Interspecific variation in herbivory level and leaf nutrients of mangroves Rhizophora

Indah Trisnawati 1,*, M Muryono 2, I Desmawati 3

123Department of Biology, Faculty of Science, Institut Teknologi Sepuluh Nopember. Gedung H Campus ITS Sukolilo Surabaya 60111, East Java, Indonesia. Tel./Fax. +62-31-5963857, email: indahtris@yahoo.com.

Abstract. Trees and saplings of two mangrove species, Rhizophora stylosa Griff. and Rhizophora mucronata Lam. were observed in herbivory level (leaf damage area and percentage of herbivory) and leaf nutrients (total nitrogen, phosphorus, potassium, water content and deterrent tannin). We observed mangrove habitat located around Institut Teknologi Sepuluh November (ITS) Campus in Surabaya, East Java dominated by Rhizophora vegetation. Herbivorous insects showed an individual abundance of 15.15% of the total insects found in mangroves around the studied areas. Seedlings tended to be significantly higher in relative leaf damage and preferred by the herbivores than trees. One way ANOVA showed a non-significant difference in leaf area damage between two species (F=0.63, p=0.43; F=2.29; p=0.13). The concentration of leaf nutrients was significantly different between two species than habitus. Total nitrogen and water content in R. mucronata were significantly higher than in R. stylosa (F=5.10, p=0.03; F=142.13, p<0.0001), whereas phosphorus and potassium (K) was significantly higher in R. stylosa than in R. mucronata (F=15.25, p=0.0002; F=75.916; p<0.0001). Descriptively, R. stylosa leaves tended to be higher in deterrent tannins than R. mucronata, especially in seedling habitus. Leaf age, represented by plant habitus, is less to be related to high leaf palatability and insect herbivore nutrition than mangrove species.

Key words: Herbivory level, leaf nutrients, Rhizophora species.

1. INTRODUCTION
As one of coastal ecosystems, mangrove ecosystems are vital to stabilize environmental nutrient cycles, as well as to support community’s livelihoods. The ability of mangrove ecosystems to maintain these functions depends on supporting factors, including environmental quality and substrate conditions (Wantasen, 2013). Yet, mangrove ecosystems have been damaged at an alarming rate. Over the past 50 years, about one-third of the world's mangrove forests have lost (Alongi 2002). It is estimated that mangroves in developing countries tend to decline by around 25 percent by 2025 (Ong and Khoon 2003). In Indonesia, which is a country with the largest mangrove forests in the world, the rate of mangrove loss is higher by 90% in Java and Sumatra (Bengen and Dutton, 2003). Besides threats due to the effects of global climate change, mangroves are also threatened by anthropogenic factors, such as agricultural and aquaculture activities. Disturbances caused by the effects of nutrient enrichment and climate change are known as the main threats occurring
simultaneously to estuary and coastal regions throughout the world (Lloret et al. 2008; Turner et al. 2009; Feller et al. 2017). The availability of nutrients can affect herbivore activities, nutrient dynamics, and the growth of mangroves which varies along tidal gradients and climates. Onuf et al. (1977) examined the interactions between nutrients and herbivores in mangrove ecosystem and found that nutrient enrichment in mangroves causes high damages due to herbivore activities. The increasing damage is due to the increasing quality of plant tissue as a source of nutrition to herbivores. Herbivory, especially leaf herbivory, have impacts ecological processes such as plant productivity, community composition, and ecosystem nutrient cycling (Turcotte et al., 2014). Any kind of herbivorous activity may cause changes in the environment, anthropogenic impacts, species extinctions, bio-invasions, or may provoke significant changes not only to the species involved in the interaction, but eventually to the system as a whole (Sebata, 2013). According to Feller (2002), herbivory can alter nutrient cycling patterns in mangrove systems. By direct removal of leaf tissue, herbivores remove resources that were directed towards detrital pathways. In addition, the tissue removed by herbivores causes a decrease in the amount of nutrients available for resorption during senescence (Feller, 2002; Neveu 2013). Mangrove herbivory impacts individual plant growth and primary productivity. Loss of leaf tissue and branch removal results in a loss of total plant photosynthetic capacity, and has also been shown to reduce mangrove reproductive output (Cannicci et al. 2008, Neveu 2013). In addition to variation in external factors that may affect plant performance, the relative success of mangrove plants will be influenced by differences in inherent growth characteristics (McKee, 1995). Tomlinson (1986) reveals that since mangroves represent a taxonomically diverse group of plants, sympatric species often differ in their morphology, phenology, physiology, and specific adaptations to the mangrove habitat. They also differ in terms of characteristics such as morphological plasticity, biomass partitioning, and defensive chemicals. An understanding of how external factors may affect mangrove plants first requires some knowledge of these plant traits, which determine a species' potential for growth and survival. This information is generally lacking for mangroves, but is necessary for an accurate interpretation of plant responses across environmental gradients. In Indonesia, the mangrove tree species belonging to the genus Rhizophora, Bruguiera, and Ceriops are considered as commercial tree species (Direktur Jenderal Kehutanan, 1978 in Kusmana et al., 1992), so that the herbivory rates and leaf nutrient composition of those species in the forests is useful information for the mangrove forest management practices. In Southeast Asia, Rhizophora spp. community, especially R. stylosa distributed more inland (Komiyama et al. 2008); and characterized by high density of prop roots (Nishino, et al. 2015). The mangrove forest of Kampus ITS Campus Surabaya is characterized by the existence of Rhizophora mangroves, i.e. Rhizophora stylosa and R. mucronata, which distributed in the campus wetland. In Southeast Asia, mangrove forest dominated by Rhizophora stands, some researcher estimated high above-ground biomass trend, such as R. apiculata in Malaysia (Putz and Chan, 1986), R. stylosa and R. apiculata in Indonesia (Komiyama et al. 2008). However, the rate of herbivory loss was not considered in the mangroves biomass and productivity, due to the paucity of studies describing herbivory on mangroves. The objective of this study was to compare two species of mangrove in terms of herbivory rates, and leaf nutrient chemical composition and to relate these characteristics to performance in a nutrient enrichment environment. The results, which indicate substantial differences among two mangrove species, provide a foundation for an evaluation of the mechanisms determining survival success or failure in the mangrove ecosystem. This study were carried out in the mangrove ecosystem managed by farmers around the ITS Campus Surabaya. The mangrove area around the ITS Campus Surabaya provides contextual example regarding the presence of nutrient enrichment activities, such as agriculture and aquaculture. Based on the results of vegetation analysis carried out at the mangrove area, Rhizophora stylosa was found as the most important mangrove species among others for each growth stage/category (unpublished data 2018). The results of this study can be used as important baseline information relating to the vulnerability status of the two mangrove species to herbivory interaction, and leaf chemical (nutrient) characteristic among the two mangrove species.
2. MATERIALs AND METHODS

2.1. Study sites
The research was carried out for three months. The data collecting time is September-November 2018 which the duration of the data was taken once every three weeks. The research location was carried out in the mangrove ecosystem managed by farmers around the ITS Campus Surabaya. (Figure 1) with an area of ± 0.5 ha.

![Figure 1](image-url) Location of research at mangrove ecosystem managed by farmers around the ITS Campus Surabaya (Modified from www.Googleearth.com) and mangrove landscape at ITS Campus. The red box shows the sampling location with an area of 0.5 ha.

Procedures

2.2. The insects herbivores assemblages
A sampling of insect as herbivores of mangrove Rhizophora area at the ITS Campus was conducted from September 2018 to November 2018. Every three week, morning survey of herbivores was carried out during the period of 07:00 am - 10:00 am on mangrove leaves of all trees in the plot of each site using sweep net (Insect Net long handled 100cm, 50cm diameter). Yellow pan trap (diameter of 40cm, height of 20cm, was set up as trap for 24 hrs) and direct hand counting (for 30 minutes). Insects were placed in insect vials containing 75% of alcohol for soft and hard insects and in collection paper for the order of Lepidoptera and Heteroptera. The sweep net method was performed to quantitatively sample insects perching or flying around vegetation (Voshell, Jr. 2002). The yellow pan trap method was performed to collect flower-visiting insects, which are usually active in the morning and afternoon (Campbell and Hanula 2007). The hand collecting was performed to collect ants, spiders and minute arthropods, such as mites. Hand collecting is the most direct technique for the inspection on foliage which may be collected the specimen alive (Basset et. al. 1997).

2.3. Measurement of herbivory level : Leaf damage area and percentage of herbivory
Herbivory was measured as degree of leaf folivory. Signs of herbivory included bites along the leaf margin, holes in the leaf, and trails/scrapes made by insects excavating leaf tissue. Leaf damage was observed in two categories of growth stage (habitus), i.e. seedlings, and trees with diameter <1.5cm and 10cm, respectively (BSN, 2011). Three mangroves per plot were randomly selected at each sampling occasion. The observation was done on three upper branches of each individual tree. To measure the damaged leaf area due to herbivory interaction, leaf pictures taken of all the leaves on the branches sampled from the outside of the canopy of each individual plant. After that, the measurement of mangrove leaf morphometry done by scanning the leaf area. The results of scanning the leaves were then processed using image analysis software ImageJ. A proportional calculation of the leaf area damage was carried out using open source application ImageJ for more-advanced image processing and analysis techniques (Schindelin et. al. 2015). ImageJ was performed to obtain a more accurate length and width of the leaves (without stems), both intact and leaves damaged by herbivores. For herbivory measurements, the leaves were differentiated by age (i.e. for tree habitus was represented old leaves and for seedling was represented by young leaves) and leaf conditions (i.e. intact or damaged). The category of leaf damage area was determined by the presence of herbivore marks, indicated by the loss of some leaf area due to bite or perforation of herbivorous insects. The analysis of ImageJ resulted in the area of lost or damaged leaves. Then the percentage of herbivory (% leaf area lost to herbivore) was calculated as follows:

\[
\frac{(PLA - ALA)}{PLA} \times 100\% = \% \text{ Herbivory}
\]

With PLA as the imaginary leaf area, and ALA is remaining leaf area. Estimation of leaf area ate by herbivores was obtained from the difference in imaginary leaf area (PLA) with the remaining damaged leaf area (ALA) (Khusna, 2008).

2.4. Leaf nutrient quality : Leaf chemical characters

Analysis of leaf nutrient quality test was carried out monthly to determine the total nitrogen, phosphorus, potassium, and water content in plant tissues. Tannin test was carried out at the end of the study. A total of 25 leaves was collected randomly from the sampling site, then they were oven-dried at 70°C until the weight is constant, and were grinded for chemical analysis. The total N was determined using Nitrogen Kjeltec unit, while other elements such as total P, K (potassium) were determined using Microwave Digestion and Inductively Coupled Plasma Mass Spectrometry (ICP-MS). Water content was determined using oven. Total tannin assay was determined using Ultraviolet-Visible Spectrophotometry. All nutrient content were expressed as percentage of dry weight of the samples, mg/kg, and mg GAE/g.

2.5. Statistical Analysis of the Correlation Analysis between Leaf Damage Area, Percentage of Herbivory with Leaf Nutrient Quality

Difference in leaf herbivory level, nutrient level (nitrogen total, phosphorus, potassium and water content) and deterrent tannin in different mangrove species and plant habitus (seedling and tree) was analysed using One-way ANOVA. Pearson correlation analysis was conducted to determine the relationship between nutrient contents in leaves and the leaf damage level, and the percentage of herbivory. All calculations were performed using IBM SPSS Statistics software version 22.0.

3. RESULTS AND DISCUSSION

3.1. The Herbivorous Insect Assemblages

In general, observations from September to November 2018 showed that herbivorous insects found in mangrove area on the ITS Campus were only less than 20% of the total mangrove insects encountered.
Mangrove area on ITS Campus tends to encounter more predator insects with a relative abundance of 62%, compared to 15% of herbivorous insects. Sixteen herbivore species from five orders, i.e. the Lepidoptera, Coleoptera, Diptera, Hemiptera, and Hymenoptera, were collected from mangrove Rhizophora at mangrove area of ITS Campus. Proximity aquaculture and agriculture activities is likely responsible for the moderate abundance of herbivorous insects, especially in mangrove Rhizophora. The composition of herbivorous species shows that the proportion of species tends to evenly distribute, especially seven species with the highest number of individuals (Figure 2). In the mangrove area around the ITS Campus, groups of pierid butterflies, hemipteran bugs, nymphaeid butterflies and coccinellid beetles, such as *Chrysis* sp., *Euthochthta galeator*, *Leptosia nina*, *Delias periboea*, *Neptis hylas*, *Minetta* sp., *Metcalfa* sp., *Zizia otis*, *Leptosia nina*, and *Harmonia axyridis* has been found abundantly in herbivorous insect assemblages. Several types of herbivorous insects are known as chewing herbivores/leaf cutters, such as Pieridae larvae, Nymphalidae, and several groups of Coccinellidae beetles.

![Figure 2](image-url)

**Figure 2.** The composition of herbivorous insects found on mangrove area in ITS Campus Surabaya.

### 3.2. Herbivory Levels: Leaf damage area and percentage of herbivory

Observation on leaf damage area and percentage of herbivory showed that there was no significant difference in the vulnerability of mangrove leaf to herbivory, particularly in Rhizophora species in the mangrove area of ITS Campus (Figure 3).
Figure 3 shows that each growth category (habitus) showing different levels of herbivory level (leaf damage area and percentage of herbivory) by herbivorous insects. The leaf damage area is significantly higher in tree habitus than seedling, especially in *R. mucronata* (*F* = 4.85, *p* < 0.03). However, the percentage of herbivory is slightly higher in seedling habitus (15.70%) than tree (14.03%), but it is not significantly different in both species (*F* = 0.64, *p* > 0.43). Tree tends to experience relatively higher leaf damage area than seedling, probably caused by leaf nutritional quality which may affect palatability by herbivorous insects. Between two species, one way ANOVA showed a non-significant difference in both leaf area damage and percentage of herbivory (*F* = 0.63, *p* > 0.43; *F* = 2.29; *p* > 0.13). The herbivory level of different habitus is more to be related to high preference of herbivorous insects on mangrove leaves.

**Leaf nutrient quality (leaf chemical characters)**

Figure 4 shows different leaf nutrient level among habitus in each Rhizophora species. Descriptively, the total nitrogen, phosphorus, and water content in seedling tended to be higher than tree habitus, except potassium (K). Two way ANOVA showed non-significant difference between habitus in total nitrogen, phosphorus and water content, except potassium (K) content in tree was significantly higher (*F* = 6.84, *p* < 0.01) than in seedling habitus.

Two way ANOVA showed that the concentration of all leaf nutrient in this study was significantly influenced by different species, *R. mucronata* and *R. stylosa*, than different habitus: total nitrogen (*F* = 11.75, *p* < 0.0008), phosphorus (*F* = 30.53, *p* < 0.0001), potassium (K) (*F* = 186.67, *p* < 0.0001), water content (*F* = 92.08, *p* < 0.0001). Among each species showed different level on nutrient level. Total nitrogen and water content in *R. mucronata* were significantly higher than in *R. stylosa* (*F* = 5.10, *p* < 0.03; *F* = 142.13, *p* < 0.0001), whereas phosphorus and potassium (K) was significantly higher in *R. stylosa* than in *R. mucronata* (*F* = 15.25, *p* < 0.0002; *F* = 75.916; *p* < 0.0001).
Figure 4. Leaf nutrient quality in the dominant mangrove species R. stylosa: a. Total nitrogen (%), b. Total phosphorus (mg/kg), c. Potassium (K) mg/kg, d. Relative leaf water content (%) between the seedling and tree habitus in the mangrove area of ITS Campus. The same letter indicates not significantly different at p-value >0.05.

Figure 5. Total deterrent tannin content (phenolics) mg GAE/g in the leaf of mangrove species among R. mucronata and R. stylosa between seedling and tree habitus at the mangrove area of ITS Campus.

The total tannin in Rhizophora leaves at the end of the study (Figure 5) showed a tendency that seedling habitus contain higher tannins than tree. The concentrations of total soluble tannins were higher in young leaves represented in seedling habitus, than mature leaves. This result suggests that herbivorous insects may not preferentially feed on leaves having higher concentration of total soluble tannin.
Descriptively, *R. stylosa* leaves tended to be higher in deterrent tannins than *R. mucronata*, especially in seedling habitus. It has been considered that, the higher phenolic contents in the plant (leaves) contributed to the greater protection of the plants towards herbivores and leaf damage (Banerjee et al., 2008, Malik et. al. 2017).

3.3. Correlation Analysis between Leaf Damage, Percentage of Herbivory with Leaf Nutrient Quality

Pearson correlation analysis was conducted to examine whether there is relationship between levels of nutrient content in leaf and leaf damage area as well as percentage of herbivory (Table 1).

**Table 1.** Results of statistical analysis to examine correlation between nutrient content in leaf, leaf damage area and percentage of herbivory at the mangrove area of ITS Campus.

| Leaf characteristic | **Rhizophora mucronata** | **Rhizophora stylosa** |
|---------------------|--------------------------|------------------------|
|                     | Seedling | Tree | Seedling | Tree |
| Leaf damage area vs % Herbivory | \( r = 0.923^* \) | \( r = 0.863^* \) | \( r = 0.828^* \) | \( r = 0.807^* \) |
|                     | \( p<0.0001 \) | \( p<0.0001 \) | \( p<0.0001 \) | \( p<0.0001 \) |
| Leaf damage area vs N total | \( r = 0.579^* \) | \( r = 0.102 \) | \( r = 0.173 \) | \( r = 0.223 \) |
|                     | \( p=0.0008 \) | \( p=0.953 \) | \( p=0.361 \) | \( p=0.237 \) |
| Leaf damage area vs phosphorous | \( r = 0.586^* \) | \( r = 0.035 \) | \( r = 0.113 \) | \( r = 0.102 \) |
|                     | \( p=0.0005 \) | \( p=0.852 \) | \( p=0.553 \) | \( p=0.592 \) |
| Leaf damage area vs potassium | \( r = 0.596^* \) | \( r = 0.079 \) | \( r = 0.076 \) | \( r = 0.327 \) |
|                     | \( p=0.0005 \) | \( p=0.882 \) | \( p=0.691 \) | \( p=0.078 \) |
| Leaf damage area vs water content | \( r = 0.332 \) | \( r = 0.105 \) | \( r = 0.237 \) | \( r = 0.046 \) |
|                     | \( p=0.073 \) | \( p=0.579 \) | \( p=0.207 \) | \( p=0.808 \) |

* significant at \( p<0.05 \), ** significant at \( p<0.0001 \)

The results of the Pearson correlation analysis showed that there was no significant correlation between leaf damage area and water content in leaves for both species (Table 1). The same data trend was also shown between leaf damage area and potassium (K) content, only for species *R. stylosa*. Only total N, phosphorous, and potassium content at seedling stage for species *R. mucronata* correlates significantly with leaf damage for seedlings. These results indicate that nutrient levels in mangrove leaves do not generally affect vulnerability of all mangrove species to herbivorous insects, especially in *R. stylosa*. However, nutrient level affect the vulnerability of species *R. mucronata* to herbivorous insect at seedling stage, especially for total N, phosphorous and potassium content.

4. Discussion

The abundance of herbivorous insects at mangrove area of ITS campus was less than 20% of the total individual insects found. The assemblage of 16 species of herbivorous insect recorded at ITS Campus mangrove area is relatively small compared to those recorded from other tropical mangrove forests. Herbivory by mangrove leaf feeders has been quantified in several locations in the neotropics (Onuf et al. 1977, Lacerda et al. 1986, Feller, 2002). Murphy (1990) reported that there are 102 species of herbivorous insects from Singapore mangrove, while Rau and Murphy (1990) reported 37 species from Thailand mangroves. Veenakumari et al. (1997) recorded 128 herbivorous species from the Andaman and Nicobar Islands of India. More recently, Burrows (2003) recorded a total of 61 species of herbivorous insect from *Avicennia marina* (31 species) and *Rhizophora stylosa* (34 species) from Queensland, Australia. The levels of defoliation reported in neotropic locations ranges from negligible in dwarf mangroves in Belize (Feller, 1995 in Feller, 2002) to 75% of the canopy leaf area in severe outbreak events in Hong Kong (Anderson and Lee, 1995), Thailand (Piyakarnchana, 1981 in Feller, 2002), Singapore (Murphy 1990), and Indonesia (Whitten and Damanik, 1986). Most studies of mangroves suggest that herbivores consume 10% of the canopy leaf area, which is similar to levels of defoliation reported from other tropical and temperate ecosystems (Coley and Aide, 1991; Feller, 2002).

This study recorded hemipteran bugs dan Lepidoptera larvae dominate insect assemblages in Rhizophora mangrove. The lower species diversity recorded in all locations is likely caused by bias in
Also found that added nutrients increase growth and alter leaf traits, tropics face the problem of increasing runoff of nutrients in coastal wetlands, which vary by feeding guild and location but are not correlated with latitude. In other study, Neveu (2013) showed that herbivory will increase in response to nutrient enrichment and this increase will be highest at the lowest level of salinity. Furthermore, Neveu (2013) reported that herbivory level was increased by phosphorus, but only in the low and medium salinity zones in Avicennia germinans. Tong et al. (2006) recorded 1.6 to 6.5% herbivory levels on Kandelia obovata in Hongkong with the highest herbivory level of 3.8% was recorded in summer (August). Leaves collected from Hongkong contained significantly higher contents of total nitrogen in two different locations in a few months (ranged 1.43 to 2.11%). Further, Tong et al (2006) reported nutritional factors of nitrogen enrichment, both naturally and artificially, have a significant relationship with the high level of leaf damage by herbivorous insects.

Based on plant habitus, leaf damage area in tree were significantly higher than that of sapling, especially for R. mucronata. Also, this study showed that leaf nutrient quality and deterrent tannin tends to be higher in seedling than in tree. Burrows (2003) suggested that two-thirds to three-quarters of the consumptive leaf area damaged by herbivorous insects were occurred on young leaves than older leaves. In general, age-based herbivory that included data from 73 tropical plant species from the Coley and Aide’s study (1991) found that 40-75% of total lifetime herbivory occurred on
young developing leaves. Further, Burrows (2003) explained that mangrove leaves change substantially in their physical and chemical composition as they aged. These attributes are likely to affect their palatability and resistance to herbivory. Therefore, it can be seen that young leaves on sapling habitus are more vulnerable to herbivory than older leaves. In recent study, Goncalves-Alvin et. al. (2011) reported that damage by herbivores on younger leaves of *Qualea parviflora* was ranged about 3-7%. When leaves were younger, it tended to less slerophyllous leaves and higher concentration of leaf nutrients and tannins. In the previous study, Coley and Aide (1991) showed that the growth of plants in habitats with excess light and nutrients would be associated with production of simple tissues and with lower concentration of phenolic and tannin compounds, where herbivore damage is negligible.

According to Schoonhoven et al. (1998), differences in herbivory level on leaves are often attributed to differences in nutrient contents, affecting their nutritive value to herbivorous insect. These chemical properties of leaves will vary as they age, with major change will occur as young leaves develop and mature leaves senescence. Tong et al. (2006) reported that young leaves of K. obovata have significantly higher concentration of nitrogen content than mature leaves in the summer (August-September). Regarding the level of vulnerability to herbivores, Burrow (2003) explained that most herbivores occurred while the leaves were young. Once past the juvenile phase, *R. stylosa* leaves were rarely attacked. It is assumed that younger habitus has better nutrient quality. Feller et al. (2017) reported that the percentage of *Avicennia marina* leaf area lost or damaged reaches the highest number (5 to 20% of the leaf area) as the nutrient content in the leaves, namely% N and% P, are relatively high. Goncalves-Alvim et al. (2011) found that nitrogen content is negatively correlated with tannin content. When nitrogen limits plant growth, carbohydrates will be accumulated in plant tissues and used for tissue differentiation products, increasing the synthesis of carbon-based secondary metabolites, such as tannins and terpenes. Furthermore, Goncalves-Alvim et al. (2011) found higher percentage of tannins and nitrogen in younger than in mature leaves of *Qualea parviflora* in Brazilian Cerrado. The synthesis of phenolic compounds such as tannins begins in young leaves and some studies reported increases and decreases of tannins and other phenolic metabolites along leaf development (Salatino et al. 1993; Goncalves-Alvim et al. 2011).

In this study, correlation analysis showed a significant relationship between leaf damage level to leaf nitrogen, phosphorous and potassium content only for *R. mucronata*, where the high level of leaf damage is likely influenced by those nutrient quality in mangrove leaves. Kathiresan (2003) reported that leaf damage was found to be maximum in *Avicennia* spp., and the difference on leaf damage has been attributed to leaf chemistry, especially tannin content. Tannin, as a deterrent against herbivores, is assumed to reduce palatability and digestibility of herbivorous insects.

**CONCLUSIONS**

In term of adaptation to herbivory, leaves of mangrove *R. mucronata* tends to be more vulnerable to herbivorous insects than *R.stylosa*, although no significant different on leaf damage area and percentage of herbivory between two Rhizophora species. There was significant difference between two species in the total N, phosphorous, potassium (K) and leaf water content. Pearson Correlation analysis generally shows that there is correlation between leaf damage due to herbivorous insects and leaf nutrient levels only for *R.mucronata*. It indicates that influence of leaf nutrient level with increased vulnerability of mangroves to herbivorous insects, especially in *R. mucronata*.

In assessing the vulnerability of mangroves to herbivorous insects, it is better to use various measurements of herbivory parameters on mangrove leaf to obtain representative effect of herbivorous insect communities at mangrove of ITS Campus. Also, deterrent leaf chemical (i.e. tannin, crude fiber, chloride, potassium) tests should be done to complement leaf nutrient test (total nitrogen, phosphorus, potassium and water content) as this study conducted. Similarly, testing the leaf nutrient content
should be expanded to various leaf ages and longevity, and mangrove species. Also, we suggest to observe the availability of nutrient quality and secondary metabolites in different mangrove habitats.

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