Effect of NPK and silicon fertilizer on growth, flowering, and nectar of *Turnera ulmifolia* L.

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**Abstract.** *Turnera ulmifolia* L. is a beneficial plant mostly planted in the oil palm plantation as it is easily adapting to a variety of environmental conditions, mainly to conserve beneficial insects. Nevertheless, the planted plants were left without proper maintenance and less study was conducted on biological control agent conservation. Thus, this study aimed to examine the effect of NPK, silicon fertilizer and their combination on the growth and development of *T. ulmifolia*. Treatments including T0 Control (No fertilization application), T1 (0.2 g NPK), T2 (0.2 g NPK+3.9 g silicon), T3 (0.2 g NPK+4.9 g silicon), T4 (3.9 g silicon) and T5 (4.9 g silicon) were applied 20 days after transplanting at monthly interval for three months. Results revealed the greatest height and number of branches were observed in T1. Overall, the chlorophyll content ranged between 27.92 to 31.18 SPAD values. All treatments gave the same effect on morphometric measurements on the first 30 days of observation. After 60 days, the greatest value for broad tube width and length was observed in T4 which differs significantly from other treatments. The application of fertilizer in T1, T2 and T3 showed 10-20% promotive effects over the control in the amount of nectar produced per flower. For all three observation periods, a similar trend was recorded for the total sugar concentration in flowers, where the mean total sugar content was between 1.37 and 1.61 mg per flower.

**Keywords:** Oil palm plantation; biological control; beneficial insects; chlorophyll content; morphometric measurements.

1. **Introduction**

The use of indigenous natural enemies of pests in conjunction with strategic cultivation techniques such as planting flowering plants that attract beneficial insects and applying organic fertilizer is becoming increasingly important. In oil palm plantation, *Turnera ulmifolia* L. was identified as a suitable species for enhancing the natural enemies of bagworm as it is favoured by predators and parasitoids. *Turnera ulmifolia* L., a member of the Turneraceae family [1] is native to Mexico and the West Indies and is commonly known as "Bunga Pukul Lapan". It is a beneficial plant that is easily adapted to a wide variety of soil and environmental conditions which makes it easier to colonize new habitats [2].

*Turnera ulmifolia* L. contains nectaries and have a high content of volatile compounds that are attractive to the natural enemies of the targeted crop [3]. Natural enemies such as parasitoids are dependent on a sufficient supply of pollen and nectar as essential dietary requirements. The quantity and quality of nectar and the number of flowers are critical for increasing parasitoid populations, and for the conservation of natural enemy populations. The sugar and amino acid composition of nectar and pollen, which varies greatly between plant species [4], is primarily responsible for floral resource food quality.

In most cases, *T. ulmifolia* were planted in oil palm plantations that were not maintained and not fertilized properly. Considering the benefits of fertilization that might alter the nectar composition of *T.*
ulmifolia, this gap in knowledge needs to be filled. This study hypothesizes that the application of fertilizers improves the growth, flowering, and nectar of *T. ulmifolia*. However, this effect depends on the type of fertilization and the amount of fertilizer applied. Fertilizers such as NPK and silicon were reported to have an advantage in flowering and quality of nectar. However, their effect on *T. ulmifolia* in the oil palm region has not yet been investigated. The present study was carried out to examine the effects of NPK, silicon fertilizer, and their combination on the growth, flowering, and nectar content of *T. ulmifolia*.

2. Materials and methods

2.1. Plant materials

Stem cuttings of *T. ulmifolia* measuring 15 cm long were placed in the plug trays with a cell size of 3 x 3 cm, containing soil and sand medium with a 1:1 ratio for rooting. The rooted cuttings were transplanted into 25 x 30 cm polythene bags filled with a 4 kg mixture of 3:2:1 topsoil, fine sand peat after a month. One plant was planted in each pot. Plants were placed under shade treatments for two weeks after planting. The basic fertilizer (NPK 15:15:15) was applied at 0.2 g as recommended by the manufacturer for each polybag. The experiment procedures were adapted from [5].

2.2. Experimental design

The study was carried out at Universiti Malaysia Sarawak. The experiment was laid out into five randomized complete block designs (RCBDs) with seven replicates for each block. The present study used 0.2 g NPK (15:15:20) as the main dosage. The fertilizer treatments were first applied 20 days after the transplant, then continued monthly for three months. There are five fertilizer treatments with one control treatment used in this study which were 0.2 g NPK (T1), 0.2 g NPK+3.9 g silicon (T2), 0.2 g NPK+4.9 g silicon (T3), 3.9 g silicon (T4), 4.9 g silicon (T5) and no fertilizer (T0 – Control).

2.3. Growth measurements and flowering

Evaluations on growth and flowering were assessed at 30, 60, and 75 days after the first fertilizer was applied. Plant height (cm) was measured from the base of the main stem to the tip of the youngest leaf using a measuring tape. The number of flowering branches was taken by counting all flowering branches. For chlorophyll content, three leaves were randomly selected and were measured using the Soil Plant Analysis Development (SPAD) chlorophyll metre. Flowering characteristics such as morphometric size was recorded.

2.4. Morphometric measurements

The morphometric measurements were based on [6] using a digital calliper (0.1 mm error). Three randomly selected flowers were chosen from each plant in the stage just after opening. The following phenotypic are shown in Figure 1: (1) narrow tube length (NL – the distance from the tube base to the base of the broad tube); (2) broad tube length (BL – the distance from the broad tube base to the top of the broad tube lobs); (3) narrow tube width (NW – quantified in the middle of the tube length); (4) broad tube width (BW – quantified in the middle of the tube length).

![Figure 1. Phenotypic of flower for measurement.](image-url)
2.5. Nectar collection
Flowers were collected at the same time of the day (08:00–11:00 am) at 30, 60, and 70 days after the first fertilizer application. The flower buds of approximately the same age and position about to open the next day were selected and covered with white plastic to prevent visitation by insects and possible contamination or nectar removal. The following day, the flowers were plucked, weighed, and placed into a falcon tube containing 10 mL distilled water to get the rinsate of the nectaries. The samples were taken into the laboratory, then stored at -20°C and kept until analyzed [6].

2.6. Sugar content analysis
HPLC was used to determine the type of sugar in *Turnera ulmifolia* L. Then, the sugar content of *T. ulmifolia* was determined using the phenol-sulfuric acid method [7] and glucose as a standard. Standard solution was prepared using different aliquots of glucose ranging from 0.1 to 1.0 ml from glucose stock solution (10 mg of pure glucose (GR Merck) dissolved in 100 ml of distilled water). The 5% phenol solution, followed by 5 mL of concentrated sulphuric acid were added to act as a catalyst for the colour development process. The absorbance was determined at 490 nm against a blank (1ml distilled water). The standard curve was prepared by plotting absorbance on excel.

Quantitative estimation of total sugars in the nectar was done according to the method described by [8]. The nectar was sampled from flowers using a washing method adapted from [9]. A total of 3-4 replications and a blank were maintained for analysis of nectar-sugars. The sugar concentration in nectar was determined from a standard curve by measurement of the absorbance at 490 nm to obtain the amount of sugar per flower (expressed as milligrams of glucose equivalent).

2.7. Statistical analysis
Using the SAS statistical software, all data collected were subjected to analysis of variance (ANOVA). Analysis of variance was performed separately for each treatment. Post hoc comparison of means was tested by the HSD Tukey test at a P < 0.05 significance level to determine if there was a significant difference.

3. Results

3.1. Effect of fertilizer on plant height, number of branches and chlorophyll content
Most of the growth parameters were significantly affected by NPK standard fertilizer practice (T1) at 30, 60, and 75 days after treatment (Table 1). The height of *T. ulmifolia* fertilized with T1 was significantly different from T0 and T4. About 13.7% increase in height for plants fertilized with T1 compared with the control plants at 30 days of observation.

| Treatment          | Plant height (cm)  | Number of branches |
|--------------------|--------------------|--------------------|
|                    | 30 days | 60 days | 75 days | 30 days | 60 days | 75 days |
| T0 (Control)       | 32.3±4.7 b      | 45.1±4.9 b      | 50.0±4.9 b | 2.7± 0.7 b | 4.0±0.7ab | 5.6±0.9abc |
| T1 (0.2g NPK)      | 37.1±3.1 a      | 50.7±3.2 a      | 56.5±2.8 a | 4.2±0.9 a  | 5.6±0.9 a  | 6.6±0.5 a  |
| T2 (0.2g NPK+3.9g Si) | 33.6±2.8 ab     | 48.5±2.7 ab     | 54.2±2.6 ab | 3.3±0.6 ab  | 4.7±0.6 ab  | 5.9±0.2 ab  |
| T3 (0.2g NPK+4.9g Si) | 33.1±3.7 ab     | 47.5±4.4 ab     | 52.8±4.1 ab | 2.6±0.7 ab  | 5.0±0.7 ab  | 6.0±0.6 ab  |
| T4 (3.9 g Si)      | 31.9±3.2 b      | 45.1±3.7 b      | 50.4±3.3 b | 2.0±0.4 b  | 2.3±0.4 b  | 4.1±0.5 c  |
| T5 (4.9 g Si)      | 34.0±3.8 ab     | 46.5±4.5 ab     | 49.9±3.8 b | 3.3±1.0 ab  | 3.9±1.0 bc  | 4.6±0.8 bc  |

Mean values followed by same letter within column did not differ significantly according to Tukey test at P < 0.05 (P < 0.05).

Number of branches per plant treated with T1 was also significant (P < 0.05) among various fertilizer treatments where the maximum values were recorded at 30, 60, and 75 days. The number of branches per plant were for T1 were increased 55.9% as compared to the control plants at 30 days of observation (Table 1).
The first 30 days of observation showed untreated (T0) *T. ulmifolia* exhibited the greatest chlorophyll content with 31.18 SPAD-units (Table 2). However, it was not significantly different with other treatments except with T3 and T4, which produced 29.10 and 27.92 SPAD-units, respectively. For all three observation periods, the chlorophyll concentration ranged from 27.92-31.18 SPAD-units.

### Table 2. The total chlorophyll content of *T. ulmifolia* at 30, 60 and 75 days after treatment.

| Treatment / Total Chlorophyll Content | 30 days        | 60 days        | 75 days        |
|--------------------------------------|----------------|----------------|----------------|
| T0 (Control)                         | 31.18±1.10a    | 30.54±0.35a    | 31.31±0.78a    |
| T1 (0.2 g NPK)                       | 29.88±0.81ab   | 29.75±0.55a    | 30.99±1.18a    |
| T2 (0.2 g NPK + 3.9 g silicon)       | 29.26±0.54ab   | 29.75±0.36a    | 30.65±0.63a    |
| T3 (0.2 g NPK + 4.9 g silicon)       | 28.10±1.02b    | 29.81±1.53a    | 31.16±1.08a    |
| T4 (3.9 g silicon)                   | 27.92±0.55b    | 30.11±0.53a    | 30.44±1.12a    |
| T5 (4.9 g silicon)                   | 29.39±0.95ab   | 29.81±0.59a    | 30.02±1.04a    |

Mean values followed by same letter within column did not differ significantly according to Tukey test at P < 0.05.

### 3.2. Effect of fertilizer on morphometric measurements

All treatments had the same effect on morphometric measurements at 30 first days of observation (Table 3). In the second period (60 days), the plants treated with T4 had a greater size of broad length and width compared to the control. For the effect on broad width, T4 was significantly different from other treatments, except with T5. For the last observation (75 days), T1, T2 and T3 treatments were only found significantly different from control on broad length.

### Table 3. Effect of NPK and Silicon on morphometric measurements of *T. ulmifolia* at 30, 60 and 75 days. (BW=Broad width; BL=Broad length; NW=Narrow width; NL=Narrow length).

| Treatment | 30 days | 60 days | 75 days |
|-----------|---------|---------|---------|
|           | BW      | BL      | NW      | NL      | BW      | BL      | NW      | NL      |
| T0 (Control) | 9.4±     | 9.5±    | 6.0±    | 9.8±    | 10.5±   | 3.9±    | 6.1±    | 10.5±   |
| T1 (0.2 g NPK) | 0.1a   | 0.2a    | 0.1a    | 0.1a    | 0.1a    | 0.1a    | 0.1a    | 0.1a    |
| T2 (0.2 g NPK+3.9 g silicon) | 0.1a   | 0.2a    | 0.1a    | 0.1a    | 0.1a    | 0.1a    | 0.1a    | 0.1a    |
| T3 (0.2 g NPK+4.9 g silicon) | 0.1a   | 0.1a    | 0.1a    | 0.1a    | 0.2a    | 0.2a    | 0.1a    | 0.1a    |
| T4 (3.9 g silicon) | 9.4±   | 10.0±   | 6.0±    | 10.3±   | 10.8±   | 4.0±    | 6.0±    | 10.6±   |
| T5 (4.9 g silicon) | 9.2±   | 10.4±   | 6.0±    | 10.8±   | 10.8±   | 4.0±    | 6.1±    | 10.6±   |

Mean values followed by same letter within column did not differ significantly according to Tukey test at P < 0.05.

### 3.3. Effect of fertilizer on nectar content

Our study reveals the main types of sugar in *T. ulmifolia* were fructose and glucose (Figure 2). The treatments did not significantly impact the nectar production compared to control (T0) (F3.53 = 3.31, P = 0.039) (Table 4). However, the application of T1, T2 and T3 showed 10-20% promotive effects over the control in the amount of nectar produced per flower. For all three observation periods, the same trend was recorded for the total sugar concentration in flowers, with the mean total sugar content between 1.37 and 1.61 mg per flower.
**Figure 2.** Chromatogram of type of sugar *Turnera ulmifolia* L. in a solution of ACN mixed with water.

**Table 4.** Effect of NPK and Silicon on nectar content of *T. ulmifolia* L. at 30, 60 and 75 days.

| Treatments                        | Total sugar content per flower |
|-----------------------------------|--------------------------------|
|                                   | 30 days | 60 days | 75 days |
| T0 (Control)                      | 1.37±1.10^a | 1.49±0.36^a | 1.43±0.78^a |
| T1 (0.2 g NPK)                    | 1.52±0.81^a | 1.70±0.55^a | 1.54±1.18^a |
| T2 (0.2 g NPK + 3.9 g silicon)    | 1.63±0.54^a | 1.43±0.36^a | 1.50±0.63^a |
| T3 (0.2 g NPK + 4.9 g silicon)    | 1.61±1.02^a | 1.71±1.52^a | 1.51±1.08^a |
| T4 (3.9 g silicon)                | 1.42±0.55^a | 1.56±0.53^a | 1.51±1.12^a |
| T5 (4.9 g silicon)                | 1.40±0.95^a | 1.61±0.59^a | 1.50±1.04^a |

Values followed by the same small letters are not significantly different among treatments within the flower trait at p < 0.05, based on HSD Tukey’s test.

4. Discussion

The present results indicated the vital role of N in plant life and its contribution to increase the height of *T. ulmifolia*. Nutrient fulfilment in the form of NPK fertilisation can improve the plant growth because elements of N, P, and K are nutrients that play an important role in the plant growth process [10].

All treatments did not have a significant effect on chlorophyll content compared to control. Numerous studies have shown that chlorophyll content in plants increases sharply following fertilisation with macroelements, particularly nitrogen [11]. The results of this study contradict prior findings, possibly due to insufficient NPK application for the formation of chloroplasts and accumulation of chlorophyll in *T. ulmifolia*. Nitrogen is a structural element of chlorophyll and protein molecules and thus influences chloroplast formation and chlorophyll accumulation in them [12].

Morphometric measurement was also not affected by the treatment applied for all observation periods in this study. Generally, the beneficial effects of Si on bud and flower growth are coupled to other variables (e.g., the species, type of soil substrate, the period and frequency of treatments, and other nutrient availability). It is also hypothesised that the effect of Si on blooming is becoming more marked if plants are under stress conditions [13]. These concepts presumably may explain why there were no effects of Si application on the flowering trait as noted in our study.

Fertilization with 0.2 g NPK with ratio 1:1:2, silicon and their combination showed a similar result in promotive influences on the nectar production of *T. ulmifolia* compared to untreated plants. This result agrees with the first attempt to investigate the effect of nutrient application on nectar secretion by [14] on non-Mediterranean plants. He concluded that nectar secretion is higher under low nitrogen which is similar to the minimum of NPK applied in the current study. This is also supported by [15] who
mentioned that the composition of nectar varies greatly depending on the plant species and the environmental conditions.

5. Conclusion
The study concluded that fertilisation of 0.2 g NPK per plant is adequate for optimum growth and showed a positive impact on flowering and nectar content of T. ulmifolia in semi-field conditions. This study can also serve as an optimised protocol for future studies on the physiological induction of flowering by nutrients. Further studies may be needed to find different levels of fertiliser application that can maximise the growth, flowering, and nectar of T. ulmifolia.

6. References
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