

## Effects of MARDI Nano-Fertilizer Application Frequency on the Growth of Rock Melon

Nadia Izati Fadzil<sup>a,b</sup>, Noor Azlina Masdor<sup>a,\*</sup>, Mohd Nor Mohd Rosmia<sup>a</sup>, Nur Sabrina Wahid<sup>a</sup>, Nor Suzaida Mohd Nor<sup>a</sup>, Muhamad Shafiq Abd. Karim<sup>a</sup>, and Mohd Firdaus Mohd Anuar<sup>b</sup>

<sup>a</sup>Biotechnology and Nanotechnology Research Centre, MARDI HQ, Persiaran MARDI-UPM, 43400 Serdang, Selangor, Malaysia.

<sup>b</sup>Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia.

\*Corresponding author. Tel.: +603-8953 6138; e-mail: azlina@mardi.gov.my

### ABSTRACT

For many years, fertilizers have been utilized in agriculture to increase the fruit yield and quality of various crops, which in turn will increase the nutritive content of products. Recently, MARDI has developed a nano-fertilizer based on nanoemulsions to increase the productivity of rock melon. The development of nanoemulsion-based fertilizer was carried out using the high-energy emulsification method which has significant advantages, such as enhancing the solubilization of active constituents while preserving biological activity. In this work, the effects of the vegetative growth of rock melon on different application frequencies (T1=0, T2=2, T3=4, T4=6, and T5=8 times) of MARDI nano-fertilizer through foliar treatment were studied. The results showed that the vegetative growth of the rock melon including plant height, stem diameter, relative chlorophyll content, and leaf area of rock melon were significantly ( $p \leq 0.05$ ) increased by 15.07%, 7.8%, 41.5% and 41.5%, respectively at T3, photosynthetic pigments of Chlorophyll a by 21.1% at T5, Chlorophyll b and carotenoid by 24.99% at T4 treatment. The dry weight biomass did not vary significantly in all treatments but showed an increase in dry biomass over the nano-fertilizers treatment compared to the control. These findings indicate that adding the foliar application of MARDI nano-fertilizer produces optimal growth in rock melon plants.

**Keywords:** Foliar, nano-emulsion, nano-fertilizer, rock melon

### 1. INTRODUCTION

Rock melon (*Cucumis melo*) is one of the most promising new fruit varieties and one of the most profitable commercial commodities, with demand potential in both domestic and international markets. Rock melon is high in potassium, beta-carotene, and vitamin C while being low in fat, salt, and cholesterol [1]. According to the Malaysian Department of Agriculture (DOA), this crop produced 5,845.81 metric tonnes on 313.4 hectares (ha) of planted land across the country in 2018, translating to an average yield of around 18.65 metric tonnes per ha of land planted. According to APCO Worldwide (2017), Malaysia is a prominent exporter, with rock melon output increasing by 13% in the five years to 2014 [2].

Fertilization is one of the most practical and efficient pre-harvest methods for controlling and improving output and nutritional quality [3]. However, most conventional fertilizers have low nutrient utilization efficacy as a result of leaching, resulting in significant economic losses [4]. To overcome all these drawbacks, nano-fertilizer offers an alternative solution to overcome the disadvantages of conventional fertilizer in an efficient way. Nano-fertilizer improved crop growth, yield, and quality parameters by increasing nutrient use efficiency, decreasing fertilizer waste, and reducing cultivation costs. [5].

Recently, MARDI has developed a nano-fertilizer where fertilizer minerals, NPK-TE held in a nanoemulsion formulation. Nanoemulsion fertilizer is one of the nano-fertilizer that is made by combining a surfactant with a liquid and mixing it with high energy to create a stable, homogeneous combination. The surfactant aids in the suspension of liquid droplets in the water-soluble matrix, allowing them to remain equally dispersed throughout the solution. The droplets are typically less than 100 nanometers (nm) in diameter, far smaller than the particles present in standard fertilizers [6]. Plants rapidly absorb and use the droplets, resulting in increased yields and crop health.

MARDI nano-fertilizer is applied as a foliar spray to enhance direct nutrient absorption into the plant system, thereby decreasing fertilizer waste [7]. However, to prevent stunted plant growth and low-quality rock melon, it is crucial to apply the correct volume of MARDI nano-fertilizer. Thus, in this study different MARDI nano-fertilizer application frequencies through foliar spray on plant growth of rock melon F1 hybrid, Sakata Glamour was investigated to determine the correct volume of fertilizer that contains sufficient nutrients for plant assimilation.

## 2. MATERIAL AND METHODS

### 2.1 Production of MARDI Nano-Fertilizer

The nano-fertilizer formulation which consists of neem oil (provided by Agrow Synergy Sdn. Bhd, Malaysia), surfactant polysorbate, and water was developed based on the selected points from the ternary phase diagram constructed and incorporated into the composition of NPK-TE using a combination of low and high-energy emulsification procedures. Neem oil was first mixed with a surfactant to form an oil phase. After that, the NPK-TE and water were added and homogenized at 1500 rpm for 25 minutes (min) (Ika T25 digital Ultra Turrax, USA). Then, the mixture was mixed and sonicated (Q500 sonicator, Qsonica, USA) for 30 min at room temperature [8], [9].

#### 2.1.1 Physicochemical Characterization of the Optimized MARDI NANO-Fertilizer

The particle size and polydispersity index (PDI) value of the nano-fertilizer were measured using Zetasizer (Brookhaven Instrument, USA) at 25±5°C. The measurement was performed using a dynamic light scattering (DLS) technique with a scattering angle of 90°. The samples were diluted with ultrapure water (1:20) and injected into the sample cell. The intensity distribution measured the mean average (z-average) particle size. The measurement was repeated in triplicate.

### 2.2 Study Site and Planting Material

The field trial was conducted in a netted rain house shelter (18.29 m x 36.58 m) at the Department of Agriculture, Serdang. In this research, the rock melon F1 hybrid, Sakata Glamour, was used. First, rock melon seeds were planted in peat moss on a seedling tray and kept in a controlled environment with 75% relative humidity and a temperature of 27°C for two weeks. After that, the seedlings were moved into polybags (16 cm x 16 cm x 30 cm) with cocopeat media in a rain house shelter according to the Randomized Complete Block Design (RCBD) arrangement.

### 2.3 Transplanting Seedlings, Plant Maintenance and Pollination Method

The two weeks of healthy and uniform seedlings were selected and transplanted into the polybags late in the evening to avoid heat stress. Seedlings were watered immediately after transplanting. A total of 400 seedlings were transplanted for the experiment.

The plant was irrigated using the fertigation technique as a channelled drip system and was watered two times daily for ten minutes (60 mL) each-fertilizer formulation used for the plant nutrients as stock (A) and (B). Stock A is a mixture of calcium nitrate (11 kg) and iron chelate (0.19 kg) in a container with 100 L of water that was stirred to dissolve into a liquid. Meanwhile, stock B is a mixture of potassium nitrate (10 kg), monopotassium hydrogen phosphate (MKP) (2.63 kg), magnesium sulphate (5.64 kg), manganese sulphate (5.64 kg), Hibor (0.033 kg), zinc

sulphate (0.012 kg), copper sulphate (0.002 kg) and molybdate (0.002 kg) in 100 L of water and was stirred to dissolve into the liquid. Then, the two-nutrient mixture was poured into the water tank to achieve the required electrical conductivity (EC).

The amount of nutrient concentration in the water tank according to days after transplanting (DAT) was measured by an EC meter. At 1-9 DAT, the EC nutrient was increased at 1.4-1.6 mS/cm, 9-15 DAT= 1.6-1.8 mS/cm, and the maximum is 3.5 mS/cm according to the procedure of the DOA, Serdang. The plants were supported by string and grew vertically at 0.61 m x 1.22 m between polybags. The plants were pruned to eight nodes and allowed to develop freely to stimulate fruit development. Standard management practices such as intercultural operations, including pruning, watering, and pesticide application, were performed using conventional methods. The rock melon plants were pollinated by hand pollination. The male flower was carefully removed from the plant, and the petals were striped to leave the stamen. The stamen was then carefully inserted into the female flower and gently rubbed on the stigma to transfer the pollen, which was done 26 DAT, soon after the female flower emerged. The pollination was carried out daily until complete pollination had been achieved.

### 2.4 Effect of the MARDI Nano-Fertilizer Application Frequency on the Growth Of Rock Melon

#### 2.4.1 Experimental Design

The experiment was conducted following the RCBD in quadruplicates. The best-performing concentration of MARDI nano-fertilizer at 0.5% (v/v) was optimized previously and selected as the foliar application in the field experiment. The solution was mixed correctly and sprayed on the rock melon leaf surface. The fertilizer was applied following the application frequency stated in Table 1 as an independent variable in addition to the standard AB fertilizer (CFs, conventional AB fertilizer) applied via the fertigation system. The flow rate from the spray valve was 0.85 L per minute. The walking speed along the net house was roughly 35.2 m per min. A mature rock melon plant with no water buds and leaves at 20–25 required an estimated 100 mL of formulation spray to cover the whole plant.

**Table 1** The frequency of MARDI nano-fertilizer treatment

Treatment	Frequency	Application (DAT)
T1	Control (CF)	-
T2	2S	7, 14
T3	4S	7, 14, 21, 28
T4	6S	7, 14, 21, 28, 35, 42
T5	8S	7, 14, 21, 28, 35, 42, 49, 56

CFs: Conventional AB fertilizer by drip irrigation.

## 2.4.2 Vegetative Growth Analysis

At 7, 21, and 35 DAT, destructive and non-destructive data were collected at three stages of plant growth: early vegetative, vegetative, and reproductive. The plants were randomly sampled from each treatment to determine plant height, stem diameter, relative chlorophyll content, leaf area, determination of photosynthetic pigments, and dry weight biomass. Measurements of plant height were taken from the surface of the media in the polyethene bag to the highest shoot tip using measuring tape. The stem diameter was measured at the lowest part of the stem using an Electronic Digital Calliper (Model SCM DIGV-6). Leaf areas were measured and recorded as the total leaf area per plant using an automatic leaf area meter (MODEL LI-300, LICOR). The plant stems and leaves were then divided, and after 72 hours at 75°C in a drying oven, the dry weight of each component was calculated (UF260, Memmert, Schwabach, Germany).

## 2.5 Statistical Analysis

The field experiment followed the RCBD in quadruplicates with 5 treatments and 20 plants per replication. Data were subjected to a one-way Analysis of Variance (ANOVA) using IBM SPSS Statistics Version 25.0 (Armonk, NY: IBM Corp). In addition, the means were compared by the Post Hoc Tukey test at the significance level of 0.05.

## 3. RESULTS AND DISCUSSION

### 3.1 Physicochemical Characterization

The particle size distribution of the optimized MARDI nano-fertilizer formulation containing NPK-TE obtained was  $90.32 \pm 0.015$  nm. The PDI was  $0.162 \pm 0.004$ , indicating a monodisperse system with a zeta potential of  $-39.3 \pm 0.781$  mV. Generally, the zeta potential value of  $\pm 30$  mV was considered a stable nanoemulsion-based nano-fertilizer formulation.

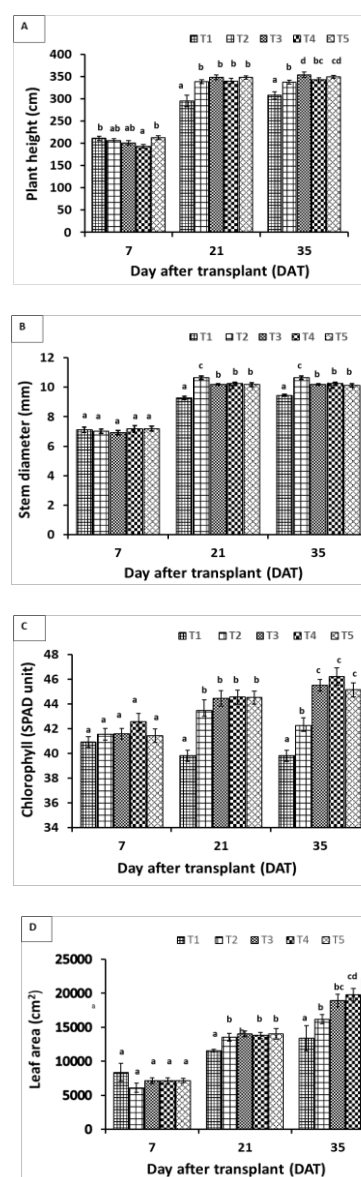
A high zeta potential value explains high repulsive electrostatic interaction between charged emulsion particles to prevent the particle from getting closer. The values provide information on the charge present on the particle surfaces where strong repulsive force prevents particle aggregation. The high stability was associated with a repulsive force which exceeded the attracting Van der Waals forces and produced dispersed particles of a deflocculated system. The interaction between the amount of surfactant and oil in the formulation could impact the zeta potential value [10]. A sufficient amount of surfactant compared to the amount of oil could affect the stabilization of formulation by manipulating surface charge due to electrostatic repulsion between particles [11].

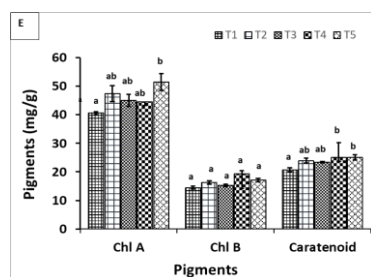
### 3.2 Vegetative Growth

The application of MARDI nano-fertilizer at different treatment frequencies contributed to the increase in vegetative growth of rock melon plants compared to conventional AB fertilizer alone (Figure 1A-E). After 35

days after transplant (DAT), the plant height (Figure 1A) increased significantly ( $p \leq 0.05$ ) by 15.07% at T3, followed by T4, T5 and T2 treatment. T2 showed the highest stem diameter (Figure 1B) at 12.7%, then T4, T3 and T5. Meanwhile, the chlorophyll content by SPAD unit (Figure 1C) was highest by T4 at 16.08% and leaf area (Figure 1D) at best of T5 at 65.88%, compared to control (T1) respectively. It was found that the treatment with T5 enhanced Chl-a pigment synthesis by 21.2% and T4 increased the Chl-b content and carotenoid at a best of 24.99% (Figure 1E) compared to T1.

These findings indicate that adding the foliar application of MARDI nano-fertilizer enhanced the growth and development of rock melon plants and enhanced the photosynthetic pigments in rock melon leaves over the growing period. However, four times the application of MARDI nano-fertilizer, T3 was chosen as the best application in this study because T3 gave no significant difference with other nano-fertilizer application frequencies. It was also considered economical as less fertilizer was used.





**Figure 1.** Vegetative growth parameters after 7, 21 and 35 DAT for A) plant height; B) stem diameter; C) chlorophyll Index using SPAD meter; D) leaf area and E) photosynthetic pigments after 70 DAT. Mean $\pm$ SEM followed by the same letter indicated no significant difference at  $p \leq 0.05$ .

control. The dry weight of leaves and stems did not vary significantly in all treatments but showed an increase in dry biomass over the MARDI nano-fertilizer treatment compared to the control.

The present study yielded comparable results to those reported by Márquez-Prieto et al. [16], indicating the absence of statistically significant variations in the biomass of green beans (cv. 'Strike') across varied dosages of K nanofertilizer. Nevertheless, the application of a concentration of 50 parts per million (ppm) of potassium (K) nanofertilizer resulted in a significant enhancement in biomass accumulation. Specifically, there was a notable increase of 23.05% compared to the application of 200 ppm of K nanofertilizer, and a modest increase of 1.18%

**Table 2** The dry weight of leaves (DWL) and stem (DWS) (g) in different treatments at different DAT

Treatments	7 DAT		21 DAT		35 DAT	
	DWL	DWS	DWL	DWS	DWL	DWS
T1	16.61 $\pm$ 1.89a	12.20 $\pm$ 0.53b	40.00 $\pm$ 1.89a	16.84 $\pm$ 1.23a	37.5 $\pm$ 6.75a	35.00 $\pm$ 3.27a
T2	18.53 $\pm$ 1.58a	10.91 $\pm$ 0.09a	43.75 $\pm$ 1.83a	24.21 $\pm$ 3.27a	66.67 $\pm$ 5.16b	36.67 $\pm$ 1.673a
T3	24.50 $\pm$ 7.62a	10.91 $\pm$ 0.476a	46.25 $\pm$ 5.32a	20.70 $\pm$ 3.26a	57.5 $\pm$ 4.91ab	37.50 $\pm$ 5.59a
T4	18.62 $\pm$ 1.10a	10.68 $\pm$ 0.04a	46.25 $\pm$ 2.63a	20.63 $\pm$ 1.58a	52.5 $\pm$ 6.75ab	37.50 $\pm$ 6.75a
T5	20.23 $\pm$ 0.96a	10.89 $\pm$ 0.09a	48.75 $\pm$ 3.98a	18.55 $\pm$ 2.54a	60.00 $\pm$ 8.02ab	38.33 $\pm$ 4.02a

Each value is an average of 4 replications, and each replication contains 2 plants (n=8). Mean $\pm$ SEM followed by the same letter indicated no significant difference at  $p \leq 0.05$ .

The nanoemulsion system's efficiency in distributing the fertilizer macro and micronutrients across rock melon leaves may be responsible for this finding. Hydroponic tomato plant development followed a similar pattern as well [12], wheat plants [13] and pot marigold (*Calendula officinalis*) plants [14] with the foliar application of macronutrients, micronutrients, and nickel (Ni), respectively. The study from Kumari et al. 2019 [9] also reported zinc-functionalized thymol nanoemulsion by the foliar application at 0.02 to 0.06% (v/v) in the field significantly increased grain yield up to 16.6% as compared with bulk thymol application and up to 50% from control. The exploited plant-based thymol and saponin as nanoemulsions could be further improved for more promising crop applications, mainly towards growth yield. The physicochemical properties of plant oil extract could be a docking site for functionalizing nanoemulsion to be used as a vehicle for the sustainability of delivering macro and micronutrients.

The crop yield is generally related to accumulating biomass as fresh and dry matter in the organs of importance for the harvest [15]. The dry weight of leaves and stems increased with plant age (Table 2). Treatment of MARDI nano-fertilizer at 21 DAT of T3 and T4 increased the dry weight of leaves to 15.63% and T5 to 21.88%. At 35 DAT, T2 gives the highest dry weight increase at 77.79% compared to the control. By 21 DAT, T2 had the highest stem dry weight; at 35 DAT, T3 and T4 had the same 7.14 % rise, and T5 had a 9.15% increase from the

compared to the application of 100 ppm of K nanofertilizer. Furthermore, the administration of the three doses resulted in a significant rise of more than 35.6% compared to the control group without any treatment.

#### 4. CONCLUSION

The utilization of MARDI nano-fertilizer has demonstrated a noteworthy positive impact on the growth, development, and nutritional characteristics of the rock melon plant. The application of MARDI nano-fertilizer at treatment levels T2 to T5 resulted in significant increases in growth metrics such as plant height, stem diameter, chlorophyll content, leaf area, and pigments.

In summary, the preferred treatment for the administration of MARDI nano-fertilizer in this study was T3, which involved four applications at 7, 14, 21, and 28 DAT. This treatment was favoured because of its reduced labour requirements and economic viability, while still resulting in optimal development and high-quality rock melon production. Therefore, the use of MARDI nano-fertilizer has the potential to serve as a viable alternative to conventional fertilizers in the field of agriculture, as it can effectively improve the efficiency of nutrient utilization, and overall crop yield, and contribute to the establishment of a sustainable ecological system.

## ACKNOWLEDGEMENTS

The author would like to thank the Malaysian Agricultural Research and Development Institute (MARDI) and Universiti Putra Malaysia (UPM) for supporting this research.

## REFERENCES

- [1] Sidik, N. J., Hashim, S. N., Mohd, Y. S., Abdullah, S. (2013). Characterization of plant growth, yield, and fruit quality of Rockmelon (*Cucumis melo*) cultivars planted on soilless culture. *Journal of Plant Sciences*. Volume: 7 Issue 2. 186-193.
- [2] Liyana, N. A., Pebrian, D. E. (2020). Energy use pattern analysis of rockmelon (*Cucumis melo*) production in Malaysia: a case study. *Food Research*. Volume: 4. 34-39.
- [3] Zulkarami, B., Ashrafuzzaman, M., Mohd Razi, I. (2010). *Journal of Food, Agriculture and Environment*. Volume: 8 Issue: 1. 249-252.
- [4] Ibrahim, I. I. (2022). Efficacy of biochar and NPK fertilizer on soil properties and yield of okra (*Abelmoschus esculentus* L.) in Guinea Savanna region of Nigeria. *Journal of Environmental Bioremediation*. Volume: 5. 6-10.
- [5] Dimkpa, C. O., Bindraban, P. S. (2016). Fortification of micronutrients for efficient agronomic production: a review. *Agronomy for Sustainable Development*. Volume: 36. 7.
- [6] Yadav, A., Yadav, K., Abd-Elsalam, K. A. (2023). *Agrochemical*. Volume: 2. 296-336.
- [7] Mali, S. C., Raj, S., Trivedi, R. (2020). Nanotechnology a novel approach to enhance crop productivity. *Biochemistry and Biophysics Reports*. Volume: 24.
- [8] Saharan, V. (2010). Effect of gibberellic acid combined with saponin on shoot elongation of *Asparagus officinalis*. *Biologia Plantarum*. Volume 54. 740-742.
- [9] Kumari, S., Choudhary, R. C., Kumaraswamy, R. V., Bhagat, D., Pal, A., Raliya, R., Biswas, P., Saharan, V. (2019). Zinc-functionalized thymol nanoemulsion for promoting soybean yield. *Plant Physiology and Biochemistry*. Volume: 145. 64-74.
- [10] Sarheed, O., Dibi, M., Ramesh, K. (2020). Studies on the effect of oil and surfactant on the formation of alginate-based O/W lidocaine nanocarriers using nanoemulsion template. *Pharmaceutics*. Volume: 12. 1-21.
- [11] Massarweh, O., Abushaikha, A. S. (2020). The use of surfactants in enhanced oil recovery: A review of recent advances. *Energy Reports*. Volume: 6. 3150-3178.
- [12] Roosta, H. R. (2011). Effects of foliar spray of different Fe sources on pepper (*Capsicum annum* L.) plants in aquaponic system. *Scientia Horticulturae*. Volume: 122 Issue: 3. 396-402.
- [13] Abdel-Aziz, H., Hasaneen, M. N., Omar, A. (2018). Effect of foliar application of nano chitosan NPK fertilizer on the chemical composition of wheat grains. *Egyptian Journal of Botany*. Volume: 58 Issue: 1. 87-95.
- [14] Sobati-Nasab, Z., Alirezalu, A., Noruzi, P. (2021). Effect of foliar application of nickel on physiological and phytochemical characteristics of pot marigold (*Calendula officinalis*). *Journal of Agriculture and Food Research*. Volume: 3. 100-108.
- [15] Barrientos Llanos, H., del Castillo Gutiérrez, C. R., García Cárdenas, M. (2015). Functional analysis of growth, biomass accumulation and translocation of dry matter eight vegetables grown in greenhouses. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*. Volume: 50 Issue:1. 1-12.
- [16] [Márquez-Prieto, A. K., Palacio-Márquez, A., Sánchez, E., Macias-López, B. C., Pérez-Álvarez, S., Villalobos-Cano, O., Preciado-Rangel, P. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*. Volume: 50. 1-12.