Bark Roughness as a Factor Affecting Cell Density of Epiphytic Microalgae (*Apatococcus Sp*)

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Abstract: *Green algae are diverse and ubiquitous in aquatic and some terrestrial habitats. They play a vital role in the global ecosystems for hundreds of millions of years. Differences between trees in bark topography can cause different epiphytic variation in community composition. The objectives of this research was to assess the response of number of algal cells towards bark roughness inhabiting 15 standing trees from field collections in the Central Region of Peninsular Malaysia, Putrajaya. Several methods were involved including field sampling, algal quantification, algal identification and physical assessment of the bark roughness. The study showed that trees with rough bark sustained higher density of microalgae (1140 $_\pm$ 405 cells/ml) compared to the smooth ones (1069 ± 434 cells/ml). The slight differences could be due to other physical parameters such as the degree of slope or inclination of the tree. As a conclusion, this study showed that trees with rough bark surfaces efficiently accumulates the atmospheric pollutants through its fissure and delayed water movement thus sustained more algal cells compared to trees with smooth bark.*

Keywords: *Bark roughness, Putrajaya, Green algae, Terrestrial algae, Alga density*

1. Introduction

Algae are known as omnipresent organisms on earth and able to grow in an extensive variety of conditions. Despite the fact that algae are typically known from marine and freshwater natural surroundings, they likewise happen in a wide variety of terrestrial environments [1], where they conspicuous growths in a few surface types, including rocks, urban walls, metals, tree barks, leaves and animal hairs [2]. Generally regular algae that can be found in basically every kind of terrestrial environment are cyanobacteria and eukaryotic algae [3], [4]. They are likewise called terrestrial algae as in they are not dependent on liquid water [5].

Since the 19th century, the variety of subaerial algae grow at tropical trees and leaves were investigated in different locations and habitat [6], [7]. Corticolous algae are plants from different altitude areas that grow on tree barks. They are exposed to the air and absorb water, minerals and other nutrients directly from the atmosphere.

Light and air humidity are considered as the most influential ecological factors on the growth and development of these organisms, while temperature is viewed as a secondary one [8], [9]. Bark pH of the surfaces has been considered one of the most important factors affecting the community structure of corticolous organisms [10]. The relationship between pH and community structure is well known for epiphytic lichens and bryophytes [11], [12] and myxomycetes [13]. The diversity of different algal groups is also influenced by light conditions [14]. Terrestrial algae have simple structures. It can efficiently accumulate the pollutant that associating with the algae. This accumulation make the algae become the good biological indicator. Aspect and inclination of tree boles also affects the degree of light exposure, but they can also influence other factors important to epiphytes

However, terrestrial microhabitats have less attention from phycologists than marine and freshwater habitats, so their diversity and community structure is still very poorly known [15], [16]. Epiphytic terrestrial green algae are the most widespread but the least known and most overlooked among all algal groups [17].

Hence, the present work has been carried out to study the effect of bark roughness towards subaerial algal density growing on tree bark.

2. Materials and Methods

2.1. Study Site

The samples were collected from Taman Wetland, Putrajaya (geographical coordinates N 02° 55.915 and E101° 40.909) (Figure 1).

Fig. 1: Map location of the sampling site in Putrajaya

The mean annual rainfall recorded at Putrajaya is about 2,000mm. The rainfall pattern reflects the annual monsoon cycles. The northeast monsoon occurs from December to March with the highest rainfall recorded around November/December. The southwest monsoon occurs from June to September. The period of lowest rainfall is around June/July. The mean annual air temperature is approximately 27 C with monthly mean air temperatures ranging from around 26 C in December to 28 C in May. The mean monthly minimum and maximum air temperatures range from about 20 C to 36 C in February, to about 22 C to 35 C in May.

2.2. Sample Collection

The bark of 15 trees with a trunk diameter of more than 10 cm was sampled for surface microbial growth at a height of 1.5 m above the soil level. Bark roughness was put into two categories (rough and smooth). Three quadrats of 10 x 10 cm were placed on 15 random trees. Algae within each quadrat were collected by scraping the bark surface with wetted cotton wool and then placed in a 100 ml sterile specimen tube containing 40 ml deionized water. The tube specimens were shaken vigorously to loosen the algae from the wetted cotton wool. All samples were stored in a refrigerator at 1-4ºC to prevent post-sampling growth. This study encompassed a collection of 45 tubes of algae (15 trees × 3 quadrats).

2.3. Sample Quantification

Algal samples on wetted cotton wool were squeezed to ensure all algal samples were transferred into sterile specimen tube. Algal samples were then shaken vigorously to dissociate clumped algae and to separate filamentous algae into fragments [18]. For quantification process, manual quantification of algal samples was conducted. The haemocytometer chamber was used as a counting chamber of algal cells. 1 ml of algal samples were pipetted into the well of haemocytometer and counted under light microscope.

2.4. Sample Identification

A Brunel digital light microscope was used to help in identification process and to count algal cells. A digital single-lens reflex (DSLR) camera was used as an external camera to capture algal images. Then, the captured images were compared to select the best algal images to facilitate algal identification during the initial stage. Photomicrograph of the algae was compared to the images in these sources in helping the identification process. [19], [20] and database from world's algal listings (www.algaebase.org) were used as references for algal identification up to the genus level. Main goal of photomicrography was to record permanently and accurately the view of an object using digital camera.

2.5. Bark roughness measurement

Bark roughness was visually estimated in two ordinal categories (1: rough, 2: smooth). They mostly reflected the interspecific differences among the tree species. The sample of bark roughness as shown in Figure 2.
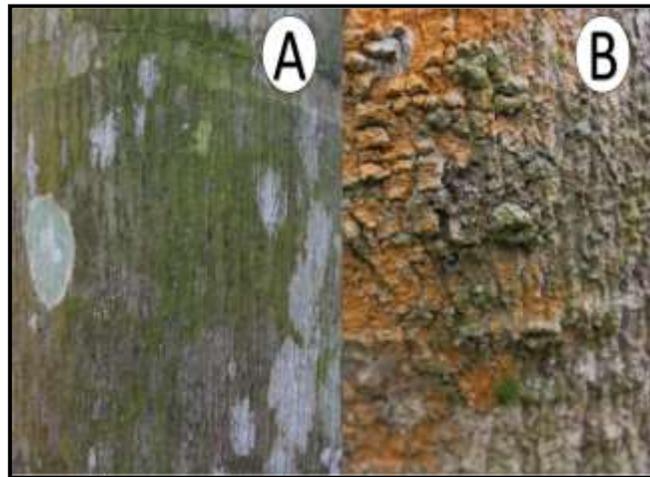

Fig 2: A. Smooth bark   B. Rough bark

3. Result

3.1. Algal species

Algae recovered from all the quadrats revealed that almost 100 % were of the green alga, *Apatococcus* sp. Another algal species namely *Trentepohlia sp* was also found but only in a very small quantity. Thus we considered *Trentepohlia* as unimportant in this study. *Apatococcus sp* as shown in Figure 3.
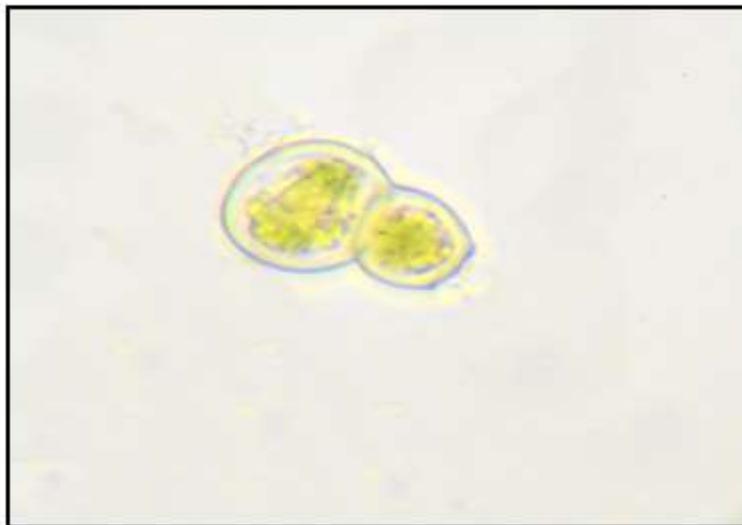
Fig. 3: *Apatococcus sp*

## 3.2. The relationship between bark roughness and algal density
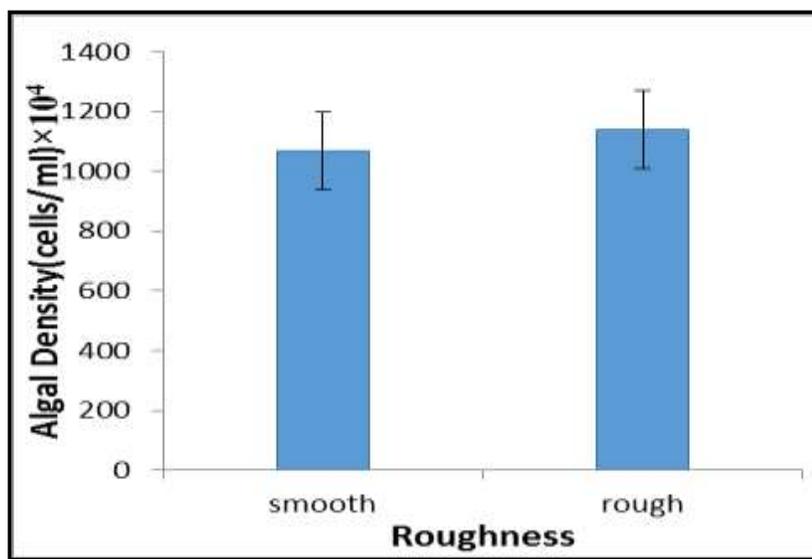
Fig. 4: Comparison of algal density between smooth bark and rough bark

From figure 4, an independent T-test was run on 15 samples to determine if there were differences in algal density based on bark roughness. Bark roughness of trees were classified into two categorical groups which smooth bark and rough bark.

There is no significant difference between algal density based on bark roughness of the trees. Although the mean of rough bark (mean=1140 cells/ml; sd=405) is higher than mean of smooth bark (mean=1069 cells/ml; sd=434), $t_{(5)} = 0.29$, p= 0.780., but this difference is not significant. All data were found to be normally distributed by using Normality Test.

4. Discussion

Based on Figure 4, algal density on the rough bark (1140 ± 405 cells/ml) was higher compared to smooth bark (1069 ± 434 cells/ml) but the overlapping of error bars shows that there was no significant difference between algal density based on bark roughness.

In this study, the rough bark shows the higher mean of algal density compared to the smooth bark. The rough bark is more efficient surface structure to grasp the water and moisture from rain and dew. In the past, [21] also found that it is often visually obvious that colonization is more extensive on rough and porous surfaces than smooth, non-porous ones. Lichen studies also showed the rough bark also provides a greater opportunity for thallus fragments of lichens to embed in its surface [22].

The growth of microalgae and lichens is known to be affected by bark topography and be an important factor for lichens growth [23], [24] probably because it affects the amount of moisture received. Differences between bark topography can cause adequate epiphytic variation in community composition.

By nature, all plants will undergo the desiccation process in any conditions. Tree bark is one of the tree components where the process will occur. The smooth bark sustained less algal density probably caused by this desiccation process. Some smooth-bark can be thin, making the bark temperature sensitive and exposed to the stress of desiccation [25]. The effect of these factors means that some smooth-barked trees typically do not support diverse or rich epiphytic assemblages.

[26] found that a rough bark surface more readily accumulates atmospheric pollutants than a smooth surface. The concentration of pollutants are highest in the surface layers of the outer bark, and decrease rapidly on moving towards the inner layers of bark. The bark crevices also affect to the pollutants accumulation in tree bark. The numerous of tree crevices, the more pollutant will accumulate in rough bark crevices. Previously, [27] and [28] claims that the higher nitrogen pollutant concentrations have often been reported to increase the rate of

algal growth. On the other hand, heavy metals and other compounds may be carried by the wind from the soil to the bark surface [29].

5. Conclusion

This study showed an increase of algal density on the rough bark compared to smooth bark. This result was supported by [30] which also stated that a rough surface more readily accumulates atmospheric pollutants than a smooth surface.

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