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ORIGINAL RESEARCH PAPER

Leaf size indices and structure of the peat swamp forest

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ABSTRACT: Leaf size indices of the tree species in the peatland of Agusan del Sur in Mindanao in Philippines was examined to deduce the variation of forest structure and observed forest zonation. Using raunkiaer and webb's leaf size classification, the leaf morphometrics of seven tree species consistently found on the established sampling plots were determined. The species includes *Ternstroemia philippinensis* Merr*., Polyscias aherniana* Merr. Lowry and G.M. Plunkett*, Calophyllum sclerophyllum* Vesque*, Fagraea racemosa* Jack*, Ilex cymosa* Blume*, Syzygium tenuirame* (Miq.) Merr. and *Tristaniopsis micrantha* Merr. Peter G.Wilson and J.T.Waterh. The LSI were correlated against the variables of the peat physico-chemical properties (such as bulk density, acrotelm thickness, peat depth, total organic carbon, nitrogen, phosphorus, and potassium, pH); water (pH, ammonium, nitrate, phosphate); and leaf tissue elements (nitrogen, phosphorus and potassium). Result showed a decreasing leaf size indices and a three leaf size category consisting of mesophyllous, mesophyllous-notophyllous and microphyllous were observed which corresponds to the structure of vegetation *i.e.*, from the tall-pole forest having the biggest average leaf area of 6,142.29 mm^2 to the pygmy forest with average leaf area of 1,670.10 mm^2. Such decreased leaf size indices were strongly correlated to soil nitrogen, acrotelm thickness, peat depth, phosphate in water, nitrogen and phosphorus in the plant tissue.

KEYWORDS: *Acrotelm; Peat swamp forest; Forest structure; Leaf size classification; Leaf size indices (LPI)*

INTRODUCTION

The variation in the leaf size of trees implies varied reasons. This could be due to light intensity and quality, adaptation mechanism and or plasticity, soil factors, seasonal variations and response to limiting factors. The size and structure of leaves vary not only with species, genotype, and habitat but also with location on a tree, between juvenile and adult leaves, between early and late leaves, and between leaves of early shoots and those of late-season shoots (Pallardy 2008). In general, leaf variation is influenced by climatic and edaphic conditions, and with altitude and latitude (Box 1981; Chabot and Hicks 1982; Givnish 1987; Grubb 1974; Liu 1993; Ohsawa and Ozaki 1992; Ohsawa 1993a; Reich *et al*. 1995; Richards 1996; Webb 1959; Whitmore 1984; Woodward 1987). Variations of leaf size also correspond to structure and physiognomy of forests on mountain ecosystems (Buot 1999). Malhado *et al.* (2009) concluded that leaf size is one of the most plastic traits of a tree. Traiser *et al.* (2005) stressed that the physiognomy of leaves can serve as an excellent tool for ecological studies hence, variation of leaf size provides direct indicators to the physico-chemical conditions operating on plants. In this paper, the pioneering work

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on leaf size classification by Raunkiaer (1934) and Webb (1959) were used to deduce the significance of leaf size indices (LSI) in relation to peat conditions and variation of forest structure and zonation in the peatland of Caimpugan with reference to the description we made which is a modification to that of Davies (2006) (*i.e.* tall pole forest, intermediate forest, sapling size, pygmy forest). The authors aimed also to further investigate the factors influencing the variation of leaf sizes by using the leaf tissue elements. This study has been carried out in Barangay Caimpugan, San Francisco, Agusan del Sur on October 2011 and December 2013.

MATERIALS AND METHODS

Study site

This study was conducted in the peat swamp forest of Barangay Caimpugan, San Francisco, Agusan del Sur on Mindanao Island, Philippines with geographic coordinates 8˚22’- 8˚27.5’N and 125˚45’-125˚55.6’E (Fig. 1). It is located approximately 33.47 kilometers away from the shorelines of Pacific Ocean. The peatland is among the major habitat types of Agusan Marshland comprising 5,630.31 hectares. The climatic condition falls under the Type II of the Corona classification characterised by evenly distributed rainfall throughout the year, with no distinct dry season and maximum rain period observed is from October to February. The peatland is categorised as oligotrophic and is bounded by two river systems, *i.e.* Gibong in the east and Agusan River in the west.

Sampling plots

Davies (2006) earlier recognised a three-vegetation zone based on forest structure comprising the tall-pole, intermediate and pygmy forest from the edge of Gibong River. However, the extent and boundary between the tall-pole forest and intermediate forest is not well-defined unlike the pygmy forest which is clearly demarcated by an abrupt change in tree heights and diameters.

In this study, sampling plots were laid-out randomly but following a straight path starting near the periphery towards the center of the peatland with consideration to the changes of stand structure and physiognomy. Hence, twelve sampling plots were established recognising a four forest zonation consisting of the tall-pole forest, intermediate forest, sapling forest, and pygmy forest. The recognised intermediate forest in this study is the forest stand characterised by a slightly

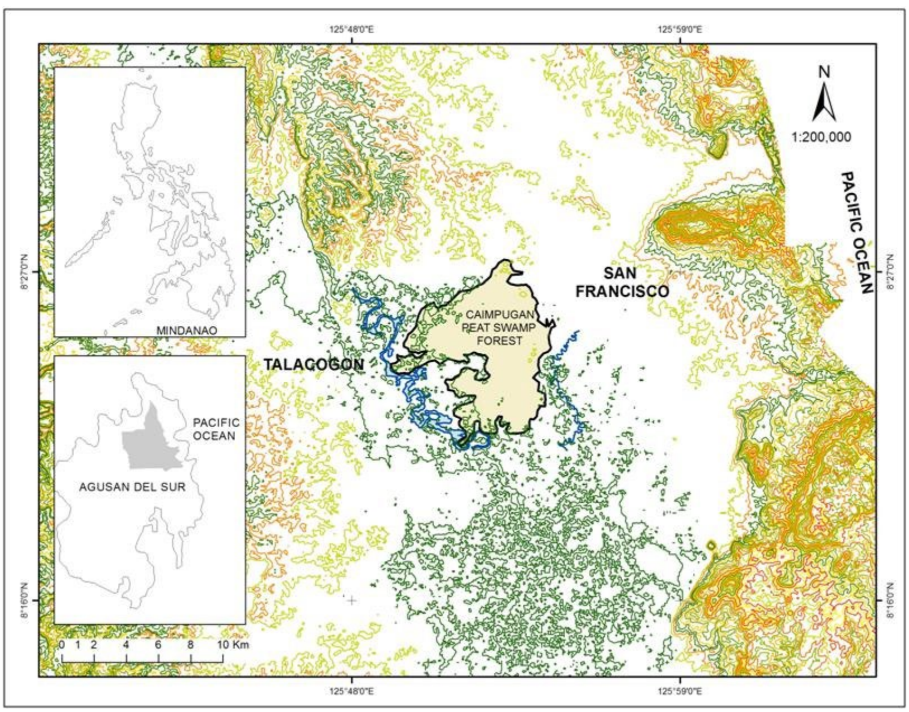

Fig. 1: Map of the study site

reduced diameters and heights compared to the trees of the tall-pole forest. This stand is situated prior to the intermediate forest recognised by Davies (2006) but now classified here as the sapling-size forest because trees have sapling-size diameters. From the tall-pole to the sapling-size forest, three 10x20m sampling plots were established while at the pygmy forest, three 10x10m was laid-out due to difficulty in access owing to the presence of void pools with water often exceeding beyond knee depth.

Tree measurement and identification

All trees with stem diameters of ≥2 cm at breast height (dbh) within the sampling plots were measured and identified. A tree altimeter was used to measure the total height of the trees from the stem base to the top of the crown. Trees were identified directly in the field however specimens were collected to reconfirm identification and for herbarium purposes. Specimens were tagged corresponding to the number entry on the data sheets.

Peat and water collection

Collections of soil samples were done randomly within the sampling plots using a probe to a depth of 30cm. Collected samples were mixed thoroughly and a composite sample of approximately 2 kg was prepared for analyses on variables such as soil pH, bulk density, total nitrogen, total phosphorus, total potassium and total organic carbon.

Peat depth and acrotelm

The thickness of acrotelm on each sampling plot was determined by measuring the surface layer up to the water table using a meter stick. Measurement was done within the holes where the soil and water samples were also collected. Similarly, the peat depth was determined using a peat sampler, a 10-meter calibrated stainless steel pole with drill-head at the tip where soil can be deposited thus enable determination of the underlying substrate.

Leaf collection and processing

The collections of leaf specimens were done on October 2011 and December 2013. Only the leaves of the tree species consistently found within the sampling plots established from the outer portion towards the center of the peat swamp forest were used in the study. This includes *Ternstroemia philippinensis*, *Polyscias aherniana*, *Calophyllum sclerophyllum*, *Fagraea racemosa*, *Ilex cymosa*, *Syzygium tenuirame*, and *Tristaniopsis micrantha*. To eliminate other potential source of variation, only the leaves from the lower portion of the crown were collected and used. These were oven-dried at 80°C for 5 days and measured to determine the LSI.

Leaf tissue analysis

A composite leaf samples consisting of five samples per specimen per vegetation zone were randomly collected to determine the variations in tissue element such as nitrogen, phosphorus, and potassium. The leaf samples were analyzed at the soil and plant analysis laboratory of Central Mindanao University.

Leaf size measurement

The one-sided leaf area was obtained using the formula of Cain and De Oliveira-Castro (1959) as used by Buot and Okitsu (1999) such as:

Leaf area = 2/3 (L x W)

where: L - full length of the leaf
W: width of the leaf at its widest portion

A table of leaf size classes by Malhado *et al*. (2009) (Table 1) earlier developed by Raunkiaer (1934) and revised by Webb (1959) was used to determine leaf categories.

Table 1: Leaf size classes (after Malhado *et al.* 2009)

Category	Dimensions (cm^2)	Raunkiaer (1934)	Webb (1959)	Merged categories
1	<0.25	Leptophyll	Leptophyll	
2	0.25 – 2.25	Nanophyll	Nanophyll	
3	2.25 – 20.25	Microphyll	Microphyll	Small leaves
4	20.25 – 45.00	Mesophyll	Notophyll	
5	20.25 – 182.25		Mesophyll	
6	182.25 – 1640.25	Macrophyll	Macrophyll	Large leaves
7	>1640.25	Megaphyll	Megaphyll	

RESULTS AND DISCUSSION

A total of 7,858 leaflets/leaves were measured from 72 voucher specimens. Species with the highest number of leaves includes *Syzygium tenuirame* and *Tristaniopsis micrantha* with 1,857 and 322, respectively (Table 2).

Variation in leaf sizes indices

Towards the center of the peat swamp forest, a decreasing trend on the average leaf area was observed from all the specimens measured (Fig. 2). The tall-pole forest obtained the highest average leaf length, width, and area of 122.65 mm, 46.39 mm, and 6,142.29 mm^2, respectively. The intermediate forest and sapling-size forest was observed to have closely similar results. In contrast, the pygmy forest had noticeable decrease in leaf size indices with the length, width and area of 77.35 mm, 32.41 mm and 1,670.10 mm^2, respectively.

Among the seven tree species, *Ternstroemia philippinensis* obtained the large variation in length ($r^2 = 0.5441$), width ($r^2 = 0.5246$) and area ($r^2 = 0.421$), respectively. This was followed by *Tristaniopsis micrantha*, with variation in length of ($r^2 = 0.3154$), width ($r^2 = 0.3706$) and area ($r^2 = 0.3419$), respectively. *Syzygium tenuirame* obtained the small degree of variations in length ($r^2 = 0.0068$), width ($r^2 = 0.0044$) and area ($r^2 = 0.00002$), respectively (Figs. 3, 4, 5).

Table 2: Number of leaves measured per specimen

Species	Number of specimens	Total number of leaves
Ternstroemia philippinensis	7	9
Polyscias aherniana	9	187
Calophyllum sclerophyllum	8	144
Fagraea racemosa	7	62
Ilex cymosa	4	112
Syzygium tenuirame	20	1857
Tristaniopsis micrantha	17	322
Total	72	7858

Variation in leaf macronutrients

The total nitrogen and phosphorus content was highest in the tall-pole forest with 1.07 and 0.468%, respectively however, the sapling-size forest had the highest total potassium content of 0.937%. The pygmy forest had the lowest values for all the leaf macronutrients (Table 3). A large variation was observed for the total nitrogen ($r^2 = 0.7503$) followed by total phosphorus ($r^2 = 0.6867$) while small variation in total potassium ($r^2 = 0.1699$) (Fig. 6).

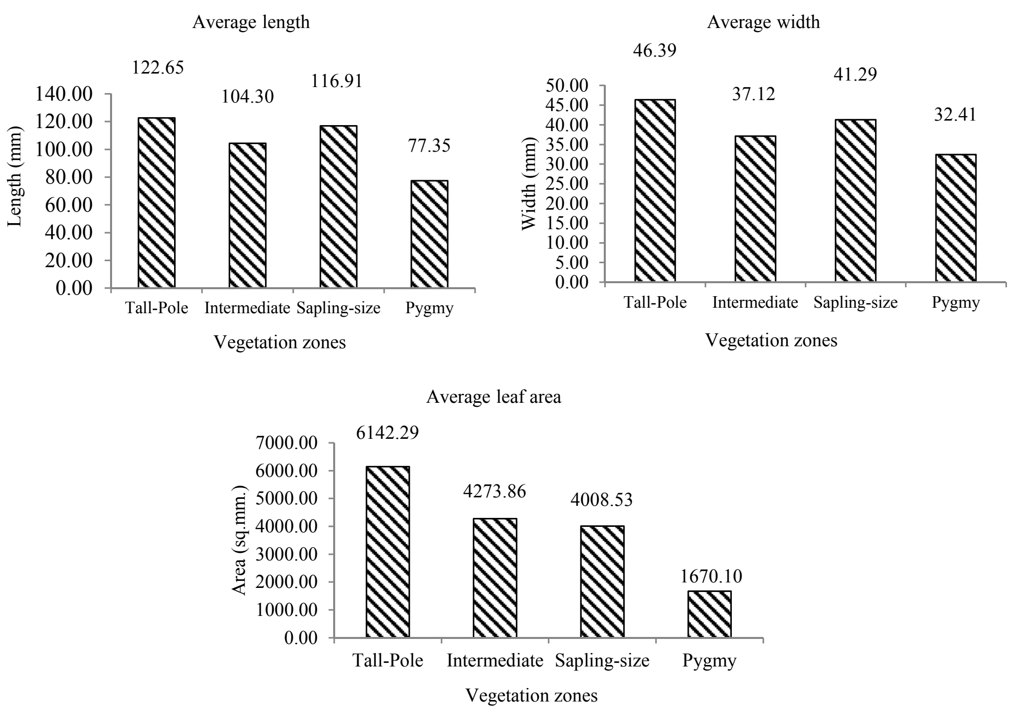

Fig. 2: Average leaf size indices of the various vegetation zones

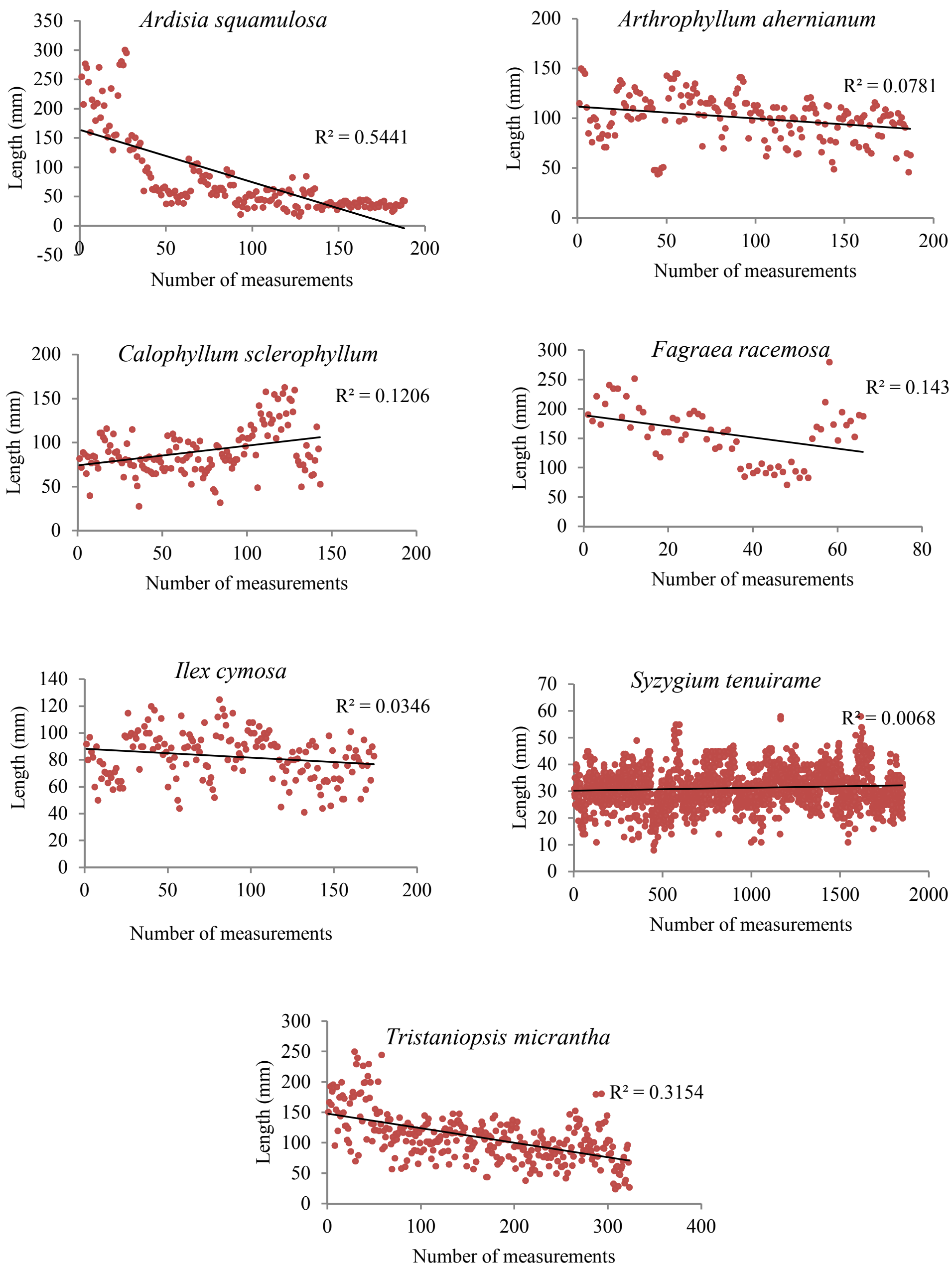

Fig. 3: Variation of leaf lengths of all the species used

Leaf-size classification

The classification of leaf size zones based on Raunkiaer (1939) and Webb (1959) system revealed that the tall-pole forest is categorised as mesophyllous, intermediate forest and sapling-size forest as mesophyllous-notophyllous, while the pygmy forest as microphyllous, respectively (Table 4).

Leaf-size indices and correlated variables

Results showed that the leaf area is positively correlated to phosphate in water, total nitrogen and phosphorus in tissue samples and acrotelm however, negatively correlated to peat depth ($P<0.05$). Also, the leaf length is positively correlated to total phosphorus and total nitrogen in tissue samples at $P<0.05$ and

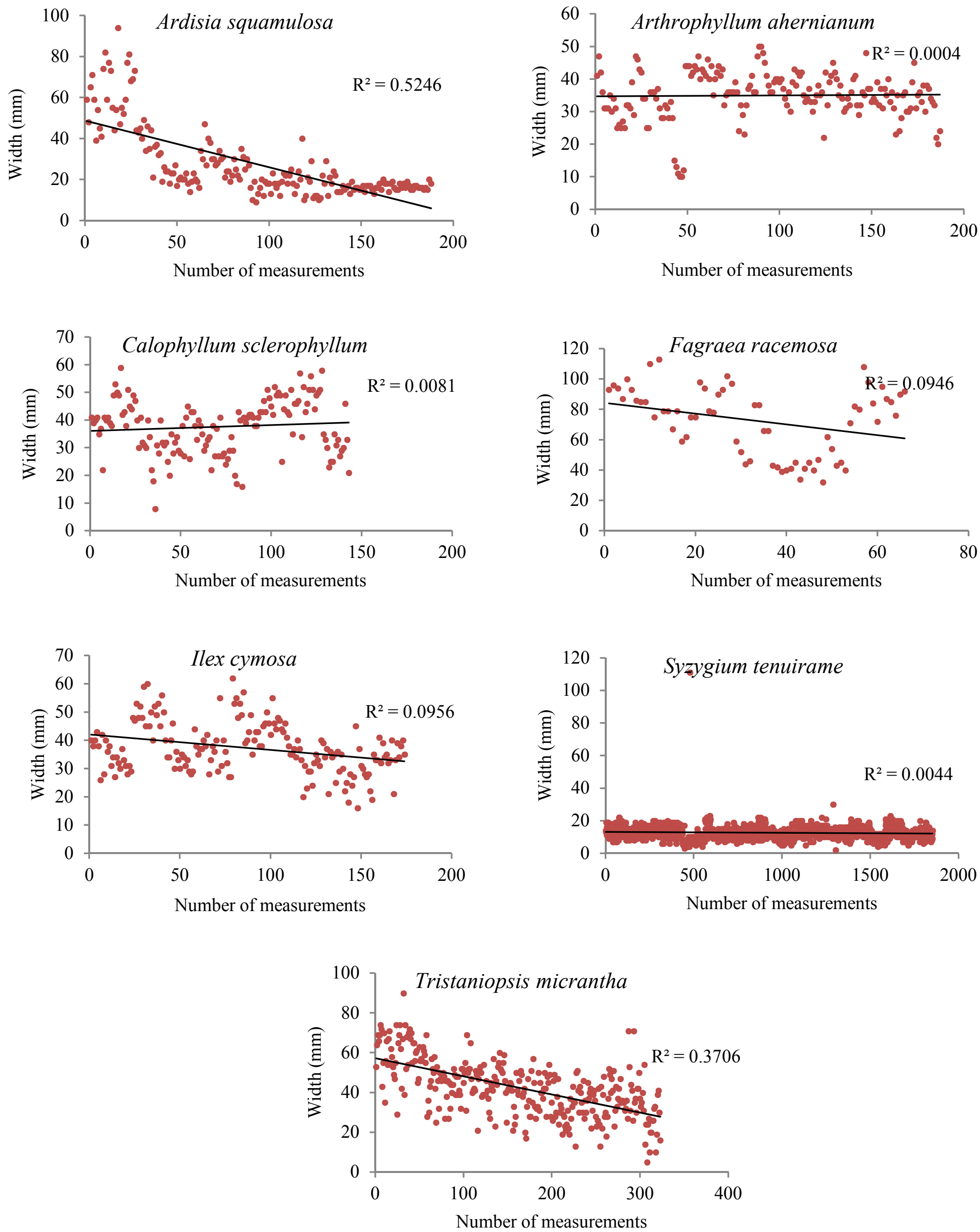

Fig. 4: Variation of leaf widths of all the species used

P<0.01, respectively. The leaf width is positively correlated (P<0.05) to total phosphorus in tissue but negatively correlated to peat depth. Leaf width also is highly negatively correlated to soil nitrogen (P<0.01).

Variation of leaf size across the peatland

The variation in leaf size indices were found considerably from one tree to another as well as from one forest zone to another. However, it is very apparent that a decreasing trend of leaf size indices from the fringing forest towards the center was observed. These variations could be strongly attributed to the harsh environment of the peat swamp forest, wherein trees predominantly exhibits smaller leaves towards center of the peatland which highly

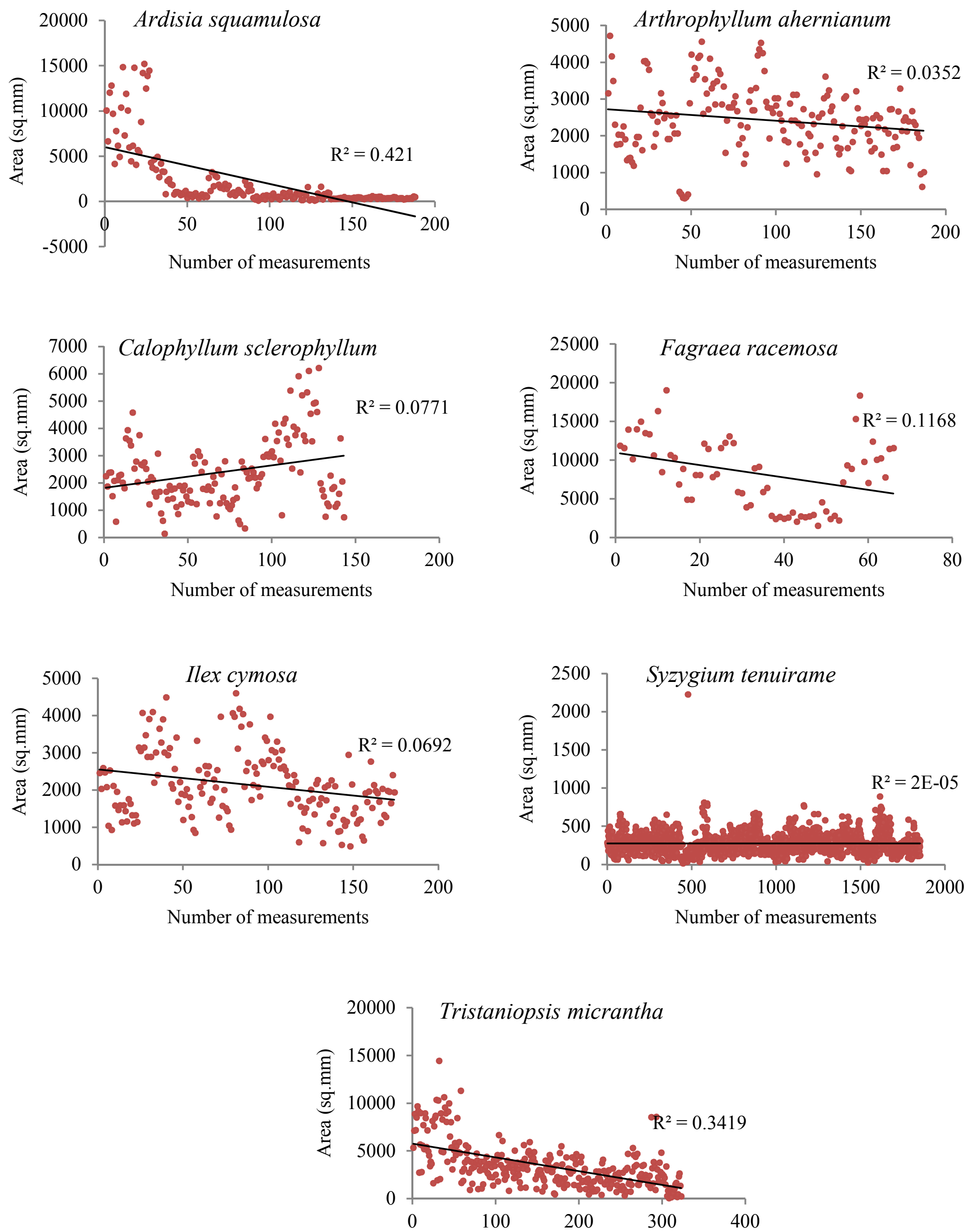

Fig. 5: Variation of leaf area of all the species used

reflects it adaptive significance to high light (Bragg and Westoby 2002) and nutrient poor condition (Beadle 1966; Cunningham *et al*. 1999; Fonseca *et al*. 2000). Further, Page *et al.* (1999) also emphasized that the differences in hydrology and nutrient availability exert strong influences on the composition, structure and physiognomy of the forest vegetation.

Leaf size classification

Based on Raunkiaer and Webb's leaf size category system, results revealed that the tall-pole forest is predominantly mesophyllous, both the intermediate forest and sapling-forest is mesophyllous-notophyllous, while the pygmy forest was predominantly microphyllous. It appears that the findings of this

Table 3: Leaf tissue analyses

Zonation	Total N (%)	Total P (%)	Total K (%)
Tall-pole forest	1.07	0.099	0.50
Intermediate forest	0.870	0.064	0.35
Sapling-size forest	0.937	0.081	0.70
Pygmy forest	0.468	0.036	0.125

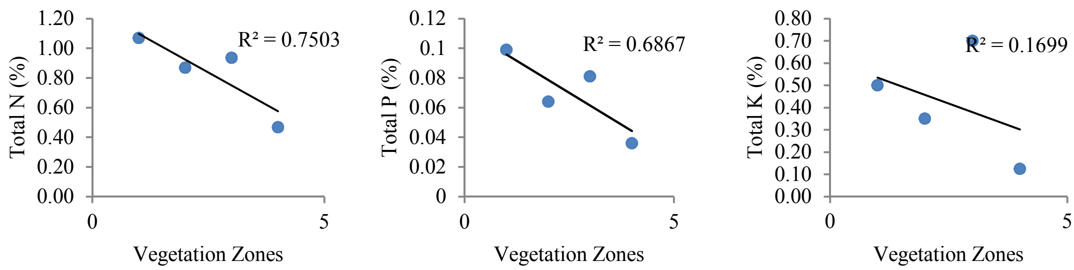

Fig. 6: Variation in leaf macronutrients

Table 4: Leaf size classification of the vegetation in Caimpugan PSF

Zonation	Leaf area (cm^2)	Leaf classification		Leaf size classification
		Raunkiaer (1934)	Webb (1959)	
Tall-Pole	61.42	Mesophyll	Mesophyll	Mesophyllous
Intermediate	42.74	Mesophyll	Notophyll	Mesophyllous-Notophyllous
Sapling-size	40.10	Mesophyll	Notophyll	Mesophyllous-Notophyllous
Pygmy	16.70	Microphyll	Microphyll	Microphyllous

study support the three vegetation zonation described by Davies (2006). However, earlier studies of Aribal (2013) and Adorador (2013) confirmed that the LSI includes predominantly mesophyllous, mesophyllous-notophyllous, notophyllous and microphyllous based on the four (4) tree species measured. Moreover, Bruenig (1990) also reported that on the mature oligotrophic peat domes in Borneo and Sarawak the accounted leaf size spectrum was predominantly mesophyll, followed by mesophyll-notophyll, predominantly notophyll, and notophyll-microphyll.

Between the intermediate and the tall-pole forest, there is an apparent disparity in LSI thus confirm that the former should not form part of the latter. This further implies that the intermediate forest could be the transition stand between the tall-pole forest and the sapling forest. However, with respect to the leaf size category, the intermediate forest could be considered as part of the sapling forest but the boundaries emanates at some part of the tall-pole forest.

Leaf size and soil nitrogen

The observed soil nitrogen in this study has strong negative correlation ($P< 0.01$) with the leaf widths based on Pearson's correlation analyses. This implies that higher amounts of soil N results in smaller leaf sizes due to toxicity. According to Vitousek *et al.* (1982), in peatland ecosystem plant species acquire nitrogen in the form of ammonium (NH_4^+) and that increasing amounts become detrimental to plants as this would led to toxicity. Rothstein and Gregg (2005) also showed that foliar nutrition uptake of nitrogen decreased with increasing proportions of NH_4^+ in nutrient medium. Goyal *et al.* (1982) emphasized that in wet and low oxygen soils coupled with more pronounced high light intensity, ammonium toxicity has the highest likelihood of occurring. Gerandas *et al.* (1997) revealed that most plant species when exposed to high ammonium concentration show reduced growth, smaller leaves and a stunted root system. While *Tristaniopsis micrantha* (Myrtaceae) as the dominant species was classified by Britto and Kronzucker (2002) as NH_4^+ tolerant however, NH_4^+ tolerant species could still suffer toxicity. Thus, ammonium toxicity coupled with greater waterlogged conditions, high light intensity and very low oxygen soils explains the apparent stunted condition of pygmy forest in contrast to tall-pole forest which has more favorable conditions. Moreover, at high water table levels the condition become more anoxic therefore making it more difficult for oxygen to diffuse through the pore spaces in peat. The conditions are said to be anaerobic and it is under these conditions that various chemical process can be inhibited.

Leaf nitrogen dynamics

A strong positive correlation ($P<0.01$) between the leaf length and total nitrogen content was observed while the leaf area was positively correlated ($P<0.05$) with the total nitrogen based on Pearson's correlation analyses. This implies that nitrogen acquired by plants influences the leaf size. Plants in nutrient poor environments produce small amount of litter and conserve large amounts of nutrients in recalcitrant tissues, thus reinforcing the infertile environment (Melillo *et al.* 1982; Hobbie 1992; Crews *et al*. 1995; Aerts and Chapin 2000). Ingram (1967) and Sparling (1967) also explained that the vegetation growing towards the periphery of the peatland dome receives an increased water flow, and consequently, an increased rate of supply of dissolved nutrients elements. Thus, leaf sizes in the tall-pole forest being located closer to the river are influences by the flow of water which contained nutrients available for the plant uptake in contrast to the pygmy forest that located at the center of the peatland.

Leaf size and dissolved phosphate

Based on Pearson's correlation analyses, leaf area is positively correlated ($P< 0.05$) to phosphate which implies that increasing amounts of phosphate affects the leaf size of the species. Basirat *et al.* (2011) stated that phosphorus (P) is absorbed only as phosphate ions (Pi) where more P allocating in shoots than in roots indicated that leaves are cumulating organ for P distribution at high Pi condition. This result conforms to the finding of Page *et al.* (1999) on the peat swamp forest of Sengai Subangau, Central Kalimantan, Indonesia where phosphate was observed at descending levels from the tall-pole forest to the pygmy forest. The tall-pole forest which had the highest phosphate concentrations had the bigger leaf area which gradually decreases as levels of phosphate diminished towards the pygmy forest. Basirat *et al.* (2011) stated that low phosphate concentration results in the reduction in the plant leaf area.

Partial decomposition rates of microbes in tall-pole forest takes place which influence the nutrient recycling of phosphorus for the nutrient uptake of the vegetation resulting in bigger leaves in contrast to pygmy forest in which greater waterlogged conditions coupled with low decomposition rate resulting into smaller leaves. However, the marked increase in phosphate activity together with decreasing nutrient concentration toward the center of the dome indicates clear increase in biological investment, by the vegetation, in the acquisition of P from organic forms (Sinsabaugh and Moorhead 1994; Wright and Reddy 2001; Cheesman *et al.* 2010).

Leaf phosphorus dynamics

Based on Pearson's correlation analyses, the leaf size indices are positively correlated ($P<0.05$) to its leaf phosphorus content. This implies that the phosphorus uptake of plants influences the leaf size of the vegetation from the outer to the inner forest zone in Caimpugan peat swamp forest.

Analysis of the leaf tissue showed a decreasing trend of leaf phosphorus content from the tall-pole forest to the pygmy forest which indicates that high phosphorus uptake results in an increase leaf size indices and gradually decreases towards the pygmy forest due to decreasing phosphorus levels. Anderson (1961a, 1964a) revealed that the phosphorus content tend to decline from the margin to the center of the peat swamp forest in Sarawak and Brunei, which is also observable in Caimpugan peat swamp forest. It was reported that only 0.1% of the total phosphorous from soil is available to plants (Peix *et al*. 2001) and available P is immediately depleted around the root zone owing to continued plant uptake (Smith *et al.* 2003). Further, it is well known that all plants exposed to different phosphate ion (Pi) concentrations show correlated responses to internal phosphorus status (Biddinger *et al.* 1998; Bucher *et al.* 2001; De Groot *et al.* 2001). Rahimi and Pouzesh (2012) revealed that plants exposed to high phosphorus level soils obtained the maximum leaf area index, stem height and root length. In addition, plants that are grown under low phosphorus level develop lower total leaf area, which adversely affect light interception and hence plant growth (Lynch *et al.* 1991; Plenet *et al.* 2000).

Leaf size and peat depth

A negative correlation was observed ($P< 0.05$) between the leaf area and peat depth as well as the leaf length and peat depth based on Pearson's correlation analyses. This implies that leaf size tend to decrease in deeper peat but increase in a shallower peat. This is well-depicted in Caimpugan peatland. The tall-pole forest is situated in shallower peat, while the intermediate forest, sapling-size forest, and pygmy forest occurs on relatively deeper peat. This environmental constraint would likely results to smaller vegetation stature and probably smaller leaf size (Page *et al.* 1999). Moreover, roots of trees in the tall-pole forest probably may penetrate through the shallow peat down to the underlying mineral soil thus obtaining more nutrients. Also, during the rainy season the tall-pole forest also receives river floodwaters containing dissolved nutrients elements thus, increased in nutrient uptake.

Leaf size and acrotelm

A positive correlation ($P<0.05$) was observed between the leaf area and acrotelm based on Pearson's correlation analyses which implies that the increase of acrotelm thickness has a corresponding increase in leaf size and vice versa. In Caimpugan peat swamp forest, a decreasing trend of acrotelm thickness was observed from the tall-pole forest towards the pygmy forest in which the former obtained the bigger leaf area and thicker acrotelm in contrast to the latter which obtained the small leaf area corresponds to the thinner acrotelm. Merryfield and Moore (1974) stated that, as the peat blanket thickens, the surface vegetation becomes insulated from underlying soils and rocks resulting in floristic changes which reflect the altered hydrology and chemistry of the peat surface. Page *et al.* (1999) also emphasized the importance of acrotelm which functioned as the rooting zone for the forest vegetation and the source of limited nutrients from organic matter decomposition that eventually influence growth of the plants. On the other hand, thicker acrotelm relates to more vigorous vegetation by means of height and diameter and consequently larger leaf area.

CONCLUSION

The variations in leaf size indices (LSI) of the plant species in Caimpugan peat swamp forest from the tall-pole forest towards the pygmy forest is strongly associated with the variations of acrotelm thickness and water table, peat depth as well as nutrient availability. Moreover, the nutrient uptake dynamics of the plant species examined was found to influence the leaf size which showed a decreasing nutrient content thus affecting the structure and/or physical appearance of the forest and eventually depicting a zonation pattern of relatively tall forest to a stunted or xeromorphic forest at the center of the peatland. The leaf size classification further clarify the extent of the forest zonation despite the findings did not conform to that of Bruenig (1990) on mature oligotrophic peat swamp forest. It is therefore concluded that leaf size and/or leaf spectrum serves as direct indicators to the physico-chemical conditions of the ecosystem and provides insights for delineating vegetation zones.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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