



## SEED PRIMING EFFECT ON GERMINATION OF DOUM PALM (*Hyphaene thebaica* Mart) AND DEVELOPMENT OF SMALL SEEDLING

[179]

Negm<sup>1\*</sup> A.H.A., Sohair E. Hassan<sup>2</sup> and Hewidy<sup>2</sup> M.

1- Arid Land Agric. Graduate Studies & Research Institute (ALARI), Ain Shams Univ., Cairo, Egypt

2- Horticulture Dept., Fac. of Agric., Ain Shams Univ., P.O. Box 68, Hadayek Shoubra, 11241 Cairo, Egypt

\*Corresponding author: [negm.a.h@agr.asu.edu.eg](mailto:negm.a.h@agr.asu.edu.eg)

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### ABSTRACT

Doum palm *Hyphaene thebaica* Mart is a very important natural flora in the Egyptian territory. It is integrated in food, drink and medicine of the Egyptian population. Seeds germination faces difficulties due to hardness and viability. The fruit (a date) is edible, the foliage is also used for local crafts, the stems for construction purposes, and the roots for phytomedicine, mainly for the treatment of bilharziasis. This study tried different priming methods after removal hard kernel. First treatment; non-treated as direct plantation (control). Second; Cold water (tap water for 48 hours), third; freezing storage in Deep-freezer at  $-18^{\circ}\text{C}$  for 12 hours then immersed in  $60^{\circ}\text{C}$  water for 1 hour, fourth; hot water at  $60^{\circ}\text{C} \pm 5$  for 3 hours then left till gradually cooled, fifth; soaking seeds in a solution of gibberellic acid 200 ppm for 48 hours, sixth; Soaking seeds in 3% solution of  $\text{KNO}_3$  for 48 hours, seventh; soaking seeds in diluted sulfuric acid ( $\text{H}_2\text{SO}_4$ ) at 30% for 15 minutes. Data collected were germination percentage %, roots length, root diameter, root fresh and dry weight, root dry weight, seedling length, wide of leaf, Fresh and dry weight of above ground parts. Freezing followed by hot water gave the highest seed germination % (90%) followed by  $\text{KNO}_3$  (80%). The lowest germination % was obtained from non-treated control and cold water in a range between 45-65%. Cold water treatment gave significant increase in root fresh weight in both seasons when compared to other treatment. Freezing and cold water treatment gave significant increase in plant height in both seasons when compared to other treatments. Moreover, the effect of various priming treatment demonstrated significant effect on characteristics of young growing

seedling. The best performance obtained from seeds treated with cold water and freezing treatment. Thus, treating Doum palm seeds after removal of hard kernel with freezing at  $-18$  for 12 hours followed by immersing seed in hot water at  $60^{\circ}\text{C}$  for one hour.

**Keywords:** Seed priming, *Hyphaene thebaica*, Seedling,  $\text{KNO}_3$ ,  $\text{GA}_3$ , Freezing.

### INTRODUCTION

Dormancy of seed due to inhibitory factors, are either due to hard seed coat (external) or physical of physiological status of interior of the seed. Seed dormancy in Doum palm (*H. thebaica* Mart) belongs to the first type. Some trees that produces seeds with hard seed coat such as Doum which is common component of flora of the southern of Egypt and other arid region are restricted in the distribution in the region because of the prolong dormancy and poor germination rate. The dormancy breaking treatment overcome the dormancy of this plant (Al-Fredan and Ali, 2008).

Zevallos and De La Cruz (1991) reported that, the seed germination rates were increased in *Acacia marcracantha* and *cyathostegia mathewsii* by soaking in cold water for 28h. In contrast, Chris (1994) reported that soaking in cold water not appreciably increase level of germination for some *Acacia* species. Kasera et al (2002) were subjected seeds to tap water for 24h in *Leptadaenia reticulata* plant and were found that, the highest germination percentage 100% obtained with this treatment. Also, Çirak et al (2007) clarified that, the soaked seeds in tap water of *Hypericum aviculavii-folium* resulted in better seed germination percentage.

Chris (1994) reported that, hot water treatment for some *Acacia* species did not appreciably increase germination level as well as the duration of water soaking treatments. Cervantes et al (1996) found that, seed germination of *leucaena exculta* and *L. macrophylla* occurred after soaking in boiling hot water for 1 min. Mechanical Scarification for seed coat can be done by scratch the seed coat by agrail, knife or shaking seeds with some abrasive materials.

Smith (1979) reported that, mechanical seed scarification improved the germination rate of *Zamia integrifolia* and *Z. furfuracea*. Also, the same results were found for scarified feeds of *Albizzia elurozz* when compared to non-scarified ones (Gogue and Emimo, 1980). Similarly, Scarified seeds of *koelreuteria paniculata* gave the best result of germination (91.0%) (Garner and Lewis 1980). Gupta et al (2001) subjected seeds of *Abutilon indicum* to different hot water treatments and found that, the highest germination percentage (82%) occurred with 70° C hot water treatment.

Witte (1977) clarified that, seeds of *Zamia* plants were collected in 1976 and cleaned, then either germinate or stored one year at 5°C. With fresh seed planted in 30°C media, germination was about equal for cracked and uncracked seed (60 to 65%), but only 43% for shelled seed. After storage, cracked seed germination better than uncracked (71% vs. 38%) and mild bottom heat increased germination of uncracked seed (49% vs. 27%) but not cracked seed. Heavier seed (29 g) germinated better than lighter one (1.69 g).

Tilki and Dirik (2007) studied the effect of cold stratification for 0 or 45 days at 4±1°C in three provenances of *Pinus brutia* and found that, stratification significantly increased germination % at all potentials under study regardless germination temperature.

Thus, we conducted this research to conduct effect of seed priming on germination of Doum palm (*Hyphaene thebaica*) seeds and growing young seedling.

## MATERIAL AND METHODS

### Plant Material

Seeds of *Hyphaene thebaica* plants (Doum Palm) were bought from El Obour Market as a source of plant material during two seasons of the study. The average fresh weight of the seeds were about 18 g and 10 cm in diameter.

### Media used & preparation

The culture media used were 1:1 sand: peat-moss (v/v). Sand was washed by water and mixed with peatmoss then treated with 1 g/l Rizolex as disinfectant for soil spores, then media were mixed with calcium nitrate to adapt pH of the soil. Plastic bags used were about (18×25 cm) in diameter as a container filled with the above mentioned medium.

### Seed treatments

Seeds of Doum Palm were treated after removal of the hard kernel with different priming treatments as follows:

1. Control treatment without soaking. (Direct plantation).
2. Soaking in tap water for 48 hours (cold water).
3. Freezing storage in Deep-freezer at -18°C for 12 hours then soaked in Hot water at 60°C for 1 hour.
4. Soaking in Hot water (60°C ± 5) for 3 hours then left till gradually cooled.
5. Soaking in a solution of gibberellic acid (GA<sub>3</sub>) 200 ppm for 48 hours.
6. Soaking in a solution of potassium nitrate (KNO<sub>3</sub>) 3% for 48 hours.
7. Soaking in sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) 30% for 15 minutes.

### Sowing date & farming procedure

The sowing date was in 12 April for the two seasons under study (2017/2018). All treatments were cultured in the plastic bags about 3 cm in depth under the surface of media then coated with sand. All treatments were then shaded with serran sheets and irrigated immediately after sowing. Irrigation of all treatments was performed every week by tap water.

### Data Recorded & Statistical analysis

The data were recorded in October of both seasons as follow.

- 1- Germination percentage % = 
$$\frac{\text{Number of germinated seeds}}{\text{total number of seeds}} \times 100 \%$$
- 2- Length of roots.
- 3- Root diameter diameter.
- 4- Root fresh weight.
- 5- Root dry weight.

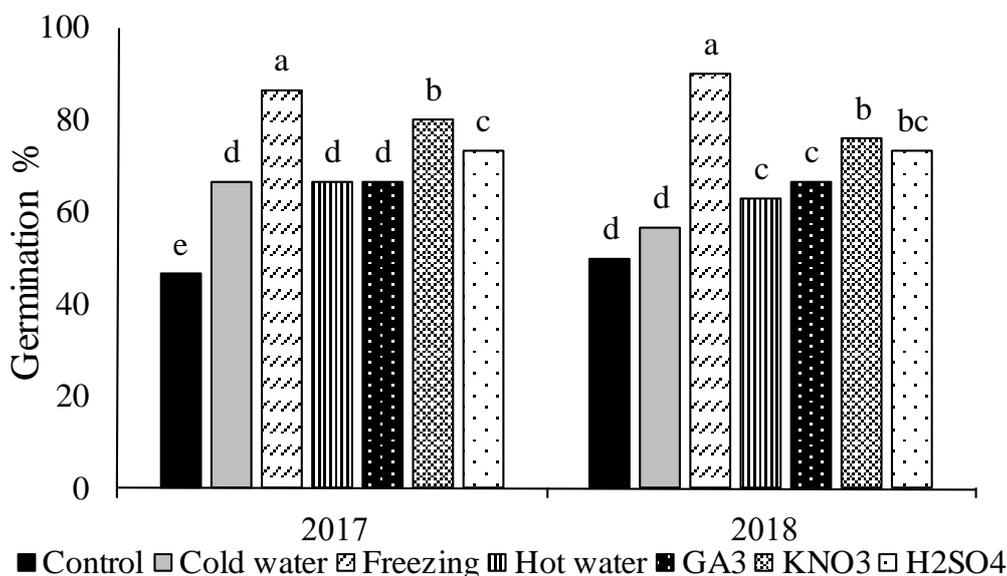
- 6- Length of seedling.
- 7- Wide of leaf.
- 8- Fresh weight of above ground parts.
- 9- Dry weight of above ground parts.

Collected data were subjected to one-way analysis of variance was carried out, as described by **Snedecor and Cochran (1989)**. The means were compared by determining for the germination experiment. Each treatment of the seed presoaking experiments consisted of 3 replicates and 5 seeds in each replicate. Duncan test ( $P \leq 0$ ) was used for analysis of the data, to evaluate differences among means (**Waller and Duncan, 1969**).

## RESULTS

### Germination percentage

Data presented in **Fig. (1)** indicate that seed germination percentage of *H. thebaica* was significantly affected by different treatments. The highest germination % was obtained from seeds treated with Freezing treatment (86.66 and 90%) and followed by soaked seeds in  $KNO_3$  (80 and 76%) in respect order by season. Meanwhile, the lowest germination % was observed when seeds were not treated with any priming methods as control (46.66 and 50%).



**Fig. 1.** Effect of various priming treatments on germination percentage of Doum palm *Hyphaene thebaica* seeds

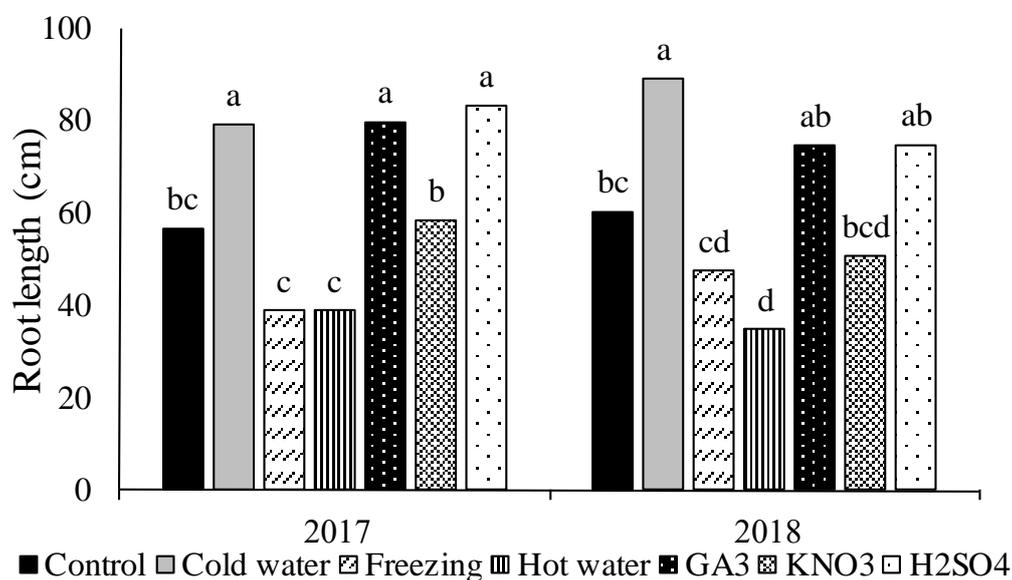
### Root length

Data presented in **Fig. (2)** showed that  $H_2SO_4$ , gibberellic acid and cold water treatments gave significant increase in root length in both seasons when compared to other treatment. In the 1<sup>st</sup> season  $H_2SO_4$ , gibberellic acid and cold water gave the longest root length (83.17, 79.67 and 79.00 cm) and in the 2<sup>nd</sup> season (89.00, 74.80 and 74.47 cm) in respect order. Whereas, the lowest root length was obtained when seeds were treated in the 1<sup>st</sup> season with hot water and freezing (38.83

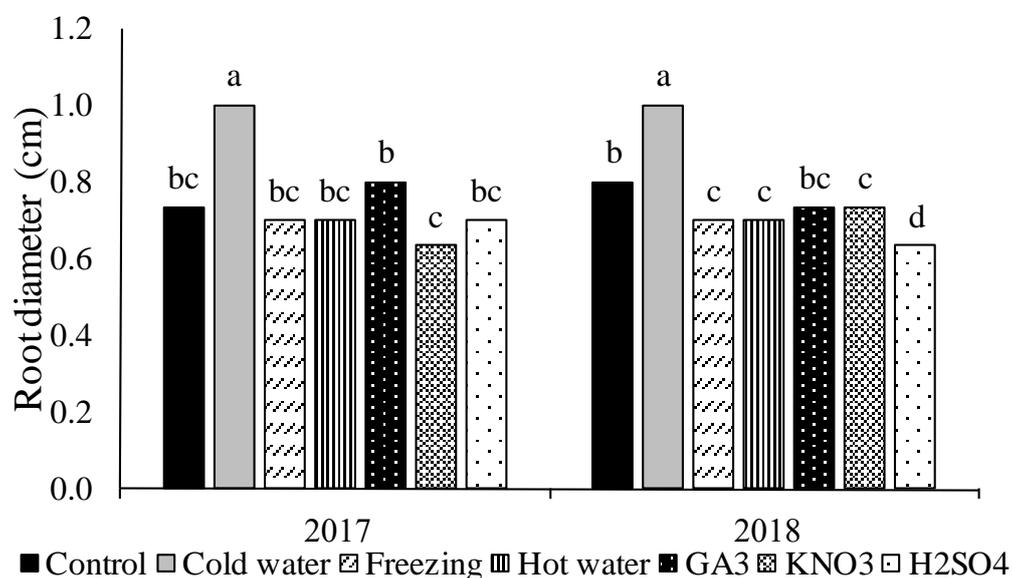
and 38.00 cm) and in the 2<sup>nd</sup> season gave (34.67, and 47.33 cm) in respect order.

### Root diameter

Data presented in **Fig. (3)** showed that cold water treatment gave significant increase in root diameter in both seasons when compared to other treatment. Whereas, the smallest root diameter was obtained when seeds were treated in the 1<sup>st</sup> with  $KNO_3$  gave the smallest root diameter (0.6 cm) and in the 2<sup>nd</sup> season  $H_2SO_4$  gave the smallest root diameter (0.63cm).



**Fig. 2.** Effect of various priming treatments on root length of young growing seedling of Doum palm *Hyphaene thebaica*



**Fig. 3.** Effect of various priming treatments on root diameter of young growing seedling of Doum palm *Hyphaene thebaica*

#### Root fresh weight

Data presented in **Fig. (4)** showed that cold water treatment gave significant increase in root fresh weight in both seasons when compared to other treatment. In the 1<sup>st</sup> and 2<sup>nd</sup> seasons, cold

water root fresh weight was (22.32 and 23.02 g/plant) in respect order. Whereas, the lowest root fresh weight was obtained when seeds were treated in the 1<sup>st</sup> season with hot water (10.29 g/plant) and with hot water and freezing (10.78 and 10.19 g/plant, respectively) in the 2<sup>nd</sup> season.

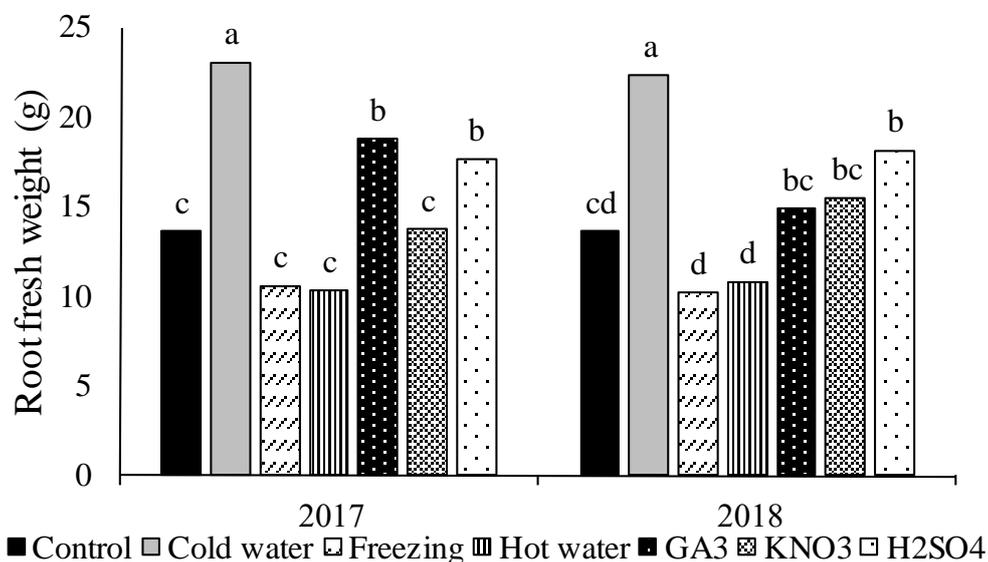


Fig. 4. Effect of various priming treatments on root fresh weight of young growing seedling of Doum palm *Hyphaene thebaica*

**Root dry weight**

Data presented in Fig. (5) indicated that cold water treatment gave significant increase in root dry weight in both seasons when compared to other treatment. Cold water treatment gave the

highest value of root dry weight (7.51 and 7.92 g/plant) in respect order by seasons. Whereas, the lowest root dry weight was obtained when seeds were non-treated (control) or hot water in the 1<sup>st</sup> season (3.97 and 3.00 g/plant) and in the 2<sup>nd</sup> season (3.77 and 3.01 g/plant) in respect order.

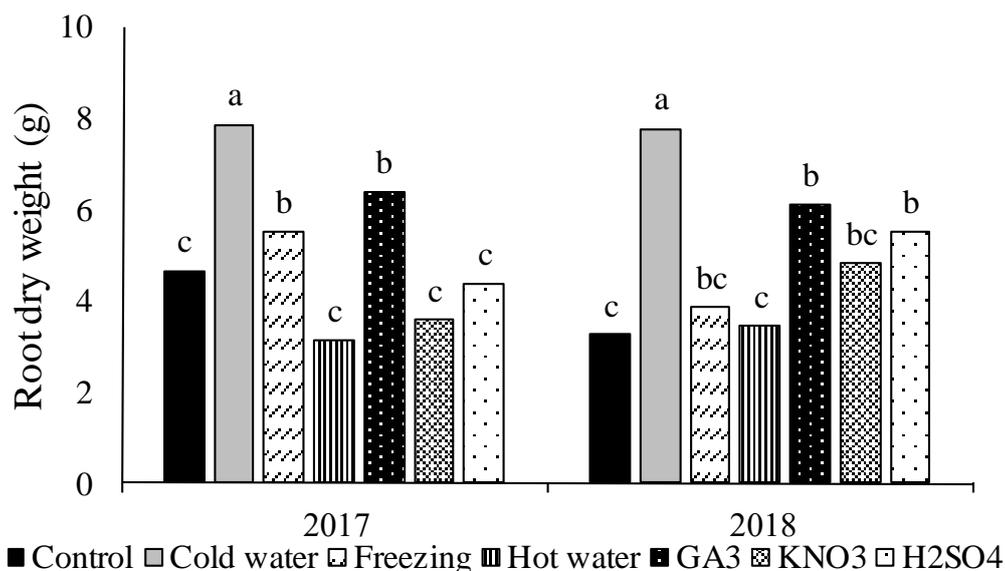
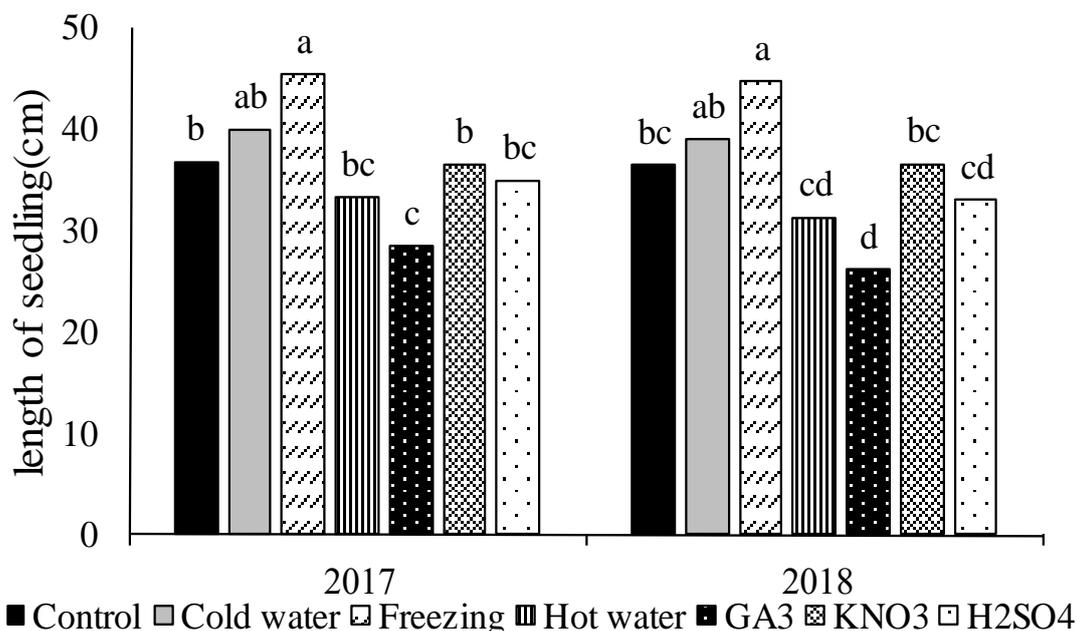


Fig. 5. Effect of various priming treatments on root dry weight of young growing seedling of Doum palm *Hyphaene thebaica*

#### 4.2.5. Plant height

Data presented in **Fig. (6)** showed that freezing, and cold water treatment gave significant increase in plant height in both seasons when compared to other treatments. In the 1<sup>st</sup> and 2<sup>nd</sup> seasons, freezing and cold water treatments gave

significant increase in plant height in the 1<sup>st</sup> season gave (45.33 and 39.97 cm) and in the 2<sup>nd</sup> season (44.6 and 26.17 cm) in respect order. Whereas, the shortest plants was obtained when seeds were treated with gibberellic acid (28.50 and 26.17 cm) in respect order by season.



**Fig. 6.** Effect of various priming treatments on length of young growing seedling of Doum palm *Hyphaene thebaica*

#### Leaf wide

Data presented in **Fig. (7)** showed that cold water, control and H<sub>2</sub>SO<sub>4</sub> treatment gave significant increase in leaf wide in both seasons when compared to other treatments. In the 1<sup>st</sup> season cold water, control and H<sub>2</sub>SO<sub>4</sub> gave larger width of leaves (2.64, 2.27 and 2.00 cm) and in the 2<sup>nd</sup>

season cold water and hot water gave larger width of leaves (2.55 and 2.50 cm) in respect order. Whereas, the smallest leaf wide was obtained when seeds were treated with gibberellic acid (1.52 cm) in the 1<sup>st</sup> and with H<sub>2</sub>SO<sub>4</sub> and gibberellic acid gave the (1.85 and 1.70 cm) in the 2<sup>nd</sup> season, respectively.

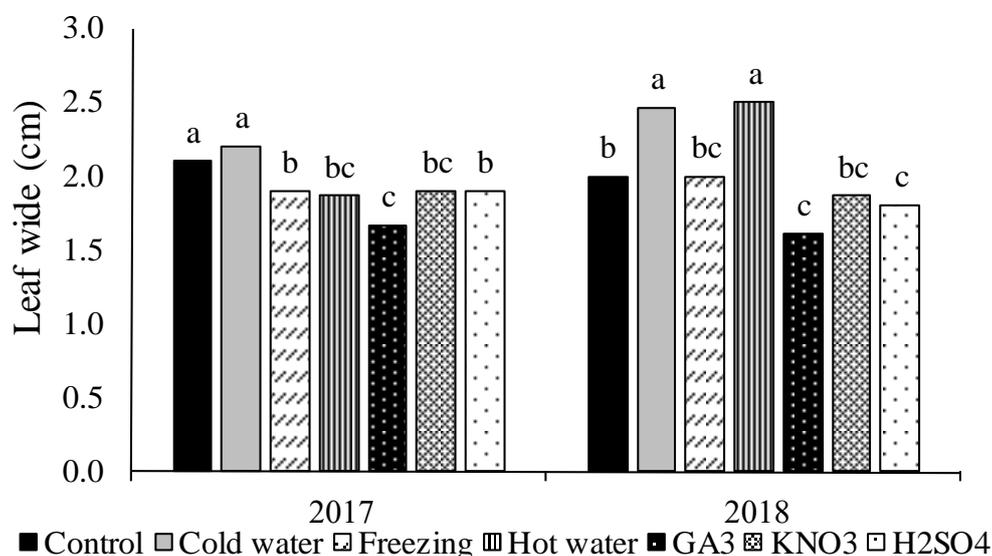


Fig. 7. Effect of various priming treatments on leaf wide of growing seedling of Doum palm *Hyphaene thebaica*

**Length of green part**

Data presented in Fig. (8) showed that freezing and H<sub>2</sub>SO<sub>4</sub> treatments gave significant increase in leaf length in both seasons when compared to other treatments. In the 1<sup>st</sup> season freezing gave the longest length of leaf (28.03 cm) and in the 2<sup>nd</sup>

season freezing and H<sub>2</sub>SO<sub>4</sub> gave the same trend (30.00 and 21.67 cm) in respect order. Whereas, the shortest of leaves were obtained when seeds were treated in the 1<sup>st</sup> season with hot water and gibberellic acid (13.67 and 12.50 cm) and in the 2<sup>nd</sup> season gibberellic acid showed the lowest value (12.85 cm).

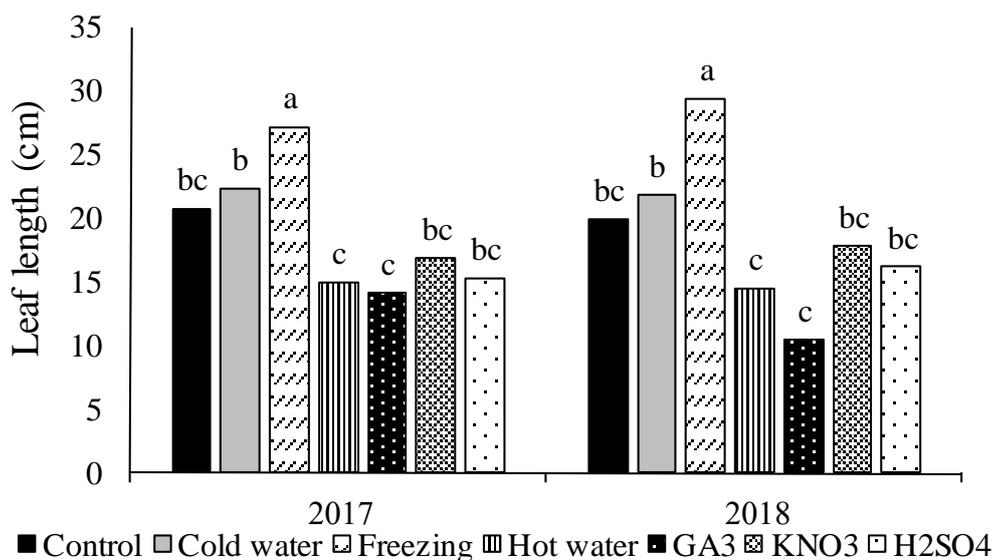
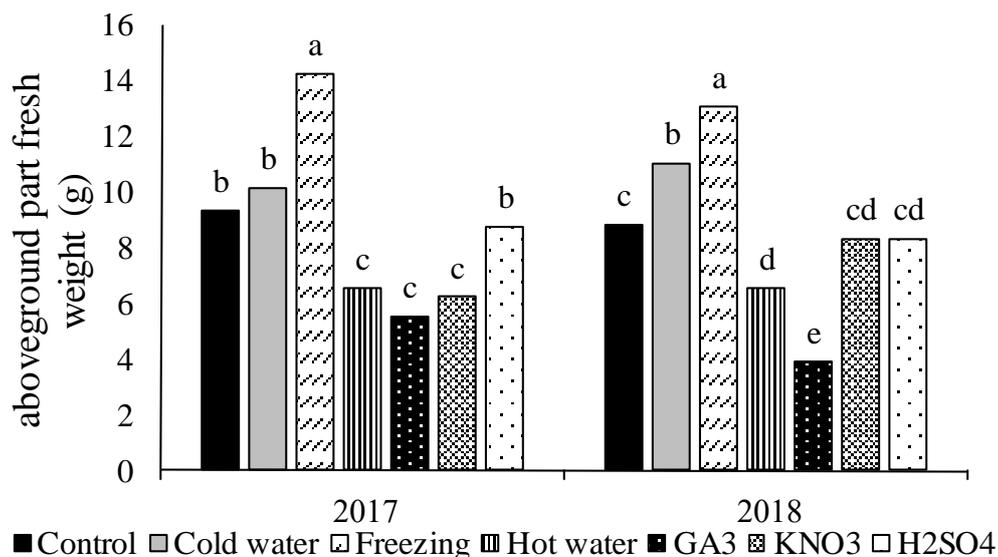


Fig. 8. Effect of various priming treatments on leaf length growing seedling of Doum palm *Hyphaene thebaica*

### Fresh weight of aboveground part

Data presented in **Fig. (9)** showed that freezing treatment gave significant increase in fresh weight of aboveground part in both seasons when compared to other treatment. Freezing gave the highest value of fresh weight for aboveground part in

both seasons (14.26 and 13.09 g/plant) in respect order. Whereas, the lowest aboveground part fresh weight was obtained when seeds were treated in the 1<sup>st</sup> with hot water, KNO<sub>3</sub> and gibberellic acid (6.56, 6.25 and 5.56 g/plant) and in the 2<sup>nd</sup> season gibberellic acid gave (3.92 g/plant).



**Fig. 9.** Effect of various priming treatments on aboveground fresh weight of growing seedling of Doum palm *Hyphaene thebaica*

### DISCUSSION

Pre-germination treatments were carried out to investigate the effect of different seed treatments on germination percentage and growth rate of young seedling of Doum palm *Hyphaene thebaica* Mart.

Results obtained here, showed that the highest percentages of seed germination of *H. thebaica* Mart were obtained when seeds were treated with freezing storage in deep-freezer at -18°C for 12 hours then soaked in hot water at 60°C for 1 hour. These findings are in agreement with those reviewed on effect of freeze-thaw scarification that breaks the seed coat by exposing seeds to temperature alternations between low and high (Stout, 1990; Rutar et al 2001). None of the previously studied research on seed priming of Doum palm tried the effect of freeze. Idohou et al (2015) on Doum *H. thebaica* Mart seeds reported that, the time of germination and water soaking had significant effects on seed germination. Also, Al-fredan

and Ali (2008) reported that on Doum palm seeds gave the highest seed germination percentage (93%) when seeds were mechanically scarificated then soaked in water for 24h. Non treated seeds gave the lowest germination percentage as well as hot water and treated seeds with gibberellic acid followed by H<sub>2</sub>SO<sub>4</sub>. The sulphuric acid treated seeds resulted in a low germination percentage (32%). Tahir et al (2007) on *H. thebaica* Mart. Çırak et al (2007) on *Hypericum aviculaviifolium*. Also, Kaseera et al (2002) found that *Leptadaenia reticulata* seeds subjected to soak in water for 24 h gave the highest seed germination percentage 100%. Sreerama et al (2000) on *Cassia fistula*, *C. spectabilis* and *Delonix regia* seeds, Moussa et al (1998) on Doum palm *Hyphaene thebaica* Mart seeds.

These above results may be explained by the fact that seed scarification might have removed the mechanical resistance of seed coat and this enabled the embryo to germinate without any mechanical obstacle.

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Mechanism behind the reduction of seed hardness by a freeze-thaw scarification is to make tiny scars on hard seed coat and make seed coat brittle to enhance germination (Busse, 1930; Stout, 1990; Hall et al 1993). A force that produces scars on seed surface through this technique is depending on the size, shape and water content of seeds and treatment intensity and durations (Kimura and Islam, 2012). Methods for cooling in freeze-thaw scarification include freezer, (Shibata and Hatakeyama, 1995); carbon dioxide (CO<sub>2</sub>) snow, dry ice, liquid air (Busse, 1930); ultra-low freezer (Stout, 1990); acetone (Rutar et al 2001); liquid N (Patane and Gresta, 2006). These findings are in agreement with those reviewed by Witte (1977) on *Zamia integrifolia*. The result of freezing on young growing seedling showed similar trend to cold water treatment. Also, similar results in agree on importance of lower temperature on seed germination Tilki and Dirik (2007) on *pinus brutia*, Millaku et al (2012) on *Gentiana lutea* L. and Kolotelo et al (2001) on some *Conifer* species.

Cracker and Barton (1953) concluded that low vigor seeds may need more water to germinate than high vigor seeds, and Hegarty (1978) contended that loss of vigor precedes loss of viability. Whereas, Martinková and Honěk, (2007) on *Taraxacum officinale* as the manner of seed storage has no effect on percentage of germination, frozen seed may be used safely for experiments studying this quality of seed materials. Thus, of lower germination % resulted from non-treated or cold water seeds, did not negative their effect for better growing young seedling.

Priming methods such as heat, freeze, mechanical and acid scarification are useful tools to soften hard seeds, improve germination and enhance seedling establishment. However, effectiveness of the methods varies depending on the duration of imposed treatments and species or cultivars to be used (Taia, 2004). Over treatment or longer time scarification may impose negative impacts on or injury to the seeds.

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## تأثير معاملات البذور علي إنبات بذور نخيل الدوم (*Hyphaene thebaica* Mart) وتطور الشتلات

[179]

أحمد حسني عبدالحى نجم<sup>1\*</sup> - سهير السيد حسن<sup>2</sup> - محمد هويدى<sup>2</sup>

- 1- معهد الدراسات والبحوث الزراعية في الأراضي القاحلة - جامعة عين شمس - القاهرة - مصر
- 2- قسم البساتين - كلية الزراعة - جامعة عين شمس - ص.ب. 68 - حدائق شبرا 11241 - القاهرة - مصر

\*Corresponding author: [negm.a.h@agr.asu.edu.eg](mailto:negm.a.h@agr.asu.edu.eg)

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### الموجز

بتركيز 200 جزء في المليون لمدة 48 ساعة، حامض الكبريتيك بتركيز 30% لمدة 15 دقيقة، التجميد علي -18°م. أظهرت معاملة التجميد أعلى نسبة إنبات (90%) يتبعه نترات البوتاسيوم (80%). وكانت أقل نسبة إنبات كانت من المقارنة والماء البارد بنسبة (45-65%). وكان لمعاملة التجميد والماء البارد تأثير جيد علي الشتلات الناتجة.

**الكلمات الدالة:** معاملة البذور، نترات البوتاسيوم، الجبريلين، التجميد، إنبات البذور، نخيل الدوم، الشتلات

نخيل الدوم يعتبر من أهم نبات الفلورا المصرية. تدخل ثمار الدوم في العديد من المنتجات الغذائية والشروبات والدواء. تواجه بذور الدوم صعوبات في الإنبات نتيجة صلابة الغلاف وحيوية البذور. في هذه الدراسة تم إختبار تأثير معاملات البذور بعد إزالة الغلاف الصلب. وكانت المعاملات كالتالي بدون معاملة وزراعة مباشرة للبذور (مقارنة)، ماء الصنبور (ماء بارد)، المعاملة بالماء الساخن لمدة 3 ساعات علي درجة حرارة  $5 \pm 60^{\circ}\text{C}$ ، النقع في محلول نترات البوتاسيوم 3% لمدة 48 ساعة، حامض الجبريليك