



## Seed Germination, Seedling Growth and Photosynthetic Responses to Temperature in the Tropical Tree *Moringa oleifera* and Its Relative Desert, *Moringa peregrina*

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*MORINGA oleifera* and *Moringa peregrina* are cultivated in many areas in Almadinah Almunawwarah, western part of Saudi Arabia. The aim of the present study is to examine the responses of seed germination, seedling growth and photosynthetic performance for two *Moringa* species to different temperature treatments; 15/12°C, 25/22°C and 35/32°C; day/night. The obtained results indicated that seeds of both species could germinate at a wide range of temperatures with optimum temperature for highest percentage of germination, at 25°C. The results showed that most parameters of growth and the photosynthetic performances of the two species were highest at 25°C. However, the chlorophyll contents of *M. oleifera* and most values of chlorophyll fluorescence for both species were similar across the three temperature treatments. Additionally, the outcomes of this study showed that *Moringa* can be cultivated in many different regions with different temperatures.

**Keywords:** *Moringa oleifera*, *Moringa peregrina*, Photosynthetic responses, Seed germination, Seedling growth, Temperature.

### Introduction

The *Moringa* genus contains 13 tree species and belongs to the Moringaceae family, most of them are found in the tropics (Olson & Carlquist, 2001). *Moringa oleifera* is the most important species and the most widely distributed in Asia, Africa, and America due to its economic importance (Ramachandran et al., 1980; Leone et al., 2015); it believed to be native to sub-Himalayan north-eastern India (Pandey et al., 2011). The second most important species is *Moringa peregrina*, which is distributed in the arid regions of the Arabian Peninsula (El-Lamey, 2015). *M. oleifera* is a medium-sized tree reached 3-15m high (Boulos, 1999); it is a fast-growing tree and is believed to be tolerant to drought and heat (Hegazy et al., 2008). *M. oleifera* is used for multiple purposes, including food, feed, fuel and

cosmetics, which has resulted in its widespread use, especially in the recent decades (Leone et al., 2015; Boukandoul et al., 2018). In Africa and Asia, *Moringa* is used as human food and in medicines because of the high content of minerals and vitamins in the different parts of the tree (Abd El-Hack et al., 2018; Khor et al., 2018). For example, it was reported that *M. oleifera* leaves contain considerable amounts of proteins, minerals, and amino acids, which make the plant a valuable fodder crop (Amaglo et al., 2010; Leone et al., 2015). The oil content in *M. oleifera* seeds, reaches approximately 35-45%, with a high content of unsaturated fatty acids and 73% oleic acid (Ayerza, 2019). The high content of oleic oil in *Moringa* oil makes it a good source for high-quality biodiesel production (Da Silva et al., 2010; Boukandoul et al., 2018). *Moringa* seeds are a valuable source of oil, which is

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promoted as a natural cosmetic emollient; it is pure, colourless, and tasteless and it is also used for medicinal purposes (Anjula et al., 2011; Abd El-Hack et al., 2018). Due to the multiple uses of the raw material from Moringa trees, their cultivation has spread worldwide to more than 80 countries (Pandey et al., 2011).

It has been reported that Moringa trees are well adapted to harsh environmental conditions, can grow in high temperatures and can highly considered drought and heat tolerant plant (Hegazy et al., 2008; El-Lamey, 2015; Wasonowati et al., 2019). The adaptation of plants to environmental stresses is usually associated with metabolic changes that resulted in the accumulation of a range of metabolites, such as sugars, phenols, and proline (Tsfay et al., 2011), that have a considerable impact on seed germination, seedling development and the productivity of plants (Tsfay et al., 2016). Tsfay et al. (2016) reported that temperature had a significant effect on the germination rate of *M. oleifera* seeds and radicle emergence was accelerated at 30/20°C, leading to germination within 48hrs. In general, plants developed adaptive strategies in response to biotic and abiotic stress, such as drought, salinity, and temperature. Although plant species differ considerably in the optimum temperature range for seed germination and growth performance, it has been reported that the optimal temperature for seed germination of *M. oleifera* is 30/20°C (Muhl, 2009; Muhl et al., 2011A). The growth and seed production of Moringa species might be affected by climatic conditions, particularly high temperatures, drought, and high levels of irradiance (Melesse et al., 2012; Hassanein & Al-Soqeer, 2017; Wasonowati et al., 2019). In a recent study on the performance of several *M. oleifera* cultivars, variations in vegetative and reproductive growth traits were observed under dry, hot climate conditions (Zheng et al., 2019).

Photosynthesis is considered one of the most sensitive physiological processes in plants. It has been found that temperature is among the uncontrollable climate variables that have the greatest effect on regulating the process of photosynthesis (Urban et al., 2017). Elevated temperatures can reduce the photochemistry of PSII ( $F_v/F_m$ ), the assimilation of CO<sub>2</sub> and the evolution of O<sub>2</sub>, resulting in a decrease in the light reactions of photosynthesis and the light saturation point of photosynthesis ( $A_{sat}$ )

(Savitch et al., 2000; Youssef, 2007). Elevated temperatures limit photosynthetic activity by affecting the transportation of electrons in the thylakoid membrane, carboxylation ability and Rubisco kinetics (Posch et al., 2019). It has also been found that the ratio of the linear quantum yield to the mean variable of fluorescence ( $F_v/F_m$ ) decreases at elevated temperatures (Maxwell & Johnson, 2000). Furthermore, the chlorophyll content was found to decrease when plants were exposed to elevated temperatures. Elevated temperatures inhibit chlorophyll biosynthesis and speed up its degradation (Ashraf & Harris, 2013).

In the Almadinah Almunawwarah region, in the western part of Saudi Arabia, *Moringa peregrina* and *Moringa oleifera* are cultivated in different areas with different temperature conditions. Temperature is one of the main factors that determine the distribution of plants in these areas. The mean temperatures of the Almadinah Almunawwarah region range between 15°C and 35°C annually (Presidency of Meteorology and Environment, Saudi Arabia). Since there is a shortage of information concerning the responses of *Moringa* species to temperature change in this region, this study is performed to investigate the responses of seed germination, seedling growth and photosynthesis performance of these two *Moringa* species to three temperature regimes: 15/12°C, 25/22°C and 35/32°C; day/night.

## **Materials and Methods**

### *Seed germination*

The seeds of two *Moringa* species, *M. oleifera* and *M. peregrina*, were used to investigate the effects of temperature on germination. The seeds of *M. oleifera* were obtained from Al-Hilali Agricultural Company, while the seeds of *M. peregrina* were donated by the Jeeda Charity Community in the Al-Ula district, where the seeds are collected from their natural habitats. The experiments in this study were carried out in the Department of Biology, Faculty of Science, Taibah University, Almadinah Almunawwarah, Saudi Arabia. For the germination test, seeds were sterilised by soaking in 0.1% mercuric chloride, washed in distilled water five times. The seeds were placed on two layers of Whatman No. 1 filter paper inside a Petri dish (9cm x 1.6cm) and watered with 10mL of distilled water. Each Petri dish contained 10 seeds, and four replicate Petri dishes were used for both *Moringa* species (*M.*

*oleifera* and *M. peregrina*). Three temperature treatments were used to test the temperature at germination (15°C, 25°C and 35°C), and the Petri dishes were placed inside incubators in darkness. The emergence of the radicle from the seeds was considered as an indicator of germination. The germination process was recorded every day for the whole incubation period during two weeks.

#### *Plant growth parameters*

Seeds of the two *Moringa* species, *M. oleifera* and *M. peregrina*, were grown in plastic pots (12cm x 20cm) containing 2kg of compost. The pots were placed inside an environmentally controlled chamber (JSR 314-240, JS Research Inc., Gumsang-Dong 40-1, city of Gongju, Korea) with a photoperiod of 14/10hrs. (light/dark) and 60% relative humidity. Fluorescent and halogen lamps were used for illumination, resulting in 400µmol m<sup>-2</sup>s<sup>-1</sup>. For each *Moringa* species, four replicates were used for the three temperature treatments (15/12°C, 25/22°C and 35/32°C; day/night). In each pot, five seeds were planted, and after emergence, the seedlings were thinned, leaving two per pot. The plants were irrigated with full-strength Hoagland solution to full field capacity. At the end of the experiment, 40 days after planting, the following growth parameters were measured: height of the plant, number of leaves, and fresh weight of the roots and shoots. To determine the dry weight, root and shoot samples were oven-dried for 48hrs. at 80°C. Leaf area was measured according to the method described by Pandey & Singh (2011), using the weight of graph paper covered by the leaves. Then, the weight was converted to area using the following equation: leaf area (cm<sup>2</sup>) =  $x/y$ , where  $x$  is the weight of the graph paper covered by the leaf and  $y$  is the weight (gm).

#### *Photosynthesis measurements*

The rates of photosynthesis were measured using a 6400XT LICOR infrared gas analyser (LI-6400, LI-COR Inc., Lincoln, Nebraska, United States of America) on the youngest fully expanded leaves, following methods described by Al-Shoaibi (2008). For photosynthesis and dark respiration, the fourth fully expanded young leaves were used. The leaf temperature was maintained at 25°C during the measurement process by using a Peltier cooling system. The leaves were illuminated steadily, with photon flux densities from 0 to 1500µmol m<sup>-2</sup>s<sup>-1</sup>. The net photosynthesis per leaf area and intercellular CO<sub>2</sub>

concentration ( $c_i$ ) were determined by applying the equations of Von Caemmerer & Farquhar (1981).  $A_{sat}$  was determined at a saturating photosynthetic photon flux density of 1500µmol m<sup>-2</sup>s<sup>-1</sup> and at an ambient CO<sub>2</sub> concentration of 410µmol mol<sup>-1</sup>.

The response of  $A$  to the intercellular CO<sub>2</sub> concentration ( $c_i$ ) was estimated using the same gas exchange system over a range of  $c_a$  = 50-550µmol mol<sup>-1</sup>, with a photosynthetic photon flux density of 1500µmol m<sup>-2</sup>s<sup>-1</sup> at a leaf temperature of 25°C. The measurement of  $A$  was first performed at a CO<sub>2</sub> concentration equal to the ambient concentration in the environment where the plants were grown. To determine the initial slope of the  $A/c_i$  response,  $c_a$  was gradually decreased over six steps to 50µmol mol<sup>-1</sup>. To check the original rate that might be regained,  $c_a$  was then restored to the ambient concentration. To complete the curve, the  $c_a$  was finally increased stepwise to 550µmol mol<sup>-1</sup>. The assumed amount of active phosphoenol pyruvate carboxylase *in vivo*, which is called 'carboxylation efficiency', was calculated from the gradient of the initial slope of the response of  $A$  to  $c_i$ , following the model of Collatz et al. (1992).

#### *Chlorophyll content determination*

The chlorophyll contents, which are expressed as the chlorophyll index, were determined by using a handheld chlorophyll content metre (Opti-Sciences Inc., CCM-200, United States). The chlorophyll contents were measured in the fourth expanded leaves, with four readings for each *Moringa* species and each temperature treatment. The average of the four measurements was used as the final chlorophyll content index.

#### *Chlorophyll fluorescence measurements*

The fluorescence of chlorophyll was measured by using a handheld fluorimeter (PEA, Hansatech, King's Lynn, Norfolk, U.K.). The measurements were performed on the youngest fully expanded leaves of the two *Moringa* species. Prior to the chlorophyll measurement, the leaves were dark-adapted for 20min. For both *Moringa* species, the variable ratio of maximum fluorescence ( $F_v/F_m$ ) was measured four times for each temperature treatment, as described by Al-Shoaibi (2008).

#### *Statistical analysis*

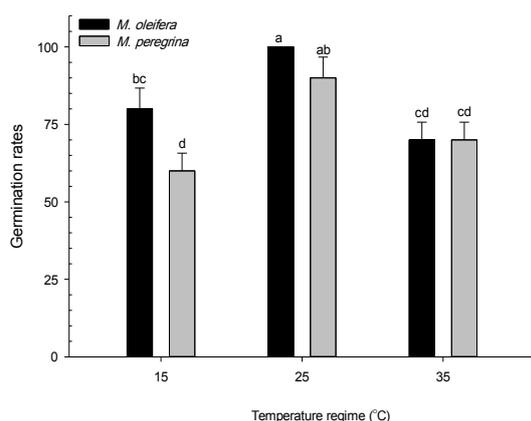
The data obtained for the different measurements were statistically analysed using

one-way and two-way analysis of variance (ANOVA) and general linear model to test the main effects and the interactions of the investigated factors (i.e., temperature and species). The significance between the different levels of the investigated factors was tested, and a multiple comparison using Tukey's test was carried out. Version 15 of Minitab (Brandon Court, Unit E1-E2, Progress Way, Coventry CV3 2TE, U.K.) was used for all analyses. For each temperature treatment, four replicates were used, and standard deviations and standard errors were determined using Microsoft Excel 2016.

## Results

### Seed germination

The effects of the three temperature treatments on the final germination percentages of the two *Moringa* species are illustrated in Fig. 1. At all the temperatures, seed germination started on the fourth day and was completed within the 14-day period of the germination trial. The effect of temperature on the germination performance was highly significant ( $P < 0.001$ ). At 15°C, *M. oleifera* had a significantly higher percentage of germination compared to *M. peregrina* ( $P < 0.05$ ). Moreover, both species showed higher percentages of germination at 25°C. The germination percentage approached 100% and 90% for *M. oleifera* and *M. peregrina*, respectively, while the lowest germination percentage (60%) was achieved at 15°C for *M. peregrina*.



**Fig. 1.** The effect of temperature treatments (15°C, 25°C and 35°C) on seed germination (as %) of the two *Moringa* species; *M. oleifera* and *M. peregrina* [n= 4, mean± S.E., means that do not share the same letter are significantly different at  $P \leq 0.05$ ]

### Plant growth parameters

Several growth parameters were measured to determine how different temperature treatments influenced the growth parameters of the two *Moringa* species; the results are summarized in Table 1. Temperature significantly affected most growth parameters of the two *Moringa* species ( $P < 0.001$ ). The results showed that, most growth parameters of the two *Moringa* species grown at 25°C and 35°C were significantly increased when compared to their relative plants grown at 15°C ( $P < 0.01$ ). The leaf number and leaf area of *M. oleifera* grown at 35°C were 631% and 547.5% higher, respectively, than those of the same species grown at 15°C; these increases were the highest observed for these indices. Similarly, the plant height and root length of *M. peregrina* grown at 35°C approached 143.2% and 198.7% higher, respectively, than those of the same species grown at 15°C, representing the maximum percentage increases for these indices. Moreover, the considerable percentage increases for most of the fresh and dry weights of both shoots and roots of *M. oleifera* were observed for plants grown at 35°C compared to those of the same species grown at 15°C ( $P < 0.001$ ; Table 1).

### Photosynthesis measurements

The results presented in Fig. 2 show the response of the uptake of photosynthetic  $\text{CO}_2$  ( $A$ ) to photon flux ( $Q$ ) for the two *Moringa* species grown in the three temperature treatments. The temperature treatments significantly affected the photosynthetic performance of the two *Moringa* species ( $P < 0.001$ ). Both *Moringa* species, the highest photosynthesis performance was observed at 25°C (Fig. 2). The rates of light saturation ( $A_{sat}$ ) for both *Moringa* species grown at 25°C were considerably higher than those of the same species grown at 35°C ( $P < 0.01$ ; Fig. 3A), with percentage increases for *M. oleifera* and *M. peregrina* of 63% and 29%, respectively. Moreover, the quantum yield ( $\phi$ ) of *M. peregrina* grown at 25°C was significantly greater than the quantum yield of the same species grown at 35°C ( $P < 0.01$ ; Fig. 3B), with a 46% increase. However, both *Moringa* species showed significantly lower  $A_{sat}$  and  $\phi$  at 15°C than at 25°C or 35°C ( $P < 0.01$ ; Fig. 3A & B). Measurements of the  $A/c_i$  curve for both *Moringa* species in the three temperature treatments are illustrated in Fig. 3. In general, the plateaus of the  $A/c_i$  curve ( $A_{max}$ ) and carboxylation efficiency for both *Moringa* species were considerably higher at 25°C than

at 35°C (P<0.01; Fig. 3C & D). For *M. oleifera*, the highest percentage increases in  $A_{max}$  and carboxylation efficiency were observed for plants grown at 25°C; these were 87% and 168% higher, respectively, than those of the same species

grown at 35°C (P<0.01; Fig. 3). On the other hand, both *Moringa* species showed significantly lower  $A_{max}$  and carboxylation efficiency values when grown at 15°C than at 25 or 35°C (P<0.01; Fig. 3C & D).

TABLE 1. Effects of temperature on the growth parameters of *M. oleifera* and *M. peregrina*

Trait	Species	15°C	25°C	35°C
Plant height (cm)	<i>M. oleifera</i>	18 <sup>bc</sup> ± 1.25	24.4 <sup>b</sup> ± 2.3	43 <sup>a</sup> ± 4.16
	<i>M. peregrina</i>	8.4 <sup>d</sup> ± 0.56	16.67 <sup>c</sup> ± 1.85	20.43 <sup>bc</sup> ± 1.45
Leaf number	<i>M. oleifera</i>	40 <sup>d</sup> ± 0.58	223.3 <sup>b</sup> ± 9.21	292.4 <sup>a</sup> ± 13.62
	<i>M. peregrina</i>	15 <sup>e</sup> ± 1.73	41 <sup>d</sup> ± 7.57	72.3 <sup>c</sup> ± 6.84
Leaf area (cm <sup>2</sup> )	<i>M. oleifera</i>	80 <sup>cd</sup> ± 1.15	446.7 <sup>b</sup> ± 18.41	518 <sup>a</sup> ± 43.55
	<i>M. peregrina</i>	22.5 <sup>d</sup> ± 2.59	62 <sup>cd</sup> ± 11.75	108.5 <sup>c</sup> ± 10.26
Shoot fresh weight (g)	<i>M. oleifera</i>	2.16 <sup>c</sup> ± 0.37	10.23 <sup>b</sup> ± 1.57	20.1 <sup>a</sup> ± 1.07
	<i>M. peregrina</i>	2.8 <sup>c</sup> ± 1.55	4.1 <sup>c</sup> ± 0.28	5.1 <sup>c</sup> ± 1.29
Shoot dry weight (g)	<i>M. oleifera</i>	0.5 <sup>cd</sup> ± 0.11	1.4 <sup>b</sup> ± 0.2	3.43 <sup>a</sup> ± 0.24
	<i>M. peregrina</i>	0.23 <sup>d</sup> ± 0.03	0.38 <sup>cd</sup> ± 0.05	0.84 <sup>c</sup> ± 0.21
Root length (cm)	<i>M. oleifera</i>	6.5 <sup>bc</sup> ± 1.25	11.83 <sup>ab</sup> ± 1.48	12.16 <sup>a</sup> ± 1.58
	<i>M. peregrina</i>	5.3 <sup>c</sup> ± 0.34	8.5 <sup>b</sup> ± 1.04	15.83 <sup>a</sup> ± 3.65
Root fresh weight (g)	<i>M. oleifera</i>	0.56 <sup>c</sup> ± 0.18	7.33 <sup>c</sup> ± 0.55	11.1 <sup>a</sup> ± 0.44
	<i>M. peregrina</i>	0.4 <sup>c</sup> ± 0.11	3.93 <sup>d</sup> ± 0.46	9.76 <sup>b</sup> ± 0.52
Root dry weight (g)	<i>M. oleifera</i>	0.06 <sup>c</sup> ± 0.02	0.8 <sup>c</sup> ± 0.17	1.56 <sup>a</sup> ± 0.03
	<i>M. peregrina</i>	0.12 <sup>c</sup> ± 0.09	0.4 <sup>d</sup> ± 0.05	1.17 <sup>b</sup> ± 0.03

- n = 4, mean±S.E.

- Means that do not share a letter are significantly different at P ≤ 0.05

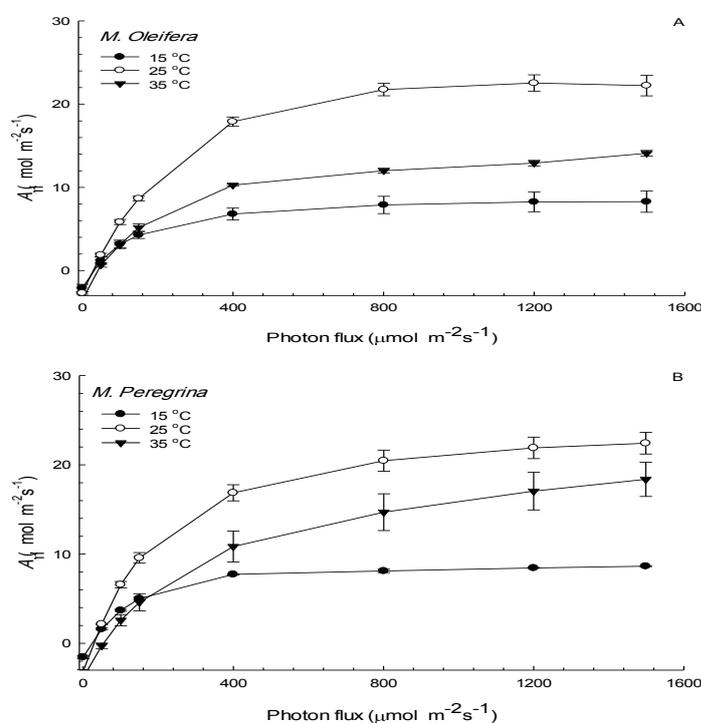
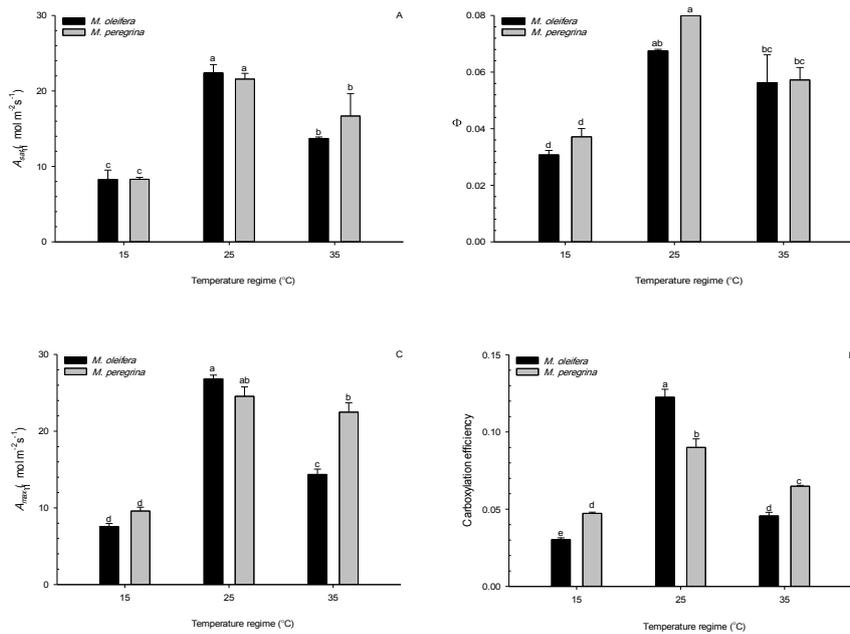


Fig. 2. The photosynthetic CO<sub>2</sub> absorption response (A) per unit area of the leaf to photon flux (Q) for two *Moringa* species; *M. oleifera* and *M. peregrina* [n= 4, mean±S.E., CO<sub>2</sub> absorption measurements were all made at 25°C and C<sub>a</sub> of 410 μmol mol<sup>-1</sup>]

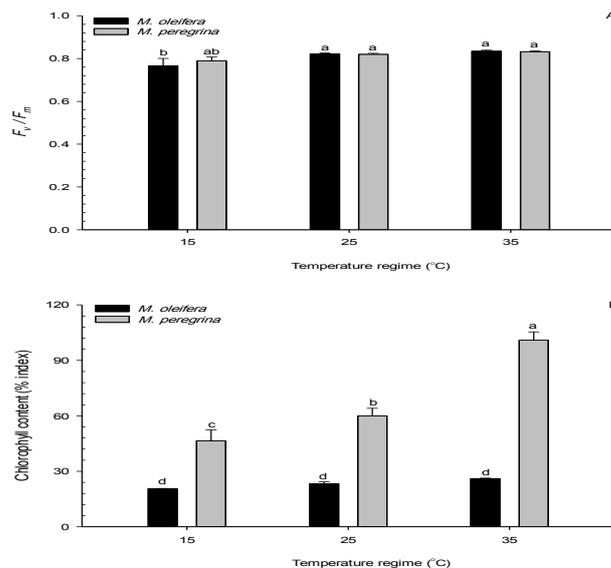


**Fig. 3.** The effect of temperature treatments (15°C, 25°C and 35°C) on physiological parameters of the two *Moringa* species; *M. oleifera* and *M. peregrina* [n = 4, mean±S.E., (A)Asat, (B) Quantum yield, (C) Amax and (D) Carboxylation efficiency. Means that do not share the same letter are significantly different at P≤0.05]

#### Chlorophyll content determination

Figure 4B illustrated the effect of the three temperature treatments on the chlorophyll content of *M. oleifera* and *M. peregrina*. The chlorophyll contents of *M. peregrina* were significantly increased by increasing the temperature from 15°C to 35°C (P<0.01). The chlorophyll

contents of *M. peregrina* grown at 25°C and 35°C increased by 29% and 117%, respectively, compared to those of the same species grown at 15°C (P<0.01). However, the three temperature treatments did not induce significant variation in the chlorophyll content of *M. oleifera*.



**Fig. 4.** The effect of temperature treatments (15°C, 25°C and 35°C) on physiological parameters of the two *Moringa* species; *M. oleifera* and *M. peregrina* [n = 4, mean±S.E., (A) F<sub>v</sub>/F<sub>m</sub> and (B) Chlorophyll content. Means that do not share the same letter are significantly different at P≤0.05]

#### *Chlorophyll fluorescence measurements*

The maximum quantum efficiency of the photochemistry of PSII ( $F_v/F_m$ ) of the two *Moringa* species grown in the three temperature treatments is shown in Fig. 4A. For both *Moringa* species, the  $F_v/F_m$  values of the plants grown at 35°C were similar to those plants grown at 25°C. However, the chlorophyll fluorescence parameter of *M. oleifera* plants grown at 15°C was significantly decreased compared to their analogues parameters of the two *Moringa* species grown at 25°C and 35°C ( $P < 0.05$ ).

#### **Discussion**

The present study examined the responses of seed germination, seedling growth and photosynthesis of two *Moringa* species to temperature. Temperature is one of the most significant factors influencing seed germination and seedling development (Zhao et al., 2014; Wang et al., 2020). The results showed that seeds of both *Moringa* species germinated over a wide range of temperatures, from 15°C to 35°C (Fig. 1). The germination of the two *Moringa* species in all the temperature treatments began on day 4 of the experimental period. The germination suppression reported in the first days of this study is consistent with some previous studies which showed germination delays for few days (Redondo et al., 2004; Yan et al., 2011A; Causin et al., 2020). This indicated the need for optimal environmental conditions to ensure a high percentage of germination and thus growth efficiency (Redondo et al., 2004). Furthermore, the results obtained showed that the optimum temperature for seed germination of both *Moringa* species was 25°C. Similar findings have been reported for *M. oleifera* (Muhl et al., 2011A; Zhang et al., 2017).

Temperature is one of the main influential and uncontrollable climate factors that regulate plant development and growth (Munir, 2004). For both *Moringa* species used in this study, all the growth parameters were higher at 35°C than 15°C (Table 1). The optimum temperature for seedling growth of both *Moringa* species was 35°C. It was previously reported that the optimum temperatures for seedling growth in *Moringa* ranged from 20°C to 35°C (Alatar, 2011; Muhl et al., 2011A). *M. oleifera* grown at 35°C showed better growth performance, especially leaf number and leaf area, than *M. peregrina*.

The considerably greater leaf area of *M. oleifera* increased the total number of stomata per plant, resulting in increased growth of vegetation and accumulation of dry matter (Muhl et al., 2011B). Furthermore, the highest percentage increase was observed for the fresh and dry weights of the shoots and roots of *M. oleifera* grown at 35°C compared with those of the same species grown at 15°C (Table 1). Genetic variations between the two studied species and their distinct origins can explain the differences observed (Mridha, 2015; Muhammad et al., 2016). Furthermore, the ability of the two *Moringa* species to develop over a large range of temperatures, from 15°C to 35°C, may support their distribution and use as important crops for medicine and nutrition (Hassanein & Al-Soqeer, 2017).

Temperature also significantly affected the gas exchange parameters of both *Moringa* species in this study. The photosynthetic measurements showed that the optimum temperature for gas exchange of both *Moringa* species was 25°C (Fig. 3). Additionally, both *Moringa* species showed nearly similar photosynthetic performances at 25°C and 35°C. However, a temperature of 15°C caused a substantial decrease in the  $A_{sat}$ ,  $\phi$ ,  $A_{max}$  and carboxylation efficiencies of both *Moringa* species compared with those of plants grown in the other temperature treatments. The inhibition of photosynthesis in *Moringa* plants grown at 15°C may be due in part to a reduction in stomatal conductance or a decrease in photosynthetic pigments. All these factors affect photosystem II (PSII) activities, clearly contributing to the deactivation of enzymes that play a key role in the photosynthetic process (Ashraf & Harris, 2013; Li et al., 2015). A decrease in the photosynthetic performance of *Moringa* was previously observed at low temperatures in controlled environments and fields (Mabapa et al., 2018). There are many physiological mechanisms that allow these two *Moringa* species to cope effectively with harsh environmental conditions, such as temperature stress (Mabapa et al., 2018). Furthermore, Chaves (1991) found that the primary processes adversely affected by drought and higher temperatures are gas exchange operations, primarily those related to photosynthetic rates. The results of this study showed that the photosynthetic parameters of the two *Moringa* species were significantly lower at 35°C than at 25°C. Similar findings were reported by Mabapa et al. (2018), who found that higher temperatures in summer months had

a negative impact on the photosynthetic rate of *M. oleifera*. This reduction in the photosynthetic process of the two *Moringa* species could be the result of stomatal limitations and reductions in the chlorophyll and carotenoid contents (Sharma et al., 2019; Morales et al., 2020). Other reasons for the reduction in the photosynthetic process could be limitations in the electron transport of the thylakoid membrane or decreases in the activity of Rubisco (Sharma et al., 2019; Morales et al., 2020).

The findings of this research showed that the chlorophyll content in the leaves of *M. peregrina* increased as the temperature increased (Fig. 4B). The highest increase in chlorophyll content was recorded for *M. peregrina* grown at 35°C. The leaves of plants grown in the highest temperature treatment area visible sign of this increase. The reason for this increase could be the rise in the number of chloroplasts within leaves developed at high temperatures. However, the chlorophyll contents in the leaves of *M. oleifera* were similar across the three temperature treatments. The chlorophyll contents of *M. peregrina* leaves were significantly higher than the chlorophyll contents of *M. oleifera* leaves in all the temperature treatments. Similar findings were reported by Hassanein & Al-Soqeer (2017), who found that *M. peregrina* accumulated greater chlorophyll contents than *M. oleifera* when grown under field conditions.

Chlorophyll fluorescence measurement has been used to determine the integrity of photosystem II when exposed to stress (Shabala & Shabala, 2002). In this study, chlorophyll fluorescence (*Fv/Fm*) was not affected by the temperature treatments in either *Moringa* species, except the 15°C treatment, in which a slight decrease was recorded for *M. oleifera* (Fig. 4A). These high *Fv/Fm* values offered clear evidence that both *Moringa* species were resistant to photo inhibition when grown under different temperature treatments. Similar results have been reported previously for *Pennisetum purpureum* grown in two temperature treatments (AL-Shoaibi, 2008). Thus, the current research shows that chlorophyll fluorescence cannot be considered one of the variables for regulating the rate of net CO<sub>2</sub> assimilation in *Moringa* species grown at different temperatures. On the other hand, the slight decrease in the *Fv/Fm* values of *M. oleifera* grown at 15°C could result from

oxidative damage to photosynthetic organelles, particularly under temperature stress (Aro et al., 1993; Yan et al., 2011B).

### **Conclusion**

The present study showed that both *M. peregrina* and *M. oleifera* have similar responses to different temperature treatments. The optimum temperature for both species, leading to the highest percentage of germination and best photosynthetic performance, is 25°C. The results also showed that 35°C was the optimal temperature for most parameters of growth of the two *Moringa* species. However, the chlorophyll contents of *M. oleifera* and most values of *Fv/Fm* for both species were similar across the three temperature treatments. Additionally, the outcomes of this study showed that *Moringa* can be cultivated in many different regions with different temperatures.

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### **References**

- Abd El-Hack, M.E., Alagawany, M., Elrys, A.S., Desok, E.M., Tolba, H.M.N., et al. (2018) Effect of forage *Moringa oleifera* L. (moring) on animal health and nutrition and its beneficial application in soil, plants and water purification. *Agriculture*, **8**(9), 145.
- Alatar, A.A. (2011) Effect of temperature and salinity on germination of *Achillea fragrantissima* and *Moringa peregrina* from Saudi Arabia. *African Journal of Biotechnology*, **10**, 3393-3398.
- AL-Shoaibi, A.A. (2008) Photosynthetic response to the low temperature in Elephant grass (*Pennisetum purpureum*) and *Zea Mays*. *International Journal of Botany*, **4**(3), 309-314.
- Amaglo, N.K., Bennett, R.N., Lo Curto, R.B., Rosa, E.A.S., et al. (2010) Profiling selected phytochemicals and nutrients in different tissues of the multipurpose tree *Moringa oleifera* L., grown in Ghana. *Food Chemistry*, **122**, 1047-1054.

- Anjula, P., Pradheep, K., Rita, G., Eroshini, N., Bhandari, D.C. (2011) Drumstick tree (*Moringa oleifera* Lam.): a multipurpose potential species in India. *Genetic Resources and Crop Evolution*, **58**, 453-460.
- Aro, E.M., Virgin, I., Andersson, B. (1993) Photoinhibition of Photosystem II. Inactivation, protein damage and turnover. *Biochimica et Biophysica Acta*, **5(2)**, 113-134.
- Ashraf, M., Harris, P.J.C. (2013) Photosynthesis under stressful environments: An overview. *Photosynthetica*, **51**, 163–190.
- Ayerza, R. (2019) Seed characteristics, oil content and fatty acid composition of moringa (*Moringa oleifera* Lam.) seeds from three arid land locations in Ecuador. *Industrial Crops and Products*, **140**, 111575.
- Boukandoul, S., Casal, S., Zaidi, F. (2018) The potential of some *Moringa* species for seed oil production. *Agriculture*, **8(10)**, 150.
- Boulos, L. (1999) "*Flora of Egypt*". vol. I (Azollaceae-Oxalidaceae). Al Hadara Publishing Cairo, Egypt.
- Causin, H.F., Bordón, D.A.E., Burrieza, H. (2020) Salinity tolerance mechanisms during germination and early seedling growth in *Chenopodium quinoa* Wild. genotypes with different sensitivity to saline stress. *Journal of Experimental Botany*, **172**, 103995.
- Chaves, M.M. (1991) Effects of water deficits on carbon assimilation. *Journal of Experimental Botany*, **42**, 1–16.
- Collatz, G.J., Ribas-Carbo, M., Berry, J.A. (1992) Coupled photosynthesis-stomatal model for leaves of C<sub>4</sub> plants. *Australian Journal of Plant Physiology*, **19**, 519-538.
- Da Silva, J.P., Serra, T.M., Gossmann, M., Wolf, C.R., Meneghetti, M.R., Meneghetti, S.M. (2010) *Moringa oleifera* oil: studies of characterization and biodiesel production. *Biomass and Bioenergy*, **34**, 1527–1530.
- El-Lamey, T.M. (2015) Ecophysiological responses of *Moringa peregrina* (Forssk) Fiori growing naturally under different habitat conditions of Eastern Desert and Fieran Oasis, Egypt. *IOSR Journal of Agriculture and Veterinary Science*, **8(4)**, 8-21.
- Hassanein, A.M.A., Al-Soqeer, A.A. (2017) Evaluation of seed germination and growth characteristics of *Moringa oleifera* and *Moringa peregrina* under laboratory, greenhouse and field conditions. *International Journal of Agriculture and Biology*, **19**, 873-879.
- Hegazy, A.K., Hammouda, O., Lovett-Doust, J., Gomaa, N.H. (2008) Population dynamics of *Moringa peregrina* along altitudinal gradient in the northwestern sector of the Red Sea. *Journal of Arid Environments*, **72**, 1537-1551.
- Khor, K.Z., Vuanghao, L., Moses, E.J., Samad, N.A. (2018) The *in vitro* and *in vivo* anti-cancer properties of *Moringa oleifera*. *Evidence-Based Complementary and Alternative Medicine*, **2018(2)**, 1-14. DOI:10.1155/2018/1071243
- Leone, A., Spada, A., Battezzati, A., Schiraldi, A., Aristil, J., Bertoli, S. (2015) Cultivation, genetic, ethnopharmacology, photochemistry and pharmacology of *Moringa oleifera* leaves: An overview. *International Journal of Molecular Science*, **16**, 12791-12835.
- Li, Z., Wu, N., Liu, T., Chen, H., Tang, M. (2015) Effect of arbuscular mycorrhizal inoculation on water status and photosynthesis of *Populus cathayana* males and females under water stress. *Physiologia Plantarum*, **155**, 192–204.
- Mabapa, M.P., Ayisi, K.K., Mariga, I.K. (2018) Seasonal effect on *Moringa oleifera* gaseous exchange and water use efficiency under diverse planting densities. *Journal of Applied Botany and Food Quality*, **91**, 219–225.
- Maxwell, K., Johnson, G.N. (2000) Chlorophyll fluorescence — a practical guide. *Journal of Experimental Botany*, **51**, 659–668.
- Melesse, A., Steingass, S.H., Boguhn, J., Schollenberger, M., Rodehutsord, M. (2012) Effects of elevation and season on nutrient composition of leaves and green pods of *Moringa stenopetala* and *Moringa oleifera*. *Agroforestry Systems*, **86**, 505–518.
- Morales, F., Ancín, M., Fakhret, D., González-Torrallba, J., Gámez, A.L., et al. (2020) Photosynthetic metabolism under stressful growth conditions as a

- bases for crop breeding and yield improvement: A review. *Plants*, **9** (1), 88.
- Mridha, M.A.U. (2015) Prospects of Moringa Cultivation in Saudi Arabia. *Journal of Applied Environmental and Biological Sciences*, **5** (3), 39-46.
- Muhammad, H.I., Asmawi, M.Z., Khan, N.A.K. (2016) A review on promising phytochemical, nutritional and glycemic control studies on *Moringa oleifera* Lam. in tropical and sub-tropical regions. *Asian Pacific Journal of Tropical Biomedicine*, **6**, 896–902.
- Muhl, Q.E. (2009) Seed germination, tree growth and flowering responses of *Moringa oleifera* Lam. (Horseradish tree) to temperature. *MSc Thesis*, Pretoria: University of Pretoria, Republic of South Africa.
- Muhl, Q.E., Toit, E.S., Robbertse, P.J. (2011 A) *Moringa oleifera* (Horseradish tree) leaf adaptation to temperature regimes. *International Journal of Agriculture and Biology*, **13**, 1021-1024.
- Muhl, Q.E., Toit, E.S., Robbertse, P.J. (2011 B) Temperature effect on seed germination and seedling growth of *Moringa oleifera* Lam. *Seed Science and Technology*, **39**, 208–213.
- Munir, M., Jamil, M., Baloch, J., Khattak, K.R. (2004) Growth and flowering of *Antirrhinum majus* L. under varying temperatures. *International Journal of Agriculture and Biology*, **6**, 173-178.
- Olson, M.E., Carlquist, S. (2001) Stem and root anatomical correlations with life form diversity, ecology, and systematics in Moringa (*Moringaceae*). *Botanical Journal of the Linnean Society*, **135**, 315-348.
- Pandey, A., Pradheep, K., Gupta, R., Nayar, E.R., Bhandari, D.C. (2011) 'Drumstick tree' (*Moringa oleifera* Lam.): A multipurpose potential species in India. *Genetic Resources and Crop Evolution*, **58**, 453–460.
- Pandey, S.K., Singh, H. (2011) A Simple, Cost-Effective Method for Leaf Area Estimation. *Journal of Botany*, Article ID 658240. <https://doi.org/10.1155/2011/658240>.
- Posch, B.C., Kariyawasam, B.C., Bramley, H., Coast, O., Richards, R.A., Reynolds, M.P., et al. (2019) Exploring high temperature responses of photosynthesis and respiration to improve heat tolerance in wheat. *Journal of Experimental Botany*, **70**, 5051–5069.
- Ramachandran, C., Peter, K.V., Gopalakrishnan, P.K. (1980) Drumstick (*Moringa oleifera*) multipurpose Indian vegetable. *Economic Botany*, **34**, 276-283.
- Redondo, S., Rubio-Casal, A.E., Castillo, J.M., Luque, C.J., Álvarez, A.A., et al. (2004) Influences of salinity and light on germination of three *Sarcocornia taxa* with contrasted habitats. *Aquatic Botany*, **78**(3), 255-264.
- Savitch, L.V., Massacci, A., Gray, G.R., Huner, N.P.A. (2000) Acclimation to low temperature or high light mitigates sensitivity to photoinhibition: roles of the Calvin cycle and the Mehler reaction. *Australian Journal of Plant Physiology*, **27**, 253-264.
- Shabala, S., Shabala, L. (2002) Kinetics of net H<sup>+</sup>, Ca<sup>2+</sup>, K<sup>+</sup>, Na<sup>+</sup>, NH<sub>4</sub><sup>+</sup>, and Cl<sup>-</sup> fluxes associated with post-chilling recovery of plasma membrane transporters in *Zea mays* leaf and root tissues. *Physiologia Plantarum*, **114**, 47–56.
- Sharma, A., Kumar, V., Shahzad, B., Ramakrishnan, M., Sidhu, G.P.S., et al. (2019) Photosynthetic response of plants under different abiotic stresses: A review. *Journal of Plant Growth Regulation*, **39**, 509-531.
- Tesfay, S., Bertling, I., Odindo, A., Workneh, T., Mathaba, N. (2011) Levels of antioxidants in different parts of moringa (*Moringa oleifera*) seedling. *African Journal of Agricultural Research*, **6**, 5123–5132.
- Tesfay, S.Z., Modi, A.T., Mohammed, F. (2016) The effect of temperature in moringa seed phytochemical compounds and carbohydrate mobilization. *South African Journal of Botany*, **102**, 190–196.
- Urban, J., Ingwers, M.W., McGuire, M.A., Teskey, R.O. (2017) Increase in leaf temperature opens stomata and decouples net photosynthesis from stomatal conductance in *Pinus taeda* and *Populus deltoides* x *nigra*. *Journal of Experimental Botany*, **68**(7), 1757–1767.
- Von Caemmerer, S., Farquhar, G.D. (1981) Some
- Egypt. J. Bot.* **61**, No. 2 (2021)

- relationships between the biochemistry of photosynthesis and gas exchange of leaves. *Planta*, **53**, 376-387.
- Wang, Q., Qu, B., Mi, J., Ma, L., Xu, Y., Shao, M. (2020) The effect of temperature on seed germination and seedling growth of two invasive plants in Roroppa. *Biotechnology Journal International*, **24**(5), 39-48.
- Wasonowati, C., Sulistyarningsih, E., Indradewa, D., Kurniasih, B. (2019) Physiological characters of *Moringa oleifera* Lamk in Madura. *International Conference on Biology and Applied Science*, **2120**(1), 030024. <https://doi.org/10.1063/1.5115628>.
- Yan, C., Wei, Y., Yang, M. (2011 A) Comparative germination of *Tamarix ramosissima* spring and summer seeds. *Experimental and Clinical Sciences Journal*, **10**, 198-204.
- Yan, K., Chen, P., Shao, H., Zhang, L., Xu, G. (2011 B) Effects of short-term high temperature on photosynthesis and photosystem II performance in sorghum. *Journal of Agronomy and Crop Science*, **197**, 400-408.
- Youssef, T. (2007) Stomatal, biochemical and morphological factors limiting photosynthetic gas exchange in the mangrove associate *Hibiscus tiliaceus* under saline and arid environment. *Aquatic Botany*, **87**, 292-298.
- Zhang, T.X., Lin, Y.H., Lin, Z.K., Chen, Z.D., Yang, J.J., Cao, M.H. (2017) Effects of different temperature treatments on seed germination, seedling growth and physiological characteristics of *M. oleifera* Lam. *Chinese Journal of Tropical Crops*, **38**, 438-443.
- Zhao, T.T., Li, Y.X., He, J.D. (2014) Effect of temperature stress on seed germination of *Chimonanthus praecox* (J). *Journal of Mianyang Normal University*, **33**, 63-68.
- Zheng, Y., Sun, H., Zhang, Y., Wu, J. (2019) Evaluation of the adaptability, productivity, and leaf powder quality of eight *Moringa oleifera* cultivars introduced to a dry-hot climate of Southwest China *Industrial Crops and Products*, **128**, 199-205.

## إنبات البذور ونمو الشتلات واستجابات البناء الضوئي لدرجة الحرارة في الشجرة الاستوائية *Moringa oleifera* والشجرة الصحراوية *Moringa peregrina*

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تزرع المورينجا أوليفيرا والمورينجا بريجرينا في العديد من المناطق في المدينة المنورة، الجزء الغربي من المملكة العربية السعودية. تهدف هذه الدراسة إلى فحص استجابات إنبات البذور ونمو الشتلات والبناء الضوئي لنوعين من المورينجا لمعالجات درجات الحرارة المختلفة. أشارت النتائج إلى أن بذور كلا النوعين يمكن أن تنبت في نطاق واسع من درجات الحرارة مع درجة الحرارة المثلى لأعلى نسبة إنبات عند 25 درجة مئوية. كان أداء البناء الضوئي للنوعين أعلى عند 25 درجة مئوية. ومع ذلك، كانت محتويات الكلوروفيل في المورينجا أوليفيرا ومعظم قيم  $Fv/Fm$  لكلا النوعين متشابهة عبر معاملات درجات الحرارة الثلاثة. بالإضافة إلى ذلك، أظهرت نتائج هذه الدراسة أنه يمكن زراعة المورينجا في مناطق عديدة بدرجات حرارة مختلفة.