Borneo Journal of Resource Science and Technology (2022), 12(2): 142-146
DOI: https:doi.org/10.33736/bjrst.4642.2022

SHORT COMMUNICATION

Terminalia catappa Leaves Effects on Aquatic Primary Productivities

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Received: 3 April 2022 Accepted: 12 August 2022 Published: 31 December 2022

ABSTRACT

Terminalia catappa leaf, also known as Ketapang leaf, is well known for its medicinal properties and long history of use in aquaculture. Studies on the effects of leaf extracts on fish productivity are scarce. This study performed a time-series observation of ground young and mature *T. catapppa* leaves on tropical phytoplankton productivity. The young and mature leaves were prepared as fine powder separately. For the time-series experiment, 10 grams of each ground leaf were added to 10 L of lake water containing phytoplankton and incubated at room temperature with ambient illumination. The chlorophyll *a* saturation optical density was quantified on alternate days using a spectrophotometer and the data were analysed using Origin 6.0 Software. On Day 11, the chlorophyll *a* percentage had decreased by 73% and 81% in the control and mature leave treatment carboys, respectively. Meanwhile, the sample treated with young leaves of *T. catappa* showed only a 29% decrease. The results have shown that phytoplankton treated with young *T. catappa* powder demonstrated a positive but weak correlation ($R^2 = 0.123$), represented by the chlorophyll saturations. This study supported the hypothesis that young *T. catappa* leaves sustained phytoplankton growth.

Keywords: Chlorophyll *a*, Ketapang, lake, phytoplankton, tropical

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Southeast Asia is the natural home of the Ketapang tree, technically known as *Terminalia catappa* Linn. (Combretaceae) (Oboh *et al*., 2009). This tree typically grows in coastal areas due to its strong tolerance to winds and saline (Mohale *et al*., 2009). This tree is a sizable deciduous tree that can reach heights of 25 m and widths of 9 m. Close spirals form the arrangement of the leaves. The leaves mature during the dry season, changing from green to brown, gold, red, or yellow (Marjenah & Putri, 2017). Young leaves may prematurely separate from their branch due to animal foraging behaviours such as nibbling and tugging on tree foliage or colonising of leaf-cutting ants. The spreading fibrous root structure of *T. catappa* plays a crucial part in the stabilisation of the coastline (Marjenah & Putri, 2017).

Numerous studies have been conducted on this plant's numerous benefits (Chen *et al*., 2000, Weerawatanakorn *et al*., 2015). It is well-known for its medical, dietary, and healing properties (Oliveira *et al*., 2000; Anand *et al*., 2015). Mature fruits are cut open and the seeds are consumed uncooked as snacks (Weerawatanakorn *et al*., 2015). The vitamin C-rich fruit's fresh leaf juice is used to make ointments or consumed orally to treat illnesses (Anand *et al*., 2015). The leaves long-standing use in ethnomedicine may be due to their abundance in antioxidant and antibacterial phytoconstituents (Christian & Ukhun, 2006; Ahmad & Baharum, 2018).

Aquarists have reported that adding *T. catappa* leaves to their ponds caused their fish to grow more quickly and healthily. Despite extensive research on this plant's medicinal effectiveness (Mohale *et al*., 2009; Anand *et al*., 2015), there is little evidence to support its link to pond fish development. Other than the tannins that were released into the water's antifungal and antibacterial properties (Earl & Semlitsch, 2015), pond fish reproduction was primarily aided by the availability of food from main phytoplankton producers (Jaeger *et al*., 2021). The effects of immature and mature *T. catappa* leaves on chlorophyll *a* saturation in tropical lake waters is examined in this study. It is hypothesised that *T. catappa* leaves stimulate phytoplankton growth which raises fish biomass.

The *T. catappa* tree's young (green) and mature (red) leaves were collected from Pagar Alam, Lahad Datu, Sabah. The leaves were cut into small pieces and dried for 48 hours at 70 °C to hasten the drying process. The dry samples were ground into fine powder using a mortar and pestle. Phytoplankton samples were collected from a freshwater lake in Bangi using a Van Dorn sampler. To reduce zooplankton grazing, the samples were promptly filtered via a 120 µm mesh size filter. A completely randomised design was performed because there were few treatments and homogeneous experimental materials. In three duplicates, separate carboys containing 10 L of lake water were added 10 g of each of the green and red leaf powders of *T. catappa* for the experiment. The carboys were then let to sit on a window workbench with ambient lighting at room temperature of 25 °C. Nutrients were not added at any point during the experiment and untreated lake water served as a control.

Chlorophyll *a* concentration was used to assess the phytoplankton density in the water. From Day 1 until Day 11, 50 ml of culture subsamples from the control carboy and the lake waters treated with *T. catappa* were collected on alternate days for analysis. The samples were filtered on Whatman glass microfibre filters GF/F. The filter membranes were then submerged overnight in 10 ml of 90% aqueous acetone. The materials were then centrifuged to mix them before estimating the chlorophyll *a* saturation thoroughly. The extract's absorbance readings were performed at wavelengths of 630 nm, 647 nm, 664 nm, and 750 nm using Spectrophotometer UV-Vis Double Beam Model UVD-2950 (Labomed Inc., United States). The blank used was 90% aqueous acetone. The calculation of photosynthetic pigments was based on Eq. (1) and Eq. (2) (Gu *et al*., 2016):

$$\text{Chl-}a\ (\mu\text{g/L}) = \frac{(11.85E664 - 1.54E647 - 0.08E630) \times v}{\text{V} \times 1} \qquad \text{Eq. (1)}$$

$$\text{Chl-}b\ (\text{ug/L}) = (20.70)\text{E647} - (4.62)\text{E664} \qquad \text{Eq. (2)}$$

E = Absorbance at different wavelength, *v* = Volume of acetone (ml), V = Volume of water filtered (L)

All calculations including the reading absorbance values were performed in Microsoft Excel. The regression graphs of chlorophyll *a* content in *T. catappa* treated lake water and control were plotted using Origin 6.0 Software. Simple regression analysis at 95% confidence interval was used. Chlorophyll *a* was measured immediately after treatments, including control, and was given a high concentration in all cultures on Day 1 (Figure 1). As the time-series experiment continued, the chlorophyll *a* concentration in culture treated with mature *T. catappa* leaves and the control showed decreasing trends ($R^2 = 0.0985$) except for culture treated with young leaves where an ascending trend was observed ($R^2 = 0.123$) (Figure 1). By Day 10, the chlorophyll *a* percentage had decreased by 73% and 81% in the control ($R^2 = 0.5751$) and mature leave treatment carboys, respectively but only decreased by 29% in the sample treated with young leaves of *T. catappa.*

Chlorophyll *a* is a valuable indicator for algal biomass in the aquatic ecosystem. The heterogeneity of phytoplankton in lake waters precluded the need for acclimatisation for this experiment (Grear *et al.,* 2017). High levels of chlorophyll *a* in all samples on the first day (Figure 1) were due to saturated photosynthesis which corresponded to high light intensity and dissolved oxygen sufficiency in lake waters at mid-day (Luo *et al.,* 2017). The changes and differences in chlorophyll *a* saturation in both treated cultures in real-time suggest that the phytoplankton growth responded positively to the addition of young *T. catappa* leaves. The non-monotonous growth relationship with chlorophyll *a* saturation was affected by phytoplankton cell size and density (Zhou *et al*., 2012). Non-monotonic responses in natural phytoplankton assemblage and species-specific cultures to various experimental conditions were common and were not statistically significant (Heiden *et al.,* 2016; Grear *et al.,* 2017). Further examination should determine the composition of phytoplankton in the lakes.

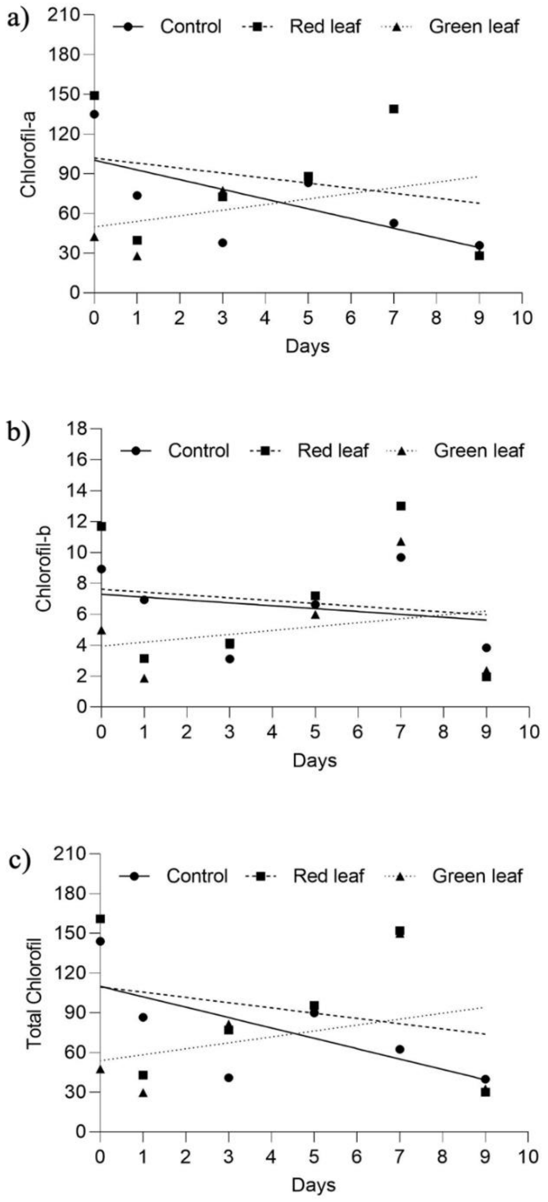

Figure 1. Chlorophyll concentrations in lake waters enriched with ground young and mature *Terminalia catappa* leaves in time-series observation. a) Chlorophyll *a* saturation against days; b) Chlorophyll *b* saturations against days; c) Total chlorophyll saturations against days.

Albeit the significant positive correlation in the sample treated with young leaves, the chlorophyll *a* saturation did not sustain. It coincided with a natural life span of phytoplankton and nutrient depletion as no additional nutrients were incorporated at the length of the experiment. In the wild, the young and mature leaves of *T. catappa* fell off the water through natural processes and animal activities. There were claims of the increased population of cultured fish in ponds by adding *T. catappa* leaves. Aya *et al.* (2019) demonstrated that providing *T. catappa* leaves and allowing them to decompose in culture tanks resulted in 48% of survival rates of Silver therapon (*Leiopotherapon plumbeus*) larvae, compared to the culture tanks without the *T. catappa* leaves which resulted in only 27% survival rates. The presence of *T. catappa* leaves would allow the colonisation of insect larvae and zooplankton on the leaf's surface which became food for the *L. plumbeus* larvae (Aya *et al*., 2019). Similarly, Ikhwanuddin *et al.* (2014) also found an increase in growth and survival rates of the Black tiger shrimp (*Penaeus monodon*) when provided with the extract of *T. catappa* leaves. It could be attributed to the decomposing leaf residues in the water, causing the rise in fish growth in ponds with a high N:P ratio (Das & Mandal, 2021).

Generally, fish growth is affected by the presence of free amino acids in fishmeal (Webster *et al*., 1995; Goswami *et al*., 2020). *Terminalia catappa* is rich in protein (Barku *et al*., 2012) with glutamic acid being the major amino acid (Ng *et al*., 2015). There was an increment in protease and lipase activities in fish fed with meals blended with *T. catappa* but it did not necessarily enhance growth (Goswami *et al.*, 2020). Instead, this study suggests the increased abundance of phytoplankton due to nutrient uptake from the young *T. catappa* extract. It boosted the primary productivities in the aquatic ecosystem and hence sustained the levels of dissolved oxygen as tannin in *T. catappa* leaves can reduce dissolved oxygen content in water (Earl & Semlitsch, 2015; Nugraha *et al*., 2021). Furthermore, the suspended solids and murkiness of water favour the proliferation of the microalgae Cynaophyceae (Miranda *et al.*, 2016). Those factors complemented with the *T. catappa* antimicrobial and antifungal abilities (Ahmad & Baharum, 2018) that protect the fish population from infection of potential pathogenic bacterioplankton in ponds exposed to the leaf detritus (Delmas *et al*., 1994).

This study concludes that over time, lake water treated with young *T. catappa* leaves showed a relative rise in chlorophyll *a* saturation, which suggested the proliferation of phytoplankton. A sustainable practice can be fostered to maximise the advantages by incorporating young *T. catappa* leaves into fish meals.

ACKNOWLEDGEMENTS

The authors would like to thank Malaysian Palm Oil Board Lahad Datu staff for the collection and preparation of Ketapang leaves. This study was funded by *Geran Galakan Penyelidik Muda* GGPM-2020-020 awarded by Universiti Kebangsaan Malaysia to Suriyanti SNP.

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