

**STUDIES ON ONION SEEDS GERMINATION:
1- EFFECT OF MOIST-CHILLING ON GERMINATION OF
ONION SEEDS STORED FOR TWO DIFFERENT PERIODS
AND SUBSEQUENT SEEDLING GROWTH.**

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ABSTRACT

Onion seeds rapidly lose its viability and vigor after short time of harvest, and therefore its germinability largely decline. Consequently, the present investigation was carried out during the period from 2010-2012 in both Plant Tissue Culture Laboratory, Vegetables & Ornamentals Department, Faculty of Agriculture, Mansoura University and greenhouse of a private nursery in production of vegetable transplants, Qlabasho village, El-Dakahlia Governorate to study the effect of Moist-chilling (cold stratification) treatment for four periods (0, 5, 10 and 15 days) at 5 ± 1 °C on improvement of the germination parameters in the laboratory (germination percentage, seedling length, fresh and dry weight and vigor index) of onion seeds cv. "Red Beheri" stored for one or two years and enhancing growth characters (bulblet diameter, transplant length, fresh and dry weights and vigor index) of the subsequent transplants. The obtained results clearly indicated that all germination parameters of onion seeds stored for one or two years (except mean germination time) and also all obtained transplant characters except leaves number were significantly increased in all moist-chilling treatments than control one. The best treatment in this regard was moist-chilling for 15 days whereas the lowest one was control (non chilled seeds).

Keywords: Onion, *Allium cepa* L., Moist-chilling, Cold stratification, Seed storage periods, Germination parameters, Transplant characters.

INTRODUCTION

Onion (*Allium cepa* L.) belongs to the family Alliaceae and is cool-season, monocotyledonous, biennial plant (that is, It requires two growing seasons to complete the cycle from seed to seed), but It is grown commercially as an annual crop (Voss et al. 1999). It is propagated by seeds. Seeds are sown in a nursery, and four to six week-old seedlings are transplanted to the field. Onion is since ancient times a valuable vegetable crop for people all over the world. In this context, a mural from Egypt, dated approximately 3000 BC, depicts already images of onions. Therefore, it can be inferred that onions were already an important food source for the people from the Ancient Egypt. The word "onion" is derived from Latin and means "large pearl". The onion was compared to a pearl not only for its shape but also for its highly valuable nutritional quality (Shigyo1 and Kik, 2008).

Onion is a popular bulb vegetable crop in temperate, tropical, and subtropical regions. Both immature and mature bulbs are eaten raw, cooked, or used in the preparation of different vegetable dishes and in food processing including pickling, chutneys, sauces and dehydration (George, 2009). The green leaves are also used in salads, and cooked as a vegetable.

Onion contains high amounts of carbohydrates, calcium, phosphorus, vitamins A and B, and the volatile substance allyl propyl disulphide. Onion has medicinal value in that it possesses antibacterial properties (Doijode, 2001).

The most famous onion varieties cultivated in Egypt are Giza 6, Beheri and Giza 20 (Currah and Proctor, 1990). In Egypt, onion cultivated area was 146453.3 feddan in 2010, which produced 2208080 ton with an average of 15.08 ton/feddan (FAO Statistics Division, 2012).

Seed deterioration is significantly reduced at lower temperatures. Cold storage of onion seeds is a very satisfactory method for conservation of high seed viability for longer periods (Doijode, 1987). Onion seeds were successfully preserved at -20°C for 15 years without decline in viability and vigor, and there were no morphological variations in the seedlings that emerged from cold-stored seeds (Doijode, 1998).

It is generally known that onion seed is one of the shortest-lived seeds of the common vegetable crops (Duvel, 1905; Pritchard, 1933). As it loses its viability and vigor after harvest more rapidly than seeds of other crops unless special precautions are taken in its storage (Ellis and Roberts, 1977; Voss et al. 1999; Amjad and Anjum, 2002; Khan et al. 2004). It is therefore generally recommended that only fresh onion seed should be used for crop production (Riekels et al. 1976), and only seed of high germination percentage should be sold. The percentage and rate of germination of onion seeds also vary considerably among seed lots (Bedford and Mackay, 1973) and this leads to difficulties in establishing optimum plant populations in the field. Nevertheless, many onion seed sellers mixed the new seeds by the old one to attain large profits. Therefore, this procedure result in various problems between onion seed sellers and onion transplant producers because of decreasing transplants required in the field.

Cold stratification or chilling under moist conditions has long been recognized and widely used as a pre-sowing treatment (a useful method) of treating seeds to improve the rate and the percentage of germination and for breaking the dormancy (Schopmeyer, 1974; Outcall, 1991; AOSA, 1992; ISTA, 1999; Wang and Berjak, 2000). This method is simple, inexpensive and effective for overcoming the seed dormancy. Though, the phenomenon is not yet fully known, the effects of moist chilling in establishing hormonal levels have been proved due to initiation of appropriate enzyme activity (Nikolaeva, 1977).

The aim of the present investigation was (1) to study the effect of some moist-chilling treatments on improvement of onion seeds germination (which stored for one or two years within refrigerator) at laboratory, (2) to determine the impact of such treatments on obtained transplants characteristics in nursery.

MATERIALS AND METHODS

The present study was carried out in both a growth chamber of the Plant Tissue Culture Laboratory, Vegetables & Ornamentals Department,

Faculty of Agriculture, Mansoura University and greenhouse of a private nursery in production of vegetable transplants, Qlabasho village, El-Dakahlia Governorate during the period from 2010- 2012.

1. Onion seeds source and storage conditions.

Seeds of onion (*Allium cepa* L.) cv. "Red Beheri" were obtained after harvest in June 2010 and 2011 years, respectively from a private farm specialized in production of onion seeds, Ein Marine Life Village, Qaleen city, Kafr El-Sheikh Governorate. Abnormal and damaged seeds were firstly removed from both of two seed samples, which harvested during either 2010 or 2011 year. While the good remaining onion seeds of both two years were well drayed and stored till 2012 year within sealed, doubled, cleared polyethylene bags which were put in sealed glass bottles and then finally preserved within refrigerator at 5-6 °C. The onion seeds which stored under these conditions for one or two years were removed from refrigerator, thereafter they remained preserved in their sealed containers overnight to equilibrate fully with laboratory temperatures before using them in moist-chilling experiment.

2. Moist-chilling treatments.

For moist-chilling (MCh) treatments, onion seeds stored either for one or two years were moisturized with distilled water then placed in a moistened cloth bags and thereafter stored in sealed plastic boxes in darkness within refrigerator at temperature of 5 ± 1 °C, for 5,10 and 15 days. Non moist-chilled onion seeds were used as the control treatment for each storage period.

3. Germination test.

Treatments were arranged in a Completely Randomized Design with four replicates, 25 seeds of each. For germination test, the onion seeds of uniform size stored either for one or two years in each treatment were sown on top of one filter paper in 12 cm diameter sterilized petri dishes. Each filter paper was moistened with 10 ml of distilled water. After pre-treatments of onion seeds, they were incubated for 14 days in a growth chamber adjusted to 20 ± 1 °C in a dark. For each treatment, 25 seeds were counted at random and placed on a moist germination paper in a petri dish. There were four replicates for each treatment. Dishes were examined daily and distilled water added as required. Every 24 hours the number of germinated seeds was counted. Germination percentage (%), mean germination time (day), seedling vigor index, seedling length (cm), seedling fresh and dry weight (mg) were registered throughout the period of germination test (14 days) according to the International Seed Testing Association (ISTA, 1993).

4. Tested measurements.

4.1. Germination parameters.

Final germination percentage (GP): It was calculated according to the germination count taken after 14 days and expressed as percentage according to the following equation which described by International Seed Testing Association (ISTA, 1993) and Ruan *et al.* (2002):

$$\times 100 \text{Germination Percentage} = \frac{\text{Number of Germinated Seeds after 14 Days}}{\text{Number of Cultured Seeds}}$$

Mean germination time (MGT): It was performed according to the following equation which described by Alvarado and Bradford (1987):

$$MGT = \frac{\sum dn}{\sum n}$$

Where (n) is the number of seeds which germinated on day (d), and (d) is the number of days counted from the beginning of germination.

Seedling vigor index (SVI): It calculated according to the following equation which described by Abdul-Baki and Anderson (1973):

$$SVI = (\text{Average Shoot Length} + \text{Average Root Length}) \times \text{Germination Percentage}$$

4.2. Seedling characters at laboratory.

- Shoot length: The shoot length of ten seedlings was registered and expressed in centimeters (cm).
- Root length: The root length of ten seedlings was recorded and expressed in centimeters (cm).
- Seedling fresh weight: The weight of ten seedlings was scored and expressed in milligram (mg).
- Seedling dry weight: The weight of ten seedlings was counted and expressed in milligram (mg) after oven drying at 70 °C for 72 hours.

4.3. Transplant characters at nursery.

Leaves number, bulblet diameter (cm), transplant length (cm), fresh and dry weight (mg) and vigor index were recorded after 4 weeks of culturing the obtained seedlings from the previous germination treatments in speeding trays contained peat moss : vermiculite at 1:1. The cultured seedlings were taken all agricultural practices respecting of this crop.

5. Experimental design and statistical analysis

The obtained data were statistically analyzed as a factorial experimental design (SPSS, 2001) applying the Least Significant Difference (LSD) at 5% for the comparison among the treatment means.

RESULTS AND DISCUSSION

A. Laboratory experiment.

A.1. Effect of moist-chilling periods on onion seeds germination and seedling parameters.

Results presented in Table (1) clearly indicate that among the investigated moist-chilling treatments for germination improvement of onion seeds, the moist-chilling treatment for 15 days was the most effective one, since it recorded the highest values for germination percentage, seedling length, seedling fresh and dry weight, and seedling vigor index (96.5 %, 6.46 cm, 29.62 mg, 3.26 mg and 623.56, respectively) followed by the moist-

chilling treatment for 10 days which registered 89 %, 5.86 cm, 27.53 mg, 3.03 mg and 521.24, consecutively for the same above parameters. Whereas, the control treatment (non moist-chilled onion seeds) scored the lowest values for all forecited parameters (68 %, 4.11 cm, 21.21 mg, 2.33 mg and 279.89, successively). On the other hand, there are no significant differences among all moist-chilling treatments and control treatment for the mean germination time.

A.2. Effect of seed storage periods on onion seeds germination and seedling parameters.

Respecting the effect of seed storage periods on seeds germination and seedling parameters, results also presented in Table (1) show obviously that there are no significant differences between effect of both onion seeds stored either for one or two years on all above mentioned seed germination and seedling parameters.

A.3. Effect of interaction between moist-chilling treatments and storage periods on onion seeds germination and seedling parameters.

Concerning the impact of interaction between moist-chilling treatments and seed storage periods on onion seeds germination, Results presented in the same table and illustrated in Fig. (1) reveal that the best treatment for enhancing seeds germination and obtained seedling parameters from both two storage periods was moist-chilling treatment for 15 days.

Table (1): Effect of moist-chilling periods, seed storage periods and their interactions on germination percentage, seedling length, seedling fresh and dry weight, mean germination time and seedling vigor index of onion seeds during 2010-2012 period.

Treatments	parameters						
	Germination percentage (%)	Seedling length (cm)	seedling fresh weight (mg)	seedling dry weight (mg)	Mean germination time (day)	Seedling vigor index	
A- Moist-chilling periods (day)							
0 (control)	68.0	4.11	21.21	2.33	6.33	279.89	
5	83.0	4.91	23.52	2.59	6.27	406.49	
10	89.0	5.86	27.53	3.03	6.26	521.24	
15	96.5	6.46	29.62	3.26	6.20	623.56	
LSD _{0.05}	6.37	0.46	1.25	0.14	0.20	54.53	
B- Seed storage periods							
One year	87.00	5.40	26.09	2.87	6.21	478.67	
Two years	81.25	5.27	24.86	2.73	6.32	436.91	
F- test	NS	NS	NS	NS	NS	NS	
C- Interaction effects							
0	One year	72	4.21	21.76	2.39	6.27	302.59
5		84	4.90	24.16	2.66	6.17	410.87
10		92	5.91	27.98	3.08	6.21	543.72
15		100	6.58	30.44	3.35	6.17	657.50
0	Two years	64	4.02	20.66	2.27	6.38	257.18
5		82	4.92	22.89	2.52	6.36	402.10
10		86	5.80	27.08	2.98	6.30	498.75

15		93	6.34	28.80	3.17	6.22	589.62
LSD _{0.05}		4.58	0.48	0.86	0.09	0.18	44.38
A × B							

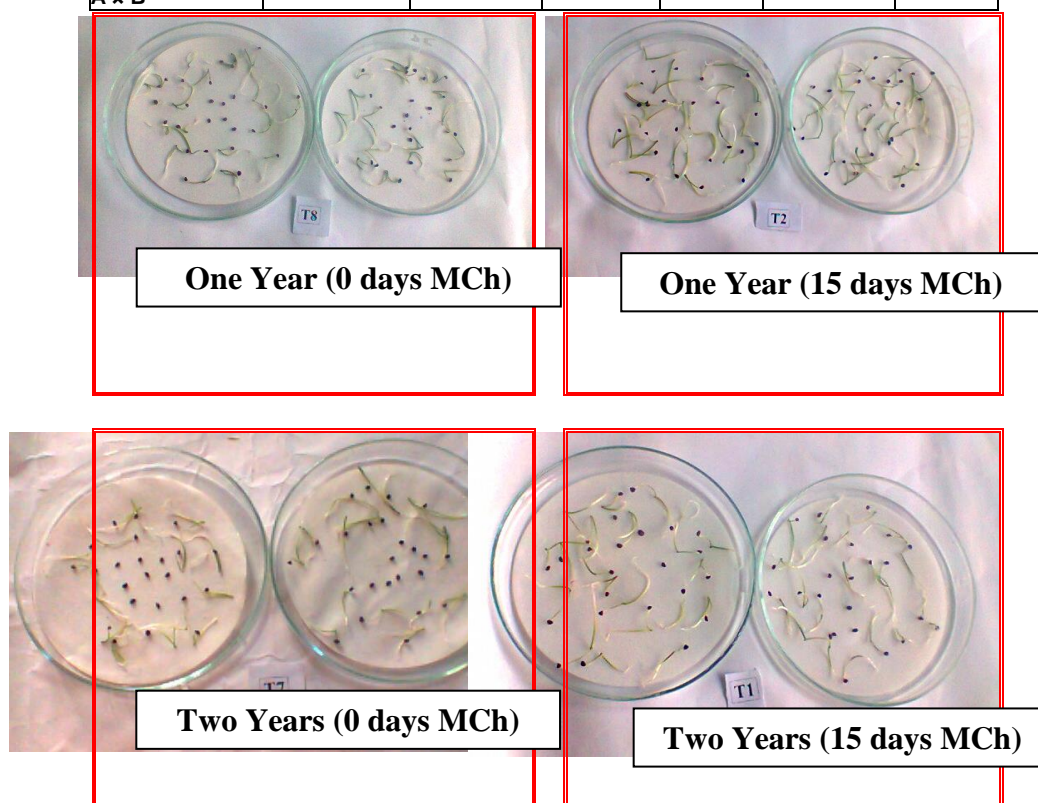


Figure (1): Effect of interaction between moist-chilling treatments and storage periods on onion seeds germination and seedling parameters. MCh, moist-chilling.

This treatment recorded the highest values (100 %, 6.58 cm, 30.44 mg, 3.35 mg and 657.50) for germination percentage, seedling length, seedling fresh and dry weight and seed vigor index, consecutively for onion seeds stored for one year and 93 %, 6.34 cm, 28.80 mg, 3.17 mg and 589.62, respectively onion seeds stored for two years. The lowest treatment for both two storage periods of seeds was control one. This treatment registered 72 %, 4.21 cm, 21.76 mg, 2.39 mg and 302.59 for onion seeds stored for one year and 64 %, 4.02 cm, 20.66 mg, 2.27 mg and 257.18 for onion seeds stored for two years for all previously mentioned parameters. The rest moist-chilling treatments gave values between these two extremes. On the contrary, there are no actual significant differences among all used treatments for mean germination time parameter.

B. Nursery experiment.

B.1. Effect of moist-chilling treatments on nursery performance of onion seedlings obtained from previous germination experiment.

Regarding the impact of moist-chilling treatments on nursery performance of onion transplants resultant from germination of onion seeds, results presented in Table (2) show that moist-chilling treatment for 15 days gave the highest values for bulblet diameter (3.70 cm), transplant length (26.38 cm), fresh weight (843.88 mg), dry weight (103.00 mg) and transplant vigor index (2546.00). Whereas, the control treatment recorded the lowest values for the above mentioned parameters in this respect (0.28 cm, 23.32 cm, 366.38 mg, 49.25 mg and 1587.00, respectively). On the other hand, there are no significant differences among all moist-chilling treatments and control respecting the leaves number parameter.

B.2. Effect of seed storage periods on nursery performance of onion seedlings obtained from previous germination experiment.

In respect to the effect of seed storage periods on nursery performance of onion seedlings, the data also existed in Table (2) clear that there are no significant differences between both onion seeds stored either for one or two years on all above mentioned seed germination and seedling parameters.

Table (2): Effect of moist-chilling periods, seed storage periods and their interactions on leaves number, bulblet diameter, length, fresh and dry weight and vigor index of transplants (4 weeks old) obtained from onion seeds during 2010-2012 period.

Treatments	parameters						
	Leaves number	Bulblet diameter (cm)	Transplant length (cm)	Transplant fresh weight (mg)	Transplant dry weight (mg)	Transplant vigor index	
A- Moist-chilling periods (day)							
0 (control)	2.25	0.28	23.32	366.38	49.25	1587.00	
5	2.25	2.03	22.79	494.13	77.25	1892.75	
10	2.50	3.11	24.75	611.38	97.63	2205.25	
15	2.63	3.70	26.38	843.88	103.00	2546.00	
LSD _{0.05}	0.72	0.74	1.84	131.99	22.99	253.79	
B- Seed storage periods							
One year	2.44	2.52	24.50	650.88	93.31	2143.63	
Two years	2.38	2.04	24.11	507.00	70.25	1971.88	
F - test	NS	NS	NS	NS	NS	NS	
C- Interaction effects							
0	One year	2.25	0.34	23.50	414.25	62.00	1693.0
5		2.25	2.55	23.13	603.25	99.25	1943.5
10		2.50	3.45	25.13	633.50	99.75	2313.0
15		2.75	3.75	26.25	952.50	112.25	2625.0
0	Two years	2.25	0.22	23.13	318.50	36.50	1481.5
5		2.25	1.50	22.45	385.00	55.25	1842.0
10		2.50	2.78	24.38	589.25	95.50	2097.5
15		2.50	3.65	26.50	735.25	83.75	2467.0
LSD _{0.05} A x B		0.77	0.60	1.95	42.91	10.16	232.08

B.3. Effect of interaction between moist-chilling treatments and seed storage periods on onion transplant characters in nursery.

With relation to the impact of interaction between moist-chilling treatments and seed storage periods on characters of onion transplants in nursery, Results presented in the same Table (2) demonstrated clearly that the moist-chilling treatment for 15 days was the most effective one in improvement of onion transplant characteristics regenerated from both two storage periods. This treatment recorded values 3.75 cm, 26.25 cm, 952.50 mg, 112.25 mg and 2625.0 for bulblet diameter, transplant length, transplant fresh and dry weight (mg) and transplant vigor index, respectively of transplants developed from onion seeds stored for one year and 3.65 cm, 26.25 cm, 735.25 mg, 93.75 mg and 2467 for the same above characters of transplants derived from onion seeds stored for two years. On the contrary, the control treatment registered the lowest values for the same previous characters, i.e., 0.34 cm, 23.50 cm, 414.25 mg, 62.00 mg and 1693.0 for transplants regenerated from onion seeds stored for one year vs. 0.22 cm, 23.13 cm, 318.50 mg, 36.50 mg and 1481.5 for transplants produced from onion seeds stored for two years. Whereas, the rest treatments gave medium values for the forecited characters of transplants resultant from germination of both two onion seed storage periods. On the other hand, there are no significant differences among all moist-chilling treatments and control for leaves number parameter.

DISSECTION

The results of this study indicated that all of the moist-chilling treatments significantly increased the germination of onion seeds stored either for one or two years and the subsequent transplant growth parameters as compared with the control one. These results are in the same line with those of El-Nabawy *et al.* (1980) on pecan seeds, El-Dengawy (1997) on peach seeds and Samaan *et al.* (2000) on apricot seeds. Likewise, Ghildiyal *et al.* (2009) concluded that cold stratification for 15 days improved the rate and percentage of germination of the non-dormant chirpine (*Pinus roxburghii*) seeds when germinated at 20 °C and 25 °C. Also, Fariman *et al.* (2011) who found that the highest germination percentage and germination rate of *Echinacea purpurea* seeds were obtained in cold stratification treatment for 21 days at 5 °C that induced about 98 % germination and the lowest mean germination time also was observed in the same cold stratification treatment. As well, Nematollah *et al.* (2011) mentioned that germination percentage of Persian shallot seeds was improved by increase of cold stratification period to the sixth week at 4 °C.

It could be interpretation of this result on the basis of that during the moist-chilling (cold stratification) treatment, some of the following biochemical changes were done, i.e., 1) some bio-chemicals and phyto-hormones were changed to be able to germinate dormant seeds by decreasing abscisic acid and increasing GAs (Mathur *et al.*, 1971; Bulard, 1985; Halinska and Lewak 1987, Hartmann *et al.*, 1990). 2) some genetic activities including changes in

mRNA and protein levels occurred and this may be due to the increased expression of genes that control biosynthesis of gibberellic acid as activator for seeds germination (Finkelstein et al., 2008).

From the obtained results in the present study, it could be concluded that the moist-chilling treatment for 15 days was the best treatment for improvement of the onion seeds germination stored for one or two years and enhancing the subsequent transplant growth at nursery.

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دراسات على إنبات بذور البصل.

١- تأثير الكمر البارد على إنبات بذور البصل المخزنة لفترتين مختلفتين والنمو التالي للبادرات

محمود محمد زغلول ، وليد علي السعدي و خديجة محمد حمد.
قسم الخضر والزينة، كلية الزراعة، جامعة المنصورة، المنصورة، مصر.

تفقد بذور البصل حيويتها وقوتها بسرعة بعد فترة قصيرة من الحصاد، لهذا السبب تضعف قدرتها على الإنبات بدرجة كبيرة. بناءً على ذلك، أجريت الدراسة الحالية خلال الفترة من ٢٠١٢-٢٠١٠ في كل من معمل زراعة الأنسجة النباتية، قسم الخضر والزينة، كلية الزراعة، جامعة المنصورة وكذلك صوبة لأحد المشاتل المتخصصة في إنتاج شتلات الخضر بقرية قلابشو، محافظة الدقهلية لدراسة تأثير معاملات الكمر البارد لأربعة فترات (صفر، ٥، ١٠ و ١٥ يوماً) علي درجة حرارة ٥ °م بغرض تحسين صفات إنبات البذور في المعمل (النسبة المئوية للإنبات، طول البادرات، الوزن الطازج والجاف ودليل القوة للبادرات) لصنف البصل البحيري الأحمر المخزنة إما لسنة أو لسنتين، هذا بالإضافة إلي تحسين صفات النمو (قطر البصيلة، طول الشتلة، الوزن الطازج والجاف وكذلك دليل القوة) للشتلات الناتجة. لقد أظهرت النتائج المتحصل عليها أن جميع صفات الإنبات لبذور البصل المخزنة لمد سنة أو سنتين (ما عدا صفة متوسط زمن الإنبات) وكذلك جميع صفات الشتلات المتحصل عليها (ما عدا صفة عدد الأوراق) قد زادت بدرجة معنوية في جميع معاملات الكمر البارد عن معاملة الكنترول. وكانت أفضل المعاملات في هذا الشأن معاملة الكمر البارد لمدة ١٥ يوم، بينما كانت معاملة الكنترول (البذور غير المكورة) المعاملة الأقل.

قام بتحكيم البحث

كلية الزراعة – جامعة المنصورة
كلية الزراعة – جامعة بنها

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