaction on Morphological Characteristics of Eight Selected *Labisia pumila* var. *alata* Clones (Kacip Fatimah) by Francis and Kannenbergs Method. *International Journal of Environmental and Agriculture Research, 11*(2).

Reich L. (n.d). The four things you need to know about soil pH. *Fine Gardening.* Retrieved from https://www.finegardening.com/article/the-four-things-you-need-to-know-about-soil-ph

P5- Sustainable management of plantation and other crops on acid soils.

P5-1

Distribution of several physical and chemicals soil in smallholder cocoa plantation konawe selatan

***Hasbullah Syaf*[1]**
[1]Department of Soil Science Faculty of Agriculture, University of Halu Oleo, , BTN UHO Blok K No 14 Anduonohu Kendari, 93232
; hassyaf@yahoo.co.id

INTRODUCTION

Cocoa is a major plantation in Southeast Sulawesi Province, including Kabupaten Konawe Selatan. Cocoa is grown in farmers' gardens so it is called the smallholder cocoa plantation. Within a period of 20 years, cocoa continues to decline in production due to aging of old plants, pests and plant diseases and less optimal land management. The results of Syaf and Kilowasid's research (2010 to 2017) indicate that the availability of physical and chemical properties of the soil data will assist in determining the biological character of the soil in aging cocoa plant management, pest and disease control and optimal land management. The objectives of this research are (1) to know the distribution of some physical and chemical properties of soil in smallholder cocoa plantation and (2) to carry out cocoa problem handling through land unit approach in Konawe Selatan Regency.

MATERIALS AND METHODS

This research uses survey method based on land unit. The land unit map used is a 1: 50,000 scale composed of 1: 100.000 Land unit detail Satellite Map through the terrain analysis approach on the Shuttle Radar Topography Mission (NASA, 2004), contour maps, topographic maps and geological maps. Image Shuttle's Radar Topography Mission interpretation (NASA, 2004) is done using Global Mapper software version 7.2 and ArcView version 3.2. Its symbolism refers to the Landform Classification Guidelines (Marsoedi et al., 1997).
Land use is prepared through Landsat imagery 7 ETM + using ER Mapper version 6.4 and ArcView version 3.2. Then the two maps are moved to the base map that has been compiled. The base map used is a scale of 1: 50,000, this map is used to illustrate the thematic maps of the results of the study, prepared using 1: 50.000 scale contour maps and 1: 50.000 scale earth map. Other data and maps which are the input of this research are land map system scale 1: 250.000. The units of land acquired are subsequently conducted overlapping with the presence of cocoa spread in South Konawe District.
The land units obtained are then collected by composite soil samples collected from several places randomly in units of land, then mixed and taken ± 1 kg. Soil sampling is done on land that has productive cocoa land (4-10 years). Furthermore, soil samples were analyzed in the laboratory to determine the physical and chemical properties of the soil. Physical properties observed and analyzed were drainage, texture, effective depth, slope, surface rock and rock outcrop. The chemical properties analyzed were soil pH, CEC, C-org, Al-dd, N-total, P-available, and K-available. Data of observation and analysis in subsequent units of land is done spatial analysis and construction of handling problem of community cocoa garden

RESULTS AND DISCUSSION

This study yielded 209 units of land based on overlay of thematic maps. Furthermore, observations were made on all units of land that have a cocoa plantation and overlayed to obtain 78 eligible land units in this study. Distribution of soil physical properties of the research location in the form of slope indicates that the unit of land is dominated by the slopes of Datar to Agak Landai (0-8%), the degree of erosion predominantly low, the depth of soil is very deep (> 100 cm) the soil varies but is dominated by textured soil Clay Dust (SiL), has a rocky surface rock class. The soil chemical properties of soil pH at the research sites are generally slightly acidic to very acid and dominated by acid-pH soils. Generally have C-org and CEC for low category, K-available is moderate, P-available is very low, N-total of medium soil. Based on the distribution of soil physical and chemical properties then carried out the construction of handling problems of smallholder cocoa plantation through the utilization of soil biology in counterbalance the availability of nutrients in the soil and create soil health.

CONCLUSION

The distribution of soil physical and chemical properties varies in the peasant garden of the people of South Konawe District. Involvement of soil biology is one of the constructs in handling the problem of smallholder cocoa plantation in South Konawe District.

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2. Planning and development agency of south konawe district

REFERENCES

Kilowasid, L.M.H. and Hasbullah S. (2013). Development of ecosystem restoration method of community cacao plantation land by ecosystem engineers ecosystem fauna (ecosystem engineers). Research Report on Competitive Grant, Kendari.

Kilowasid, L.M.H. and Hasbullah S. (2013). Integration of the soil fauna community in the analysis of community cocoa land resources. Report MP3EI, Kendari.

Syaf, H. (2010). Analysis of cocoa plantation resources in Kolaka District, Southeast Sulawesi Province, Doctoral Dissertation, Post Graduate Program, Padjadjaran University., Bandung.

Syaf, H. (2013). Restoration of Cocoa Conformity Classification Criteria By Minimizing Land Characteristics Used In Evaluating Land In Southeast Sulawesi. Fundamental Research Report. Decentralization of Higher Education. Jakarta

Syaf, H., M. Arifin, A. Suriadikusumah, and R. Harryanto. (2011). The classification of cacao land suitability uses the maximum limiting factor in Kolaka District, Agroteknos Journal, **1** (3): 127 - 134.

Syaf, H. and Laode M.H.K. (2013). Technique of Increasing Soil Fertility on Cocoa People's Land by Farmers in Konda Subdistrict of South Konawe Regency with Ecosystem Engineering Ecosystem Fauna. Report BOPTN UNHALU, Kendari

Syaf, H., Syamsu A., Laode M.H.K., Gusti R.S., Mardin and Jufri K (2015). Report on Preparation of Master Plan for Plantation Plantation Development in Konawe Selatan District. Cooperation between Bappeda Kabupaten Konawe Selatan and Faculty of Agriculture Universitas Halu Oleo. Kendari.

P5-2

Performance of phosphate efficient oil palm genotypes seedlings grown under moisture stress condition

***Siti Nasuha Suboh*[1], *Tan Ngai Paing*[1], *Zaharah Abdul Rahman*[1], *Siti Nor Akmar Abdullah*[1], *Adibah Mohd Amin*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
43400 UPM Serdang, Selangor, Malaysia
; nasuhasuboh92@gmail.com

INTRODUCTION

Most of the phosphate (P) fertilizer applied in soils is fixed by sesquioxides, therefore, it is unavailable for plant uptake. Thus, a higher rate of application of phosphate fertilizer is often required for oil palm cultivation. As Malaysians soils are low in P, P fertilizer is required throughout the production cycle of oil palm. Malaysian oil palm industry is also faced with climate uncertainty as a result of climate change that has resulted in drier weather which has caused moisture stress in oil palm production. This has resulted in low yields of fruitlet production. In view of the limited resources of P in the world coupled with the high price of fertilizer, it is more strategic to identify the P efficient and drought tolerant genes of oil palm genotypes.

MATERIALS AND METHODS

A total of 108 oil palm seedlings (3 months old) from three genotypes were planted inside polyethylene bags for a period of 7 months and 10 days. Only half of seedlings were applied with phosphorus during the study, while the remaining acted as controls. Moroccan rock phosphate (MRP) is used. After 7 months of phosphate starvation, the water was withheld from the seedlings for 30 days, except the control seedlings which were watered daily according to field water capacity. After 1 month of moisture stress, the seedlings are harvested to determine the performance of the genotypes. The P content and the leaf gas exchange measurement in the seedlings are analyzed.

RESULT AND DISCUSSION

The longer the time of treatment, the decline in the moisture content in the plant. As the water is insufficient for the plant uptake, the stomatal closure are occurred and reduced the photosynthetic and transpiration rate due to the decreased of CO_2 diffusion to the chloroplast. Meanwhile, the plants that undergo phosphate starvation, the P content in the plants are lowere compared to the control.

CONCLUSION

This research will introduce the adoption of plants with good nutrient uptake efficiency and drought tolerant with a more environmentally friendly as an ecological practicable strategy to increase the growth of plant in phosphorus and moisture deficient soils.

ACKNOWLEDGEMENTS

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REFERENCES

A.M Azzeme, S.N.A. Abdullah, M.A. Aziz, P.E.M. Wahab. 2017. Oil palm drought indicuble DREB1 induced expression of DRE/CRT- and non-DRE/CRT-containing genes in lowland transgenic tomato under cold and PEG treatments, Plant Physiology et Biochemistry, doi:10.1016/j.plaphy.2016.12.025.

P5-3

Evaluation of different rates of moroccon phosphate rocks for oil palm (*Elaeis guineensis*) production

***Norakmal Khairuanuar*[1], *Tan Ngai Paing*[1], *Mohamed Hanafi Musa*[1], *Adibah Amin*[1], *Zaharah Abdul Rahman*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
43400 UPM Serdang, Selangor, Malaysia
; norakmalkhairuanuar@yahoo.com

INTRODUCTION

Malaysia is currently the world's second largest palm oil producer and has a total of 5.39 million ha of oil palm planted area. Despite this prosperous growth, oil palm cultivation depends heavily on external nutrient source. This is due to the plant's high demand for nutrients as well as the inherently low soil fertility in Malaysia. Phosphorus is one of the most essential elements for plant growth. Phosphorus deficiency is considered to be one of the major limitations in the crop production on a global scale, especially in the tropical acidic soil (George et al., 2006; Raghothama and Karthikeyan, 2005). To ensure a good production, oil palm plantation in Malaysia has been relying heavily on the use of phosphorus (P) fertilizer in particular phosphate rocks. Among the different varieties of phosphate rocks, Christmas Island Rock Phosphate has been extensively used. To evaluate the effectiveness of other phosphate rocks, this study aims to investigate the effect of different rates of Moroccan Rock Phosphate on oil palm growth performance (the uptake on N, K and Mg as affected by phosphate rocks application).

MATERIALS AND METHODS

This study was conducted in Ladang 10, Faculty of Agriculture, UPM Serdang for 8 months. Two different genotypes of oil palm seedlings were used in this study. They were Felda Yangambi (ML161) and Sime Darby Avros. Five-months-old oil palm seedlings from two genotypes were arranged in Randomized Complete Block Design (RCBD). The top 30 cm of a Bungor soil series was collected, air dried and prepared for nursery planting of oil palm seedlings. This soil sample was analysed for their physical and chemical properties prior to the planting. Twenty kg of mixture of Bungor soil series (70%) and sand (30%) were weighed and placed in each polythene bags (20 cm x 20 cm). This soil was allowed to incubate for a week.Grade B MPR was used in this study at different rates. Five different rates were used which then were replicated into 4 replicates (4 plants per replicate). They were 0 mg/kg P, 25 mg/kg P, 50 mg/kg P, 100 mg/kg P and 200 mg/kg P. A total of 160 polythene bags were prepared (2 genotypes x 5 rates x 4 replicates x 4 plants).

RESULTS AND DISCUSSION

The soil has a pH_{water} of 3.98, Bray-2 extractable P of 2.57 mg/kg, organic carbon of 0.292%, exchangeable K 0.122 $cmol^{(+)}$/kg, exchangeable Ca 0.251 $cmol^{(+)}$/kg and exchangeable Mg 0.072 $cmol^{(+)}$/kg. The soil texture analysis showed 58.38% clay, 9.12% silt, and 32.48% sand which gave a textural class of sandy clay. A higher rate of phosphate rock applied to soil will result in higher amount of phosphorus in soil. Whereas the solubility of phosphate rock is dependent on its particle size and types of organic acids released by oil palm roots**.** The optimum rate of phosphate rock used will result in optimum amount of P in soil.

Details of Moroccan Rock Phosphate

P sources		Total P (%)	Solubility as percent of rock	
			2% Formic Acid (FA)	2% Citric acid (CA)
Moroccan phosphate rock	Grade B	30.83	16.83	10.20

A higher rate of phosphate rock applied to soil will result in higher amount of phosphorus in soil. Whereas the solubility of phosphate rock is dependent on its particle size and types of organic acids released by oil palm roots**.** The optimum rate of phosphate rock used will result in optimum amount of P in soil.

CONCLUSION

The effectiveness of phosphate rock in supplying P to acid soil is also dependent on the grade and solubility rate of the phosphate rock itself. The highest grade of phosphate rocks will give the highest effectiveness of phosphate rock in supplying P to acid tropical soil, and vice versa. Thus, the problem faced by the acid tropical soil can be solved and also the growth of the oil palm crops will be as good.

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REFERENCES

George, T.S., Turner, B.L., Greogory, P.J., CadeMenun, B. J. and Richardson, A.E. (2006). Depletion of organic phosphorus from Oxisol in relation to phosphatase activities in the rhizosphere. European Journal of Soil Science, **57**, 47-57.

Sharifuddin, H.A.H and Zaharah, A.R (1980): Phosphorus in Malaysian agriculture. In Pushparajah, E. (ed.) *Proc. Conf. Soil Sci. and Agric. Devt. In Malaysia*. Malaysian Soc. Soil Sci. pp: 108-119.

Tan, N. P., Zaharah, A. R., Akma, A. S. N., & Jamaluddin, N. (2010). Evaluating the variability of Gafsa phosphate rock uptake by oil palm genotypes at nursery stage. *Editorial Board*, ***33***(2), 223-231.

P5-4

Effect of different types of organic materials on low soil pH in cocoa (*Theobroma cacao* L.) yield production at Madai, Kunak, Sabah

***M. Boney*[1], *M. Dandan*[2] *and O. Rozita*[3]**
[1]MCB Cocoa Research and Development Centre, Mile 10, Apas Road,
P.O. Box No. 60237, 91012, Tawau, Sabah, Malaysia
[2]Faculty of Sustainable Agriculture, Universiti Malaysia Sabah,
Locked Bag No 3, 90509, Sandakan, Sabah, Malaysia
[2]MCB Cocoa Research and Development Centre, Jalan Jengka 23,
P.O Box 34, 28000, Temerloh, Pahang Darul Makmur
; boneymuda@koko.gov.my

Fertilizers are organic or inorganic materials of natural or synthetic origin which are added to the soil to supply certain essential elements to plants for grown. With the recent increase price of inorganic fertilizers and environmental issues effects, the utilization of organic materials could be an alternative fertilizer with optimally use. However, organic fertilizers are low in nutrient concentration or solubility or both, yet the slow release of nutrients makes them are available for a longer period of time, thus could affects the soil pH.

This study has been made to determine the effects of different type of organic materials on low pH soil in Madai, Kunak, Sabah for cocoa yield production. The treatments were mixture fertilizer, composted chicken manure, composted cow manure, composted empty fruit bunch and composted cocoa pod husk. Study was conducted using the Randomized Complete Block Design (RCBD) with the block perpendicular to the soil gradient, topography and the slope, whilst the parameters studied were yield production and its soil chemical properties. Statistical analysis was carried out for one way ANOVA (Analysis of Variance) and Tukey's multiple comparison tests using Statistical Product and Service Solutions (SPSS 21.0) software for means comparison if the treatments were significantly different.

Soil analyses preliminary conducted at untreated soil in experimental site (Table 1). The results indicated that the soils initially have low soil pH, thus could affect the availability of some plant nutrients. Despite that, total N, available P and exchangeable potassium were within the adequate range for cocoa.

Table 1: The initial soil chemical characteristics of Madai, Kunak, Sabah

Variables	Adequate range*	Topsoil (0-20 cm)
pH (H_2O)	5.5 -6.5	4.167
Total N (%)	> 0.16	0.18
Available P (mh/kg)	> 15	32.13
Exchange K ($cmol^{(+)} kg^{-1}$)	> 0.24	0.243

*Source: Wong, I. F. T. (1974) (revised) – Soil-Crop Suitability Classification for Peninsular Malaysia, Soils and Analytical Services Bulletin Nr.1, Ministry of Agriculture, Kuala Lumpur

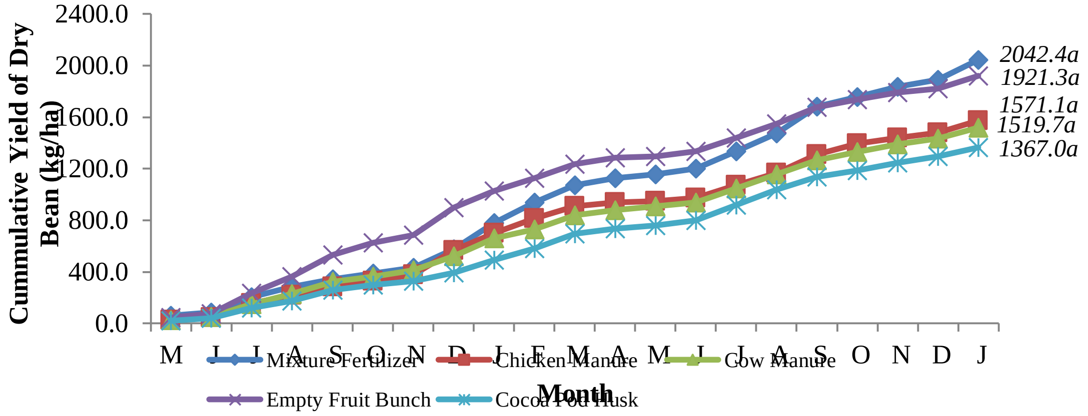
The effects of different types of fertilizer nutrient analysis of topsoil are showed in Table 2. After treatments, results indicated that there are no significant difference ($p>0.05$) of nutrient contents in the soils between treatments. However, the soil pH at the experimental site after treatment has slightly increased up to 4.60 or 12.07% than the preliminary analysis. Although lime was applied onto the soil twice annually in correcting the soil pH, it often take a year or more before a response can be seen even under perfect conditions. As for nitrogen, the concentration in all treatments was lower

(0.09% to 0.15%) than adequate range. The P concentration for all treatments indicated less than adequate range (>15.0 mg/kg) with mean values between as low 2.80 mg/kg to 13.25 mg/kg. As for potassium, all treatments showed higher nutrient concentration than adequate range (0.24 cmol $^{(+)}$/kg) which ranged between 0.27 cmol$^{(+)}$/kg to 0.59 cmol$^{(+)}$/kg.

Table 2: Total mean of nutrient in soil for different types of fertilizer

Treatments	pH (H_2O)	Total N (%)	Available P (mg/kg)	Exch. K(cmol $^{(+)}$ kg^{-1})
Mixture fertilizer	4.60 ± 0.15^a	0.12 ± 0.01^a	8.40 ± 3.20^a	0.56 ± 0.05^a
Chicken manure	4.46 ± 0.03^a	0.14 ± 0.02^a	13.35 ± 6.05^a	0.56 ± 0.12^a
Cow manure	4.37 ± 0.08^a	0.09 ± 0.006^a	2.80 ± 0.70^a	0.59 ± 0.15^a
Empty fruit bunch	4.67 ± 0.16^a	0.15 ± 0.01^a	6.60 ± 0.70^a	0.49 ± 0.04^a
Cocoa pod husk	4.37 ± 0.03^a	0.13 ± 0.01^a	8.80 ± 2.70^a	0.57 ± 0.10^a

For the yield production, the effect of treatments on the yield production for total cumulative dried bean can be seen 21 months after its first application (Figure 1). The highest production was in mix fertilizer (2042.4 kg/ha) followed by oil palm empty fruit bunch (1921.3 kg/ha), chicken manure (1571.1 kg/ha), cow manure (1519.7 kg/ha) and the lowest production, cocoa pod husk (1367.0 kg/ha). These varying results however, gave no significant differentce (p>0.05) between the treatments.



Means with same letter were not statistically different using Tukey's at p>0.05 probability level.
Figure 1: Cumulative yield production for different types of fertilizer

CONCLUSION

The yield consistency of cocoa (*Theobroma cacao* L.) production have helped in the better understanding of the important role of organic materials as fertilizer to improve the crop yield, particularly, in cocoa plantation even under the acidic soil conditions.

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REFERENCES

Allen, V. B. 2010. Science and Technology of Organic Farming. CRC Press, Taylor and Francis Group.

Akanbi, O. S. O, Ojeniyi, S. O, Famaye, A. O, Ipinmoroti, R. R, Oloyede, A. A, 6: Oyewumi, I.K, Ayegbonyin, K, Adejobi, K. B, Idrisu, M, 2014. Soil nutrients and cocoa seedling performance as influenced by plant residue ash and NPK fertilizer addition on a depleted soil in Ibadan, South Western, Nigeria. International Research Journal of Agricultural Science and Soil Science (ISSN: 2251-0044) Vol. 4(1) pp. 1-xxx, January, 2014

Ann, M., Clain, J. and Kathrin, O. R., 2017. Nutrient Management : Soil pH and Organic Matter. Module 8. Montana State University Extension.

Ayeni, L.S., 2009. Integrated application cocoa pod husk ash and poultry manure on soil and nutrient contents and maize yield. Am. Eur. J. Sustainable Agric.., **2**: 92-97.

Sharifuddin, H.A.H. and A.R. Zaharah. 1987. Utilization of natural and agricultural waste products in Malaysian agriculture. Paper presented at the Organic Farming Conference, Khon Kaen, Thailand.

Wong, I. F. T. (1974) (revised) – Soil-crop suitability classification for Peninsular Malaysia,

Soils and Analytical Services Bulletin Nr.1, Ministry of Agriculture, Kuala Lumpur

Yadav A., Gupta R. and Garg V. K., 2013. Organic manure production from cow dung and biogas plant slurry by vermicomposting under field conditions. International Journal of Recycling of Organic Waste in Agriculture 2013, **2**:21.

P5-5

Growth performance and biomass production of Tongkat Ali (*Eurycoma longifolia*) on low acid soil

***K. Wan Rasidah*[1], *Ho, W.M*[1]. *and I. Mohamad Fakhri*[1]**
[1]Forest Research Institute Malaysia (FRIM), 52109 Kepong, Selangor, Malaysia
; howaimun@frim.gov.my ; rasidah@frim.gov.my

INTRODUCTION

The medicinal properties of *Eurycoma longifolia* or locally known as tongkat ali have been known for centuries in traditional remedies. Each part of the plant, especially the roots, is used among others as an afterbirth tonic, reducing fevers, curing mouth ulcers and to treat intestinal worms (Burkill 1993). The roots contain active chemical compounds such as quassinoids, canthin, alkaloids, tirucallane-type triterpenes, eurycolactone, laurycolactone and eurycomalactone (Bhat & Karim 2010). Some quassinoids isolated from *E. longifolia* have been identified to have the potential as anti-tumor promoting and anti-parasitic. Despite all these therapeutic properties, *E. longifolia* is more precious to the public for its aphrodisiac properties. On the retail market, products containing *E. longifolia* particularly pre-mixed drinks are available widely in Malaysia. Increased manufacturing volume has led to the excessive harvesting of this species from its natural habitat. Thus, the Malaysian government had placed *E. longifolia* on the list of protected plants in Malaysia, while in Singapore it is under the critically endangered category on the Red List of threatened plants.

In order to reduce pressure of harvesting from natural forests, it is important to cultivate *E. longifolia* either in plantations or small scale farms for sustainable production of quality raw materials. As such, understanding optimum conditions for the growth of this species is vital. Several unpublished reports claimed that *E. longifolia* prefers well-drained acid and sandy soils at low altitude up to 700 m above sea level, with partial shade and no water deficit. Bhat and Karim (2010) reported that *E. longifolia* grows well in sandy, well-drained soil. This study aims to investigate the growth as well as biomass production of *E. longifolia* on acidic soil with different planting densities.

MATERIALS AND METHODS

The study site is located at Jengka 16 and Jengka 23 in the state of Pahang. These sites were selected as a central region area as part of a larger study. *Eurycoma longifolia* planted in study plot of Jengka 16 has different survival rates while Jengka 23 has a density of 225 trees/plot. Trees in Jengka 16 were planted in October 2005 in the open while those in Jengka 23 were planted in March 2004 under coconut trees. Diameter at breast height (DBH) and height of tree were measured. Selected trees were harvested for determination of biomass.

RESULTS AND DISCUSSION

Soil pH for all studied plots recorded values below pH 5 and is thus considered as acidic soil. At Jengka 16, lower survival rate (56%) provided a condition similar to low density planting which resulted in slightly better growth as observed in DBH and height of *E. longifolia* (Table 1). Besides growth parameters, determination of biomass in *E. longifolia* also revealed lower values in high density planting. Table 2 shows differences in biomass accumulation in roots and leaves, both of which are actively growing parts. This clearly indicates that farming practice of having high density planting may not be suitable for this species probably due to competition for water and nutrients.

Table 1: Growth of *Eurycoma longifolia* with different planting densities at Jengka 16

Plot	Average DBH (cm)	Average Height (m)
High Survival (81%)	5.10	5.82
Low Survival (56%)	5.17	6.03

Table 2: Biomass of *Eurycoma longifolia* with different planting densities at Jengka 16

Plot	Biomass (kg)			
	Leaf	Stem	Root	Fruit
High Survival (81%)	1.03	10.10	2.00	-
Low Survival (56%)	1.43	10.15	2.38	0.40

Table 3: Growth of *Eurycoma longifolia* at Jengka 23

Plot	Average DBH (cm)	Average Height (m)
A	4.88	6.14
B	5.00	6.49
C	5.20	6.51

Table 4: Biomass of *Eurycoma longifolia* at Jengka 23

Plot	Leaf	Stem	Root
Average	1.33	10.35	1.55

On the other hand, the 8-year-old *E. longifolia* in Jengka 23 have slightly lower DBH but better height performance (Table 3). The average DBH and height of trees in Jengka 23 are 5.03 cm and 6.39 m, respectively. The results showed much lower root biomass as compared to those planted in Jengka 16 although biomass of leaves and stem are similar.

CONCLUSION

Through our findings, we conclude that this precious medicinal plant grows well in most types of soil including slightly acidic soil but partial shade may be preferable particularly at the initial years. In addition, planting with high density number of *E. longifolia* is discouraged due to competition for water and nutrients causing slower growth and lower biomass accumulation.

ACKNOWLEDGEMENTS

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REFERENCES

Bhat R & Karim A. A. 2010. Tongkat Ali (*Eurycoma longifolia* Jack): A review on its ethnobotany and pharmacological importance. Fitoterapia **81**: 669-679.

Burkill, I. H. 1993. A dictionary of the economic products of the Malay Peninsula. Publication Unit, Ministry of Agriculture, Malaysia, Kuala Lumpur. Volume 1: 1-1240; Volume **2**: 1241-2444.

P5-6

Biomass production and essential oil yield of *Cymbopogon nardus* applied with different levels of nitrogen fertilizer on two contrasting acid soils

Wan Rasidah K[1]., Nor Azah M. A[1]., Rosazlin A[2]. & Rozita A[1].
[1]Forest Research Institute Malaysia, 52109 Kepong, Selangor
[2]Institute of Biological Sciences, University of Malaya, Kuala Lumpur.

; rashidah@frim.gov.my

INTRODUCTION

Herbal based healthcare products are gaining market popularity prompting demand for higher quantity of quality raw materials. One such plant species is serai wangi or scientifically known as *Cymbopogon nardus*. Methodology for essential oils extraction and product development have been well progressing (Ganjewala 2009, Muhammad Hazwan et al. 2014). Field trials were set up to study the nitrogen demand and effect of urea fertilizer on essential oil yield. Biomass production and essential oil yield of *C. nardus* clumps planted on two contrasting soils, coastal sandy beach and inland sedentary soils were quantified. Nitrogen is known to induce higher leaf biomass thus different rates of N fertilizer input were examined.

MATERIALS AND METHODS

Study sites

The major part of the study was focused on BRIS soil in Setiu, Terengganu. A study was established on inland sedentary soil in Maran was meant for comparison. The two sites were distinguished through contrasting soil properties, sandy and mineral soils. The present experiments were carried out at the FRIM Research Station in Setiu, Terengganu (Jambu series soil) and FRIM Research Station in Maran, Pahang (Bungor series soil). Properties of Jambu Soil Series had been described by Amir et al. (1993) having soil pH range from 3.6 to 4.6 while the Bungor Soil Series (Typic Paleudult) in Maran has pH below 5 (Rozita et al. 2011, 2012).

Plot establishment

Seven treatments including the control were established in Setiu (Table 1) while in Maran, fertilizer rates were the same but no organic mulch applied due to limited clumps availability. Experimental layout was a complete randomized block design with four replicates, having 30 measured clumps for each plot in Setiu and 18 measured clumps in Maran. The first stage foliar sampling of *C. nardus* was carried out three months after fertilizer application. The whole leaves were sampled and their fresh yield recorded, and then sub-sampled for chemical and essential oil analysis (only from Setiu).

Table 1. Details of fertilizer treatments on serai wangi

Treatment	Fertiliser rate	Note
T1	Control	
T2	50 kg/ha urea	P & K added*
T3	75 kg/ha urea	P & K added*
T4	100 kg/ha urea	P & K added*
T5	50 kg/ha urea + EFB mulch mat (initial)	Apply on top of fertiliser
T6	75 kg/ha urea + EFB mulch mat (initial)	Apply on top of fertiliser
T7	100 kg/ha urea + EFB mulch mat (initial)	Apply on top of fertiliser

* P and K rate at equivalent amount as available in mulch mat

Data collection and analysis

Above ground biomass of *C. nardus* clumps were harvested at three months after sowing. The re-growth was sampled again on three-monthly interval, repeated until clumps produced flowers. Biomass data were recorded.

For essential oil extraction, the harvested plant materials were left to dry for two days and were subjected to water distillation in Clevenger-type apparatus for 8 hours. The oily layers obtained were separated and dried over anhydrous sodium sulphate. The yields were averaged over two measurements and calculated based on the dry weight of the plant materials. The citronella oils obtained were also tested for physical properties; specific gravity and refractive index. The chemical compositions of the oils were determined using GC and GC-MS.

RESULTS AND DISCUSSION

The average foliar biomass produced from each treatment is shown in Table 2 for trial in Setiu and Figure 3 for trial in Maran. Urea fertilizer application resulted in significant improvement in foliar biomass yield in both areas. The use of organic mulch in Setiu further improved biomass yield, possibly due to better soil microenvironment and improved efficiency of urea when applied in combination with organic material. The effect subsided through the second sampling as EFB mulch decomposed. Urea fertilizer at 50 kg/ha seemed sufficient to improve biomass yield of serai wangi on sandy soil in Setiu, Terengganu. At the fifth sampling, the clumps have reached unproductive cycle, producing low leaves biomass due to vigorous flowering. Replanting is necessary after the fourth sampling. Throughout the cycle, T4 had the lowest yield on BRIS soil.

Because of better soil properties, the yield obtained in Maran was much higher than Setiu (Figure 1). However, yield patterns are somewhat similar for both sites. Application of 50 kg/ha urea led to significant increase in yield while higher rate of fertilizer gave similar yield compared to lower rate.

Table 2. Foliar biomass yield (kg fresh) of serai wangi planted in Setiu following different fertilizer treatments

Treatment	Sampling					
	1st	2nd	3rd	4th	5th	
	Leaves	Leaves	Leaves	Leaves	Leaves	Flower*
Control	5.18 a	9.15 a	11.86 a	10.58 a	8.48 b	26.1
T2	9.51 b	13.02 b	21.42 b	20.94 c	7.13 b	45.3
T3	8.84 b	13.05 b	22.28 b	22.26 c	9.50 b	43.1
T4	7.66 b	7.50 a	14.69 a	15.92 b	5.14 a	37.3
T5	12.03 c	14.20 b	21.44 b	20.43 c	9.20 b	44.2
T6	13.17 c	15.20 b	20.77 b	19.25 c	8.08 b	54.5
T7	11.39 c	13.37 b	21.32 b	22.16 c	8.00 b	49.2

Note: sampling intervals at every three months; Fertilizers were applied at 6-months intervals; Biomass yield are for 30 measured clumps and average of four plots; * - flower + stalk; Means in columns with the same letter are not significantly different at 5% probability level.

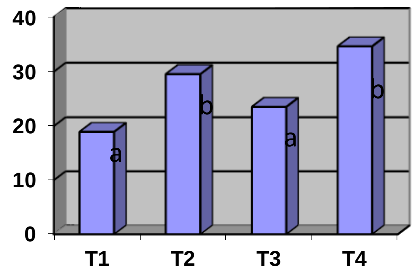

Figure 1. Foliar biomass yield (kg/18 clumps) of serai wangi in Maran

The main chemical composition of the citronella oils is shown in Table 3. Major constituents identified from the oils were geraniol, citronellol and citronellal with varying percentage composition. For non-treated samples (control), the citronella oils comprised of geraniol (57.4-62.4%), citronellol (12.0-14.4%) and citronellal (11.2-13.7%). In the first harvest, samples treated with T6 and T7 showed higher oil yields, thus the percentage composition of the essential oils were compared. Overall, T6 and T7 samples from all R1, R2, and R4 plots showed a decrease in geraniol content and an increase in citronellol content (R3T7, R4T6) and citronellal content (R3T6, R4T7, R2T7, R1T7). Higher citronellol content were also observed in samples R3T2 (21.6%) and R3T4 (29.2%). Major constituents of citronella oils from the second harvest were still made up of geraniol, citronellol and citronellal with varying percentage composition. The presence of *Z*-citral was noticeable and overlapped with retention time of citronellol. Although the chemical profiles of each citronella oils are similar, the influence of granular urea performance on the quality of the oils is still inconclusive at this stage.

Table 3. Major constituents in citronella oils extracted from serai wangi leaves

Chemical compound	Quantity, %
Citronellal	10 - 12
Citronellol (+ z-citral)	13 - 18
Geraniol	53 - 58
Geranyl acetate	3 - 4

Table 4. Essential oil production from serai wangi leaves harvested in Setiu as influence by different urea application rates

Treatment	Essential oil yield*			
	1st sampling		2nd sampling	
	ml/30 clumps	litre/ha	ml/30 clumps	litre/ha
Control	36.3	12.1	64.1	21.4
T2	66.6	22.2	91.1	30.4
T3	61.9	20.6	91.4	30.5
T4	53.6	17.9	52.5	17.5
T5	84.2	28.1	99.4	33.1
T6	92.2	30.7	106.4	35.5
T7	79.7	26.6	93.6	31.2

* - calculated based on average pilot scale production of 7 ml citronella oil for every kg fresh weight leaves

The most important indicator for serai wangi planting is the essential oil production. Even though percentage of oils are highest in the control samples (Tables 3 & 4), the total oil obtained calculated based on biomass produced showed an increase of 83% in the first sampling and 42% in the second sampling due to fertilizer input

(Table 4). Fertilizers were applied at 6-monthly intervals while leaves harvesting were carried out at 3-monthly intervals, thus for second sampling there was no fertilizer cost incurred.

REFERENCES

Amir H.M.S, Suhaimi W.C., Adzmi Y. & Mohd Ghazali H. 1993. Which canopy tier should be sampled to determine the fertility (nutritional) status of *Acacia mangium* on BRIS soils? Journal of Tropical Forest Science 6(1): 48-55.

Ganjewala D. 2009. *Cymbopogon* essential oils: Chemical compositions and bioactivities. International Journal of Essential Oil Therapeutics (2009) **3**, 56-65

Muhammad Hazwan H., Hasfalina C.M., Zurina Z.A. & Hishamuddin J. 2014. Comparison of Citronella oil extraction methods from *Cymbopogon nardus* grass by Ohmic-heated hydro-distillation, hydro-distillation, and steam distillation. Bioresources **9**(1): 256-272.

Rozita A, Wan Rasidah K, Tumirah K, Rosazlin A & Mohamad Fakhri I. 2011. Effects of soil properties on heavy metals content and growth performance of two herbal plants on different soils. Pp 177-185 in Mahmus S. et al. (Eds.) Proceedings SOILS 2011 Conference: Soil Fertility and Plantation Productivity. 19-21 April 2011 Kota Kinabalu Sabah

Rozita A, Wan Rasidah K, Tumirah K, Rosazlin A & Mohamad Fakhri I. 2012. Heavy metals assessment in two medicinal plants grown on different soils. Pp 354-360 in Wan Rasidah K et al. (Eds.) Proceedings SOILS 2012 Conference: Soil quality towards sustainable agriculture production. 10-12 April 2012, Kota Bharu, Kelantan.

P5-7

Effects of site improvement techniques on survival and growth of four selected tree species grown on an ex-landfill in Ara Damansara environmental park

L.K. Tang[1]*, *L.H. Ang[1] and W.M. Ho[1]
[1]Ecophysiology Branch, Forest Plantation Program, Forest Biotechnology Division, Forest Research Institute Malaysia, 52109 Kepong, Selangor Darul Ehsan.

; treefriend2011@gmail.com ; anglh@frim.gov.my

INTRODUCTION

Ara Damansara Environmental Park (ADEP) was established from greening and beautifying Ara Damansara Flood Mitigation Pond (AFMP) (JPZSB, 2009). The AFMP was built using dump materials from household, construction and industrial solid wastes. The soil pH of the landfill ranges from 3.5 to 6.5. The foundation of AFMP was constructed using compaction to pack the solid wastes together with unknown origins of excavated soil materials to form the structure of the detention pond. This presentation aims to highlights the effects of site improvement on the survival and vegetative growth of four selected rainforest tree species grown on an ex-landfill.

MATERIALS AND METHODS

Ordinary big-hole planting point of 1 x 1 x 1 m had 80 to 100% mortality in the park. Hence, only two site improvement techniques were tested on four rainforest tree species, namely Site Improvement Techniques SI (0.7 m) and SI (2 m). SI (0.7 m) included loosening the site to 0.5 m depth and large planting hole of 0.7 m depth x 0.5 m radius and applied 3 kg soil conditioner comprising a mixed coco-peat and charcoal (4:1) at each planting point. Whereas, SI (2 m)+organic included loosening the site to the depth of 1.5-2.0 m and followed by an application of burnt-rice husk and coconut peat of ratio 1:1 at 10 kg m^{-2}. An addition of organic enriched mineral soils at 0.6m^3 m^{-2} at site where household waste plastic and polystyrene materials occupied > 30% surface of the soil pit.

RESULTS AND DISCUSSION

The survival of the four tree species treated with SI (0.7 m) had survival of 60 to 86.7% at 40 months after planting. Whereas, the four tree species treated with SI (2 m) had survival of 82.4 to 100%. SI (2 m) treatment had higher survival than SI (0.7 m) for all the four rainforest species. All the four tree species grown treated with SI (2 m) had significantly better growths than SI (0.7 m) treated trees (Table 1).

Table 1: Comparison of survival and vegetative growth parameters of four selected rainforest tree species from NA, VU and DD categories of IUCN Red-List classification grown in ADEP at 40 months after planting. MH, MAIH, RGRH, MD, MAID, RGRD and LCR denotes mean top height, Mean annual increment of top height, Relative growth rate of height, Mean diameter at breast height (dbh), Mean annual increment of dbh (MAID), Relative growth rate of mean dbh and live-crown ratio, respectively.

Species	Survival (%)	MH (m)	MAIH (m/y)	RGRH (m/m/y)	MD (cm/y)	MAID (cm/y)	RGRD (cm/cm/y)	LCR (%)
Milletia atropurpurea (NA) - SI (0.7m)	77.7	3.79a (1.05)	1.13a (0.32)	0.12a (0.15)	7.6a (2.5)	2.28a (0.76)	0.426a (0.165)	43.4 (15.9)
Milletia atropurpurea (NA) -SI (2m)+organic	88	5.9b (0.8)	1.78b (0.26)	0.31b (0.07)	11.98b (2.3)	3.58a (0.69)	0.61b (0.07)	58.5 (10.7)
Shorea macrophylla (VU) -SI (0.7m)	60	1.8a (0.1)	0.53a (0.03)	0.017a (0.001)	2.8a (0.9)	0.84a (0.28)	0.098a (0.103)	62.2 (8.8)
Shorea macrophylla (NA) -SI (2m)+organic	87.5	4.12b (0.75)	1.23b (0.22)	0.28b (0.06)	5.56 b (1.19)	1.66a (0.35)	0.33b (0.10)	62.0 (8.6)
Alstonia angustiloba (NA) -SI (0.7m)	86.9	4.08a (0.89)	1.20a (0.27)	0.15a (0.08)	16.6a (7.0)	4.98a (2.11)	0.599a (0.188)c	59.1 (9.3)
Alstonia angustiloba (NA) –SI (2m)+organic	100	4.91a (1.1)	1.5a (0.32)	0.17a (0.05)	15.7a (4.4)	4.72a (1.33)	0.54a (0.12)	73.4 (14.2)
Ochanostachys amentacea (DD) -SI (0.7m)	76.9	2.88a (0.84)	0.86a (0.25)	0.14a (0.10)	3.6a (1.2)	1.08a (0.35)	0.284a (0.138)	72.9 (9.9)
Ochanostachys amentacea (DD) -SI (2m)+organic)	82.4	3.78b (0.77)	1.13b (0.23)	0.19b (0.77)	4.9b (1.31)	1.48b (0.39)	0.338b (0.134)	70.5 (7.7)

CONCLUSION

The trees species grown at site treated with a loosening of soils to 2 m depth and treated with organic materials and organic mineral soils have higher survival and growth compared to the other treatment.

REFERENCES

Jurutera Perunding Zaaba Sdn Bhd (JPZSB). 2009. Final Report Vol 1&2. Executive Summary and Main Report. 91p.

P5-8

Survival of ten endemic, endangered and threatened tree species grown on slime tailings at six months after planting

L.H. Ang[1], W.M. Ho[1] and L.K. Tang[1]
[1]Forest Research Institute Malaysia (FRIM), 52109 Kepong, Selangor, Malaysia
; anglh@frim.gov.my ; treefriend2011@gmail.com

INTRODUCTION

Asian Forest Cooperation Project (AFoCo) has funded a collaborative project between Malaysia-Thailand on domestication of endangered, endemic and threatened tree species (EETs) on degraded terrestrial ecosystem. The project planted some EETs classified by national and IUCN Red List (Chen, 2004; Chua *et al*. 2010). This paper aims to share the plot establishment experiences including site amelioration technique, planting and tending experiences. In addition, early survival of the mixed stand at six months after planting is also reported. The objective of this study is to share establishment technique and early survival of the planting of 10 selected tree species grown on slime tailings.

MATERIALS AND METHODS

Study site, site preparation and planting stock

The study site is located in Tin Tailings Afforestation Centre, SPF Bidor, Perak. The slime tailings had pH range of 4.5–6.5. It has 95–99% of composition of silt and slime. Each planting hole was spaced at 5 x 4 m and enriched soils with a mixture soil conditioner comprising of coconut-peat with burnt-rice husk (biochar) at a ratio of 1:2 or 5 kg coconut-peat with 10 kg burnt-rice husk. The planting stock of the ten species and their conservation value, size and quantity are listed in Table 1.

Table 1: Planting stock of the 10 selected tree species

Species	Conservation Value	Size	Quantity
Aquilaria malaccensis	*Vulnerable	Sapling	300
Dipterocarpus chartaceus	#Lowland timber species	Seedling	140
Dryobalanops oblongifolia	#Lowland timber species	Sapling/seedling	50
Hopea ferruginea	*Critically Endangered	Seedling	56
Hopea helferi	*Critically Endangered	Seedling	179
Lagerstroemia langkawiensis	*Endangered	Sapling	301
Neobalanocarpus heimii	*Vulnerable	Sapling/seedling	164
Palaquium maingayi	#*Lower Risk	Sapling/seedling	60
Shorea glauca	*Endangered	Seedling	200
Shorea sumatrana	*Critically Endangered	Sapling/seedling	50
Total			1,500

# denotes criteria classified as threatened timber species from lowland forest (Chen 2004),
* denotes classification using IUCN Red List species version 2.3

Tending regime

Watering for the planting was carried out immediately and continued daily till heavy rainfall, then, the planting was watered at two dry-day interval. The soil of 50 cm radius of the planting point was loosened, climbers were removed monthly. Fertilizer application is carried out quarterly. The application of a mixture of 60g compound fertilizer NPK (17:17:17) + trace elements with chicken manure at 80% maturity was carried out in June, September and December 2017.

RESULTS AND DISCUSSION

The survival count at six months after planting shows that only *Lagerstroemia langkawiensis* has not suffered from any mortality even subjected to the dry period of middle of May till early July 2017. *Hopea ferruginea* suffered the most during the dry period (Table 2). The mortality of the saplings and seedlings of EETs are mainly due to drought and heat stresses during middle of May till middle of June 2017, even though watering was being carried out but some of them still could not survive the heat load and dryness of the air which create high ambient vapour pressure demand (VPD). High VPD would increases the loss of water during photosynthesis and eventually drying up the plants if there is no leaf shedding. *Lagestroemia langkawiensis* and *Aquilaria malaccensis* shed their leaves at the onset of drought and heat stresses, and they survived through the dry period. Low survival of *Hopea ferruginea* was properly due to root damage caused by wild pigs.

Table 2: The survival count and percentage of the 10 tree species at six months after planting

Species	**Quantity**	**Apr**	**May**	**Jun**	**Jul**	**Aug**	**Sept**	**Oct**	**Oct (%)**
Aquilaria malaccensis	300	300	299	268	268	265	260	260	86.7
Dipterocarpus chartaceus	140	140	140	138	138	138	138	137	97.8
Dryobalanops oblongifolia	50	50	50	50	49	49	49	49	98.0
Hopea ferruginea	56	56	36	26	24	24	24	24	42.8
Hopea helferi	179	179	179	179	177	177	176	174	97.2
Lagerstroemia langkawiensis	301	301	301	301	301	301	301	301	100.0
Neobalanocarpus heimii	164	164	164	154	154	154	154	154	93.9
Palaquium maingayi	60	60	60	59	59	59	59	59	98.3
Shorea glauca	200	200	191	190	190	190	190	190	95.0
Shorea sumatrana	50	50	49	49	47	47	47	47	94.0

CONCLUSION

The preliminary survival of the EETs shows that *Lagerstroemia langkawiensis* is more adaptable to the site condition of the slime tailings and had 100% survival through the mid-year short dry period in Bidor.

REFERENCES

Chen Hin Keong. 2004. A Malaysian assessment of a world list of threatened trees. Traffic Southeast Asia. 59p.

Chua L.S.L, M. Suhaida, M. Hamidah & L.G. Saw. 2010. Research Pamphlet No. 129. Malaysia Red List. Peninsular Malaysia Dipterocarpaceae. 210p.

IUCN Red List version 2.3. http://www.iucnredlist.org/search.

P5-9

Assessment of two selected biofertilizers on soil pH, organic carbon and nitrogen of a paddy field

Norziana, Z.Z[1].**, **Emmyrafedziawati Aida, K.R[1]., Illani Zuraihah,I[1]. and Theeba,M[1].
[1]Crop and Soil Science Research Centre, Malaysian Agricultural Research and Development Institute (MARDI), MARDI Headquaters Serdang, Malaysia
; zianazz@mardi.gov.my

INTRODUCTION

Through intensive agriculture, which involves various high yields of rice and other crops, that has caused in discriminate high usage of chemical fertilizer , has led to the loss of nutrients from the soil; fertility imbalance and resulted soil health deterioration (John et al., 2001). Recently, environmentally-friendly bio-fertilizer application is used as an alternative to minimize the impact of continuous usage of chemical fertilizers to the soil and the environment. The use of biofertilizers is believed to sustain soil fertility and sustainability over the long term and subsequently produce and higher yield crops. The objective of this study was to assess the application of the two selected commercial biofertilizers on changes in soil chemical properties such as pH, soil organic carbon and nitrogen in paddy fields.

MATERIALS AND METHODS

Field experiment was conducted in a paddy rice field in MARDI Seberang Prai, Penang Malaysia, for two cropping seasons (off season 2016 and main season 2016/2017). The study was conducted in research plots size 5x5 m for each treatment and was arranged in randomized complete block design with five replicates. Management of field for cultivation of rice is based on normal activity practiced by farmers. Five treatments used in this study were :T1 : Subsidy fertilizer only ; T2 : Subsidy fertilizer + Biofertilizer A ; T3:Subsidy fertilizer + Biofertilizer B; T4 : Subsidy fertilizer + Biofertilizer A + Biofertilizer B ; T5: Biofertilizer A + Biofertilizer B. Soil sampling (0-20 cm) was done before planting of the first season crop and at the end of planting for two seasons. The samples were taken to the laboratory and ground to size 2mm before they were analyzed for nutrient content and soil chemical properties. Analysis of N content was determined by the Kjeldahl method (Bremner and Mulvaney, 1982). Meanwhile, the organic carbon was determined by Walkley and Black using the titration method (Nelson and Sommers, 1982). Soil parameters data were subjected to analysis of variance and means were compared with Duncan Multiple Range Test using the SAS statistical package.

RESULTS AND DISCUSSION

The effects of Biofertilizer A, Biofertilizer B, and combination of both Biofertilizer A and Biofertilizer B either applied with subsidy or without subsidy fertilizers on soil properties are shown in Figure 1 to 3. Results in Figure 1 show that there was improvement of pH in Biofertilizer A with combination of Biofertilizer B + subsidy fertilizer plot (T4) at the end of planting of season two compared to others treatment. It is also shown that plots that were applied with Biofertilizer A have improved the soil acidity at the end of planting of season two when compared to before planting (initial). T4 plots increased pH from 4.73 at initial to pH 5.14 at the end of planting season two. The application of Biofertilizer A, Biofertilizer B, and combination of both Biofertilizer A and Biofertilizer B either applied with subsidy or without subsidy fertilizers have remarkably improved total nitrogen and organic carbon content in the soil (Figure 2 and 3). Specifically, T4 and T5 treatments resulted in the highest of total nitrogen content (0.17%) compared to T1 treatment at the end of season two. However, the additional application of Biofertilizer A, Biofertilizer B or combination of both Biofertilizer A and Biofertilizer B, either applied with subsidy or without subsidy fertilizers on soil total N resulted in no significant difference at the end of season 2 (Figure 3) . Results show that T4 gives the highest soil organic carbon content at the end of season two compared to other treatments. However, results in Figure 4 and Figure resulted in no significant difference among treatments on soil organic carbon at the end of two planting season. In this trial, total nitrogen and soil organic carbon contents increased considerably in all the fertilization treatments suggesting that the fertilizers are beneficial towards the accumulation of soil organic matter and thus improve soil fertility.

CONCLUSION

In conclusion, the application of Biofertilizer A, Biofertilizer B and combination of both Biofertilizer A and Biofertilizer B either applied with subsidy or without subsidy fertilizers resulted in a slight increase of soil pH, total nitrogen and soil organic carbon contents. However, there are no significant differences among different fertilization treatments in soil fertility of the study plots. It is found that total nitrogen and soil organic carbon will increase with time in all treatments, suggesting the effects of the biofertilizers application do increase nutrients content in the soil but the effectiveness might be seen after long-term study of the biofertilizers.

ACKNOWLEDGEMENT

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REFERENCES

Bremner, J.M. and Mulvaney, C.S. (1982). Methods of soil analysis, Part 2: Chemical and Microbiological properties, 595-624

John P. S., M. George and R. Jacob, 2001. Nutrient mining in agro– climatic zones of Kerala. Fertilizer News, 46: pp. 45-52 & pp. 55-57

Nelson, D.W. and Somners, L.E. (1982). Total carbon, OC and organic matter. In: *Methods of Soil Analysis, Part 2. Chemical and Microbiological Properties. Agronomic Monograph*, vol.9, Second Edition, (Page, A.L., Miller, R.H.and Keeney, D.R., eds.), p. 539-577. Madison, WI: Agronomy Society of America and Soil Science Society of America

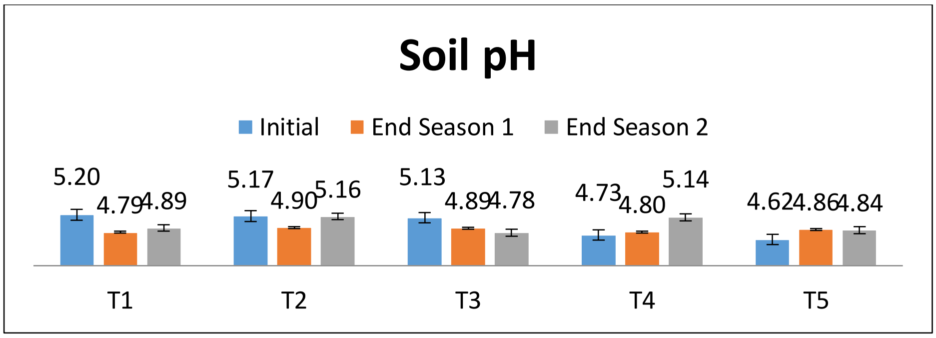

Figure 1: Effects of different biofertilizers application on soil pH before and after planting in season one and season two (*Vertical line represent standard error bar, n=5)*

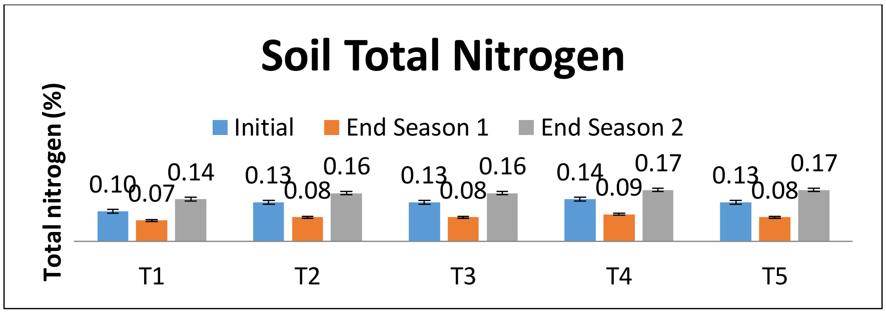

Figure 2: Percentage of soil total nitrogen before and after planting in season one and season two after different biofertilizers application (*Vertical line represent standard error bar, n=5)*

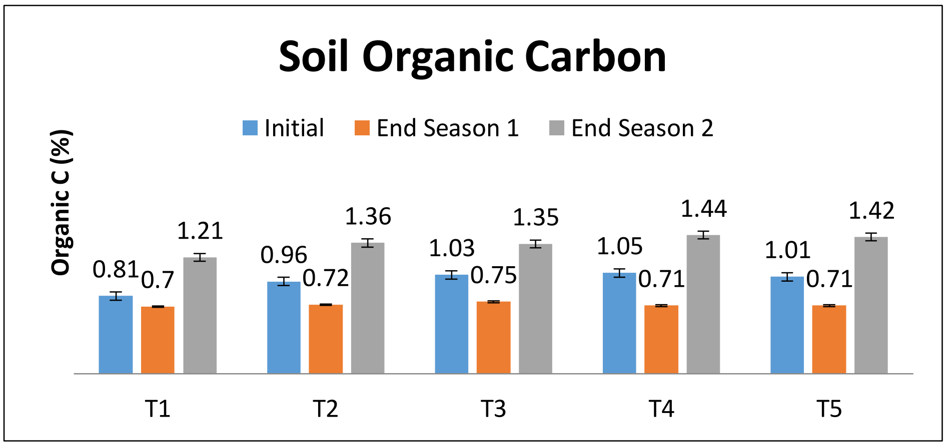

Figure 3: Percentage of soil organic carbon before and after planting in season one and season two after different biofertilizers application (*Vertical line represent standard error bar, n=5)*

P5-10

Adsorption-desorption of aminomethylphosphonic acid (AMPA) in Alfisols amended with cow dung and rice husk ash

Jamilu Garba[1,2], Samsuri Abd Wahid[2], Radziah Othman[2], Muhammad Saiful Ahmad Hamdani[3]
[1]Department of Agricultural Education, College of Education, 1002 Maru, Zamfara, Nigeria
[2]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia,
[3]Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia
; samsuriaw@upm.edu.my

INTRODUCTION

Soil pollution is an important environmental issue affecting ecosystem, as its consequences is associated to soil degradation, surface and ground water contamination. Soil contaminated with pesticide is of great environmental concern. Glyphosate is the common organophosphate herbicide widely used for weed control in both agricultural and non-agricultural land. Aminomethylphosphonic acid (AMPA) is a metabolite of glyphosate microbial degradation in soils. Therefore, substantial use of glyphosate leads to accumulation of AMPA in the soil environment. Recent studies revealed that AMPA is toxic to humans and animal cells (Alexis et al. 2017). Explaining the behaviour of AMPA requires an assessment of the processes influencing its fate, transport, sorption, desorption, degradation and persistence in soils. On the other hand, organic wastes are continuously applied to soils to increase fertility and productivity but less attention was given on their alternate use for soil decontamination study. The objective of this research was investigates the adsorption-desorption of AMPA by Alfisols amended with cow dung or rice husk ash.

MATERIALS AND METHODS

The soil used in this study was from Benta series, an Alfisols collected from Sementa Hulu (Lat. 3.841663 ^{0}N, Long. 101.947251 ^{0}E), Raub district, Pahang Malaysia. Aminomethylphosphonic acid (AMPA) of 99% purity was purchased from Sigma Aldrich® (Seelze, Germany). All the other chemicals used were of analytical reagent grade. One gram of soil was mixed with 0.1 g of either cow dung or rice husk ash in a centrifuge tube and incubated for 1 week at field capacity. At the end of the incubation period, the mixture was allowed to dry to a constant weight and then used for the sorption study. The sorption study was performed by weighing 1 g of sample into a centrifuge tube and then added with 20 mL solution of different concentration of AMPA prepared in 0.01 M $CaCl_2$ containing 200 mg L^{-1} of $HgCl_2$ as bioinhibitor. The initial concentrations of AMPA were 0, 4, 8, 17, 25, 33, 42, 50 mg L^{-1}. The centrifuge tubes were shaken for 24 h on a rotary shaker at 100 rpm at room temperature and then centrifuged at 10,000 rpm for 10 minutes. The supernatants were later decanted and passed through 0.45 µm HmbG syringe filter model P0377 prior to analysis. Desorption study was performed immediately after the supernatants in the adsorption study were decanted. Into each of the centrifuge tubes, 20 mL solution containing only 0.01M $CaCl_2$ and 200 mg L^{-1} $HgCl_2$ were added to each centrifuge tube and managed similar to the adsorption study. The analysis of AMPA was achieved using HPLC-FLD method earlier developed (Garba et al. 2018). The adsorptive removal of ions from the adsorbents were calculated while adsorption isotherm data was fitted to Freundlich and

Langmuir models. Similarly, the separation factor (R) was calculated from the constant of Langmuir isotherm model.

RESULTS AND DISCUSSION

Adsorptive removal of AMPA was high in soil amended with cow dung (67.28%) followed by soil amended with rice husk ash (62.41%) then control soil (62.27%). Therefore, when compared with the natural soil, application of cow dung or rice husk ash resulted in 5.00% and 0.14% increase respectively in AMPA removal from the aqueous solution. Equally, application of cow dung had 4.87% increase in adsorptive removal compared to rice husk ash. The adsorption isotherm of AMPA was S-type for both control and soils amended with cow dung or rice husk ash. This indicates high affinity of AMPA for the adsorption surfaces on the soil at low concentration but this affinity decreased as concentration increases. The experimental isotherm data of AMPA for both control and amended soils best fitted to the Freundlich model ($r^2 \geq 0.701$). This indicates the existence of heterogeneous surface coverage. The Freundlich's constant (K_F) for AMPA was highest in the soil amended with rice husk ash (7.27 mg g^{-1}) indicating its high affinity and maximum adsorption by this soil added with rice husk ash. This followed by the cow dung amended soil (5.69 mg g^{-1}) and then control soil (4.91 mg g^{-1}). The 1/n value for AMPA in both soils was >1 indicating cooperative adsorption. As a result, the adsorption first occurred at high energy site then followed by low energy site (Yu & Zhou, 2005). The Langmuir separation factor (R) indicated that the adsorption of AMPA by the adsorbents was favourable (Guo et al., 2009).

The results of desorption study revealed that, addition of cow dung and rice husk ash increased desorption of AMPA from this soil by 4.22% and 1.78% respectively, compared to the control soil. This was attributed to an increase in pH due to application of either cow dung or rice husk ash which resulted in more net negative charges (Sidoli et al. 2016) and repulsion between soil surfaces and AMPA leading to greater desorption compared with control.

CONCLUSION

Addition of organic wastes such as cow dung and rice husk ash may increase the mobility of AMPA in soils making it more susceptible to microbial degradation. If the soils have insignificant microbial populations, the addition of organic wastes to the soils may results in ground water contamination of AMPA.

REFERENCE

Alexis Grandcoin, Stephanie Piel, E. B. (2017). AminoMethylPhosphonic acid (AMPA) in natural waters : Its sources , behavior and environmental fate. *Water Research*, *117*.

Garba, J., Samsuri A.W., Othman R., and Ahmad Hamdani, M. S. (2018). Simplified method for derivatization of extractable glyphosate and aminomethylphosphonic acid and their determination by high performance liquid chromatography. *Environmental Research & Technology*, ***1***(2), 19–30.

Guo, S., Li, W., Zhang, L., Peng, J., Xia, H., and Zhang, S. (2009). Kinetics and equilibrium adsorption study of lead(II) onto the low cost adsorbent-Eupatorium adenophorum spreng. *Process Safety and Environmental Protection*, ***87*(**5), 343–351.

S**idoli, P., Baran, N., and Angulo-Jaramillo, R. (2016).** Glyphosate and AMPA adsorption in soils: laboratory experiments and pedotransfer rules. *Environmental Science and Pollution Research*, ***23*(**6), 5733–5742.

Yu, Y., and Zhou, Q. X. (2005). Adsorption characteristics of pesticides methamidophos and glyphosate by two soils. *Chemosphere*, ***58***(6), 811–816.

P5-11

Effect of Vermicompost on the Growth Performance and yield of Kaempferia Galanga. L

***Halijah, I[1]*, Rosazlin, A[1]., Noor, Z.M[1]. and Norsyuhada, J[1].**
[1]Institute of Biological Sciences, Faculty of Science, University of Malaya 50603 Kuala Lumpur, Malaysia
; rosazlin@um.edu.my

INTRODUCTION

Vermicompost is well recognized as one of the organic approaches that contribute to increment of crop's yield, improving soil structure and soil fertility. Its properties of high nutrient contents in plant-available forms such as nitrates, phosphates, and exchangeable calcium and soluble potassium, lead to the less usage of chemical fertilizer and application of pesticides (Orozco et al., 1996; Vasnie et al., 2018). Previous studies (e.g. Ansari & Kumar, 2010 and Joshi et al., 2015) suggested the application of vermicompost improved the quality of crops as well as developed high crop yield, number of leaf/fruits, leaf area, chlorophyll content, root length and nutrients content among others. In cognizance, the aim of this study is to evaluate and subsequently to compare the growth performance and yield of *K. galanga* grown in with and without vermicompost.

MATERIALS AND METHODS

The study was carried out at the selected site (3°03'12.8"N, 102°03'50.4"E) located in Pusat Penyelidikan Bioteknologi Glami Lemi (PPBGL), UM, Jelebu, Negeri Sembilan. A total of 160 rhizomes of *K. galanga* were planted in polybags (length 50 cm, height 40 cm, one rhizome per bag) filled with peat moss and sand mix. The polybags were then divided into two parts with each part represented a treatment. Two treatments were used: (1) NPK fertilizer; and (2) Vermicompost fertilizer. Width (cm), length (cm), leaf surface area (cm^2), shoot number and leaf number were evaluated once a week for four weeks during plant growth.

Preparation of vermicompost fertilizer was conducted according to a method from previous study (see Adi and Noor, 2008) with ratio 80:20 of goat manure (GM):spent mushroom compost (SMC). 8 kg of GM and 2 kg of SMC were mixed together with water to maintain the moisture content on wet basis in the range 60% to 70% and added into a plastic container with size of 45 cm x 34 cm x 27 cm. 21 days of pre-composting period required to make sure the substrates stable in terms of pH, temperature and moisture before vermicomposting. This is followed by introduction of 220 g of earthworms from *Lumbricus rubellus* species into the 10 kg substrates. Subsequently, 70 days of vermicomposting process was taking place in order to produce good quality of vermicompost fertilizer.

RESULTS AND DISCUSSION

Table 1 indicates the growth performance of *K. galanga* planted in soil treated with chemical fertilizer and soil treated with vermicompost. Soil treated with vermicompost significantly increased leaf width (cm), leaf length (cm) and leaf surface area (cm^2) over 4 weeks planting, compared to soil treated with chemical fertilizer. The highest reading of leaf width (cm) of *K. galanga* is 8.15 cm, represented by vermicompost treatment on week 4 while chemical fertilizer treatment towards *K.*

galanga on week 4 is 4.98 cm, slightly lower than vermicompost treatment. The same trend shown by vermicompost treatment on leaf length (cm) and leaf surface area (cm^2) which both has the highest reading on week 4 (12.38 cm and 72.48 cm^2) respectively, compared to chemical fertilizer treatment.

Significantly higher growth parameters were observed when *K. galanga* were grown in vermicompost-based media compared to NPK-based substrate. In addition, vermicomposted *K. galanga* also yielded a comparatively higher number of leaf and shoots as shown in Table 2.

The number of leaf and shoot of *K. galanga* were observed for 4 weeks, from the day after planting (DAP). For soil treated with vermicompost, the number of leaf ranged from 3 to 8 while for soil treated with chemical fertilizer, the number of leaf ranged from 1 to 4. The same trend also shown by number of shoot of *K. galanga* when soil treated with vermicompost results between ranges 5 to 12 while soil treated with chemical fertilizer ranges 1 to 8. The plants with soil treated with vermicompost were also significantly higher in leaf and shoot number compared to chemical fertilizer treatment.

Table 1: Comparison of growth performance of *K. galanga* in each treatment (1)

Treatment	Leaf Width (cm)				Leaf Length (cm)				Leaf Surface Area (cm^2)			
	W1	W2	W3	W4	W1	W2	W3	W4	W1	W2	W3	W4
Vermicompost	3.30	5.64	6.32	8.15	5.11	9.10	9.56	12.38	13.82	38.19	44.27	72.48
NPK Fertiliser	1.26	2.76	3.10	4.98	1.97	4.88	5.32	8.69	2.51	10.20	12.78	33.69

Table 2: Comparison of yield of *K. galanga* in each treatment (2)

Treatment	Number of Leaf						Number of Shoot					
	W1	W2	W3	W4	Average	SD	W1	W2	W3	W4	Average	SD
Vermi-compost	3.30	5.64	6.32	8.15	12.93	8.58	5.11	9.10	9.56	12.38	2.68	1.53
NPK Fertiliser	1.26	2.76	3.10	4.98	5.01	4.24	1.97	4.88	5.32	8.69	2.28	1.29

The data recorded was in accordance to previous literatures that suggest vermicomposting has potential for improving plant growth significantly. From the experiment conducted, amendment of soil with combination of GM, SMC, and *L. rubellus* produced higher growth percentage of *K. galanga* at 97.5% compared to those treated with NPK fertiliser at 37.5%.

CONCLUSION

In the present study, the combination of goat manure and spent mushroom compost as vermicompost was applied to *K. galanga* to evaluate its effect on the plant growth and productivity. Higher plant growth and yield were observed in *K. galanga* that received vermicompost as nutrient supplier. This indicated that vermicomposting is beneficial in supporting the growth of *K. galanga* and its potential for future sustainable production of such plant should be considered.

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REFERENCES

Adi, A. J., Noor, Z.M., & Noorlidah, A. 2012. Waste recycling: Feasibility of saw dust based spent mushroom substrate and goat manure in vermicomposting. Sains Malaysiana **41**(11): 1445-1450.

Ansari, A. A., Kumar, S. 2010. Effect of vermiwash and vermicompost on soil parameters and productivity of okra (*Abelmoschus esculentus*) in Guyana. Afr J Agric Res 5(14):1794–1798

Joshi, R., Singh, J. & Vig, A.P. 2015. Rev Environ Sci Biotechnol **14**: 137. https://doi.org/10.1007/s11157-014-9347-1

Orozco, F. H., Cegarra. J., Trujillo, L. M., and Roig, A. (1996). Vermicomposting of coffee pulp using the earthworm Eisenia fetida: Effects on C and N contents and the availability of nutrients. Biology and Fertility of Soils **22**, 162-166

Vasnie, G., Sirpaul, J., Abdullah, A. A., & Oudho, H. 2018. The effect of vermicompost and other fertilizers on the growth and productivity of pepper plants in Guyana. In Hussman, S. (Ed.), Automation in Agriculture (167-187). https://doi.org/10.5772/intechopen.73262

P6-Agroforestry, Environment, Marginal Acidic Soil Soil Evaluation Method DevelopmentmEnvironmental Monitoring of Low pH Soil

P6-1

Amending bris soil using rare earth by-product: effect on soil properties and environment

Faridah, M[1]., ***Wan Abdullah, W.Y[1]., Illani, Z.I[1]., Theeba, M1., Noorsuhaila, A.B[1]., Sahibin, A.R[1]., Wan Razi, I[1]., and Aznan, F.I[2]***
[1]Plant and Soil Science Research Centre, Malaysian Agricultural Research and Development Institute (MARDI), 43400 Serdang, Selangor, Malaysia
[2]Faculty of Science and Technology, University Kebangsaan Malaysia, 43600 Bangi, Selangor, Malaysia
; faridah@mardi.gov.my

INTRODUCTION

The BRIS (Beach Ridges Interspersed with Swales) soil is defined as one of the problematic soils in Malaysia. The major constraints of BRIS soils are high nutrient leaching, low CEC of 0.61-2.58 meq/100 g with pH 4.3-4.4, perched water table, low nutrient status and poor moisture retention capacity. This BRIS soils spread along the east coast of the Peninsular Malaysia and the coastal area of Sabah. They cover an area of 155,400 ha in Peninsular Malaysia and 40,400 ha in Sabah.

A new material produced from by-products of the rare-earth extraction which is rich in phosphorus (P), calcium (Ca), magnesium (Mg) and iron (Fe) mixed with organic filler was used as soil conditioner to improve the properties of BRIS soil. This project also emphasises on the product's safety for agriculture and environmental uses in terms of heavy metals and radioactive contents.

MATERIALS AND METHODS

This experiment was conducted on BRIS soil (Baging series) at the MARDI station, Cherating, Pahang using sweet corn as a test crop. Three treatments involved; NPK without soil conditioner as control (T1), rare earth by-product soil conditioner + NPK (T2) and ground magnesium limestone (GML) as current practice (T3). Soil conditioner and GML was applied at 10 t/ha as basal while fertilizer (NPK) was applied at 2-weeks interval throughout the cropping season. Harvesting was at 75 days after sowing. Routine agronomic practices for pest control were followed. Fresh yield was recorded at harvest. All data were statistically analysed using ANOVA. Treatment means were compared using Tukey test at $p < 0.05$ with SAS version 9.4.

Heavy metals analysis

The soil was dried at room temperature, pounded and sieved to pass a 2-mm sieve. For heavy metals determination, the soil was pulverized to pass 63-micron sieves. Plant samples were cleaned in running water and rinsed with deionized water at least three times before cut into small pieces, dried and ground using agate mortar. Heavy metals content in samples (e.g. As, Cd, Pb, Cr, Ag, Ba, and Hg) were measured using the ICP-MS Perkin Elmer Elan 9000 Model.

Nuclear analysis for determination of radionuclide uptakes by plant

The transfer of ^{226}Ra, ^{228}Ra,^{238}U,^{232}Th and ^{40}K from soil to plant was determined using a parameter known as the transfer factor (TF) as follows:-

$$TF = \frac{C_P}{C_S}$$

Where C_p and C_s are the concentrations of radionuclides of interest in plant and soil (Bq kg^{-1}) (IAEA, 2010). The transfer factor is used to describe the soil-to-plant transfer of radionuclide through the plant roots.

RESULTS AND DISCUSSION

Effect of byproduct on soil propertieson soil properties

In BRIS soil, the pH, capacity of soil to retain essential nutrients (CEC) and nutrient content are the dominant limiting factors for its soil health. The changes in soil properties are summarised as the overall amount of soil nutrients increased from the initial state after the treatments application. Carbon (C) content from the soil conditioner had highly influenced the amount of soil organic matter (SOM). Howaver soil bulk density was not significantly different for the treatment (Table 1). Soil available-P (>30 mg/kg) after treatments were higher compared to its initial value (1.07 mg/kg). The soil pH and cation exchange capacity (CEC) which is an important indicator of soil fertility were also found to have increase after the application of soil conditioner from rare earth by-product compared to the untreated soil (Table 2). For microbial population studies, it showed that bacteria make up the most abundant group of microorganisms in the soil (3.0 x 10 ^{6} – 5.0 x 10 ^{8} per gram of soil), followed by actinomycetes (1.0 x 10 ^{6} – 2.0 x 10 ^{7}), fungi (5.0 x 10 ^{3} – 9.0 x 10 ^{6}), yeast (I.0 x 10 ^{3} – 1.0 x 10 ^{6}), algae and protozoa (1.0 x 10 ^{3} - 5.0 x 10 ^{5}) and nematodes (50 – 200 counts per gram of soil).

Table 1: Soil bulk density of the sweet corn plot during initial (before treatment) and after treatment application

Treatment	Bulk density (g/cm^3)	
	Depth (0-15) cm	**Depth (15-30) cm**
Initial	1.43	1.47
T1	1.29	1.46
T2	1.30	1.46
T3	1.34	1.48

Table 2: Soil chemical status of sweet corn plot after 75 days of treatment application (at harvest)

Treat-ment	pH	CEC*	C	N	Avail-P	K	Na	Ca	Mg	Fe	Al
		(Cmol/kg soil)	**-----(%)-----**		**--------------------------(mg/kg)-----------------------------------**						
Initial	4.77	0.85	0.13	0.03	1.07	0.05	0.36	9.20	1.93	0.02	0.02
T1	6.90a	1.38a	0.11b	0.03a	36.46a	0.05b	0.42a	16.57b	2.30c	0.02a	0.01b
T2	6.92a	1.79a	0.20a	0.03a	34.54a	0.29a	0.56a	28.56a	5.69a	0.02a	0.02a
T3	6.99a	1.28b	0.22a	0.03a	28.47a	0.22a	0.46a	19.15a	3.20b	0.01b	0.02a

Means with the same letter in the column are not significantly different by Tukey's test at $p \geq 0.05$;
*Note: CEC = Cation Exchange Capacity

In term of yield, the application of soil conditioner from rare earth by-product (T2) produced 7.5 t/ha of sweet corn which was as good as farmer's practice of using GML (T3) with 8.0 t/ha, while control (T1) with 6.2 t/ha.

Environmental monitoring

Heavy metals

Heavy metals content in soil and plant (corn grain) treated with T2 were comparable with the control (T1) and were not significant different ($p>0.05$) for certain elements (Table 3). From the baseline data, there is no indication of heavy metals accumulation in soils and plants after the treatments This show that the use of rare earth by-product as soil conditioner in BRIS soil is safe due to no accumulation of heavy metals and the contents assessed to be below or within the standard target value in soil and corn grain.

Table 3: Properties of soil and plant (corn grain) heavy metals after treatment application

Heavy metals (mg/kg)	Baseline soil	T1	T2	Allowable Value[1] (mg/kg)	T1	T2	Food Regulations[2] (mg/kg)
	---------- Soil ----------				---------- Plant ----------		
As	0.32	0.63a	0.18b	29.0	0.06a	0.02b	2.0
Cd	0.02	0.01b	0.08a	0.8	0.08a	0.04b	1.0
Ce	108.52	7.55a	6.67a	nd	0.20b	10.69a	nd
Pb	18.39	1.70b	3.38a	50	0.36a	0.17b	2.0
Cr	83.75	2.88a	2.60a	100	0.39a	0.10b	40
Ag	0.14	0.21b	0.36a	nd	0.09b	0.63a	nd
Hg	0.09	0.03a	0.02a	0.5	0.003a	0.003a	0.05
Ba	45.30	11.21a	9.03a	200	0.21b	12.38a	nd

Source of standard:
[1]Kabata-Pendias, A. (2011)
[2]Fourteenth Schedule (Regulation 38) Maximum permitted proportion of metal contaminant in specified food Table 1 (Vegetable product and fruit prduct (Food Act 2009 Edition)
nd = not detected

Radiological Hazard Index and Soil-to-Plant Transfer Factor (TF)

Radium equivalent (Ra_{eq}) is defined as radiation hazard index which is used to assess the radiation hazard due to the natural radionuclides. The calculated radium equivalent for soil treated with T2 was 71.1 Bq kg^{-1}, which was much lower than the recommended limit of radium equivalents in soil (370 Bq kg^{-1}) (UNSCEAR 1982). The TF of ^{226}Ra, ^{228}Ra, ^{238}U, ^{232}Th and ^{40}K obtained in this study were comparable to that observed in several types of plant (rice, vegetables and other main crops in Malaysia) with ranging from 63.2 – 414.4 Bq kg^{-1}. Therefore, the radiological concern due to ^{226}Ra, ^{228}Ra and ^{40}K in soil can be ignored, hence the application of T2 did not show any significant contribution to the accumulation of radionuclides in soil.

CONCLUSION

From this project, rare earth by-product soil conditioner is a new alternative soil conditioner that showed comparable performance with current practices and has a potential for increasing fertility of BRIS soil with no impact to the environment.

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REFERENCES

AL-Areqi, W. M., Majid,A. Ab., and Sarmani, S., 2014. Thorium, uranium and rare earth elements content in lanthanide concentrate (LC) andwater leach purification (WLP) residue of Lynas advanced materials plant (LAMP). AIP Conference Proceedings 1584, American Institute of Physics, Melville, NY, 2014, pp. 93-96.

Aswood, M.S., Jaafar, M.S., Sabar, B., 2013. Assessment of Radionuclide Transfer from Soil to Vegetables in Farms from Cameron Highlands and Penang, (Malaysia) Using Neutron Activation Analysis. *Applied Physics Research*, 5(5), 85-92.

Beretka, J., Matthew, P.J., 1985. Natural Radioactivity of Australian Building Materials, industrial waste and by products. Health Physics, 48, 87-95.

Gaffar, S., Ferdous M.J., Begum, A., Ullah, S.M., 2014. Transfer of natural radionuclides from soil to plants in North Western Parts of Dhaka. Malaysian Journal of Soil Science 18, 61-64.

I**AEA (2010)** - IAEA Technical Report No. 472. *Handbook of Parameter Values for the Prediction of Radionuclide Transfer in Terrestrial and Freshwater Environment*. Vienna: IAEA.

Masitah, A., Zaini, H., Saat, A., 2005. Determination of ^{226}Ra, ^{228}Ra and ^{40}K in Soil from Jengka-15 Oil Palm Plantation. *Journal of Analytical Sciences*, 9(1), 126 – 132.

Maria Greger, 2004. Technical Report TR-04-14: Uptake of Nuclide by Plant. SKB, Sweden.

Md. Sarwar, J., Isa, M., Mohammad, M.K., 2014. Effects of Soil Amendments on BRIS Soil Health, Crop Physiology and Production. *International Journal of Research and Innovations in Earth Science*. Vol.1: 1375 – 2394.

Nikolaos, G., Ioannis, V., Alexandros, P., Eleftheria, S., Nikolaos, C., Adamantia, C. 2013. Chemical and Biological Properties of a Sandy Loam Soil Amended with Olive Mill Waste, Solid or Liquid Form. *In Vitro. International Journal of Recycling of Organic Waste in Agriculture*, 2: 13

Priharti, W., & Supian S., 2016. Radiological Risk Assessment from The Intake of Vegetables and Fruits in Malaysia. *Malaysian Journal of Analytical Sciences,* **20** (6), 1247 – 1253.

P6-2

Nutrient recovery of solid waste from spent bleaching earth and biosolids in asean countries

Kamarulzaman, M. I[1]., A. Sukor[2], and S. S. Zaharah[3]
[1]Institute of Plantation Studies, Jalan Maklumat, Universiti Putra Malaysia
[2]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia
[3]Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia
; shairah@upm.edu.my

INTRODUCTION

Sewage sludge is one of the solid waste, generally refers to the settled solids removed from domestic wastewater (Lwin et al., 2015). Spent bleaching earth (SBE) is another solid waste of palm oil refinery and it has been used to absorb dark colour matters and odour-causing substance in crude palm oil (CPO). About 5-10 kg of bleaching earth per tonne of CPO is used in the refinery process resulting in the generation of solid waste up to 170,000 tonne of spent bleaching earth SBE per annum in Malaysia (MPOB, 2016). The most common practice to discard SBE and biosolids is disposal at landfills, which is economical due to its high disposal cost and creates environmental pollution due to its effect of greenhouse gas emissions upon its disposal (Lwin et al., 2015). The conventional method of disposing SBE and biosolids are at landfills. Howaver, SBE and biosolids has essential minerals and beneficial elements for potential use as a soil supplement for crop growth.

Since SBE consists of natural clay (either bentonite or montmorillonite), so, it can be potentially used as fertilizer binder. The powdered form of SBE can be potentially used as fertilizer binder due to its small particle size and very fine powdered material. However, it can be a challenge dealing with a very fine powdered form of SBE, which creates dust during handling. Therefore, it has to be mixed with biosolids and converted into a pelletized form for handling stability, ease of transportation and easier post-processing. Pelletization is a process of increasing bulk density of a material, reduces dust during handling, easy transportation, allows steady release of nutrient to plant rooting zone. The main objective of this study is to achieve zero-waste, reduce environmental hazards and high disposal cost from the generation of up to 170,000 t of SBE per year and 5,000,000 t of dewaterd sewagesludge per year being disposed to landfills. The recovery of useful resource from SBE and biosolids can be converted into a pelletized form which can then be commercialized for agricultural use.

OBJECTIVES

The objectives of this study are to (i) convert fine powdered form SBE and dewaterd sewagesludge into pelletized form according to the desired ratio to reach the sufficient cohesion, and (ii) to evaluate the chemical properties of the pelletized SBE and dewaterd sludge.

MATERIALS AND METHODS

Bulk material of SBE will be sourced from EcoOils Sdn. Bhd. palm oil refinery at Nilai, Negeri Sembilan on a regular basis, while bulk material of a dewatered sewage sludge will be supplied by Indah Water Konsortium Sdn. Bhd at Putrajaya. The raw material of SBE and dewaterd sewagesludge were analysed for their physical and chemical properties. The powdered form of SBE and dewaterd sewagesludge were mixed in a mixer at 2,400 rpm and was pelletized using a modified pelletizer involving steps such as feeding, pelletizing, separation, drying and packaging (Munkholm and

Kay, 2002). The powdered form of SBE and dewaterd sewagesludge were fed manually to the pelletizer at a ratio based on the theoretical calculation to achieve the desired $N:P_2O_5:K_2O$ content for crop requirement (Loh et al., 2013). The pelletized form of SBE was then analysed for the -chemical properties (Table 1).

RESULTS AND DISCUSSION

Table 1. Chemical characterization of pelleted SBE and dewaterd sewagesludge.

Indicators	Mean ± Standard Deviation
pH (H_2O)	6.4 ± 1.3
Electrical conductivity (mS/cm)	4.2 ± 0.9
Total Carbon (%)	20.3 ± 2.6
Total Nitrogen (%)	2.8 ± 0.5
Phosphorus, P (%)	1.2 ± 0.3
Potassium, K (%)	0.4 ± 0.1
Iron, Fe (%)	2.5 ± 0.9
Copper, Cu (mg kg^{-1})	115 ± 26
Zinc, Zn (mg kg^{-1})	750 ± 160
Cadmium, Cd (mg kg^{-1})	2.3 ± 1.1
Lead, Pb (mg kg^{-1})	29 ± 6.3

Dewaterd sewagesludge consists of permissible amounts of heavy metals, but were found to be lower than the recommended limit of European Union (EU) and Environmental Protection Agency (EPA) (Rosazlin et al., 2005). However, the public is still concern about heavy metal contents in dewaterd sewagesludge due to its bio-accumulative nature once it enters the food chain. Spent bleaching earth (SBE) has been reported to serve as adsorbents for the efficient removal of heavy metals ions such as Cu, Pb and Cd in biosolids (Loh et al., 2013). Further studies need to be conducted to test for the efficacy of SBE as a binder and adsorbent for heavy metal in dewaterd sewagesludge, dissolution rate, incubation study under glass house and fill application need to be investigated.

CONCLUSION

Dewaterd sewagesludge and SBE has a potential to be developed as a pelletized supplementary fertilizer.

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REFERENCES

Loh, S. K., S. James, M. Ngatiman, K. Y. Cheong, Y. M. Choo, and W. S. Lim. 2013. Enhancement of palm oil refinery waste – Spent bleaching earth (SBE) into bio organic fertilizer and their effects on crop biomass growth. Industrial Crops and Products **49**:775-781.

Lwin, C. M., K. N. Maung, and S. Hashimoto.2015. Future sewage sludge generation and sewer pipeline extension in economically developing ASEAN countries. Journal of Material Cycles Waste Management **17**:290-302.

MPOB. 2016. Malaysian oil palm industry performance 2016 and prospects for 2017.
Malaysian Palm Oil Board (MPOB).

Munkholm, L. J. and B. D. Kay. 2002. Effect of water regime on aggregate-tensile strength, rupture energy, and friability. Soil Science Society of America Journal **66**: 702-709.

Rosazlin, A., C. I. Fauziah, A.B. Rosenani, and S. Zauyah. 2005. Domestic sewage sludge application to an acid tropical soil. Malaysian Journal of Soil Science **9**:39-52.

P6-3

Flat optical scanner technique to measure oil palm root production on peat

***Rumpang.E*[1], *Ohashi.M*[1]., *Kume.T*[1]. *and Kho.L.K*[1].**
[1]Tropical Peat Research Institute, MPOB Research Station Belaga, Biological Research Division, Malaysian Palm Oil Board.
; elisa@mpob.gov.my

INTRODUCTION

Root productivity plays an important role in terrestrial ecosystems. The production and mortality of roots contribute substantially to the carbon dynamics in the soil system. Approximately fine roots constituted about 33% of terrestrial net primary productivity (Jackson et al. 1997; Gill and Jackson. 2000). Some of the decomposed roots will release C to the atmosphere or remained in soil as a soil organic matter (Norby and Jackson. 2000). Understanding roots mortality and production is important to develop a standard approach for roots estimates and modelling (Harun and Md Noor. 2004). In particular, because of their short life span, fine roots can be indicator to environmental changes (Hirano et al. 2007). Researcher use several techniques to measure root production and behaviour. The existing methods are divided into two techniques: destructive and non- destructive methods (Yuan and Chen. 2012). It is unable to estimate the amount of dead fine roots (Satomura et al. 2007) using destructive method (coring, digging and augers). But, still there is no perfect methodology to measure the roots without causing disturbance on roots. There is limited information available for root processes on peat swamp forest and oil palm in peat ecosystems due to the inaccessibility of root systems. It is still unclear how perfect it is to spatially determine growth and mortality of root processes in peat swamp forest and oil palm ecosystem to understand belowground carbon dynamics, particularly on tropical peat soils due to the complexities of soil properties and condition.

Therefore, this study aims to observe and assess root parameters (e.g. life span, root production) for oil palm roots and peat swamp forest. We also discuss how this new economical technique by using the scanner may allow robust and extensive assessment of roots. Scanner techniques provides a non-destructive, *in situ* method to record the roots. It is one of the best tools available to directly study roots.

MATERIALS AND METHODS

The study was conducted in peat swamp forest and oil palm plantation located in Sarawak Oil Palm Berhad (SOPB), Sebungan and Sabaju, Bintulu, Sarawak. In this study, four transparent scanner boxes were installed vertically in oil palm plantation on peat soil - palm based management (January 2017). Meanwhile, in peat swamp forest the transparent boxes were installed randomly during May 2017. Technically, the installation should be done in the dry season to avoid the rainfall from eroding the contact between the soil and the screen. The procedure was conducted carefully to prevent supplementary root damage and losses. The scanner boxes were leaft for three months to allow roots to grow and recover from the disturbance during installation. Roots images were taken monthly using a Charge Coupled Device (CCD) flatbed image scanner (GT-S630, Epson, Japan). We scanned images at monthly interval. This method enables us to observe larger images across wider spatial area and at a lower cost.

RESULTS AND DISCUSSION

The investigation and data images analysis is currently in progress. However, the scanner images for peat swamp forest was collected around September 2017. From the initial observation on oil palm roots in peat, more live roots appears than dead roots on the third month after the scanner box was installed. The roots development (length and diameter) of oil palm can be seen clearly below (Figure 1). Roots can be more easily analyzed by enlarging the image (Dannoura et al. 2008), changing the resolutions and brightness.

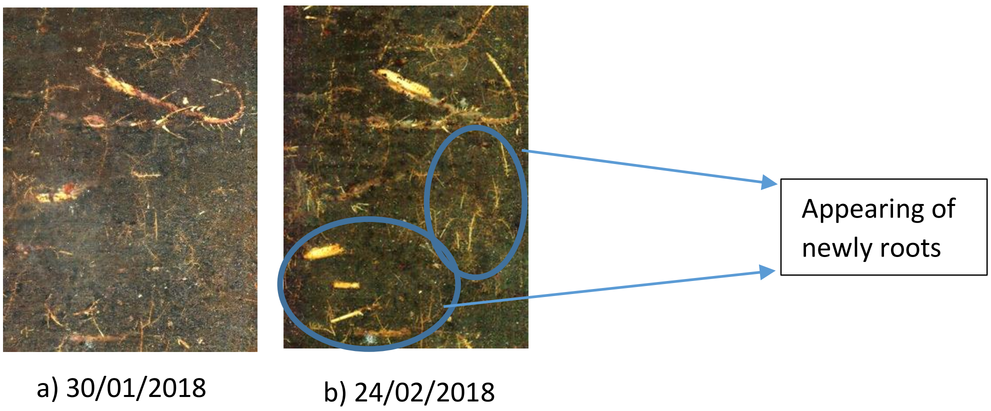


Figure 1: Oil palm roots images collected on the same observation frame at different dates.

This scanner images show new roots tips and hairs growing and increasing along the scanner wall. Using this images, growth and dead root can be determined by estimating the changes in roots length, diameter, appearance and disappearance. A white root is consider as an active growth root (Figure 1) while brown colour or no roots appearance is considered as dead roots. Scanner method provide a simple yet effective method to measure root behaviour. However, we believe the pattern and behaviour of root production may be influenced by soil conditions (Noguchi et al. 2005), soil moisture (Joslin et al. 2000), physic-chemical properties, nutrient availability, parent material and soil management.

CONCLUSION

Scanner imaging technique enable us to observe live and dead roots in the soil clearly following a wider scale of view from one sampling point. This is an ideal yet cheap and effective method to characterize root processes in peat soil ecosystems as well as other soil types. The scanner method has advantages to minimizing the soil disturbance and allow to monitoring the roots production in the long term. Besides, it can measure root production rates with less error and help to deepen our understanding of belowground biomass and productivity in peat swamp forest and oil palm in peat.

REFERENCES

Dannoura M, Kominami Y, Oguma H, Kanazawa Y (2008) The development of an optical scanner method for observation of plant root dynamics. *Plant Root* **2**: *14-18. (JSRR).*

Hooshang Majdi, Kurt Pregitzer, Ann-Sofie Moren, Jan-Erik Nylund and Goran I. Agren (2005) Measuring fine root turnover in forest ecosystems.

Harun M H and Md Noor M R (2004) Oil palm roots study using the mini-rhizotron technique. *Oil Palm Belletin. P. 17-23.*

Hirano Y, Mizoguchi T, Brunner I (2007) Root parameters of forest trees as sensitive indicators of acidifying pollutants: a review of research of Japanese forest trees. J. For. Res.12: 134-142.

J. D. Joslin, M. H. Wolfe, P. J. Hanson (2000) Effects of altered water regimes on forest root systems. *Plant and Soil.* ***276****:1- 8.*

K. Noguchi, T. Sakata, T. Mizoguchi, M. Takahashi (2005) Estimating the production and mortality of fine roots in a Japanese cedar *(Cryptomeria japionica D. Don*) plantation using a minirhizotron technique.

Richard J N and Robert B J (2000) Root dynamics and global change: seeking an ecosystem perspective. *New Phytol.* ***147,*** *3–12.*

Richard A G and Robert B J (2000) Global patterns of root turnover for terrestrial ecosystems. *New Phytol.* ***147****, 13–31.*

Satomura T, Fukuzawa K, Horikoshi T 2007 Considerations in the study of tree fine-root turnover with minirhizotrons. Plant Root **1**: 34-45

Yuan ZY, Chen HYH (2012) Indirect Methods Produce Higher Estimates of Fine Root Production and Turnover Rates than Direct Methods. *PLoS ON*

P6-4

The influence of pH and soil biota on cu contamination under different soil type

Haryati M[1]., Surya M[2]., Yves D[2]., and Intan Nadhirah M[1].
[1]Malaysian Agricultural Research & Development Institut
[2]Wageningen University and Research
; ytmansor@mardi.gov.my

INTRODUCTION

Copper (Cu) is a micronutrients which is important for plant in small amounts. However, the presence in high amount of Cu in plant will cause toxicity, also for human and animals. The effect of Cu in differents soil conditions were analyse both biologically and chemically using rye grass (*Lolium perenne*) as indicator plant. The goals of this experiment were to monitor the effects of heavy metals on rye grass growth and the influences of soil pH, soil type and biota on the contamination.

MATERIALS AND METHODS

The experiment was conducted to evaluate the effects of heavy metals on the growth and composition on rye grass, and the influences of soil type, pH and soil biota on the heavy metals. A five factorial design was conducted in pots. The soil types used for this experiment include sandy soil with pH-$CaCl_2$ of 4.5 and 6.5, and a clay soil with pH-$CaCl_2$ of 5.0 and 7.0. The heavy metals studied were Cu and Zn, and the pots were treated with either Cu or Cu and Zn. To evaluate biological effects, pots were inoculated with and without mycorrhiza (*Rhizophagus irregularis*). There were no replicates for each treatment.

After a six-week growth period, visual evaluation of the pot experiment was conducted before shoots and roots were harvested and weighed. Soil samples of each plot were also taken. Root and shoot samples were dried at a temperature of 70 °C for 24 hours and weighed again to determine dry-weight. The soil samples were also dried at a temperature of 40°C for 24 hours, which were then sieved through a two millimetre screen to remove gravel and roots.

In all samples, Cu and Zn were measured. The soil was extracted with 0.43 M HNO_3 to determine the total reactive Cu- or Zn-content and 0.01M $CaCl_2$ was used to determine the bioavailable amount. Methods to quantify the effects of Cu contamination under altered soil pH and mycorrhizal fungi conditions include the effective concentration and the mycorrhizal effect. The effective concentration tells at what metal concentrations the yield has no effect (NOEC), is eighty percent of the original yield (EC_{20}), and is half of the original yield (EC_{50}). The ratio of ln (W+/W-) helped to identify more yield effects of mycorrhizal fungi. This ratio tells if the mycorrhizal fungi contribute significantly to the heavy metal uptake of the plants.

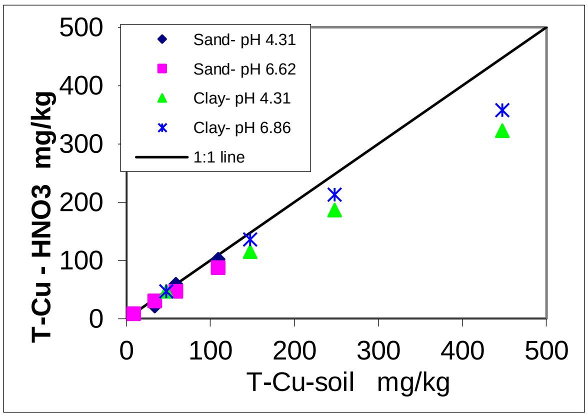
The extraction with $CaCl_2$ was conducted through the addition of 3 gram of soil and 30 milliliters of 0.01 $CaCl_2$ in a centrifuge tube. A reference sample and a blank were also included in the procedure. The samples were shaken for one hour and the suspensions were allowed to settle before measurement of the pH. The procedure continued by centrifuging the solution using a bench centrifuge at 3000 rpm for ten minutes. Next, 10 milliliters of the clear solution were filtered using a 0.45μm membrane filter. The filtered solutions were measured for Cu or Zn using the AAS. Depending on the treatment, the solution should be diluted before measurements are done.

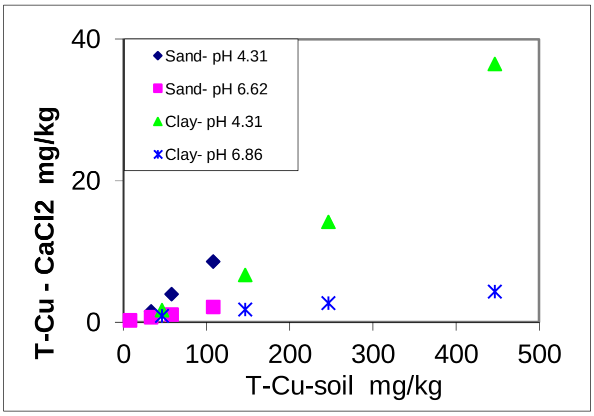
RESULTS AND DISCUSSION

Extractable Copper Concentrations

The Cu measurements are more accurate (close to the 1:1 line) at lower concentrations. At higher concentrations (greater than 150 mg/kg), the measured amounts are lower than the actual amounts (Figure 1). When mycorrhizal fungi are added, the patterns are similar to without it, however, the points fall slightly closer to the 1:1 line. The total amount of Cu present in the soil cannot be completely recovered by the soil extraction with 0.43 M HNO_3. This is evident because, in all pots, the measured amount was underestimated relative to the 1:1 line.

As with the HNO_3 extraction, the clay soils tend to deviate farther from the 1:1 line than sandy soils at high Cu concentration. The sand at pH 4.31 has the highest bioavailability, and clay at pH 6.86 has the lowest. Both clay and sand reveal higher availability at a lower pH than at a higher pH. Soil organic matter (SOM) is the reactive surface for heavy metal binding, and phenolic and carbocyclic groups are pH dependant; high pH results in the protonation of the phenolic and carbocyclic groups. At high pH, SOM becomes more negatively charged, and therefore, binds more heavy metals and decreases the bioavailability. Therefore, higher bioavailability is expected at a lower pH. Mycorrhiza has no effect on the bioavailability of Cu, as it only influences the uptake of the heavy metals.





Effect Concentration Analysis

With the single metal Cu treatment, the results did not show any clear effect of mycorrhiza. The mean NOEC for both the $CaCl_2$ extractable as well as the HNO_3 extractable Cu differ slightly when treatments with and without mycorrhiza are compared. From our results, clay soils have higher EC_x values than sandy soils. With high pH, the EC_x values are higher, which means that Cu effects the plants less at higher pH. As the EC_x values are higher with mycorrhiza, the mycorrhiza is able to protect the plant from excessive heavy metals. The presence of mycorrhiza enables the plant to take more of the metal, but at higher concentrations, the presence of mycorrhiza reduces the uptake of Cu. The bioavailability of heavy metals is controlled by soil characteristics, such as pH. The pH affects the chemical speciation of the heavy metals in the soils and the metal binding to the active sites on biota. The $CaCl_2$ extraction measures the Cu in the soil solution, which the plants ultimately take up. The $CaCl_2$ extraction is a better parameter than HNO_3 extraction to set a critical NOEC or EC_x because the metal uptake of the plant comes from the soil solution and not directly from the soil solid phase.

The mean value, standard deviation (SD) and coefficient of variation (CV) show that, overall, the mycorrhizal effect occurs at higher concentrations. In sandy and clay soils with HNO_3 extraction and with or without mycorrhiza, the EC_{20} is higher in high pH. The same pattern exists with $CaCl_2$ extraction.

Table 1 – The EC_x values for the two extractions methods with and without mycorrhiza for pots treated with Cu.

With mycorrhiza

HNO_3-extractable Cu in soil (mg/kg)	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	8.2	8.2	47.3
Sand-pH 6.62	8.7	8.7	--
Clay-pH 4.31	49.3	49.3	49.3
Clay-pH6.86	139.5	242.6	--
Mean	51.4	77.2	48.3
SD	61.8	111.9	1.4
CV	1.2	1.5	0.03

$CaCl_2$-extractable Cu in soil (mg/kg)	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	0.3	0.3	4.2
Sand-pH 6.62	0.3	1.1	--
Clay-pH 4.31	1.8	1.8	1.8
Clay-pH6.86	2.1	2.6	--
Mean	1.1	1.5	3.0
SD	1.0	1.0	1.7
CV	0.9	0.7	0.6

Without mycorrhiza

HNO_3-extractable Cu in soil (mg/kg)	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	8.7	8.7	58.9
Sand-pH 6.62	8.5	30.9	--
Clay-pH 4.31	47.0	47.0	115.6
Clay-pH 6.86	136.0	358.4	--
Mean	50.1	111.3	87.3
SD	60.1	165.5	40.1
CV	1.2	1.5	0.5

$CaCl_2$-extractable Cu in soil (mg/kg)	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	0.3	0.3	4.0
Sand-pH 6.62	0.2	0.7	--
Clay-pH 4.31	1.7	1.7	6.7
Clay-pH6.86	1.8	4.3	--
Mean	1.0	1.8	5.4
SD	0.9	1.8	1.9
CV	0.9	1.0	0.4

Mycorrhiza effect

In pots with Cu (Table 2), there is a significant effect of mycorrhiza at a low pH in sandy and clay soils. At a low pH, there is an increase of metal uptake by mycorrhiza. Conversely, at a high pH in both soils there is no significant effect of mycorrhiza on the yield. It means that mycorrhiza did not seem to have any beneficial effect on plant growth.

Table 2: Mycorrhiza influence on plant yield. The ln(W+/W-) values for sand and clay for Cu at both low and high pH (left). The ln(W+/W-) values for sand and clay for Cu/Zn interactions at both low and high pH (right).

	Cu			**Cu/Zn**		
	ln(W+/W-)	Mean	Significant	ln(W+/W-)	Mean	Significant
Sand+ pH 4.27	0.03	0.17	Yes	0.13	0.08	No
	0.30			0.05		
	0.12			0.4		
	0.23			-0.27		
Sand+ pH 6.84	-0.03	0.01	No	0.03	-0.01	No
	-0.06			-0.02		
	0.07			0.00		
	0.08			-0.03		
Clay+ pH 4.27	-0.03	-0.17	Yes	-0.04	0.28	Yes
	-0.43			0.69		
	-0.54			0.39		
	0.33			0.07		
Clay+ pH 6.90	-0.01	0.04	No	-0.04	-0.03	No
	-0.01			-0.04		
	0.01			-0.05		
	0.17			0.00		

Cu-Zn Interactions

Compared to the values in Table 1, the mean values are lower for the HNO_3 extraction but are higher for the $CaCl_2$ extraction. This means that although there are higher concentrations of heavy metals, less of them are bioavailable. Furthermore, with the ln (W+/W-) test, mycorrhiza is only significant with clay at a low pH, unlike with just Cu when it was significant in all soil types at low pH. Comparing the results for $CaCl_2$ extraction between single treatment and interaction treatment, we see only a small difference between treatments.

In sandy soils with both Cu and Zn, there is a larger reduction in biomass than pots with Cu only, especially at low pH. Pots with Zn and Cu in sandy soil with mycorrhiza show a larger reduction in biomass in low pH, although mycorrhiza still does not have a significant effect in sandy soils with the interaction of Cu and Zn. Pots with the addition of Cu and Zn in clay soils, have a higher yield in higher pH than in low pH and show higher reduction in biomass than pots with Cu only. Pots with Zn and Cu in clay soils with mycorrhiza show a higher yield in high pH than in low pH, so mycorrhiza does not affect the pots.

Table 3 – The EC_x values for the two extractions methods with and without mycorrhiza for pots treated with Cu. The mean value, standard deviation (SD) and coefficient of variation (CV) show that, overall, the patterns from Table 1 still hold, however, with the HNO_3 extraction, the EC_x values are lower.

Without mycorrhiza

Cu in HNO_3-extraction (mg/kg)			
	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	8.7	8.7	28.7
Sand-pH 6.62	8.5	25.5	--
Clay-pH 4.31	47.0	47.0	47.0
Clay-pH6.86	47.5	114.8	--
Mean	27.93	49.00	37.85
SD	22.32	46.58	12.94
CV	0.80	0.95	0.34

With mycorrhiza:

Cu in HNO_3-extraction for Cu/Zn (mg/kg)			
	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	8.2	8.2	27.0
Sand-pH 6.62	8.7	39.3	--
Clay-pH 4.31	49.3	49.3	119.8
Clay-pH6.86	118.0	143.8	--
Mean	46.05	60.15	73.4
SD	51.69	58.45	65.62
CV	1.12	0.97	0.89

Cu in $CaCl_2$-extraction (mg/kg)			
	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	0.3	0.3	2.9
Sand-pH 6.62	0.7	1.3	--
Clay-pH 4.31	1.7	1.7	1.7
Clay-pH6.86	1.9	3.4	--
Mean	1.15	1.68	2.30
SD	0.77	1.29	0.85
CV	0.87	0.77	0.37

Cu in $CaCl_2$-extraction Interaction of Cu and Zn (mg/kg)			
	NOEC	EC_{20}	EC_{50}
Sand-pH 4.31	0.3	0.3	3.6
Sand-pH 6.62	0.3	2.1	--
Clay-pH 4.31	1.8	1.8	6.7
Clay-pH6.86	2.4	4.0	--
Mean	1.20	2.05	5.15
SD	1.07	1.52	2.19
CV	0.89	0.74	0.43

CONCLUSION

Results of this study show growth can by limited by low pH. Cu concentration in the soil had a strong effect on the bioavailable amount in the soil. With low pH, there was more Cu bioavailable. The yield was more strongly affected at lower pH. For sandy soil with low pH, the yields were clearly affected by the Cu contamination but the clays soils could handle it longer at higher Cu concentration. Mycorrhiza only has a significant effect on the Cu uptake at low pH for both soil types. Mycorrhiza did not seem to have any beneficial effect on plant growth under heavy metal contaminated soil. From studying the interaction of Cu and Zn, it is evident that Cu is more toxic than Zn and is responsible for the corresponding reduction in biomass. Rye grass grows better in clayey soil.

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P6-5

Effetcs of water management practices on yield and nutrient composition of rice cultivars

Hossain, M.[1], G.J. Norton[2], A. Travis[2], A. Price[2], A.A. Mahmud[1] and M.R. Islam[1]
[1]Department of Soil Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh
[2]School of Biological Sciences, Cruickshank Building, University of Aberdeen, AB24 3UU, UK.
; mahmud.ss@bau.edu.bd ; mrislam58@yahoo.com

INTRODUCTION

Rice is the staple food for half of the world's population and currently rice is supplying 20% of the daily calorie intake of this population. The ever-increasing population in the world is posing a challenge of increasing rice production from 676 million tons in 2010 to 852 million tons by 2030.Scientists are trying to exploit every possible means of increasing rice yield with minimum inputs. Irrigation water is one of the major expenses for rice production and in many parts of the world, irrigation water is contaminated, especially with arsenic. Hence, adoption of techniques that decrease water usage while maintaining or increasing crop yield are needed. Alternate wetting and drying (AWD) is one of these techniques. AWD is a method in which the rice field is allowed to dry until the water level in the rice soil goes down to 15 cm depth and then the field is reflooded therefore, a strong redox cycle is introduced in rice growth period. The degree to which AWD affects rice cultivars differently in acid soil- has not been adequately addressed to date. The present research was designed to find the effect of water saving rice culture, AWD on yield and nutritional value of rice in acid soil.

MATERIALS AND METHODS

Twenty-two boro rice cultivars were grown during dry season of 2014 at Mymensingh (MN) (24°42′58″; 90°25′26″) and Madhupur (MD) (24°35′19″; 90°02′22″) of Bangladesh Soil pH of Mymenssingh was almost neutral (6.7) while that of Madhupur was acidic (6.0). Conventional rice irrigation system termed as continuous standing water (CSW) and water saving rice irrigation system termed as alternate wetting and drying (AWD) were tested on 22 rice cultivars, and each treatment was replicated four times. Rice was transplanted in December 2014. The bunds in the AWD plots were lined with polythene sheet to check water movement into the plots. AWD pipes were placed in the AWD plots and the plots were irrigated when the water level in the AWD pipes went below 15 cm depth. Plots were fertilized with 120 kg N/ha, 15 kg P/ha, 50 kg K/ha, 15 kg S/ha and 3 kg Zn/ha. Intercultural operations were done as per requirement of the crop. Plants were harvested at maturity and yield was recorded at 14% moisture content. Elemental analysis of rice straw and grains were conducted as described in Norton et al. (2017). Total arsenic, phosphorus, iron and zinc in grain and straw were analyzed by ICP-MS. Nitrogen content in shoot was determined on the powdered samples by CNS analyser.

RESULTS AND DISCUSSION

The straw biomass production significantly varied between the two field sites, and a site by treatment interaction ($P < 0.001$). Higher straw biomass was observed at Madhupur (89.0 g) than at Mymensingh (74.3 g) (Fig. 1A).

Overall, there was no significant difference in straw biomass for plants grown under AWD (82.6 g) compared to CSW (80.7 g). Grain mass was significantly affected ($P < 0.001$) between the two locations. The grain mass was higher in Madhupur (58.3 g) than at Mymensingh (50.6 g) (Fig.1A). Overall, there was a significant difference ($P = 0.02$) in grain mass for plants grown under AWD compared to CSW, with the AWD plants on average having a grain mass of 57.5 g compared to 50.3 g for the CSW plants. There was no significant site by treatment interaction. The concentration of straw nitrogen was significantly affected by the location when they were grown under AWD or CSW (Fig. 1B). Nitrogen concentration in the straw was observed higher at the Mymensingh site (0.76%) compared to the Madhupur (0.53%). AWD had lower shoot nitrogen (0.62%) compared to CSW (0.67%) between the locations.

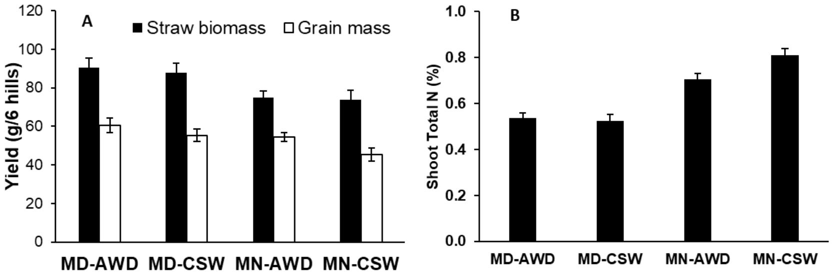

Figure 1. A. Straw biomass and grain mass, and B. shoot N of 22 boro rice cultivars at two sites under AWD and CSW. MD = Madhupur and MN = Mymensingh. AWD, Alternative wetting and drying; CSW, continuous standing water.

The concentration of straw arsenic was significantly affected by the sites. The average straw arsenic concentration was higher at Mymensingh (1.25 mg/kg) compared to Madhupur (0.76 mg/kg). AWD significantly decreased straw arsenic by 16.7% compared to CSW. There were significant effects of site, treatment and a site × treatment effect for grain arsenic with AWD on average decreasing grain arsenic by 3.2% between the sites. The straw phosphorus was significantly affected by site and treatment as well as interaction between site × treatment (Figure 2B). The higher shoot P concentrations were found in plants grown at the Madhupur site (631.8 mg/kg) followed by Mymensingh (550.3 mg/kg). The AWD treatment caused a significant decrease in straw phosphorus between the sites (AWD average concentration 459 mg/kg and CSW average concentration 590 mg/kg). The plants grown at Madhupur had significantly higher concentrations of grain phosphorus (4390 mg/kg) compared to Mymensingh (4197 mg/kg). AWD reduced grain phosphorus by 5.9% compared to plants grown under CSW between the sites. (Figure 2B).The zinc concentration in straw and grain was significantly affected by the locations where rice was grown. The higher shoot zinc concentrations were found in plants grown at the Madhupur site (61.04 mg/kg) followed by Mymensingh (30.19 mg/kg (Fig. 2C). The grain Zn concentration was also

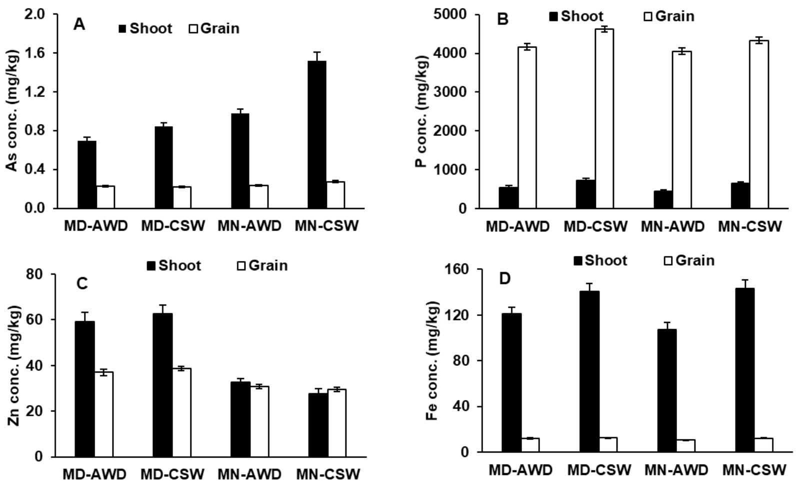

Figure 2. Rice elemental concentration grown at two sites under AWD and CF. (a) arsenic, (b) phosphorus, (c) zinc, (d) iron. MD = Madhupur, MN = Mymensingh. AWD, Alternative wetting and drying; CF, continuous flooding.

higher in Madhupur (average 37.8 mg/kg) than in Mymensingh (30.2 mg/kg) where the rice experiment was done. There was no significant effect of AWD treatment on grain zinc compared to CSW. Straw and grain iron was also affected between the sites, with AWD on average causing a 12.9% and 12.4% decrease in straw and grain iron, respectively, compared to CSW (Fig. 2D).

CONCLUSION

AWD slightly increased grain mass compared to plants grown under CSW conditions. Shoot N concentration decreased under AWD compared to plants grown under CSW. The elemental concentrations in rice were also affected when plants were grown under AWD compared to CSW; iron increased, but phosphorus and arsenic decreased in the grains of plants grown under AWD.

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REFERENCES

Norton, G. J., M. Shafaei, A. J. Travis, C. M. Deacon, J. Danku, D. Pond, et al. 2017. Impact of alternate wetting and drying on rice physiology, grain production, and grain quality. *Field Crop Res.* 205:1–13.

P6-6

A Rapid Method to Generate Tea Cuttages

Liu, X. M[1]., L. L. Sun[1], and H. Liao[1]
[1]Root Biology Center, Fujian Agriculture and Forestry University, 350002 Fuzhou, Fujian, China
; lilionsun@163.com

INTRODUCTION

As one of the three major beverages in the world, tea is an important economic crop. Tea plants are well known for their great adaptation to acidic soils (Gülen *et al*., 2010). However, the underlying mechanisms are still unclear. For better studies on tea plants, uniform seedlings are much helpful. For the complex genetic background (Xia *et al*., 2017), tea cuttages are more popular for its stable inheritance compared with sexual reproduction. Traditionally, tea cottages are produced through trimming branches into soils for root generation (Sun *et al*., 2001). However, there are many limitation factors in this traditional method, such as growth stage and soil type. Therefore, a rapid method is demanded.

MATERIALS AND METHODS

The tea variety, Fuding Dabai, was used as the experimental material. Tea branches with healthy leaves, stem diameter larger than 3 mm and axillary buds over 0.5 cm were collected from tea plantation. Then, the branch was trimmed into 2 cm with one leaf and one bud. The incisions on the stem were smooth and chamfered at 45 degree parallel to leaves. After cutting, tea branches were placed in distilled water. Then, the surface of the stem was cut off with a length of about 1 cm and a width of about 2 mm using a surgical blade.

Since moisture and oxygen were important for tea plant rooting from the cut, sponge was used to generate root. The blocks of sponges with 3×3×2 cm were prepared as culture medium. Two cutting branches were placed upright and packed with sponges. The treated branches were placed in a wide-diameter glass bottle, and cultured by 20 mL nutrient solution specially made for tea plants every 3 days. After rooting, the seedlings were transplanted into hydroponics. The condition of growth chamber was about 28°C and 60% of humidity.

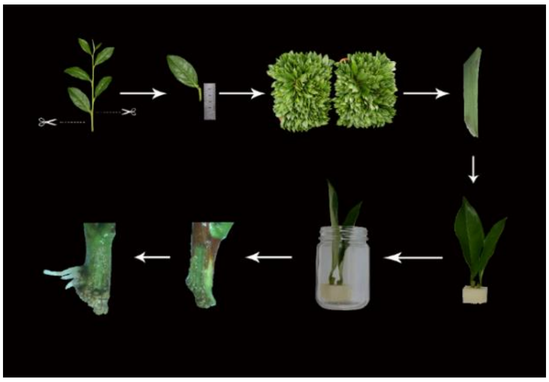

Fig.1 Process of tea plant cuttages

RESULTS AND DISCUSSION

Fifteen to twenty days after treatment, the stems formed callus on the cutting place. When the growth rates of the callus slowed down, roots came out from the callus. Roots were growing in the sponge. After transferring to hydroponic system, the roots emerged more and grew longer.

Sponge with porous characteristics could protect the cutting place from water loss and promote the formation of callus. Compared with the complicated conditions of soils, the use of sponges can easily break the limitation of time and space, and shorten the rooting time.

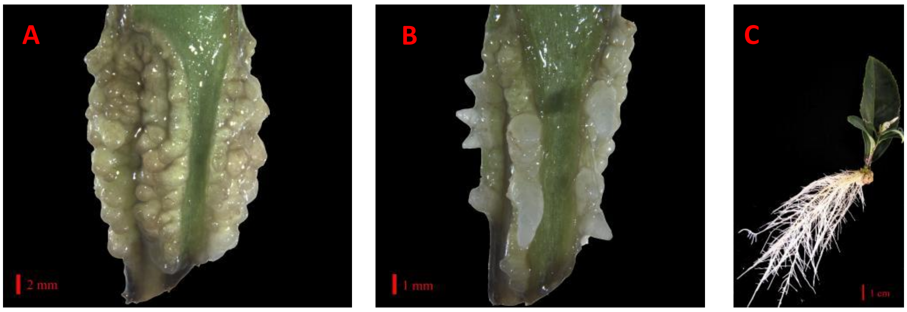

Fig.2 Pictures of tea plants in rooting process

CONCLUSION

Compared with the traditional method, the use of sponge as culture medium significantly increases the amount of callus and promotes root formation. The cultivated cutting seedlings using this method can produce well-developed root systems.

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REFERENCES

Gülen, Ö., A. Ö. Mehmet, Ö. Osman, and S. Abdulkadir. 2010. Some physical a chemical properties of tea grow soils in Rize and Artvin Provinces. Anadolu Tarım Bilimleri Dergisi **25**(2):94-99.

Sun, Z. X., J. Liu, Z. R. Liu, and Z. L. Qiu. 2001. Research of technology on cutting plant of tea tree in Shandong Province. Journal of Shandong Agricultural University **32**(3):285-288.

Xia, E. H., H. B. Zhang, J. Sheng, et al. 2017. The tea tree genome provides insights into tea flavor and independent evolution of caffeine biosynthesis. Molecular Plant **10**(6):866-877.

P6-7

Earthworm population as an indicator of soil health under organic farming system

Intan Nadhirah, M[1]., Noor Haslizawati, A. B[1]., Mohamed Hafeifi, B[1]., Norziana, Z. Z[1]., Illani Zuraihah, I[1]., Mohamad Fakhri, M[1]., and Azrizal A. R[1].
[1]MARDI, P. O. Box 12301, Pejabat Pos Besar, 50774 Kuala Lumpur, Malaysia
; *intannadhirah@mardi.gov.my*

INTRODUCTION

Soil health is normally viewed in terms of biomass production (McBratney et al., 2012) or productivity indices relative to fundamental soil properties. Many studies were conducted based on increasing yields from a profit standpoint regarding the fitness of soils for crop production. According to Tilley in 2016, specific plant or crop varieties that are used to improve soil quality are known as green manure. Growing green manure crops is also regard as feasible way to control nitrate leaching (McDaniel et al., 2014). Earthworms are one of the most important groups of soil invertebrates and are known to improve soil fertility by enhancing the physical, chemical and biological characteristics of the soil (Lee, 1985).

This paper is aimed at understanding the influence of soil condition on earthworm population under organic-managed system.

MATERIALS AND METHODS

Site description:

The study was carried out at soil health management research plot which was established in Integrated Organic Farm, MARDI Serdang. Total of 20 beds consisted of five (5) treatments and four (4) replications each measuring 2m x 3m. Treatments were described in the Table 1. Plot was irrigated sufficiently through sprinkler watering system throughout four (4) months of study period. Earthworm sampling were conducted during 3 stages (early, middle and final) of treatments cultivation.

Table 1: Types of treatments

Treatments	Green manure mode of application
T1: Control	-
T2: *Arachis pintoi*	Planted in beds
T3: *Medicago sativa*	Planted in beds
T4: *Gliricidia sepium*	Leaf as mulch was applied every 1 month
T5: *Moringa oleifera*	Leaf as mulch was applied every 1 month

Earthworm sampling:

A square-foot marking was measured, and the soil was dug about 12 inches depth by using hand trowel. Earthworms were removed by hand sorting. Numbers of earthworm were counted, and further soil breaking was done to extract maximum number of worm.

RESULTS AND DISCUSSION

Initial earthworm sampling resulted in 7 per square foot of soil (70 earthworms/m^2). After 4 months of treatments incorporated into the soil, except for treatment 4 showing no changes in earthworm count, the rest of the treatments showed increment in number of worms per square foot of soil (Table 2).

Table 2: Average number of worm according to treatments

Treatments	**Worm count (worms/sq ft)**	
	Initial	After 4 months
T4: *Gliricidia sepium*	7	7
T3: *Medicago sativa*		9
T5: *Moringa oleifera*		11
T1: Control		15
T2: *Arachis pintoi*		17

About 10 earthworms per square foot of soil (100 worms/m^2) is generally considered a good population in agricultural system. Populations generally do not exceed 20 per square foot of soil (200 worms/m^2) in cultivated systems (Edwards, 1983). In grassland systems, population can generally range up to about 50 per square foot of soil (500 worms/m^2) (Edwards, 1983). It was expected that T3 would give low number of earthworm count due to tillage management during cultivation of *Medicago sativa*. According to Curry in 1998, tillage generally kills 25% of the earthworm population. However, in the case of *Arachis pintoi*, its fast-growing leguminous characteristic provides quick recovery and the nodules in the soil provide better aeration for earthworm population to multiply. Depth of aeration in soils affects the deep-burrowing species (Curry, 1998). It was also expected that earthworm in T1 (control) would multiply as well due to the soil being undisturbed. Earthworm populations are generally higher in undisturbed soil systems (Curry, 1998).

All the soil texture of the reaserch plots fall under the categorie of sandy clay loam. The number of earthworms collected during 4 months of study was based on the actual condition of the plot on the sampling day and time. For instance, we found that beds that are not fully irrigated were too dried and hardened. Hence the number of earthworm sampled was the lowest in T4 and quite low in T3 even though the beds were minimally disturbed. In terms of soil texture, medium textured soils are more favourable for earthworms than sandy or clayey soils (Curry, 1998). Moreover, earthworm foraging, and casting behaviours depend of intrinsic soil parameters (e.g. texture, moisture), climate conditions and soil organic matter availability or palatability (Butt et al., 2005; Lowe and Butt, 2002).

CONCLUSION

Based on the study, improved soil physical condition will result in increased number of earthworm population. Therefore, growing *Arachis pintoi* as green manure gives the best result in improving soil health in terms of increasing number of earthworm.

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REFERENCES

Butt, K.R., Nieminen, M.A., Sirén, T., Ketoja, E., Nuutinen, V., 2005. Population and behavioural level responses of arable soil earthworms to boardmill sludge application. Biol. Fertil. Soils **42**, 163–167.

Curry, J. P. 1998. Factors affecting earthworm abundance in soils. P.37-64. In: C. A. Edwards (ed.) Earthworm ecology. CRC Press, Boca Raton.

Edwards, C. A. 1983. Earthworm ecology in cultivated systems. P.123-137. In: J. E. Satchell (ed.) Earthworm ecology. From Darwin to Vermiculture. Chapman and Hill, London.

Lee, K. E. (1985). Earthworms: Their ecology and Relationships with Soils and Land Use. Academic Press, Sydney, Australia.

Lowe, C.N., Butt, K.R., 2002. Influence of organic matter on earthworm production and behaviour: a laboratory-based approach with applications for soil restoration. Eur. J. Soil Biol. 38, 173–176

McBratney, A., Minasny, B., Wheeler, I., Malone, B.P., 2012. Frameworks for digital soil assessment. In: Minasny, B., Malone, B.P., McBratney, A.B. (Eds.), Digital Soil Assessment and Beyond. Taylor & Francis Group, London, pp. 9–14.

McDaniel, M.D., Tiemann, L.K., Grandy, A.S., (2014). Does agricultural crop diversity enhance soil microbial biomass and organic matter dynamics? A meta-analysis. Ecol. Appl. **24** (3), 560–570.

Tilley, N. 2016. Gardening Know How. March 20. Accessed April 17, 2018. https://www.gardeningknowhow.com/edible/vegetables/cover-crops/green-manure-2.htm.

P6-8

Assessment of agricultural landuse on water quality at Berembun Valley, Cameron Highlands.

Noor Haslizawati A. B.[1]***, Mohd Shahid S.***[1]***, Intan Nadhirah M.***[1]***, Mohamad Fakhri M.***[1]***, Mustafa Kamal H.***[2]***, Azrizal A. R.***[1] ***and Nor Azhar A.***[1]
[1]Crop & Soil Science Research Centre, MARDI Headquarters, 43400 Serdang, Selangor
[2]MARDI Agrotechnology Park, Cameron Highlands, 39000 Tanah Rata, Pahang
; haslizawati@mardi.gov.my

INTRODUCTION

Farmers in the Cameron highlands are some of the primary producers of Malaysia agricultural products, particularly fresh vegetables (Hamdan *et al.,* 2014). Farming in tropical highlands commonly causes serious on-and off-farm environmental impacts (Allen *et al.,* 1995; Rerkasem *et al.,* 2002; Forsyth 2007). The expansion and intensification of farming in the Cameron Highlands has seriously polluted streams and groundwater with sediment, manure enriched runoff, agrichemicals and sewage (Amminuddin *et al*,. 1990; Midmore *et al.*, 1996; Wan Abdullah *et al.*, 2001; Wong *et al.*, 2002). Our objective for this paper was to determine the effect of agricultural land use on water quality at highland area.

MATERIALS AND METHODS

Sampling locations: This study will be conducted at Berembun Valley, Cameron Highlands with GPS coordinates N 4.472213 E 101.389822. This area was intensive open farming area where it where it is planted with a variety of vegetables such as cabbage, French bean, chili, eggplant, zucchini etc. There is a small stream known as the Berembun river which is the source of irrigation for smallholders here. Water samples will collected at 17 points along the Berembun river (Figure 1).

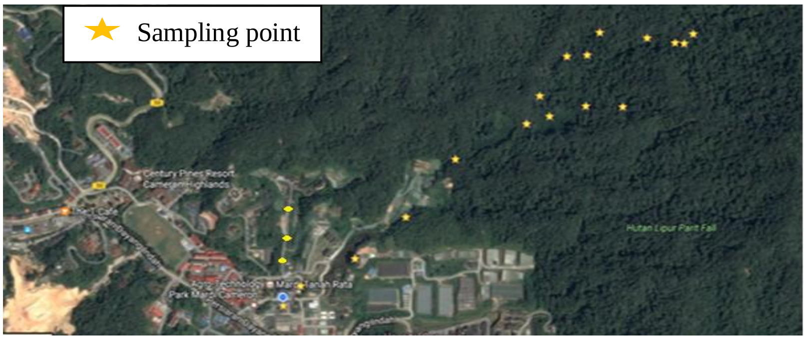

Figure 1: Sampling point at Berembun Valley, Cameron Highlands

The value of each parameter *in situ* measurements of pH, electrical conductivity (EC), total dissolved solids (TDS) and stream flow of each river were taken using relevant field equipment. Laboratory analysis was also carried out on the water samples for other physico-chemical parameters. Samples were taken from the main watercourse after homogenizing the addition of (polluted) sources along river Berembun. During average water flow (AWF) and high water flow (HWF) represented the status of the rivers during the average or dry water flow periods and during the high water flow period as experienced during the rainy season. This depends on the water rates and riverbed structure of the water streams (Eisakhani and Malakahmad, 2009).

RESULTS AND DISCUSSION

The water samples collected were analysed and the physic-chemical parameters are shown in Table 1.

Table 1: Physico-chemical parameters of water at Berembun river and permissible standards limits of WHO, EC and BIS for comparison.

Parameters	Unit	Min	Max	WHO Standard	EC Standard	BIS Standard
pH		5.72	8.02	6.5-8.5	6.5-8.6	6.5-8.5
EC	µS/cm	0.01	58.50	-	-	-
TDS	mg/L	0.01	0.11	1000	1000	500
BOD	mg/L	0.00	6.41	20-30	-	-
COD	mg/L	0.00	132.00	50	-	150
Phosphate	mg/L	0.00	112.50	10	5	-
Ammonia	mg/L	0.00	6.09	1.5	0.5	-
Sulphate	mg/L	0.00	250.00	400	25	100
Nitrate	mg/L	0.01	24.50	50	10	45
Potassium	mg/L	0.95	4.08	10	10	-
Sodium	mg/L	6.34	15.39	200	175	-
Calcium	mg/L	24.13	52.17	75	-	75
Magnesium	mg/L	3.86	8.01	30	-	30
Iron	mg/L	314.83	1079.91	0.3	0.2	0.3
Boron	mg/L	7.00	146.99	-	-	-
Copper	mg/L	0.86	11.26	-	-	-
Cadmium	mg/L	0.97	9.63	-	-	-
Manganese	mg/L	4.15	10.12	0.05	-	0.1
Zinc	mg/L	-12.01	38.25	-	-	-
Lead	mg/L	-9.58	172.04	-	-	-
Chromium	mg/L	1.50	15.00	-	-	-
Cobalt	mg/L	-0.02	0.05	-	-	-

* WHO=World Health Organization, * EC=European Community, * BIS=Bureau of Indian Standard

The pH value of water sample in the study area ranged from 5.72 to 8.02 during average water flow and during high water flow. Chau and Jiang (2002) have indicated that natural river water is slightly acidic because of its origin of rain water and because of tannin and leave acids released from the forest floors. Any increase in the pH is thus likely as consequence of acid-forming substances such as sulphate, phosphate, and nitrates release into the river basin. These substances as abundance in fertilizer usage might have altered the acid-base equilibria, resulted in the reduced acid-neutralizing capacity and hence raising the value of pH (Razak *et al.,* 2009).

Electrical conductivity is valuable indicator of the amount of material dissolved in water; and was found from 0.01 to 58.50 during average water flow. While the amount of total dissolved solids (TDS) was 0.01 to 0.11 mg/L during this time, which was relatively low. TDS indicates the general nature of water quality or salinity. The undisturbed and unpolluted rivers of the Cameron Highlands flowing through the forests display a very low EC and TDS. It can be supported by similar result in the research that was done by Hashim and Wan Yusoff (2003). This is mainly due to the origin of the river water of rain water and due to the inert stream bank material (mainly granite). In rainy season, due to floods and rains, water level in river increases which contains more electrolytes.

Water analysis parameters such as chemical oxygen demand (COD) in the range 0.00 - 132.00 mg/L was higher than WHO standards due to originates from sewage and fertilizers (animal fertilizers as chicken manure), through point sources (hardly treated domestic sewage) and non-point sources as agricultural runoff. While biological oxygen demand (BOD) is within the permissible limits as shown in Table 1 but the addition of nutrient-content of the water in some forms of nitrogen and phosphorus will increase BOD and COD.

Higher concentrations of nutrients and heavy metal were expected in high water flow because of agricultural runoff by comparing with WHO norms, these parameters were much higher than the permissible limits (Table 1). During agricultural runoff; nutrients stored in the surface soil layers are released with rain into the river. The concentration of nutrient in average water flow is likely to be almost solely from domestic wastewater and will be more diluted in high water flow. Nitrates are considered better indicators in average water flow, due to their relatively good solubility and non-reactive behaviour. Phosphate however is tightly bonded in the surface soil layers and to sediment and thus found in higher concentration during high water flow (Hashim and Wan Yusoff, 2003). This might be due to over-application of fertilizer, improper manure management practices, improper operation and maintenance of septic system and the decomposition of organic debris added from soil erosion and the river load. The newly submerged plants and grasses, the garbage and agricultural waste might also have an impact on the nutrient enrichment of soil sediment (Devi *et al.*, 2008). Beside deforestation the natural environment was degraded by mechanical excavations and other earthworks, and compensate for the negative effects of soil erosion on productivity by extensive use of irrigation, chicken manure, lime, and inorganic fertilizers.

CONCLUSION

It is important to understand the relationship between human induced disturbances and their effect on water resources. Water pollution sources are grouped as point sources (sewage and solid waste) and non-point sources (agricultural and urban runoff). The water quality of the Berembun river deteriorates because of the huge increase of suspended solids, the high concentrations of nutrient and heavy metal including COD, which cause very significant enrichment. Proper management of river and stream systems must be based upon a comprehensive monitoring strategy that is able to detect degradation in streams water quality due to human disturbance.

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REFERENCES

Abdul-Razak, A., Asiedu, A.B., Entsua-Mensah, R.E.M. and deGraft-Johnson, K.A.A. 2009). Assessment of the Water Quality of the Oti River in Ghana. *West African Journal of Applied Ecology*, **15**, 1-12.

Allen, B.J., Bourke, R.M. & Hide, R.L. 1995. The sustainability of Papua New Guinea agricultural systems: the conceptual background. *Global Environmental Change*, **5**(4): 297-312.

Aminuddin, B.Y., Chow, W.T. & Ng. T.T. 1990. Resources and problems associated with sustainable development in upland areas in Malaysia. Blair, G. & Lefroy, R. (eds.). *Technologies for sustainable agriculture in marginal uplands in Southeast Asia:* 55-61. Proceedings No. 33 Australian Centre for International Agricultural Research, Canberra.

Devi, R., Tesfahune, E., Legesse W., Deboch, B., Beyene. A., 2008. Assessment of siltation and nutrient enrichment of Gilgel Gibe dam, Southwest Ethiopia. *Bioresource Technology* **99**:975–979

Eisakhani, M., and Malakahmad, A. 2009. Water quality assessment of Bertam river and its tributaries in Cameron Highlands, Malaysia. *World Applied Science Journal* 7(6):769-776

Forsyth, T. 2007. Sustainable livelihood approaches and soil erosion risks: who is to judge? *International Journal of Social Economics,* **34**(1-2): 88-102.

Hashim G.M. and Wan Yusoff, W.A. 2003. Environmental Issues In Highlands Agriculture, *Malaysian Agricultural Research and Development Institute (MARDI).*

Hamdan, M. E., Man, N., Md. Yassin, S., D'Silva, J. L. and Mohamed Shariff, H. A., (2014). Farmers sensitivity towards the changing climate in the Cameron HIghlnads. *Agricultural Journal* **9** (2): 120-126.

Chau, K. W. and Jiang, Y. W. 2002. Three-dimensional pollutant transport model for the Pearl River Estuary. *Water Research,* **36**(8):2029-2039.

Midmore, D.J., Jansen, H.G.P. & Dumsday, R.G. 1996. Soil erosion and environmental impact of vegetable production in the Cameron Highlands, Malaysia. *Agriculture, Ecosystems and Environment* **60**(1): 29-46.

Rerkasem, K., Korsamphen, C., Thong-ngam, C., Yimyam, N. & Rerkasem, B. 2002. Agrodiversity lessons in mountain land management. *Mountain Research and Development,* **22**(1): 4-9.

Wan Abdullah, W.Y., Aminuddin, B.Y. & Zulkifli, M. 2005. Modelling pesticide and nutrient transport in the Cameron Highlands, Malaysia agroecosystems. *Water, Air, & Soil Pollution: Focus,* **5**(1-2): 115-123.

Wong, N.C., Lee, B.S., Yuen, P.M., Wan Abdullah, W.Y. & Ridzuan, M. 2002. Effects of continuous monocropping of chrysanthemum flowers under rain shelters on soil quality. Shamshuddin J., Hamdah J. & Samsuri A.W. (eds.). *Sustainable land management*: 236-292. Kulala Lumpur: Malaysian Society of Soil Science.

P6-9

Paddy soil nutrient classification using geospatial interpolation

Muhammad Zamir A. R.[1], ***Illani Zuraihah I.[1], Theeba M.[1], Nor Ziana Z. Z.[1], Noorsuhaila A. B.[1], Mohd Naim F. R.[1], Mohd Najib M. Y.[1], Hishamuddin A.[2], Asnita A. H.[2], Masni M.[2], Noranizam M. S.[2]***
[1]Malaysian Agricultural Research and Development Institute (MARDI), Serdang Selangor
[2]Department of Agriculture Malaysia (DOA), Putrajaya
; zamirar@mardi.gov.my

INTRODUCTION

Soil fertility is one of the main factors associated with the increased productivity of rice which is influenced by several key factors such as soil pH, cation exchange capacity (CEC), and nutrient availability. Spatial interpolation studies with regards to surface soil nutrients is a norm in many parts of the world. These include Inverse distance weightage (IDW) and ordinary kriging (OK) methods that are some of the many types of soil property mapping (Wu, et al., 2017). It is often a challenge to select best spatial interpolation based on available sets of data mainly because there are no consistent findings for best end results where their best performance depends on many factors (Liu, Du, Zhao, & Zhang, 2016) (Shope & Maharjan, 2015). IDW is also used for various interpolation such as for climate (Shope & Maharjan, 2015) (Rathnayake, De Silva, & Dayawansa, 2016). IDW is a simple and basic linear method (Lee, Yoo, Choi, & Engel, 2016) performs by using weighted function onto cell values of the sampling points whereby neighboring cells will have most influence of the interpolation, allowing more detail (Yang, Xie, Liu, Ji, & Wang, 2015), and fast to compute values (Islam, Shen, Hu, & Rahman, 2017).

This study under the development project of Soil Profiling (Site specific nutrient management for paddy) Program is expected to address this issue by increasing paddy crop yield by applying sufficient and efficient nutrient inputs in the form of fertilizer application with the aid of spatial interpolation.

MATERIALS AND METHODS

a. Study site and soil sampling

This study was conducted in two separated locations, which is in IADA Pulau Pinang, focusing in Seberang Prai Selatan region and IADA Kemasin Semarak, covering of approximately 1,357 hectares and 2,258 hectares of paddy land concurrently. A total of 355 soil sampling points in IADA Pulau Pinang and 512 soil sampling points in IADA Kemasin Semarak were taken, analyzed, and compiled by the Department of Agriculture Malaysia. Summary of the results can be found on Table 1.

Table 1: Soil sampling results for IADA SPS and IKS

		pH	CEC (cmol/kg)	N	P (mg/kg)	K (cmol/kg)
SPS	Mean (average)	5.95	25.52	0.27	42.65	0.87
	Min	4.40	17.39	0.07	6.00	0.17
	Max	7.70	36.99	0.51	514.00	2.68
	Variance (s²)	0.4035	13.0599	0.0041	2,185.04	0.1818
	Std Dev (s)	0.6352	3.6138	0.0638	46.7444	0.4263
IKS	Mean (average)	4.74	10.34	0.23	39.95	0.25
	Min	3.20	0.69	0.01	1.00	0
	Max	6.91	34.40	7.00	424.00	2.14
	Variance (s²)	0.4082	28.32	0.1024	3,467.69	0.0702
	Std Dev (s)	0.6389	5.3216	0.3200	58.8871	0.2649

b. Classification scheme

Soil classification scheme concept as illustrated in Figure 1.

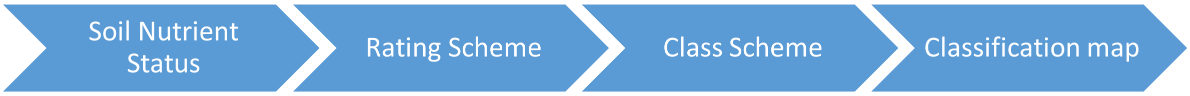

Figure 1: Classification outline workflow

Table 2: Soil nutrient rating scheme

Category	N	P	K	Rating
High	≥ 0.25	≥ 17	≥ 1.00	3
Medium	0.11 - 0.24	11 – 16	0.31 – 0.99	2
Low	≤ 0.10	≤ 10	≤ 0.30	1

The 3 classes, are as shown in Table 3.

Table 3: Overall classification scheme

Classification	NPK Rating	pH	CEC
1	7 – 9	≥ 5.0	≥ 20
2	5 – 6	4.5 – 5.0	11 – 19
3	3 – 4	3.5 – 4.5	≤ 10
4	-	≤ 3.5	-

RESULTS AND DISCUSSION

In IADA Pulau Pinang (Seberang Prai Selatan), it shows that the dominant soil pH above 5 at 96% (Figure 2a), and soil NPK class 1 (rating 7-9) values at 99% (Figure 2c). This indicate that most of the soil elements under study was dominantly high in all ranges and classes.

Under similar circumstances, IADA Kemasin Semarak shows variation and spread of classes throughout the district. For soil pH, it shows that the classification coverage is almost evenly distributed at 3.5-4.5 (35%), 4.5-5.0 (32%) and above 5.0 (31%) with exception of pH value lower than 3.5 (0.40 %) (Figure 2b). Most of the higher pH value covers the North Eastern and South Western part of the study area. For soil NPK ratings (Figure 2d), lower class 3 (rating 3-4) covers 8% of the total area, class 2 (rating 5-6) shows area coverage of 58%, and higher class 1 (rating 7-9) covers 32% from the total area under study.

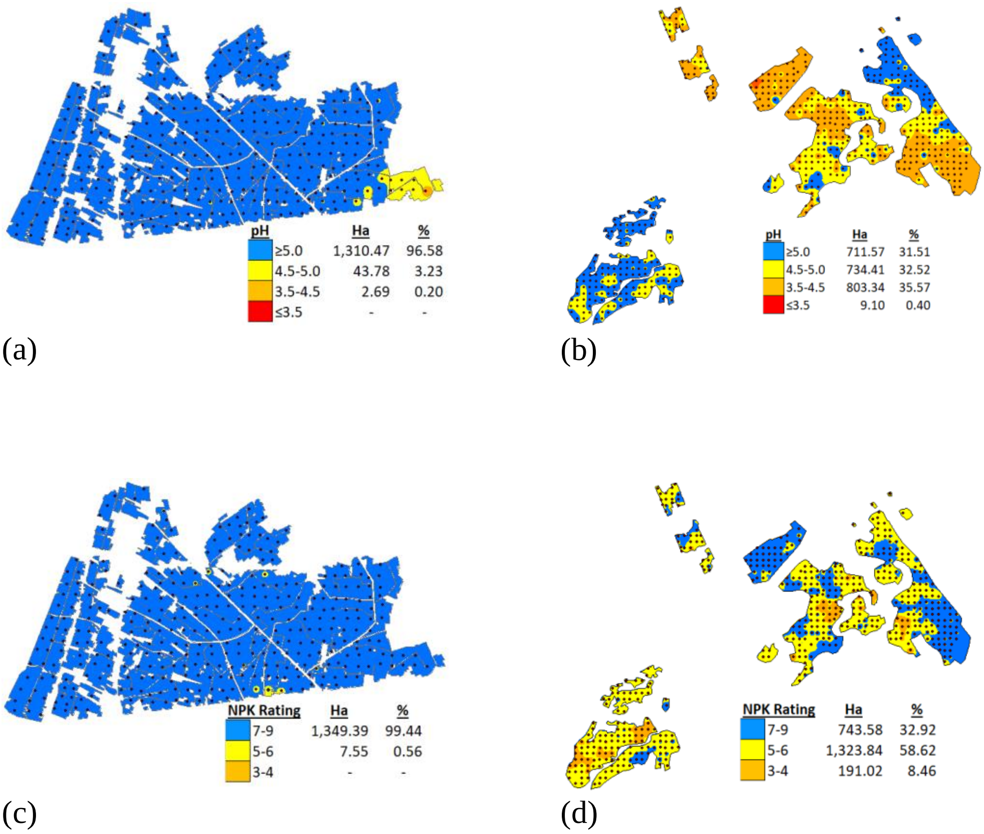

Figure 2: Results for IDW interpolation for IADA Seberang Prai Selatan (a), (c) and for IADA Kemasin Semarak (b), (d).

CONCLUSION

From this study, it can be concluded that IADA Pulau Pinang can be classified under singular class 1 soil because of its overall dominant higher values and area coverage in soil pH, and NPK ratings. Whereas in IADA Kemasin Semarak, the soil condition shows a wide variation of classes. It can be summarized that IADA Kemasin Semarak can be considered under two separated soil classes; class 2 and class 3 soils according to its location.Overall, the interpolation maps can be of use for further analysis especially concerning soil inputs for paddy cultivation, such as using soil pH maps for liming application and soil NPK maps for fertilizer application.

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REFERENCES

C**hen, R., Zhong, L., Jing, Z., Guo, Z., Li, Z., Lin, X., & Feng, Y. (2017).** Fertilization decreases compositional variation of paddy bacterial community across geographical gradient. *Soil Biology & Biochemistry*, 181-188.

I**slam, A. R., Shen, S., Hu, Z., & Rahman, M. A. (2017).** Drought Hazard Evaluation in Boro Paddy Cultivated Areas of Western Bangladesh at Current and Future Climate Change Conditions. *Advances in Meteorology*, *2017*, 12.

Ja**mil, Z. (n.d.).** Penyaluran bantuan dan insentif kepada pesawah-pesawah. *Lahir Peladang Progresif*, pp. 11-13.

Lee, S.-H., Yoo, S.-H., Choi, J.-Y., & Engel, B. A. (2016). Effects of climate change on paddy water use effciency with temporal change in the transplanting and growing season in South Korea. *Irrig Sci*, 21.

L**iu, W., Du, P., Zhao, Z., & Zhang, L. (2016).** *An Adaptive Weighting Algorithm for Interpolating the Soil Potassium Content.* www.nature.com/scientificreports/.

Rathnayake, W., De Silva, R., & Dayawansa, N. (2016). Assessment of the Suitability of Temperature and Relative Humidity for Rice Cultivation in Rainfed Lowland Paddy Fields in Kurunegala District. *Tropical Agriculture, 27*(4), 370-388.

S**hope, C. L., & Maharjan, G. R. (2015).** Modeling Spatiotemporal Precipitation: Effects of Density, Interpolation, and Land Use Distribution. *Advances in Meteorology*, *2015*, 16.

W**u, Q., Li, Q., Gao, J., Lin, Q., Xu, Q., Groffmanand, P. M., & Yu, S. (2017).** Non-Algorithmically Integrating Land Use Type with Spatial Interpolation of Surface Soil Nutrients in an Urbanizing Watershed. *Pedosphere*, *27*(1), 147-154.

Yang, X., Xie, X., Liu, D. L., Ji, F., & Wang, L. (2015). Spatial Interpolation of Daily Rainfall Data for Local Climate Impact Assessment over Greater Sydney Region. *Advances in Meteorology*, *2015*, 12.

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The effect of landfill leachate on surface water quality at ayer hitam sanitary landfill, Puchong, Selangor, Malaysia

***Nurul Atigah Ahmad*[1], *Daljit Singh Karam*[1], *Keeren S. Rajoo*[1]**
[1]Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, Selangor, Malaysia
; atiqahahmad_94@yahoo.com

INTRODUCTION

Landfills are site deposition of waste materials or also call as dumping site of waste materials. Landfill site can produce environmental pollution resulted from their landfill leachates and gas emission (Ismail and Manaf, 2013). Landfill leachate is the runoff water that occurs from operational or closed landfills. This leachate is in liquid form that flow from the landfills and contain either dissolved and suspended materials (Raghab *et al.*, 2013). The leachate or water flow will bring along heavy metals and also other chemicals from the contaminated soil and transfer them to the nearby water source such as river, lake or sea and cause water pollution. River at Ayer Hitam Sanitary Landfill (AHSL), Puchong, Selangor is surrounded by residential areas. The water from the river gave foul smell to the area. These have attracted insects and caused health disruption to people around the area. The people from the residential areas had filed a complain of this situation a long time ago but some people still use the water for living purposes. Therefore, this study is carried out to investigate the effect of landfill leachate on surface water quality after the sanitary landfill had been closed.

MATERIALS AND METHODS

The study was conducted at the river connected to the Air Hitam Sanitary Landfill, Puchong, Selangor, Malaysia. Air Hitam Sanitary Landfill (AHSL) is located near the Air Hitam Forest Reserve in Mukim Petaling, Daerah Petaling, Puchong, Selangor with the longitude 101° 39’ 55” E and latitude 03° 0’ 10” N. The water samples were collected along the river of AHSL and labelled as Station 1 until Station 10. The starting point of water collection (Station 1) was started from the point of origin of the waste water flow to the end point of water collection (station 10) which is towards the residential areas. The distance between each station is 120 cm and the water sample was taken 10 cm below the depth of surface water. The in-situ measurement such as pH and electrical conductivity (EC) and the ex-situ measurement such as ammoniacal nitrogen (NH_3-N), biochemical oxygen demand (BOD), chemical oxygen demand (COD), total suspended solid (TSS), heavy metal content such as cadmium (Cd), copper (Cu), arsenic (As) and chromium (Cr) were analysed. The in-situ measurement of water was conducted for pH and EC. The ex-situ measurement of BOD was analysed using 5 days test (HACH, 2003). The NH_3-N was analysed using Nessler reagent and COD was analysed using DRB 200 Reactor. The heavy metals in water was analysed using ICP-OES. The data of BOD, NH_3-N, TSS, DO, pH and COD were used for water quality index (WQI) classification. The statistical analysis of parameters and heavy metals of water samples were executed by using SAS 9.4 (One-way ANOVA, CRD method). The significance of the data ($p<0.05$) were analysed by using post hoch tests with Tukey’s method.

RESULTS

The average of pH, EC, TSS and NH_3-N did not exceed the permissible limits used by DOE. However, BOD and COD content in the water exceeded the suitable limits for direct use and needed extensive or advanced treatment before it can b utilized. The average of WQI at AHSL were classified as Class III where the water can be used as water supply but with some extensive or advanced treatment and the result of WQI was categorised as slightly polluted by DOE. The Cr, Cd, Cu and As of water did not exceed permissible limit stated by DOE.

Table 1: Water value of AHSL

	STATION									
PARAMETER	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
pH	6.74^{a}	6.22^{c}	6.77^{a}	6.54^{abc}	6.68^{ab}	6.76^{a}	6.70^{ab}	6.31^{bc}	6.43^{abc}	6.71^{ab}
EC (µS/cm)	196.00^{f}	208.67^{e}	223.33^{d}	235.33^{c}	226.00^{d}	242.67^{a}	240.67^{ab}	242.33^{a}	237.33^{bc}	24.67^{ab}
NH_3-N (mg/L)	$0.27c^{d}$	0.18^{e}	0.32^{bc}	0.20^{ed}	0.89^{a}	0.05^{f}	0.16^{e}	0.35^{b}	0.22^{de}	0.01^{f}
TSS (mg/L)	142^{h}	291^{d}	137^{h}	1015^{b}	198^{f}	225^{e}	178^{g}	337^{c}	1379^{a}	177^{g}
BOD (mg/L)	3.93^{bc}	4.23^{ab}	4.60^{a}	4.23^{ab}	3.30^{c}	1.60^{d}	0.33^{f}	1.00^{de}	0.76^{ef}	0.63^{ef}
COD (mg/L)	38.67^{e}	35.00^{f}	50.33^{bc}	27.67^{h}	32.33^{g}	51.67^{ab}	53.67^{a}	48.33^{c}	42.67^{d}	38.33^{e}
As (mg/L)	0^{d}	0^{d}	0.005^{c}	0^{d}	0^{d}	0^{d}	0^{d}	0.009^{b}	0.016^{a}	0.006^{bc}
Cu (mg/L)	0.03^{a}	0.002^{b}	0.001^{b}	0.002^{b}	0.004^{b}	0.006^{b}	0.007^{b}	0.001^{b}	0.001^{b}	0.001^{b}
Cd (mg/L)	0.003^{a}	0^{b}	0^{b}	0^{b}	0.001^{ab}	0.001^{ab}	0^{b}	0^{b}	0^{b}	0^{b}
Cr (mg/L)	0.002^{ab}	0^{b}	0.003^{ab}	0^{b}	0.002^{ab}	0^{b}	0.003^{ab}	0.003^{ab}	0.005^{a}	0.003^{ab}

Note: Means with different letters within the row indicate significant difference $p<0.05$

CONCLUSION

Landfill site still contaminates the river even after the site was closed. The water was not contaminated with heavy metals but the water quality index (WQI) showed the water still need to be treated before being used. The order of heavy metal contents from entire river water samples are Cu > As > Cr > Cd.

REFERENCES

Ismail, S.N.S and Manaf, L.A. (2013). The challenge of future landfill: A case study of Malaysia. *Journal of Toxicology and Environmental Health Sciences*, **5**(6): 86-96.

DOE (2007). Malaysia Environmental Quality Report 2006, pp: 24.

HACH, DR/500 spectrophotometer procedure manual, Loveland, Colorado: Hach Company **(2003).**

Raghab, S.M., El Meguid, A.M.A. and Hegazi, H.A. (2013). Treatment of leachate from municipal solid waste landfill. Housing and Building National Research Center, Cairo, Egypt, **9**:187

NOTES

LIST OF PARTICIPANTS

Amal A.M. AL-GHAMDI
Department of Botany, Environment Program, Faculty of Biological Sciences, King Abdul Aziz University, Jeddah, Saudi Arabia
amalalgamdi@gmail.com

Aida Azwani Abdul RAHIM
Department of Land Management, Faculty of Agriculture, UPM
aidaazwani@gmail.com

Aiza Shaliha JAMALUDDIN
Department of Forest Management, Faculty of Forestry, UPM
aiza.jamaluddin@gmail.com

Amirul Farid Bin Abd MALEK
FELDA Global Venture (FGV), Lot 775, Lorong Hussein, KM8, Kandang, 75460, Melaka
amirulfarid.am@feldaglobal.com

Abdul Razak bin IJANG
Sime Darby Research Sdn Bhd (Company No. 560590)
KM10, Jalan Banting-Kelanang
42700 Banting, Selangor
abdul.razak.ijang@simedarbyplantation.com

Amelia Md SOM
UniKL MICET, Bandar Vendor, Taboh Naning, 78000 Alor Gajah, Melaka
ameliamds@unikl.edu.my

Adibah Mohd AMIN
Department of Land Management, Faculty of Agriculture, UPM
adibahamin@upm.edu.my

Arina Shairah Abdul SUKOR
Department of Land Management, Faculty of Agriculture, UPM
shairah@upm.edu.my

Ali Tan Kee ZUAN
Department of Land Management, Faculty of Agriculture, UPM
tkz@upm.edu.my

Akio MORITA
College of Agriculture, Academic Institute Shizuoka University, 836 Ohya, Suruga-ku, Shizuoka, 422-8529 Japan
morita.akio@shizuoka.ac.jp

Azeri Bin AZAMI
Bhgn. Padi, Tanaman Industri & Florikultur, Pusat Pertanian Negeri, 21800 Ajil, Terengganu
ptif_trg@yahoo.com ;
azeri@doa.gov.my

Abdullah bin Abdul RAHMAN
Human Resources & Administration, Sime Darby Research Sdn Bhd KM 10., Jalan Banting-Kelanang, 42700 Banting Selangor
abdullah.rahman@simedarbyplantation.com

Abdullah Al MAHMUD
Dept. of Soil Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh
mkuddin@upm.edu.my

Abdul Haziq Qayyum Bin. OTHMAN
MADA Rice Training Center,
KM 4, Jalan Langgar,
05400 Alor Setar, Kedah
komplekslatihanmada@gmail.com

Akitomo KAWASAKI
CSIRO Agriculture and Food, Canberra, ACT 2601, Australia
Akitomo.Kawasaki@csiro.au

Asnita bt. Abu Harirah
Bahagian Pengurusan dan Pemuliharaan Sumber Tanah, No. 30, Persiaran Perdana, Presint 4, 62624 Putrajaya
asnita@doa.gov.my

Ahmad Saif bin Mohd Salmizi
Unit Pengurusan dan Pemuliharaan Sumber Tanah, Kompleks Pertanian Kuala Terengganu, KM. 07, Jalan Kuala Berang, 20050 Kuala Terengganu, Terengganu
ahmadsaif@doa.gov.my

Boney MUDA
MCB Cocoa Research and Development Centre, Mile 10, Apas Road, P.O. Box No. 60237, 91012 Tawau, Sabah, Malaysia
boneymuda@koko.gov.my

Brigitta TóTH
Institute of Nutrition,
University of Debrecen
Egyetem Square 1.
Debrecen 4032
Hungary
btoth@agr.unideb.hu

Che Ku Hafeez Che Ku DENANG
Ladang Rakyat Trengganu Sdn. Bhd., KM1, Jalan Cherul, Bandar Cheneh Baru, 24000 Kemaman, Terengganu
hrdiv@lrtsb.com.my

Chao WANG
State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing, China
chwang@issas.ac.cn

Clea Anne V. CORSIGA
Visayas State University- Villaba, Villaba, Leyte, Philippines
vallejera_19@yahoo.com

Dar-Yuan LEE
College of Bioresources and Agriculture
Distinguished Professor, Dept. of Agricultural Chemistry
National Taiwan University
dylee@ntu.edu.tw

Dian FIANTIS
Department of Agrotechnology, Faculty of Agriculture, Andalas University, Indonesia
dianfiantis@yahoo.com

Che Fauziah ISHAK
Department of Land Management, Faculty of Agriculture, UPM
cfauziah@upm.edu.my

Daljit Singh KARAM
Department of Land Management, Faculty of Agriculture, UPM
daljitsingh@upm.edu.my

Elisa RUMPANG
TROPI Belaga
elisa@mpob.gov.my

Evans OUMA
Rongo University, Kenya
samgudu2002@yahoo.com

Eliyas b. SAAD
IADA Barat Laut Selangor,
Kuala Selangor
eliyas@moa.gov.my

Frankie LANYING
Soil Management Branch, Department of Agriculture, Sarawak, Jalan Badruddin, 93400 Kuching, Sarawak
f.lanying@sarawak.gov.my

Frazer MIDOT
Sarawak Tropical Peat Research Institute (TROPI), Lot 6035 Kuching-Kota Samarahan Expressway,
94300 Kota Samarahan, Sarawak
frayzm@gmail.com

Frena Monti GERAI
Sarawak Tropical Peat Research Institute (TROPI), Lot 6035 Kuching-Kota Samarahan Expressway,
94300 Kota Samarahan, Sarawak
frenaena@gmail.com

Farah Fazwa Md ARIFF
Plant Improvement Prog. Forestry Biotech Division, FRIM, 52109 Kepong, Selangor
farah@frim.gov.my

Faridah Binti MANAF
Plant and Soil Science Research Centre, Malaysian Agricultural Research and Development Institute (MARDI), 43400 Serdang, Selangor, Malaysia
faridah@mardi.gov.my

Frisa Irawan GINTING
Department of Soil Science, Faculty of Agriculture, Andalas University, Kampus Limau Manis, Padang 25163 Indonesia
dianfiantis@yahoo.com

Faustina Elfrida SANGGOK
Sarawak Tropical Peat Research Institute (TROPI), Lot 6035 Kuching-Kota Samarahan Expressway,
94300 Kota Samarahan, Sarawak
faustina270187@gmail.com

GUI Jie Lei
Institute of Plant Science and Resources, Okayama UniversityChuo 2-20-1, Kurashiki 710-0046, Japan
leiguijie@126.com

Haryati MANSOR
Program Teknologi Baja,
Pusat Penyelidikan Tanaman & Sains Tanah, Ibu Pejabat MARDI
ytmansor@mardi.gov.my

Hishamuddin bin Ahmad
Bahagian Pengurusan dan Pemuliharaan Sumber Tanah, No. 30, Persiaran Perdana, Presint 4, 62624 Putrajaya
hishamuddin@doa.gov.my

Hiroto YAMASHITA
Graduate School of Integrated Science and Technology, Department of Agriculture,Shizuoka University, Laboratory of Plant Functional Physiology
yamashita.hiroto.16@shizuoka.ac.jp

Hasbullah SYAF
Department of Soil Science, Faculty of Agriculture, University of Halu Oleo, Indonesia
hassyaf@yahoo.co.id

HOANG Han Nguyen
Graham Center for Agricultural Innovation (Charles Sturt University & NSW Department of Primary Industries)
hnguyen@csu.edu.au

Hiroyuki KOYAMA
Faculty of Applied Biological Sciences, Gifu University, Japan
koyama@gifu-u.ac.jp

Huiyong LV
Root Biology Center, Fujian Agriculture and Forestry University, 350002 Fuzhou, China
2446468167@qq.com

Hari Mohan MEENA
Department of Soil Science & Agricultural Chemistry, UAS, GKVK, Bengaluru, India
harimohanmeena46@gmail.com

Haruka SHIGETA
Graduate School of Horticulture, Chiba University, Japan
ahwa4256@chiba-u.jp

HOANG Huu Chien
Kuroshio Science Program, Graduate School of Integrated Arts and Sciences, Kochi University Japan
chienbk87@gmail.com

Hondong XIAO
Department of Horticulture, Foshan University, Foshan, 528000 Guangdong, China
xuewen.li@foxmail.com

HO Wai Mun
FRIM, 52109 Kepong, Selangor
howaimun@frim.gov.my

Imran SHABBIR
Department of Land Management, Faculty of Agriculture, UPM
imranshabbir111@gmail.com

Izzah binti Abd HAMID @ GHAZALI
Department of Crop Science, Faculty of Agriculture and Food Sciences, UPMKB, Sarawak Malaysia
izzahabdhamid@gmail.com

Isharuddin Md ISA
Department of Land Management, Faculty of Agriculture, UPM
imi@upm.edu.my

Illani Zuraihah IBRAHIM
Soil Science and Water Programme
Crop and Soil Science Research Centre
Malaysian Agricultural Research and Development Institute (MARDI)
MARDI Headquarters Serdang
illani@mardi.gov.my
illaniz@yahoo.com

Intan Nadhirah MASRI
MARDI, P.O. Box 12301
Pejabat Pos Besar, 50774
Kuala Lumpur, Malaysia
intannadhirah@mardi.gov.my

Ingrid HAGAROVÁ
Comenius University in Bratislava, Faculty of Natural Sciences, Institute of Laboratory Research on Geomaterials Mlynska dolina, Ilkovicova 6, 842 15 Bratislava 4, Slovakia
ingrid.hagarova@uniba.sk

Jicqueline Mitchell Varonica RATAI
Sarawak Tropical Peat Research Institute (TROPI), Lot 6035 Kuching-Kota Samarahan Expressway,
94300 Kota Samarahan, Sarawak
jicqueline@gmail.com

Joanna Sieciῃka
Institute of Agrophysics Polish Academy of Sciences in Lublin, Poland
j.wrobel@ipan.lublin.pl

John Anak JADOL
Pusat Penyelidikan Pertanian Semonggok, KM20, Jalan Puncak Borneo, Peti Surat 977, 93720 Kuching Sarawak
johnj2@sarawak.gov.my

Jian Feng MA
Institute of Plant Science and Resources, Okayama University, Chuo 2–20–1, Kurashiki 710–0046, Japan
maj@rib.okayama-u.ac.jp

Ji Feng SHAO
Institute of Plant Science and Resources, Okayama University, Chuo 2-20-1, Kurashiki, 710-0046, Japan
87445362@qq.com

Jeyanny VIJAYANATHAN
Soil Management Branch, Forest Plantation, Forest Research Institute Malaysia (FRIM), 52109 Kepong Selangor
jeyanny@frim.gov.my

Jiakun ZHENG
Root Biology Center, Fujian Agriculture and Forestry University, 350002 Fuzhou, China
17750297807@163.com

Jing CHE
Institute of Plant Science and Resources, Okayama University, Kurashiki 710-0046, Japan
wonderful1122@gmail.com

Jiuyu LI
Institute of Soil Science,
Chinese Academy of Sciences,
71 East Beijing Road
Nanjing 210008, China
Email: jyli@issas.ac.cn

Jun WASAKI
Graduate School of Biosphere Science,
Hiroshima University, Japan
junw@hiroshima-u.ac.jp

Kazuyuki INUBUSHI
Soil Science Lab, Faculty of
Horticulture, Chiba University,
Matsudo, CHIBA 277-8510 Japan
inubushi@faculty.chiba-u.jp

Kengo YOKOSHO
Institute of Plant Science and
Resources, Okayama University, 2-20-1
Chuo, Kurashiki, Okayama 710-0046,
Japan
k-yokosho@okayama-u.ac.jp

Khairul Hafiz bin Mohd Yusoff
Department of Land Management,
Faculty of Agriculture, UPM
my_hafiz@upm.edu.my

Kumaran M. RAJAGOPAL
Sime Darby Research Sdn Bhd
(Company No. 560590)
KM10, Jalan Banting-Kelanang
42700 Banting, Selangor
kumaran.rajagopal@simedarbyplantation.com

KONG Goon Siong
WFM Plantation Sdn Bhd, No.86-90,
Pusat Pedada, Jalan Pedada, 96000
Sibu, Sarawak
gsk3838@gmail.com

Khasifah MUHAMAD
Malaysian Agricultural Research and
Development Institute (MARDI),
Persiaran MARDI-UPM
43400 Serdang, Selangor
khasifah@mardi.gov.my

Kaori MATSUOKA
Institute of Fruit Tree and Tea
Science, NARO, Department of
Radioecology, Institute for
Environmental Sciences, 033-0123
Arusuhori 203, 17-126, Horikawa,
Misawa, Japan
kaori.matsuoka@hotmail.co.jp

Laynil Hakih bt. SULHAN
IADA Barat Laut Selangor,
Kuala Selangor
laynil@moa.gov.my

Leon V. KOCHIAN
Plant Physiologist, University of
Saskatchewan, Canada;
Associate Director and Canada
Excellence Research Chair in Global
Food Security, Global Institute for Food
Security
leon.kochian@gifs.ca

Lynette K. ABBOTT
UWA School of Agriculture and
Environment M079 & UWA Institute
of Agriculture,The University of
Western Australia, Perth,
WA 6009 Australia
lynette.abbott@uwa.edu.au

Lulie MELLING
Sarawak Tropical Peat Research
Institute (TROPI), Lot 6035 Kuching-
Kota Samarahan Expressway,
94300 Kota Samarahan, Sarawak
luliemelling@gmail.com

Ludan ZHANG
Root Biology Center, Fujian Agriculture
and Forestry University, Fuzhou,
350002, China
zcchen@fafu.edu.cn

LEE Chin Tui
FELDA Global Ventures Holdings
Berhad (FGV), Level 42, Menara
Felda, Platinum Park, Persiaran
KLCC 50088 Kuala Lumpur
ct.lee@feldaglobal.com

Min YU
Department of Horticulture, Foshan
University, Foshan 528000 Guangdong,
China
yumin0820@hotmail.com

M. JAHIRUDDIN
Department of Soil Science,
Bangladesh Agricultural University,
Mymensingh 2202, Bangladesh
m_jahiruddin@yahoo.com

Mohd Kamarulzaman ISMAIL
Department of Land Management,
Faculty of Agriculture, UPM
shairah@upm.edu.my

Mohd Zulhilmy ABDULLAH
SARAWAK Tropical Peat Research
Institute, Lot 6035, Kuching-Kota
Samarahan Expressway,
94300 Kota Samarahan,
Sarawak MALAYSIA.
mdzoolhillmy@gmail.com

Mohamed Helmi Bin SHARI
Malaysian Cocoa Board, 5th, 6th & 7th
Floor, Wisma SEDCO, Lorong Plaza
Wawasan Off Coastal Highway, Locked
Bag 211, 88999 Kota Kinabalu, Sabah
Mohamed.helmi@koko.gov.my

Muhamad Zaid bin Mohamed KHALID
Sime Darby Research Sdn Bhd,
(Company No. 560590), KM10,
Jalan Banting-Kelanang,
42700 Banting, Selangor
muhammad.zaid@simedarbyplantation.com

Milton Ferreira de MORAES
Federal University of Mato Grosso
(UFMT), Graduate Program of Tropical
Agriculture (UFMT), Research Fellow of
CNPq-Brazil (Ministry of Science and
Technology
moraesmf@ufmt.br

Muhammad Zamir bin Abdul RASID
Pusat Penyelidikan Tanaman dan Sains
Tanah, Ibu Pejabat MARDI,Persiaran
MARDI-UPM, 43400 Serdang
Selangor Darul Ehsan
zamirar@mardi.gov.my

Mohamed Hanafi MUSA
Department of Land Management,
Faculty of Agriculture, UPM
mmhanafi@upm.edu.my

Mohd. Yusoff bin Abd SAMAD
Department of Land Management,
Faculty of Agriculture, UPM
myusoffas@upm.edu.my

Mohd Firdaus Mohd ANUAR
Department of Land Management,
Faculty of Agriculture, UPM
m.firdaus@upm.edu.my

Muhammad Firdaus SULAIMAN
Department of Land Management,
Faculty of Agriculture, UPM
muhdfirdaus@upm.edu.my

Md Kamal UDDIN
Department of Land Management,
Faculty of Agriculture, UPM
mkuddin@upm.edu.my

Mohd Izuan Effendi HALMI
Department of Land Management,
Faculty of Agriculture, UPM
m_izuaneffendi@upm.edu.my

Mohd Rizal ARIFFIN
Department of Land Management,
Faculty of Agriculture, UPM
ra_rizal@upm.edu.my

Mohamed Hafeifi Bin BASIR
Program Pertanian Bandar dan
Florikultur, Pusat Penyelidikan
Hortikultur, MARDI Ibu Pejabat
43400, Serdang Selangor
hafeifi@mardi.gov.my

Mutsutomo TOKIZAWA
Global Institute for Food Security,
University of Saskatchewan, Canada
mutsutomo.tokizawa@gifs.ca

M. Abdulaha-Al BAGUY
State Key Laboratory of Soil and
Sustainable Agriculture, Institute of
Soil Science, Chinese Academy of
Sciences, Nanjing, China
mabaquy@gmail.com

Mohammad H. GOLABI
College of Natural and Applied Sciences, University of Guam, Mangilao, Guam-USA
mgolabi@triton.uog.edu

Mohd Khanif YUSOP
Department of Land Management
Faculty of Agriculture, UPM
ymkhanif@gmail.com

Mahmud Hossain SUMON
Dept. of Soil Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh
mahmud.ss@bau.edu.bd

Muhammad Rafiqul ISLAM
Dept. of Soil Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh
mkuddin@upm.edu.my

M. Mazibur RAHMAN
Dept. of Soil Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh
mkuddin@upm.edu.my

Mohamad Zin bin Mohd Supaat
Bahagian Pengurusan dan Pemuliharaan Sumber Tanah, No. 30, Persiaran Perdana, Presint 4, 62624 Putrajaya
mohdzin@doa.gov.my

Md. Mahmudul HASSAN
Department of Land Management, Universiti Putra Malaysia, Malaysia
mmhasanlimon93@gmail.com

Mohamad Sofian bin Omar
Bahagian Pengurusan dan Pemuliharaan Sumber Tanah, No. 30, Persiaran Perdana, Presint 4, 62624 Putrajaya
mohamadsofian@doa.gov.my

Mohd Rossli bin Yusoff
Unit Pengurusan dan Pemuliharaan Sumber Tanah, Jabatan Pertanian Lundang, 15990 Kota Bharu, Kelantan
rossli@doa.gov.my

Mohd. Nizar Khairuddin
Institute of Agrobiotechnology, Universiti Sultan Zainal Abidin, Campus Besut, 22200 Besut, Terengganu, Malaysia
mohdnizar7@yahoo.com

Noraini Md. JAAFAR
Department of Land Management, Faculty of Agriculture, UPM
j_noraini@upm.edu.my

Nur Qursyna Boll KASSIM
Fakulti Perladangan & Agroteknologi, Universiti Teknologi MARA (Melaka), Kampus Jasin, 77300, Merlimau
MELAKA
qursyna@uitm.edu.my

Noor Azlina Binti Mhd. ROSLI
MADA Rice Training Center
KM 4, Jalan Langgar
05400 Alor Setar, Kedah
komplekslatihanmada@gmail.com

Norakmal KHAIRUANUAR
Department of Land Management, Faculty of Agriculture, UPM
norakmalkhairuanuar@yahoo.com

Nurfadzilah Binti MARDIAN
Malaysian Cocoa Board, 5th, 6th & 7th Floor,
Wisma SEDCO, Lorong Plaza Wawasan Off Coastal Highway, Locked Bag 211, 88999 Kota Kinabalu, Sabah
nurfadzilah@koko.gov.my

Nurul Atiqah AHMAD
Department of Land Management, Faculty of Agriculture, UPM
atiqahahmad_94@yahoo.com

Norhaleza bt. SHAHBUDIN
Sime Darby Research Sdn Bhd
(Company No. 560590)
KM10, Jalan Banting-Kelanang
42700 Banting, Selangor
norhaleza.shahbudin@simedarbyplantation.com

Nur Azima BUSMAN
Sarawak Tropical Peat Research Institute,
Lot 6035, Kuching-Kota Samarahan Expressway,
94300 Kota Samarahan,
Sarawak MALAYSIA
zima_b67@yahoo.com

Noor Haslizawati Binti Abu BAKAR
Program Sains Tanah dan Air,
Pusat Penyelidikan Tanaman & Sains Tanah, Ibu Pejabat MARDI
Persiaran MARDI-UPM
43400 Serdang, Selangor
haslizawati@mardi.gov.my

Nur Liyana ISKANDAR
Soil Science and Water Programme,
Crop and Soil Science Research Centre, MARDI
liyanais@mardi.gov.my

Norziana Zin ZAWAWI
Program Sains Tanaman, Tanah dan Air, Pusat Penyelidikan Tanaman dan Sains Tanah, Ibu Pejabat MARDI, Serdang
zianazz@mardi.gov.my

Noor Azilah bt. Mohidin
Unit Pengurusan dan Pemuliharaan Sumber Tanah,
Kompleks Jabatan Pertanian Titi Gantong, 32600 Bota, Perak
noorazilah@doa.gov.my

Nurhajati HAKIM
Faculty of Agriculture, Andalas University, Indonesia
nhakimsa@yahoo.co.id

Osumanu Haruna AHMED
Department of Crop Science, Faculty of Agriculture and Food Sciences, UPM Bintulu, Sarawak Campus,Sarawak, Malaysia
osumanu@upm.edu.my
; osumanuharuna@gmail.com

Prakash NAGABOVANALLI
Department of Soil Science & Agricultural Chemistry, UAS, Bengaluru, India
nagabovanalliprakash@rediffmail.com

Pupathy Uthrapathy THANDAPANI
Sime Darby Research Sdn Bhd, (Company No. 560590-X), KM 10, Jalan Banting-Kelanang, 42700 Banting, Selangor, Malaysia
pupathy.uthrapathy@simedarbyplantation.com

Peter MATÚŠ
Comenius University in Bratislava, Faculty of Natural Sciences, Institute of Laboratory Research on Geomaterials, Mlynska dolina, Ilkovicova 6, 842 15 Bratislava 4, Slovakia
peter.matus@uniba.sk, matus1@uniba.sk

POONG Ken Heong
Omya Asia Pacific Sdn. Bhd.,
Unit 2-1, Level 2, UOA Corporate Tower, Avenue 10, The Vertical, Bangsar South, No. 8 Jalan Kerinchi
59200 Kuala Lumpur,

Premkumar TAMILARASAN
Sime Darby Research Sdn Bhd , (Company No. 560590), KM10, Jalan Banting-Kelanang, 42700 Banting, Selangor

Padmini KARANANIDI
Universiti Kuala Lumpur Malaysia, Institute of Chemical and Bioengineering Technology, Bandar Vendor Taboh Naning, Alor Gajah
78000 Melaka, Malaysia

Malaysia
Brian.Poong@omya.com

prem.tamilarasan@simedarbyplantation.com

padmini.karananidi@s.unikl.edu.my

Qian LI
Graduate School of Horticulture, Chiba University, Matsudo 648, Matsudo-shi 271-8510 Japan
liqian0223@yahoo.co.jp

Radziah Binti JACK
Soil Management Branch, Department of Agriculture, Sarawak, Jalan Badruddin, 93400 Kuching, Sarawak
radziahj@sarawak.gov.my

Renyong SHI
State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing, China
ryshi@issas.ac.cn

Rozita Binti OSMAN
Pusat Penyelidikan & Pembangunan Koko, Jalan Jengka 23, Peti Surat 34, 28000 Temerloh, Pahang
orozita@koko.gov.my

Rozilawati bt IKHSAN
Sime Darby Research Sdn Bhd (Company No. 560590), KM10, Jalan Banting-Kelanang, 42700 Banting, Selangor
rozilawati.ikhsan@simedarbyplantation.com

Ryan T. SARIMONG
Capiz State University, Burias, Mambusao, Capiz, Phillipines
ryansoiler4@yahoo.com

Rakiba SHULTANA
Department of Land Management, Faculty of Agriculture, UPM
rakiba_83@yahoo.com

Rosazlin ABDULLAH
Institute of Biological Science, Faculty of Science, Universiti Malaya, 50603 Kuala Lumpur
rosazlin@um.edu.my

Rosenani Abu BAKAR
Department of Land Management, Faculty of Agriculture, UPM
rosenani.abubakar@gmail.com

Ren Fang SHEN
State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing, China
rfshen@issas.ac.cn

Renkou XU
State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences, Nanjing, China
rkxu@issas.ac.cn

Radziah OTHMAN
Department of Land Management, Faculty of Agriculture, UPM
radziah@upm.edu.my

Suhaili bt. SAHARI
IADA Barat Laut Selangor, Kuala Selangor
suhaili@moa.gov.my

Shamshuddin JUSOP
Department of Land Management, Faculty of Agriculture, UPM
shamshud@upm.edu.my

Shyama Malika MALWALAGEE
Post Graduate Institute of Agriculture, University of Peradeniya, Kandy, Sri Lanka
shyamamalika@gmail.com

Siti Nasuha SUBOH
Department of Land Management, Faculty of Agriculture, UPM
nasuhasuboh92@gmail.com

Sudharsan SELVARAJA
FELDA Global Ventures Holdings Berhad (FGV), Level 42, Menara Felda, Platinum Park, Persiaran KLCC 50088 Kuala Lumpur
sudharsanselvaraja@gmail.com

Syafiqah Nabilah Samsul BAHARI
Plant Improvement Prog. Forestry Biotech, Division, FRIM, 52109 Kepong, Selangor
syafiqah@frim.gov.my

Sharon LAU Yu Ling
Sarawak Tropical Peat Research Institute (TROPI), Lot 6035 Kuching-Kota Samarahan Expressway, 94300 Kota Samarahan, Sarawak
lauyuling@gmail.com

Sheng HUANG
Institute of Plant Science and Resources, Okayama University, Chuo 2–20–1, Kurashiki 710–0046, Japan
pdqr7242@s.okayama-u.ac.jp

Susilawati KASIM
Department of Land Management, Faculty of Agriculture, UPM
susilawati@upm.edu.my

Samuel GUDU
Ag. Vice Chancellor, Rongo University, P.O. Box 103-40304 RONGO
samgudu2002@yahoo.com; vc@rongovarsity.ac.ke

Shilin Wen
Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing, China
caizejiang@caas.cn

Samsuri Abd. WAHID
Department of Land Management, Faculty of Agriculture, UPM
samsuriaw@upm.edu.my

Syaharudin ZAIBON
Department of Land Management, Faculty of Agriculture, UPM
syaharudin@upm.edu.my

SU Chong Ming
Sarawak Oil Palms Berhad, No. 124-126 Jalan Bendahara, P.O. Box 547, 98007 Miri Sarawak
monica.lok@sop.com.my

Syamsul Zuraida@Fadhilah bt. ZAINUDIN
IADA Barat Laut Selangor, Kuala Selangor
syamsulzuraida@water.gov.my

Shahril Shamsir@Fadly bin AMRAN
IADA Barat Laut Selangor, Kuala Selangor
shahrilsf@gmail.com

Syaliza Hanom bt. ZAKARIA
IADA Barat Laut Selangor, Kuala Selangor
syalizaiada@gmail.com

Tao LIANG
Guangdong Key laboratory of Agricultural Environment Pollution Integrated Control, Guangdong Institute of Eco-Environmental Science & Technology, Guangzhou 510650, P.R. China
taoliang@soil.gd.cn

Thilaga a/p BATUMALI
IADA Barat Laut Selangor, Kuala Selangor
thila81@yahoo.com

TAN Swee Sian
Advanced Agriecological Research Sdn. Bhd., No. 11, Jalan Teknologi 3/6, Taman Sains Selangor, 47810 Petaling Jaya, Selangor

Tasren Nazir MAHAMOOTH
Advanced Agriecological Research Sdn. Bhd., No. 11, Jalan Teknologi 3/6, Taman Sains Selangor, 47810 Petaling Jaya, Selangor

jobs@aarsb.com.my

jobs@aarsb.com.my

Takuo ENOMOTO
United, Graduate School of Agricultural Science, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan
v6103002@edu.gifu-u.ac.jp

TAN Ngai Paing
Department of Land Management, Faculty of Agriculture, UPM
ngaipaing@upm.edu.my

Takashi IKKA
College of Agriculture, Academic Institute Shizuoka University, 836 Ohya, Suruga-ku, Shizuoka, 422-8529 Japan
ikka.takashi@shizuoka.ac.jp

TANG Lai Kuen
Ecophysiology Branch, Forest Plantation Program, FRIM
tlkuen@frim.gov.my

Toru FUJIWARA
Laboratory of Plant Nutrition and Fertilizers, Department of Applied Biological Chemistry,Graduate School of Agricultural and Life Sciences,
The University of Tokyo
Yayoi, Bunkyo-ku, Tokyo 113-8657
atorufu@mail.ecc.u-tokyo.ac.jp

Wan Mohd Rusydan bin Wan Ibrahim
Unit Pengurusan dan Pemuliharaan Sumber Tanah, Jabatan Pertanian Negeri Selangor, Tingkat 3, Bangunan Darul Ehsan, No. 3, Jalan Indah, Seksyen 14, 40632 Shah Alam, Selangor
wmrusydan@doa.gov.my

Wan Asrina binti Wan YAHAYA
Department of Crop Science, Faculty of Agriculture and Food Sciences, UPMKB, Sarawak Malaysia
asrina@upm.edu.my

Wan Rasidah Bt. Wan. A KADIR
FRIM 52109 Kepong, Selangor
rashidah@frim.gov.my

Xiao Fang ZHU
State Key Laboratory of Soil and Sustainable Agriculture, Institute of Soil Science, Chinese Academy of Sciences
xiaofangzhu@issas.ac.cn

Xuewen LI
Department of Horticulture, Foshan University, Foshan, 528000 Guangdong, China
xuewen.l@hotmail.com

Xue Qiang ZHAO
State Key Laboratory of Soil and Sustainable Agriculture,
Institute of Soil Science, Chinese Acade my of Sciences, Nanjing 210008
China
xqzhao@issas.ac.cn

Xiaomei LIU
Root Biology Center, Fujian Agriculture and Forestry University, 35002, Fuzhou, Fujian, China
liuxiaomeitx@yeah.net

Yang LIU
Root Biology Center, Fujian Agriculture and Forestry University, 350002 Fuzhou, China
329129309@qq.com

Yuki NAKANO
The United Graduate School of Agricultural Science, Gifu University, 1-1 Yanagido, Gifu 501-1193, Japan
v6103005@edu.gifu-u.ac.jp

YAP Mui Lan
Sarawak Tropical Peat Research Institute (TROPI), Lot 6035 Kuching-Kota Samarahan Expressway, 94300 Kota Samarahan, Sarawak
maylan90@gmail.com

Yahya Awang
Department of Plant Science, Faculty of Agriculture, UPM
yahya_awg@upm.edu.my

Zaifiwan b. MAT ZIN
IADA Barat Laut Selangor, Kuala Selangor
zaifiwan@doa.gov.my

Zejiang CAI
Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing, China
caizejiang@caas.cn

ZhiChang CHEN
Root Biology Center,Fujian Agriculture and Forestry University, 350002, Fuzhou, Fujian, China
zcchen@fafu.edu.cn

Zhi ZHOU
Root Biology Center, Fujian Agriculture and Forestry University, 35002, Fuzhou, Fujian, China
2456850435@qq.com

Zulkifli Hj. Shamsuddin
Malaysian Society of Soil Science (MSSS)
zulsham08@gmail.com

Annur Bin Mohd Razib
Department of Land Management
Faculty of Agriculture, UPM
annurrazib@yahoo.com

Dayang Safinah
Department of Land Management
Faculty of Agriculture, UPM
cfauziah@upm.edu.my

Rabeatuladawiah
Department of Land Management
Faculty of Agriculture, UPM
cfauziah@upm.edu.my

Vajidah binti Sunoto@Hj. Faisal
Unit Pengurusan dan Pemuliharaan Sumber Tanah, Jabatan Pertanian, 86100 Ayer Hitam, Johor
vajidah@doa.gov.my

Mohan A/L Perumal
Sarawak Land Consolidation & Rehabilitation Authority (SALCRA)
No. 1, Lot 2220, Block 26, MTLD
Jalan Dato Mohd Musa
94300 Kota Samarahan, Sarawak
ndcg@salcra.gov.my
info@salcra.gov.my

Adelinna Libin
Sarawak Land Consolidation & Rehabilitation Authority (SALCRA)
No. 1, Lot 2220, Block 26, MTLD
Jalan Dato Mohd Musa
94300 Kota Samarahan, Sarawak
ndcg@salcra.gov.my
info@salcra.gov.my

Agatha Rengga
Sarawak Land Consolidation & Rehabilitation Authority (SALCRA)
No. 1, Lot 2220, Block 26, MTLD
Jalan Dato Mohd Musa
94300 Kota Samarahan, Sarawak
ndcg@salcra.gov.my
info@salcra.gov.my

Julian Patrick
Sarawak Land Consolidation & Rehabilitation Authority (SALCRA)
No. 1, Lot 2220, Block 26, MTLD
Jalan Dato Mohd Musa
94300 Kota Samarahan, Sarawak
ndcg@salcra.gov.my
info@salcra.gov.my