The anatomic response of the mangrove vegetation due to the changing in land functions

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Abstract. The mangrove forest in Indonesia have transformed into conservation area, tourist objects, and fishponds, causing the environmental changing. The purposes of this research are to find out the condition of the environment, the varieties of the species and the anatomy of the leaves. The locations of this research are determined based on the Karimun Java in December 2019. The locations are conservation area in Menjangan Besar Island, fishpond area in Kemujan Island and Mangrove Tracking area. The environment parameters such as temperature, pH, DO, TDS, and salinity. Photomicrograph is used to check stomata. The data are analyzed descriptively. The environmental conditions in those three locations have different condition, except temperature. The species found in Menjangan Island are A.marina and R.stylosa, the species found in Kemujan fishpond are C.tagal, R.apiculata and E.agallocha meanwhile the species found in tracking Kemujan are C.tagal, R.apiculata, R.stylosa and L.racemosa. The result of the observation towards the anatomy of the leaves in those three locations has not showed responses to the environment stress, but the condition of the environment in the fishpond showed the rising in salinity level. Therefore, it is suggested to grow Avicennia because it is more adaptable towards high level of salinity.

1. Introduction
Mangrove belongs to woody plants growing in tropical areas and subtropical areas which have extraordinary natural conditions, one of its important functions is to stabilize and to decrease the abrasion of the coast [1][2]. Indonesia has more than 20% mangrove areas of the total mangrove areas throughout the world and owns the most types of mangrove species compared to other countries [3]. In 2009, it was stated Indonesia owned 3.244.018 hectares of mangrove forests, in details, 30.7% was in good condition, 27.4% was slightly damaged and 41.9% was seriously damaged [4]. The damage of the mangrove forests in Indonesia was caused by human activities in making the land’s degradation becoming fishponds, roads and constructions [5].

The conversion of mangrove lands into fishponds and residential areas caused the changing of natural conditions [6][7]. There are some environmental factors that might affect the growth of mangrove. For instance, temperature, radiation, salinity, the tidal dynamic, wind speed, and soil aeration [8, 9]. The unstable quality of environmental parameters will result in decaying or even dying to mangrove plants [10]. Salinity is the main factor for mangrove to allow adaptation [11]. Different salinity can be seen from the adaptation of its leaves, for example stomata and the spreading zones of mangrove [12][13].

The parameters of the stomata anatomy on the leaves in order to find out the effect of salinity towards mangrove are the number and the size of stomata [14]. The research held by [15], resulted that mangrove has different adaptation on the anatomy of the leaves in every species in every different zone. Based on this research, we can find out that there are different anatomy of the leaves in every different zone, so that we can draw a conclusion that the anatomy of the mangrove leaves is affected by different natural conditions. The purpose of this research is to observe the different anatomy of the mangrove leaves, for...
example the number, the width and the types of stomata on mangrove in three different areas in Karimun Java Island. The mangrove observation areas in Karimun Java are divided into three stations according to the differences of natural conditions, they are Menjangan Besar Island, fishponds areas in Kemujan Island and tracking mangrove Kemujan Island. The aims of this observation are to know the natural conditions, the types of species and the anatomy of the leaves.

2. Methods

2.1 The determining and the making of research plot
The mangrove areas, which are used for the research plot, are divided based on the natural conditions. The observation was held in Karimun Java Island in December 2019. The natural conditions observed in mangrove areas were conservation areas, fishponds areas, and tracking mangrove areas. The conversation areas are located in Menjangan Besar Island, meanwhile, the fishponds areas and tracking mangrove areas are located in Kemujan Island. Menjangan Besar Island is labeled as Location A, while the fishponds areas in Kemujan Island are labeled as Location B and Tracking Mangrove areas in Kemujan are labeled as Location C. The following table is the coordinate points where the samples are taken (Table 1).

| Locations                        | The coordinate points                                      |
|----------------------------------|------------------------------------------------------------|
| A (Menjangan Besar Island)       | 5°53′15.7″S, 110°25′52.2″E                                   |
|                                  | 5°53′00.8″S, 110°25′28.6″E                                   |
|                                  | 5°53′17.1″S, 110°25′22.1″E                                   |
| B (The fishponds in Kemujan)     | 5°49′42.2″S, 110°28′59.2″E                                   |
|                                  | 5°49′40.4″S, 110°28′57.7″E                                   |
|                                  | 5°49′39.4″S, 110°28′55.5″E                                   |
| C (Tracking Mangrove in Kemujan) | 5°49′35.4″S, 110°28′03.3″E                                   |
|                                  | 5°49′32.6″S, 110°27′57.8″E                                   |
|                                  | 5°49′22.7″S, 110°27′53.7″E                                   |

2.2 The analysis of environmental quality
To analyze the quality of the environment, we measured the water quality using Horiba- Multiparameter Water Quality Checker. The water parameters, namely the temperature, pH, Dissolve Oxygen (DO), Total Dissolve Solid and Salinity were checked carefully. Each location would be measured three times in repetition.

There are some steps how to operate the Horiba-Multiparameter Water Quality Checker according to [16], first, open the lid/the cap of the probe by rotating or spinning it. Next, open the lid/the cap of DO sensor. Then turn on the Water checker and immerse it down into the water until it is completely dipped. The results of the measurement will be shown right on the screen. Wait for a moment until the result numbers shown on the screen are stable, afterward press meas (measure) to save the data. To maintain the stability of the sensor for the next using, the Probe water checker should be calibrated by washing it using aquades. The DO sensor should also be calibrated by applying few drops of KCl before the probe is closed. The steps how to use the Water Quality Checker must be repeated everytime we use it.
2.3 Identifying the types of the species
The identification of the species in mangrove areas is done according to [17], by observing the morphology characteristics of the species, like roots, leaves and flowers. After identifying the types of the species, the next step is to write down all the names of the species. The identification of the species are done in these three stations, which are Menjangan Besar Island, fishponds in Kemujan, and the tracking mangrove in Kemujan Island. The manual book to carry out the process of identification by [18] uses Handbook of Mangroves in Indonesia.

2.4 Sampling and leaves analysis
The mangrove leaves of 3-4 individuals from each species were picked out, after that the fully-grown leaves which were fully exposed by sunlight were chosen. The samples of the leaves were taken out from the fourth row to the fifth row from the base of the tree trunk [19]. The samples of the leaves were fixed with alcohol 70%. Next, the leaves were coated by nail polish liquid on the upper part and the down part of the leaves. When the liquid had set, the next step was the leaves were covered with adhesive tape and then it would be pulled out very slowly. The adhesive tape which had the epidermis stick on it was attached on the glass cup and marked to recognize the upper part and the down part of the leaves.

The parameters observed for the leaves anatomy were the types of the stomata, the length of the stomata, the width and also the density of the stomata. The observation was held using photomicrograph. The measuring for length and width were repeated three times. The repetitions consisted of two different wide fields of views of the microscope randomly. Based on [20], the formula for counting stomata is:

\[ \text{Density of the stomata} = \frac{\text{The number of stomata}}{\text{per unit of wide fields of views of the microscope}} \]

3. Result and discussion

3.1 Environmental factors

| Stations | Temperature | pH  | DO (mg/L) | TDS (g/L) | Salinity (ppt) |
|----------|-------------|-----|-----------|-----------|----------------|
| A        | 29.90       | 8.09| 6.74      | 23.36     | 24.32          |
| B        | 28.59       | 7.10| 2.11      | 26.63     | 28.16          |
| C        | 28.03       | 6.72| 1.16      | 24.26     | 25.41          |

Mangrove is a halophytic intertidal species, in the forms of trees and bushes spreading throughout tropical areas [21]. Some coastal areas had experienced some changes dynamically because of the land conversions which affected the decreasing number of mangrove areas [22]. The results of the environmental factor analysis towards water (Table 2.) showed that the average temperature in three stations is between 28-30°C. Station A (Menjangan Besar) has the water temperature for 29.90 °C, meanwhile the water temperature in Station B (tambak Kemujan) is 28.59 °C and the water temperature in Station C (tracking mangrove Kemujan) is 28.03 °C. Every mangrove species has distinctively optimal temperature. The optimal temperature for mangrove to grow well is between 28-32 °C [23]. The rising temperature can increase the fixation of CO₂ on mangrove plants, but it can be obstructed whenever the temperature is above 40 °C [24]. Low temperature is one of obstacles for mangrove plants to thrive and spread, in the meantime, the increasing temperature along the time and the changing of rainfall will bring effect to the distribution of mangrove plants [25].

The results of pH measurement in three stations (Table 2.) showed different pH. Station A (Menjangan Besar) has pH 8.09, while Station B (tambak Kemujan) has pH 7.1 and Station C (tracking Kemujan) has pH 6.72. The best pH for mangrove to grow is around 6-8.5 that will influence to aquatic organisms and the decomposer activities [26]. The pH values in water territory are divided into three categories, for pH 5.5-6.5 is categorized into an unproductive water territory, if the pH is 6.5-7.5 so it belongs to a productive water territory, and for pH 7.5-8.5 is categorized as a highly productive water
territory [27]. The water territory in Station A is categorized as a highly productive water territory, on the other hand, the water territories in Station B and Station C belong to productive water territories.

The DO (Dissolve Oxygen) level on each station differed one to another. The result for DO (Table 2.), indicated that Station A (Menjangan Besar) has the highest DO level, that is 6.74 mg/L. While the results on Station B (Fishponds in Kemujan) and Station C (tracking Kemujan) showed the levels of DO 2.11 mg/L and 1.16 mg/L. DO is one of the water parameters and controls the growth of mangrove plants [28]. The low DO value is caused by the high level of organic pollution and the nutrition along the mainstream rises because of the respiration process during the degrading of organic matter [29]. The concentrate of DO is changing everytime and is influenced by some Physics factors, Biology and Chemistry factors such as pH, temperature, atmosphere pressure and salinity [30]. The ammonia compound produced by leftovers shrimp food and shrimp dirt which are accumulated deep down in the sea water decreases the quality of DO [31]. The lower the Do concentrate is, the lower the pH in the water is [32].

The TDS (Total Dissolve Solids) in Station A is 23.36 g/L, while the TDS (Total Dissolve Solids) in Station B and Station C are 26.63 g/L and 24.26 g/L. (Table 2.). The parameter showed that the highest TDS was in Station B, which is fishponds areas. The highest level of TDS in Station B is because of the conversion land into fishponds areas, as the result, the density of the mangrove areas is decreasing. TDS can be used as the indicator to find out the level of the water pollution because TDS contains some dissolved matters such as organic matters, inorganic matters and any other mineral matters which have diameter < 10^-3 µm in length [33]. The lower density of mangrove areas will increase the TDS value [34]. Mangrove areas have some functions to slow down the water current and to improve the level of sedimentation so that the concentrate of TDS will decrease [35].

The measurement of the salinity in every station shows different salinity level (Table 2.). The salinity in Station A and Station C have almost the same level, which are 24.32 ppt dan 25.41 ppt. The highest salinity level is found in Station B, the fishponds areas, for about 28.16 ppt. The differences in salinity levels are caused by the environmental condition. Mangrove plants can grow well with the level of salinity approaching 0 ppt or 35 ppt, it depends on the types of the species [36]. If the water salinity is beyond 35 ppm, it can cause negative osmotic pressure, as the result, it will bring some effects to the vegetation of the mangrove plants [10]. The threat of high salinity level can make some changing on photosynthesis activities and the structures of cells [36].

3.2 The identification of the species

| Stations | The Types of The Species |
|----------|--------------------------|
| A        | R.stylosa, A.marina, C.tagal |
| B        | R.apiculata, E.agallocha, C.tagal, R.apiculata |
| C        | R.stylosa, L.racemosa |

The observation towards the types of the species in Station A (Table 3.) can identify two species, they are R. stylosa dan A.marina. Station A is a mangrove conservation area which is located in Menjangan Besar Island. A.marina is the type of mangrove species with the largest distribution areas among other types of mangrove species, so it easily adapts to any conditions and environments [37]. A.marina can adapt to the changing of the environment, for example, temperature, salinity, and the nutrient content P [38]. The strategies how A.marina adapts to the high salinity are by secreting 95% of the salinity level through its roots in order to prevent the salty water enters the transpiration process,
keeping balance of the intracellular osmotic pressure in the vacuole or cytoplasm, adapting the anatomy of the wood and the salt excretion cells on leaves [39].

Meanwhile, in Station B (the fishponds areas in Kemujan Island), it is found three types of species, they are *R.apiculata*, *C.tagal* dan *E.agalloca* (*Table 3*). Both the Species of *R.apiculata* and *C.tagal* are found in Station C (tracking mangrove in Kemunjan Island). The Species of *R.stylosa* in Station A is also found in Station C. The best adaptation towards the water salinity based on the types of the species are *A.marina* p, the most tolerant, and followed by *R.stylosa* and *R.apiculata*, *C.tagal*, *L.racemosa* and *E.agallocha* [40]. The species of *R.apiculata* and *C.tagal* belong toRhizophoraceae that do not have a morphology adaptation to excrete salt or non-secreting [41]. The ability to secrete salt for the species of *C.tagal* is done by root ultrafication [42]. Mangrove plants have various kinds of roots for adapting the environmental conditions [43].

### 3.3 The anatomy of stomata on mangrove leaves

**Table 4.** The anatomy of stomata on mangrove leaves in three different stations

| Stations | Species  | Upper Stomata | Lower Stomata | Vast (µm) | Density(Num ber/mm²) |
|----------|----------|---------------|---------------|-----------|----------------------|
|          |          | Length (µm)   | Width (µm)    | Length (µm) | Width (µm)          |
| A        | *R.stylosa* | -             | -             | 35.67±1.1  | 11.55±1             | 325.67±29.3          | 193.78±76          |
|          | *A.marina* | -             | -             | 20.27±2.3  | 7.34±1              | 116.48±3.12          | 201.78±32.0        |
|          | *C.tagal*  | -             | -             | 37.01±3    | 10.94±2.4           | 321.85±83.6          | 251.56±55.5        |
| B        | *R.apiculata* | -            | -             | 30.03±4.9  | 7.84±1.23           | 185.73±48            | 242.67±18.5        |
|          | *E.agalloca* | -            | -             | 32.43±1    | 10.17±2             | 259.59±43.8          | 360±311.91         |
|          | *C.tagal*  | -             | -             | 32.75±0.5  | 9.21±2.73           | 238.76±34.1          | 186.67±34.7        |
| C        | *R.apiculata* | -            | -             | 31.88±4.0  | 9.15±0.53           | 230.76±34.1          | 282.67±25.4        |
|          | *R.stylosa* | -             | -             | 34.44±1.0  | 9.63±3.37           | 263.32±98.7          | 237.33±86.3        |
|          | *L.racemosa* | 23.49±2.1    | 9.35±1.5      | 27.51±10.1 | 9.81±0.79           | 209.91±62.5          | 252.44±53          |

The parameters used for doing the anatomy of leaf are the length, the width, the vast, and the number of the stomata (Table 4).

All types of species have stomata on the lower part of the epidermis, except for *L.racemosa* whose stomata is located on the upper part of epidermis and under the leaves. *Lumitzera racemosa* has stomata on the leaves and under the leaves [44]. The stomata is located randomly or neatly on the surface of the upper and lower epidermis of the leaf [45].

The longest stomata is discovered on the species of *C.tagal* in Station B (the fishponds areas in Kemujan Island) (Table 4). The widest stomata is found on the species of *R.stylosa* in Station A, followed by the species of *C.tagal* and *E.agalloca* in Station B. The length and the width of stomata on each type of the species in different areas vary in results. The most density of stomata is on the species of *E.agalloca* (Table 4). The density of stomata relies on the environmental factors such as the water supply, the light intensity and the temperature [46]. Stomata will close if the water salinity is high in order to minimalize the loss of water caused by the transpiration by reducing the carbon bonds [47][48]. Stomata will keep closing until the condition enables it to absorb water [49]. The size of the stomata and the numerous quantity of the stomata are the process of water efficiency and to prevent the loss of water [50].
3.4 The types of stomata

Table 5. The types of stomata based on the types of the species

| Stations | Species   | The Types of Stomata |
|----------|-----------|----------------------|
| A        | R.stylosa | Anomocytic           |
|          | A.marina  | Diititic             |
|          | C.tagal   | Parasitic            |
| B        | R.apiculata | Anomocytic      |
|          | E.agollaca | Diititic           |
|          | C. tagal  | Parasitic            |
| C        | R.apiculata | Anomocytic      |
|          | R.stylosa  | Anomocytic          |
|          | L.racemosa | Parasitic           |

There are three types of stomata possessed by the types of the species in three observation stations. They are anomocytic, diititic and parasitic. The type of anomocytic is found on the species of R.stylosa (Figure 1) and R.apiculata (Figure 5). Rhizophoraceae normally has the type of anomocytic [51]. The anomocytic type is the type in which the closure cells are surrounded by neighbouring cells [52].

The type of diititic Stomata is found on the species of A.marina (Figure 2) and E.agalloca (Figure 4). The leaves of A.marina have the type of diititic stomata [41]. E.agalloca is a small mangrove tree that belongs to Euphorbiaceae which has the type of diititic stomata [53]. The type of parasitic stomata is found on the species of C.tagal (Figure 3) and L.racemosa (Figure 6). Having neighbouring cells which have inline cells or in the same direction as the closure cells is the characteristic of parasitic stomata [54].

Figure 1. R.stylosa with anomocytic stomata
Figure 2. A.marina with diititic stomata
Figure 3. C.tagal with parasitic stomata
Figure 4. E.agalloca with diititic stomata
Figure 5. R.apiculata with anomocytic stomata
Figure 6. L.racemosa with parasitic stomata
4. Conclusion
The types of the species discovered in Menjangan Besar Island are A.marina and R.stylosa, while in the fishponds in Kemujan, the species found are C.tagal, R.apiculataand E.agallocha.next in tracking Mangrove in Kemujan, C.tagal, R.apiculata, R.stylosaand L.racemosa are found. The result relating to the observation of the leaf anatomy in these three locations has not to correlate significantly towards the threats of the environment, yet the environmental conditions in the fishponds show the rising in water salinity. That is why, it is recommended to grow mangrove with the type of Avicennia karimunjawa island for a reason it is more adaptable to high salinity on its leaves.

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