STUDIES ON THE EFFECT OF CAFFEINE ON THE GROWTH RATE OF *Ipomoea aquatica*

Nur Nazihah Mohd Hanafi[1], Nur Azizah Nadhirah Azni[1], Hanan Hasina Jahabar Maraikan[1], Wan Nur Farisha Alya Wan Mokhtar[1], Nor Azwin Nor Azam[1], Ahmad-Faris Seman-Kamarulzaman[1*]

[1]*Faculty of Applied Sciences*
Universiti Teknologi MARA Pahang, 26400 Bandar Jengka

[*]*Corresponding author: afkamarulzaman@uitm.edu.my*

Abstract

Ipomoea aquatica is a type of plant full of nutritional values. In increasing its production, chemical fertilizers are frequently used by farmers. An uncontrolled usage of chemical fertilizers, however, may cause environmental damage. Recently, large amounts of coffee residues are generated and reported to be used as a greener alternative fertilizer. The effect of caffeine in coffee residue has not yet been tested on *I. aquatica*. Hence, this research aims to determine the effect of caffeine in coffee residue towards *I. aquatica* and soil pH. The *I. aquatica* seedlings are divided into 5 groups with 5 seedlings in each group. Three groups are treated with 0.35% (w/v), 0.20% (w/v) and 0.05% (w/v) of caffeine while the other two groups are watered with AB fertilizer and tap water as positive and negative controls respectively. The caffeine treatment in this study shows no significant difference in the growth of *I. aquatica* even though it has changed the pH of the soils. However, the change of pH value still falls within the range of an optimum pH for *I. aquatica*, which is between 5.5 to 7 pH. The results of this study, on the other hand, still suggest an excessive use of caffeine. Future studies should be conducted with a lower level of caffeine in obtaining a positive effect of caffeine towards *I. aquatica*.

Keyword: Caffeine, Coffee residue, Potential fertilizer, *Ipomoea aquatica*, Soil pH

Introduction

Vegetables are a part of the agricultural crop that takes about 44, 000 hectares of land in Malaysia. Among the many types of vegetables, *Ipomoea aquatica* is one of the ten productive vegetables which consume majority of the land (Ibrahim et al. 2018). *I. aquatica* is commonly known as water spinach and popularly called as “kangkung” by Malaysians (Dua et al, 2015). The characteristic of having carotenes, flavonoids and some alkaloids has brought attention to this plant, as a source of natural product in drug discovery against diseases (Alkiyumi, 2012). The plant has also been used in China and Bangladesh for a long time as a traditional medicine to fight against various diseases such as diabetes, constipation, liver malfunction, and also in food poisoning treatment (Dua et al, 2015). The high content of crude fibre in *I. aquatica* is reported to help in lowering the serum cholesterol level, hypertension, breast cancer, and also the risk of coronary heart disease (Umar, 2007). Moreover, *I. aquatica* is also a potential functional food for humans and animals as it is full of nutritional goodness such as carotenes, minerals, amino acids, sugars, fibre, lipid, and also vitamins (Prasad, 2008). Various therapeutic properties in *I. aquatica* are founded while several researches have also been conducted in proving that the plant is an excellent source of medication (Kaur, 2016). Compared to tomatoes, *I. aquatica* has four times higher content of dietary fibre and protein (Umar, 2007). The various benefits of *I. aquatica* have led to an increase in its demand by the consumer. In meeting the demand, the farmers have started using chemical fertilizers to speed up the production of *I. aquatica*. Nevertheless, uncountable amount of chemical fertilizers may increase the risk of soil deterioration, greenhouse gas emissions and water contamination. Soil degradation is where the soil loses its agricultural quality due to the loss of organic matter and brings damage to the crops (Hillel, 2005; Ritsema, 2005). Excessive usage of chemical fertilizers may also lead to eutrophication where the nitrogen leached from the soil to the environment thus polluting groundwater (Wang et al., 2018). The coffee industry, in the meantime, has experienced constant growth. As a consequence, large amounts of residues are generated worldwide (Mussatto, 2011). One of the main coffee residues is the spent coffee ground which refers to the solid residues obtained after preparations of the coffee beverages. Several studies report that coffee waste has been synthesized and characterized as compost and fertilizer (Kasongo, 2011). Vigardt (2012) has founded that the usage of coffee residue as a type of fertilizer significantly improved the growth of *Spinacia oleracea* and increased the nutrients in the soil. The same result is obtained from the study on *Darnel ryegrass* and *Mentha piperita* (Hashimi, 2019). The growth of the treated plant has improved due to the characteristics of the coffee residue which contains nitrogen, phosphorus and potassium (Darnaudery et al. 2018). As it is generally known, nitrogen is an important type of nutrient for the growth of the plant (Zhang *et al*., 2018). One of the major components in coffee residue which is rich in nitrogen is the caffeine itself (Campos-Vega et al. 2015). However, the effectiveness of caffeine in the spent coffee ground is not yet tested as a potential fertilizer. Thus, this study aims to determine the effect of caffeine in coffee residue towards *I. aquatica* and soil pH.

Materials and Methods

Seed Germination

The nursery medium is prepared by weighing the soil ratio of 2: 1: 1 which is 2 for cocoa peats, 1 for organic soil and 1 for river sand. All of them are mixed well. Then, the mixture is filled in the seedling germination tray with a size (4.0 × 4.0 × 4.0) cm, ¾ of each pot. The *I. aquatica* seeds are planted with a thin layer of soil on top. Five seeds are planted in each pot of the tray. The seeds are then watered using tap water; 20ml for each pot. The process of watering the seeds is repeated twice a day using a syringe (once in the morning and once in the evening). The tray of seeds is covered with black plastic until the cotyledon leaf emerged out from the soil during seed germination. Then, the plastic is removed for the plants to grow. After the seedlings have produced two true leaves, 25 seedlings are chosen to be transplanted into 25 polybags.

Plant Growth and Treatment

Five seedlings are used for each treatment. The seedlings chosen are equal in height and number of true leaves. One seedling is transferred into each polybag. The seedlings are treated with tap water (negative control) and three different concentrations of caffeine. The concentrations of caffeine used are 0.35% (w/v), 0.20% (w/v) and 0.05% (w/v). The amount of caffeine supplied is 250mL, for four times per week. The plants are then observed weekly, by measuring the number of leaves and the height of the plants. The heights of plants are measured from the shoot to the stem using a ruler and a thread. The pH of soil is determined after four weeks of treatments.

Soil pH Determination

The pH values of the soils are determined by a pH meter. The pH meter is calibrated prior use. The pH of the soils is measured before and after all treatments.

Statistical Analysis
All the characterizations on the growth parameter of I. aquatica are conducted in five replicates. The data collected are presented as means ± standard deviation. The statistical analysis is performed using Minitab 18 software (Minitab, Inc., State College, Pennsylvania, USA). Statistical comparisons are made using the one-way analysis of variance (ANOVA), followed by Tukey's multiple-comparison test. The significant difference is determined with 95% confidence interval ($P < 0.05$).

Result and Discussion

The parameter to measure the growth of *I. aquatica* are recorded in **Table 1**, where all values are indicated as Mean ± Standard deviation (n=5).

Table 1 Parameters on *I. aquatica* of different treatments

Treatment	Height	No. of Leaves	Soil pH
-ve control	14.10 ± 2.63^a	11.00 ± 1.73^a	4.36 ± 0.08^a
0.05% (w/v)	14.24 ± 1.86^b	11.40 ± 0.55^a	5.07 ± 0.03^b
0.20% (w/v)	13.70 ± 1.83^b	10.00 ± 1.23^a	5.52 ± 0.02^c
0.35% (w/v)	11.24 ± 2.14^b	9.00 ± 1.41^a	5.23 ± 0.04^d

^{a,b,c,d} Means with different letter are significantly different from each other (P<0.05)

The Height and Number of Leaves of *I. aquatica*.
According to **Figure 1**, the highest *I. aquatica* plant is 14.24 cm (0.05%), followed by 14.10 cm (negative control), 13.70 cm (0.20%), and 11.24 cm. (0.35%). The same pattern is shown in **Figure 2**, where the highest mean of *I. aquatica* plant leaves number belongs to 0.05% (11.4 leaves). This is followed by a negative control (11.0 leaves), 0.20% (10.0 leaves), and 0.35% (9.0 leaves). The results show that there is an increase of growth throughout the 4 weeks of treatment. However, negative control has shown a significantly higher plant compared to the treated plant. There is no significant difference in height between all treated plants with the different concentration of caffeine. There is also no significant difference between the numbers of leaves for all treated plant.

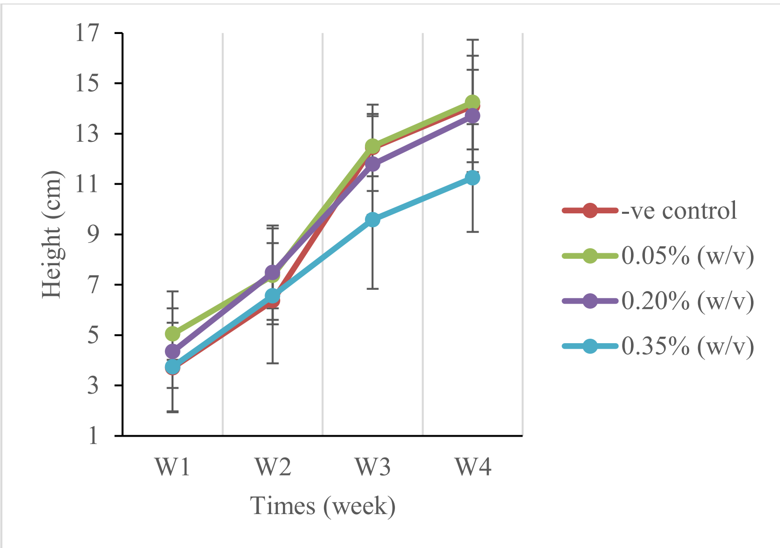

Figure 1 The graph on mean height of *I.aquatica* of different treatments in periods of four weeks.

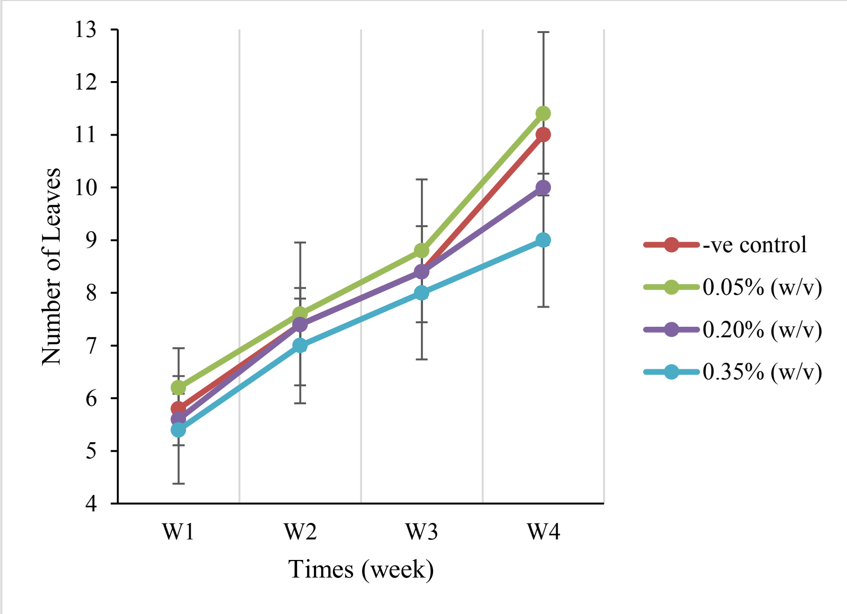

Figure 2 The graph on mean number of leaves of *I. aquatica* of different treatment in period of four weeks.

Treated plants are found to be inhibited as they are eventually smaller than the negative controlled plants. The inhibited growth is proposed to be caused by the allelopathic role of caffeine by millimolar amounts once it leaches into the soil with plant debris (Tanti, 2016). Meanwhile, Smyth (1992) has also testified that caffeine concentration greater than 0.05% (w/v) can inhibit the growth of plants. Therefore, the results obtained in this study indicate that using caffeine concentrations lower than 0.05% (w/v) can improve the plant's growth.

Soil pH

Figure 3 shows that all pH of the soils from the treated samples in this study shows a higher pH compared to the negative control. However, all of the soil pH falls within the range of 5.1-5.5 which is still in the optimum range for soil pH of *I. aquatica*. The higher pH of the soil is implied to be caused by the contribution of caffeine which is able to reduce the acidity of soil and improves the soil composition (Darnaudery at al. 2018).

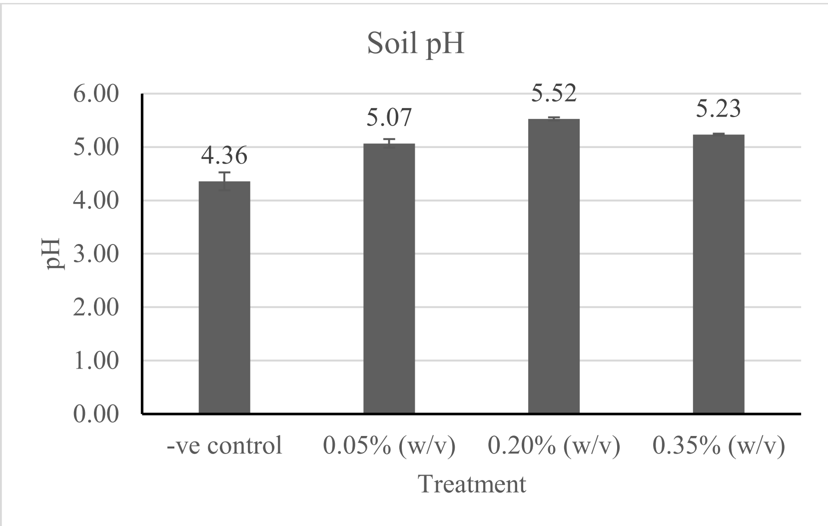

Figure 3 The graph of soil pH of different treatments

Conclusion

The caffeine which is used in this experiment shows no significant effect towards the growth of *I. aquatica* when compared to the negative control. The results suggest that the caffeine used in this study is too high; indirectly inhibiting or not promoting any growth effect towards the *I. aquatica* plant. The usage of caffeine has changed the pH of the soil, but the pH value still falls within the range of an optimum pH for *I. aquatica,* which is from 5.5 to 7.0. Hence, future studies should be conducted using a much lower caffeine concentration (lower than 0.05% (w/v)) in order to measure the positive effects of caffeine towards the growth of *I. aquatica*.

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Conflict of interests

Author declares no conflict of interest.

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