**Effects of cold and warm stratification on seed germination and early seedling growth of Mazari palm (*Nannorrhops ritchieana* Griff.)**

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**Abstract**

Arecaceae, the palm family, is a monophyletic group including 183 genera and 2364 species. The family includes palm trees of economical importance both as a source of agricultural produce and ornamental components as landscape objectives. There exists three genus of Arecaceae family in Iran among them, *Nannorrhops* commonly known as Mazari palm is native. Tow verities for this species have recorded in Iran namely; *N.ritchieana var. daz* (locally called Daz) and *N.ritchieana var. pork* (locally called Pork) in Sistan and Baluchestan province. This research was conducted to assess the effects of warm and cold stratification on seed germination and early seedling growth in Mazari palm. The results indicated that both seed germination percentage and seedling characteristics such as shoot length, root length, shoot fresh and dry weight and root fresh and dry weight were affected by the applied treatments particularly in Pork.The effects of cold stratification were more pronounced on Pork than Daz.

Key words: Stratification, seed germination, *Nannorrhops ritchieana* Griff.

**Introduction**

Arecaceae, the palm family, is a monophyletic group including 183 genera and 2364 species (Gomez-Navarro *et al*., 2009). At present, most palms are distributed in the tropics with a few species reaching subtropical areas. Fossil records from the tropics are less common, however, than those from middle latitudes. The palm fossil records reflect a broader latitudinal distribution of palms in the late Mesozoic and early Cenozoic because of warmer global climate, but also lack of study in the tropics (Gomez-Navarro *et al*., 2009). The main diversification centers are found in the equatorial coast of Africa, Oceanic, the Brazilian coast, the Amazon, Indonesia and the Antilles (Rivas *et al*., 2012). The palms are a globally important family of socio-economic plants. The family includes palm trees of economical importance both as a source of agricultural produce and ornamental components as landscape objectives (Iossi *et al*., 2006). The diversity in palm family is manifest in numerous growth forms, textures, colors and cultivation requirements. Palms are used in various interior and exterior landscape settings because of their assortments of features that add a touch of elegance and stimulate the senses (Perez, 2009). Although, a few palms are amenable to propagate through division of rhizomes, offshoots or tissue culture, the vast majority of landscape palms are propagated by seeds. However, commercial production of many palm species is hampered by delayed and inconsistent germination. Such sporadic germination is due to dormancy mechanisms (Perez, 2009). Palms are unique among woody ornamental plants, because with relatively, few exceptions, palm species can only be propagated from seed. Palms are also notorious in the nursery trade for slow and uneven seed germination. It has been estimated that over 25% of all palm species require over 100 days germinating and having less than 20% total germination (Meerow, 2004). The propagation of palms for agricultural and landscape purposes is accomplished typically by seeds. Because of irregular germination in most species, seed propagation may be a challenge (Robinson, ?)

*Nannorrhops ritchiana*, a gregarious perennial palm, commonly known as Mazari palm, is a distinctive flora of Saharo-Sindian Region (Naseem *et al*., 2005). The worldwide distribution of this genus is restricted to Iran, Afghanistan, Pakistan and Saudi Arabia (Mozaffarian, 2006), Oman (Dransfiel, 2004) and some other surrounding countries (Naseem *et al*., 2005). There exists three genus of Arecaceae family in Iran among them, *Nannorhops* is native (Roohani, 1989). The only species of this genus, *N.ritchieana* (Griff.) Aitchison, locally named Daz or Iranian dwarf palm, is naturally grown in Sistan and Baluchestan, Kerman and Hormozgan provinces (Emtehani, 2009). Tow verities for this species have recorded by Emtehani (2009) namely; *N.ritchieana var. daz* (locally called Daz) and *N.ritchieana var. pork* (locally called Pork) in Sistan and Baluchestan province.

About 15% of local people's income depends on the commodities, art crafts and handicrafts made of Daz leaves. More than 30 kinds of instruments such as ropes, baskets, cradles, hats, hand fans, cupboards, banns, bags, shoes and recently a beautiful cover for cell phones are made of Daz leaves which are attractive to domestic and international tourists. Some similar instruments made of Daz leaves are reported from Pakistan (Gibsson and Spanner, 1995). The leaves are also used for constructing a special rural house called "Kapar" in Persian. A cheese-like substance is derived from the core of inflorescence stem. The leaves and trunks could also be processed and used as palmpeat or for paper production. The species has some medicinal and nutritional applications in human’s diet and also used for animal feeding. In addition to vital ecological importance in ecosystems of southeastern parts of Iran, they also are very important from ornamental perspective considering sustainable landscape purposes.

Seed dormancy is an ecological adaptation that allows seasonal timing of germination for seeds in a population (Genevee, 2003). The challenge for seed ecologists is to determine why there is a delay in germination and why seeds eventually germinate at some specific seasons. Information on the environmental conditions required to promote germination provides valuable insights into what controls timing of germination in the fields and ultimately into the seed/seedling stage is adapted to a particular habitat (Chien *et al*., 2011). The positive effect of GA3 and alternate warm-cold stratification on seed germination has reported in *Taxus* *mairei* (Chien *et al*., 1998) and *Empetrum hermaphrodicum* (Baskin *et al*., 2002). Warm and cold stratification have found to be effective in overcoming seed dormancy in *Jasminu fruticans* (Pipinis *et al*., 2009). A specific germination response to warm-cold stratification has reported for *Rosa* taxa (Alp *et al*., 2009). The stimulatory effect of a sequence of cold-warm stratification has also reported for *Stewartia pseudocamellia* (Oleksak and Struve, 1999). The positive effects of GA3 on seed germination percentage has reported in *Helleborus* (McElhanno, 2008). Cold stratification increased seed germination rate in *Hyppophae rhamnoides* L. (Olmez, 2011). Positive effects of KNo3 on seed germination, seedling survival and seedling total biomass has reported for *Prunus armenaca* L. (Bhan and Sharma, 2011). Seedling height, seedling survival, higher seed germination and higher seedling biomass was obtained when seeds of *Prunus armenaca* L. were subjected to stratification (Bhan and Sharma, 2011). The application of KNO3 at 3% resulted in the highest germination percentage, seedling survival and shoot / root ratio in *Prunus armeniaca* (Bhan and Sharma, 2011). The highest germination percentage was obtained when *Cottinis ciggygria* seeds were subjected to sulphuric acid (50 min) and cold stratification for 15 days (Olmez *et al*., 2009). Cold stratification was found to be effective in increasing germination percentage and germination rate in some drought tolerant species such as, *Arbutus andrachne*, *Colutea* *armena*, *Cotoneaste*r *numullria*, *Elaeagnus* *angustifolia*, *Pyracanth*a *coccinea*, *Rhus coriaria*, etc. (Olmez *et al*., 2007).

The exploitation of *N.ritchieana* (Griff.) Aitchison, over the recent decades and also fire and particularly successive drought occurrences in native habitats have resulted in significant reduction in the vegetation. Meanwhile, there is little scientific information available on seed germination of these two varieties of *Nannorhops* naturally grown in Iran. Consequently, this research was conducted to investigate germination behavior of these two varieties in response to warm and cold stratification with the aim to find the best germination treatment for its large-scale sexual propagation.

**Materials and methods**

The fruits were collected from two natural stands in Sistan and baluchesnan province.After the seeds were removed from the fruits by hand; they were soaked in sodium hypochlorite for 30 min for disinfestations treatment. Then they were thoroughly washed and were kept in sterile cotton globes as medium and maintained for 20, 40 and 60 days at refrigerator at 5-7 ºC for stratification treatment. For warm stratification, the same procedure was followed and the seeds were kept at room temperature. The cotton globes were sprayed with distilled water occasionally to provide the moisture. At the end of warm and stratification periods, the seeds were sown in plastic pots contained cocopeat: perlite (1:1 V:V). The pots were watered as required. After 40 days, seed germination percentage, germination rate were measured at a 6-day interval for 5 times. Finally, the seedlings were taken out from the pots and shoot and root fresh and dry weights were measured. Data analysis was performed using SAS.9.1 software and the means were compared at 1 and 5% of probability using DMRT.

**Results**

Germination percentage in both varieties was affected by both cold and warm stratifications but this response was more pronounced in Pork than Daz (Fig. 1). Although the highest germination percentage was obtained in Daz with 20 days of cold stratification, but it was not different compared to control and some other treatments. All treatments increased germination percentage in Pork and the lowest germination percentage was gained in control treatment for this variety. The different response of these two varieties to the applied treatments, may reflect genetic differences as already reported for some other plant species such as…….... Shoot length was significantly increased in both varieties in comparison with control (Fig.2). Although the response to warm and cold stratification treatments followed no specific trend but it appears that Pork is more responsive to cold stratification than warm treatment. Some populations of Pork are naturally grown at higher altitudes than Daz and this characteristic may, in part, be related to this type of response. In the case of root length, the same trend was observed as was the case for shoot length and the lowest root length was measured in Pork control. All treatments significantly increased root length in Pork compared to control and cold stratification (40days) resulted in the highest root length. As was the case for shoot length, root length was more affected by cold stratification than warm stratification in Pork (Fig.3). All treatments significantly increased shoot fresh weight compared to control in Pork. The results also indicated that the longer the length of cold stratification, the higher the shoot fresh weight in Pork (Fig. 4). It seems that Daz follows no trend in this regard and the response is somewhat obscure. Generally, shorter periods of cold stratification (20 and 40 days) and warm stratification period (60days) significantly increased shoot fresh weight in this variety. Generally speaking, Daz is naturally grown at lower altitudes in plains and river beds and banks. Thus, this feature may, in part, explain its preference for shorter periods of cold stratification or longer warm stratification periods as seen in Fig.4. Root fresh weight follows the same trend observed in shoot fresh weight. All treatments significantly increased root fresh weight in Pork in comparison with control. In general, warm stratification resulted in higher root fresh weight in Daz than Pork (Fig. 5). But as mentioned for shoot fresh weight, root fresh weight followed no specific trend in Daz than Pork. A decrease in root fresh weight is seen in Daz, as the period of cold stratification increase. Shoot dry weight was not greatly affected by the applied treatments. Although not significant, but warm stratification decreased shoot dry weight in Pork than control. The highest shoot dry weight was gained in Daz with 60 days warm stratification but the difference was not significant compared to most of the treatments with the exception of warm stratification for 40 and 20 days in Pork. In contrast to shoot fresh weight, shoot dry weight in Daz is higher than Pork as shown in Fig. 6. It should be noted that the leaves of Pork is naturally thicker than Daz and for this reason, the leaves are not used for handicrafts. Therefore, this higher shoot dry weight may be associated with lower leaf water content in this variety. The interaction of variety and treatment indicated that this trait was not greatly affected by treatments (Fig.7). Although not significant, but root dry weight in Pork was higher than Daz. In general, Pork is larger than Daz and in some cases it grows as a small tree reaching to even 10 m in natural habitats. It might be postulated that larger root system are required for this type of growth habit. On the other hand, Daz prefers to grow on river beds and banks where bed rock is close to soil surface and water is more available. In contrast, Pork is naturally found in drier conditions where a massive root system is required for water uptake from deeper parts of soil. Consequently, the difference between these two varieties in root fresh and dry weights could most probably be associated with the ecological niches and natural habitat they commonly grow.

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Fig. 1- The interaction of variety and treatment on germination percentage.

Fig. 2- The interaction of variety and treatment on shoot length.

Fig. 3- The interaction of variety and treatment on root length.

Fig. 4- The interaction of variety and treatment on shoot fresh length.

Fig. 5- The interaction of variety and treatment on root fresh length.

Fig. 6- The interaction of variety and treatment on shoot dry length.

Fig. 7- The interaction of variety and treatment on root dry length.