DEVELOPMENTAL CONTROL OF SOME PHYSIOLOGICAL FACTORS ON REPRODUCTIVE BIOLOGY AND RUDIMENTARY EMBRYOS PHENOMENON IN CARROT SEEDS

Aseel M.H. H. Al-Khafaji **Kadhim D. H. Al-jubouri**
Lecturer **Prof.**
Dept. Hort. Landscape Gard. Coll. Agric. Engin. Sci./ University of Baghdad
aseel.m@coagri.uobaghdad.edu.iq **kadhim.aljubouri@coagri.uobaghdad.edu.iq**

ABSTRACT

This study was aimed to investigate delayed germination and immature embryos of carrot seeds; This research was implemented at vegetable field of College of Agricultural Engineering Sciences - University of Baghdad during two sequent seasons (fall and spring). The experiment was conducted using factorial within Randomized Complete Block Design arrangement with three factors and replicates (3X3X2). The aqueous barley sprouts extract (B_0, B_1) (0, 100 g.L^{-1}) represented the first factor. Trehalose (T_0, T_1, T_2) (0, 50, 75 mmole.L^{-1}) represented the second factor. Calcium (C_0, C_1, C_2) (0, 1, 2 ml.L^{-1}) represented the third factor. Results showed the effectiveness of third order interaction treatment $B_1T_2C_1$ in increasing germination speed and percent for the seeds of royal umbel (92, 97%); secondary umbel (90.67, 94.67%); and tertiary umbel (75.68, 86.33%). The mentioned treatment increased embryo length for the seeds of primary (royal) umbel (1628.3 µm), secondary umbel (1620.3 µm); and tertiary umbel (874.7 µm). in compare with the lowest percents that found in $B_0T_0C_2$ treatment.

Keywords: trehalose; calcium, barley sprouts, immature embryos; delayed germination; embryogenesis; embryo/endosperm proportion
***Part of Ph.D. dissertation for the 1st author**

السيطرة التطورية لبعض العوامل الفسلجية على الحياتية التكاثرية وظاهرة الاجنة الاثرية في بذور الجزر

أسيل محمد حسن هاتف الخفاجي **كاظم ديلي حسن الجبوري**
مدرس **استاذ**
قسم البستنة وهندسة الحدائق/كلية علوم الهندسة الزراعية/جامعة بغداد

المستخلص

بهدف حل مشكلة تأخر انبات وعدم اكتمال نضج اجنة بذور نبات الجزر نُفذت تجربة حقلية في حقول كلية علوم الهندسة الزراعية/جامعة بغداد بموسمين متتاليين (خريفي وربيعي، دورتي النمو الاولى والثانية). طُبقت التجربة باستعمال تصميم القطاعات الكاملة المعشاة على وفق ترتيب التجارب العاملية وبثلاث عوامل وبثلاث مكررات (2X3X3)، مَثَّل الرش بالمستخلص المائي لحبوب المستنبتة لنبات الشعير (0، 100 غم حبوب جافة.لتر$^{-1}$) العامل الاول والذي رُمز له (B_0 وB_1)، اما العامل الثاني فتضمن الرش بسكر التريهالوز (0، 50 ، 75 ملي مول.لتر$^{-1}$) والذي رمز له (T_0 وT_1و T_2)، واشتمل العامل الثالث على الرش بالكالسيوم على هيئة كالسيوم مخلبي (0 و 1 و 2 مل.لتر$^{-1}$) والذي رمز له (C_0 وC_1و C_2)،. اظهرت النتائج التفوق المعنوي لمعاملة التداخل الثلاثي $B_1T_2C_1$ في زيادة النسبة المئوية لسرعة ونسبة الانبات لرتب النورات الملكية (92، 97%) والثانية (90.67، 94.67 %) والثالثة (75.68، 86.33 %)، فضلاً عن زيادة طول الجنين لبذور كل من رتب النورات الاولى (الملكية) (1628.3 مايكرون) والثانية (1620.3 مايكرون) والثالثة (874.7 مايكرون) مقارنة باقل النسب التي وجدت عند معاملة $B_0T_0C_2$.

الكلمات المفتاحية: تريهالوز، كالسيوم، مستنبتات الشعير، اجنة غير ناضجة, انبات متأخر, التخلق الجنيني، نسبة الجنين الى السويداء
***مستل من اطروحة دكتوراه للباحث الاول**

Received:11/2/2023, Accepted:7/5/2023

INTRODUCTION

Carrot *Daucus carota* that belongs to Apiaceae; has very sophisticated reproductive biology because of its distinctive features that could be summarized by First; umbels consequent appearance, Second; carrots protandrous flowers (20), and finally; carrot rudiment embryos (23). The main cause of this problem is a defect in carrot embryogenesis that illustrated by the growth and enlargement of endosperm over embryo. As a results; carrot seed embryo forms 2-3% only from endosperm (21). What mentioned prior leads to cause a cascade of different problems; their final result is producing seeds with different qualities and large percent of weak seeds (23). Sprouting grains sharply increase GA_3 and rapid decrease in ABA levels, which could be a good reason to activate embryogenesis in carrot seeds (24). Moreover; they considerably increase nutrients bioavailability, i.e. breaking down proteins to amino acids, starch to sugars, and fats to fatty acids (17). Hence, their aqueous extracts could be sprayed on plant and have a good impact (3, 4, 5). Trehalose has many functions in plant such as, carbon metaboliztion, plant protection from abiotic stresses (2 , 6, 13, 22) and signalization (8, 11, 16). In addition, it has proven that trehalose organizes florigen transporting to shoot tips in order to evocate floral induction. Moreover; trehalose activates *SPL* gene that also stimulates flowering (Cho) (22). Eastmond et al (9) demonstrated that trehalose metabolism is necessary for embryogenesis stages completion. Calcium an essential element in plant reproductive biology since it directly affects fertilization. It considers a directional cue for pollen tube. Even more; calcium establishes the polarity of pollen tube. As for the ovary and ovules; calcium abundance in micropyler region and embryo sac attracts the pollen tube entry (7, 10, 12). Consequently, this study was aimed to target carrot seeds embryogenesis by using different treatments (sprouts aqua extract, trehalose, and calcium) that are targeting the same issue but with different mechanisms of action.

MATERIALS AND METHODS

This experiment was conducted during two sequent seasons, fall -first growth cycle (roots production), and spring -second growth cycle (seeds production) at researches station (A) College of Agricultural Engineering Sciences, University of Baghdad (Al-Jadiryah). Table 1 shows the chemical and physical characteristics of the soil for the two seasons. For the first season; the seeds of carrot var. Nantes were sowed on rows on terraces in 15/September. The field was under drip irrigation system. The seeds were planted in a plant density 1,000,000 plants.ha^{-1}. The entire plots harvested after 115 days of the planting date. The produced stecklings (roots) stored in a dark place at 4ºC for 28 days to induce vernalization (21). After that (the second season); carrot stecklings transplanted on terraces with two rows. The spacing between one plant and another was 0.3 m. Mineral fertilizer was added as recommended for carrot plants (120 kg.ha^{-1}, 120 K_2O_5 kg. ha^{-1}, 40 K_2O kg. ha^{-1}) to all plots before transplanting (1). The experiment was implemented using randomized complete block design with factorial arrangement (2X3X3) with three replicates. Spraying barley aqueous sprouted grains extract was represented the first factor with two levels (0, 100g.L^{-1} DW) which symbolized (B_0, B_1). The second factor is spraying with three levels of trehalose (0, 50, 75 mmol.L^{-1}) which symbolized (T_0, T_1, T_2). The third factor is spraying with three levels of calcium (0, 1, 2 ml.L^{-1}) (as chelated calcium 30% Ca), which symbolized (C_0, C_1, C_2). The plants foliarly sprayed for three times. For the first season (roots production); The first spraying was ten days after thinning and the duration between one spraying and another was 15 days. For the second season (seeds production); the first spraying was after emergence completion. The period between sprayings was 30 days.

Table 1. Physical and chemical characteristics of the soil for the two seasons

Character	Values 1st season	2nd season
pH	7.44	7.41
$EC_{1:1}$ (ds.m^{-1})	2.39	2.36
Total N (mg kg^{-1})	55.0	45.6
P (mg kg^{-1})	13.7	12.1
K (mg kg^{-1})	170	166
Ca (mg kg^{-1})	187	177
Mg (mg kg^{-1})	170	130
Fe (mg kg^{-1})	2.60	2.10
Na (Meq L^{-1})	61.0	63.0
Cl^- (Meq L^{-1})	51.0	55.0
SO_4^{-2} (Meq L^{-1})	207	209
HCO_3^- (Meq L^{-1})	477	453
O.M. (%)	10.3	9.10
Gypsum (%)	320	327
Sand (%)	12.0	15.0
Silt (%)	40.0	45.1
Clay (%)	48.0	39.9
Texture	Clay Loam	

Barley aqueous sprouted grains extract was prepared by hydroponically germinating 100 g of barley grains, var. Ebaa class 265, (until radical penetration stage). After that, the germinated grains blended in an electric blender until the ingredients were mixed well. Then the solution was filtered by whatman filter paper 42 pore size and the volume was completed to 1 liter. The extract was sprayed on the plants directly after preparation. An aqueous extract was prepared from the quiescent barley grains (for the purpose of comparison and calculating the nutritional bioavailability). Table 2 shows the chemical and physical properties and conversion ratio of barley aqueous sprouted and grains extract.

Table 2. Physical and chemical characteristics of the aqueous extract of quiescent (Q), sprouted (S) barley grains and conversion ratio

character	Values Q	S	CR
pH	7	6.9	—
$EC_{1:1}$ (ds.m^{-1})	1.70	1.80	—
Total N (g L^{-1})	1.52	2.01	1.32
P (mg L^{-1})	219	232	1.05
K (mg L^{-1})	278	266	0.95
Ca (mg L^{-1})	29.5	39.1	1.32
Mg (mg L^{-1})	76.3	88.7	1.16
Fe (mg L^{-1})	2.50	6.00	2.40
Zn (mg L^{-1})	2.00	4.01	2.00
Gibberellin (µg L^{-1})	2	304	152

(Q) aqueous extract of quiescent barley grains
(S) aqueous extract of sprouted barley grains
(CR) conversion ratio: calculated by dividing Q/S for each nutrient

The Study traits were Determination of the following;

1- Anatomical (histological) traits: Anatomical traits were carried out on carrot seeds in Nematode Laboratory/Plant Protection Department/College of Agricultural Engineering Sciences. Carrot seeds were placed in water for two hours, and then longitudinal sections of the seeds were taken from the first third of the seeds (figure 1) under an optika electron microscope. Then dye it with a salt solution of 2,3,5-Triphenel Tetrazolium Chloride, commercially known as tetrazolium salt or TZ, at a concentration of 1 g. 100 ml^{-1} distilled water and leave it in the dark for two hours at room temperature for the reaction to complete (15), as the salt dyes. The living tissue in the seed (embryo) is colored red, making it easy to distinguish from the endosperm, which is very similar in color. It was then examined under an electron microscope using a graduated lens.

Figure 1. The longitudinal section of carrot seed from the first third of the seed

The collected data analyzed using analyses of variance and the means were compared according to L.S.D. test under 5% probability.

RESULTS AND DISCUSSION

1.Embryo and endosperm length (µm) of primary, secondary, tertiary umbels of carrot seeds: The results of the statistical analysis of the individual factors show the significant superiority of B_1 in producing the longest embryo in primary secondary, and tertiary umbels (1335.9, 1255.3, 704.3 µm) respectively. In compare with the shortest embryos which found in B_0 (Table 3A). Plants that received foliar trehalose application (75 mmol/l^{-1}) exhibited longer embryos in all

umbels orders than plant sprayed with distilled water (Table 3A). Spraying calcium (1 ml.l^{-1}) produced the longest embryo P compared to the shortest embryo in C_1 treatment (Table 3A). All sole factors exhibit non-significant effect on endosperm length.

Table 3A. embryo length (ML) and endosperm length (NL) (µm) of primary, secondary, tertiary umbels of carrot seeds after treatment for individual factors

treatments	Primary umbels		Secondary umbels		Tertiary umbels	
	ML	NL	ML	ML	ML	NL
			B			
B_0	1235.3	2203	1081.9	2239	654.8	2019
B_1	1335.9	2199	1255.3	2304	704.3	2021
LSD $_{0.05}$	32.25	N.S.	22.20	N.S.	7.97	N.S.
			T			
T_0	155.1	2263	1012.8	2335	597.3	2024
T_1	339.8	2177	1230.8	2246	705.1	2012
T_2	362.0	2163	1262.1	2234	736.2	2024
LSD $_{0.05}$	39.5	N.S.	27.19	N.S.	9.76	N.S.
			C			
C_0	1324.7	2179	1201.4	2264	686.2	2018
C_1	1457.3	2250	1354.9	2303	784.1	2034
C_2	1074.9	2174	949.9	2248	568.4	2008
LSD $_{0.05}$	39.5	N.S.	27.19	N.S.	9.76	N.S.

Table 3B. Table 3 A. embryo length (ML) and endosperm length (NL) (µm) of primary, secondary, tertiary umbels of carrot seeds after treatment for two ways interaction

treatments	Primary umbels		Secondary umbels		Tertiary umbels	
	ML	L	ML	ML	L	ML
			BXT			
B_0T_0	1046.9	2232	808.10	2268	582.8	2023
B_0T_1	1311.8	2208	1229.1	2233	682.9	2028
B_0T_2	1347.3	2168	1208.4	2217	698.7	2008
B_1T_0	1263.3	2294	1217.6	2402	611.9	2025
B_1T_1	1367.8	2146	1232.6	2258	727.2	1997
B_1T_2	1367.8	2158	1315.8	2251	773.8	2041
LSD $_{0.05}$	55.86	N.S.	38.45	N.S.	13.81	N.S.
			BXC			
B_0C_0	1316.1	2214	1100.4	2247	680.8	2024
B_0C_1	1395.7	2227	1266.6	2253	751.3	2041
B_0C_2	994.2	2167	878.70	2219	532.2	1994
B_1C_0	1333.3	2144	1302.4	2281	691.6	2013
B_1C_1	1518.9	2274	1443.3	2352	816.8	2027
B_1C_2	1155.6	2180	1020.1	2278	604.6	2022
LSD $_{0.05}$	55.86	N.S.	N.S.	N.S.	13.81	N.S.
			TXC			
T_0C_0	1157.2	2266	936.50	2348	553.3	2016
T_0C_1	1244.0	2272	1146.8	2287	726.7	2041
T_0C_2	1064.2	2250	955.20	2370	512.0	2015
T_1C_0	1339.8	2178	1317.8	2269	732.8	2039
T_1C_1	1531.5	2207	1414.0	2284	792.0	2003
T_1C_2	1148.0	2146	960.70	2184	590.3	1995
T_2C_0	1477.2	2092	1350.0	2174	772.3	2001
T_2C_1	1596.3	2272	1504.0	2337	833.5	2058
T_2C_2	1012.5	2125	932.30	2192	602.8	2014
LSD $_{0.05}$	96.76	N.S.	47.09	N.S.	16.91	N.S.

The results of Table (3 B) indicated that there were significant differences in second order interaction between the aqueous extract of barley sprouts and trihalose. (B_1T_2) produced the longest embryo for the seeds of primary, secondary, tertiary umbels (1367.8, 1315.8 and 773.8 µm) respectively. With reference to the interaction between aqueous germinated barley grains extract and calcium (Table 3B); B_1C_1 showed the longest embryo for the seeds of primary, secondary, tertiary umbels (1518.9, 1443.3, and 816.8 µm) respectively in compare with the shortest embryos that found in B_0C_2. The results of the second order interaction between calcium and trehalose (Table 3B) shows the consistent differences between treatments, T_2C_1 treatment increased embryo length in primary, secondary, tertiary umbels (1596.3, 1504, and 833.5 µm) respectively compared to the lowest percentages in T_0C_2. The results of 3B table also show that the endosperm length in all umbels order didn't reach to significant level.

Table 3 C. embryo length (ML) and endosperm length (NL) (µm) of primary, secondary, tertiary umbels of carrot seeds after treatment for three ways Interaction

traits	Primary umbels		Secondary umbels		Tertiary umbels	
treatments	ML	L	ML	NL	ML	NL
$B_0T_0C_0$	1047.7	2183	637.7	2258	561.3	2001
$B_0T_0C_1$	1124.6	2262	1025.7	2262	710.7	2075
$B_0T_0C_2$	968.30	2250	761.00	2283	476.3	1992
$B_0T_1C_0$	1346.3	2323	1343.7	2331	732.7	2092
$B_0T_1C_1$	1498.0	2183	1386.3	2211	751.0	2013
$B_0T_1C_2$	1091.0	2118	957.30	2159	565.0	1978
$B_0T_2C_0$	1554.3	2135	1320.0	2151	748.3	1977
$B_0T_2C_1$	1564.3	2235	1387.7	2287	792.3	2035
$B_0T_2C_2$	923.20	2133	917.70	2214	555.3	2012
$B_1T_0C_0$	1266.7	2348	1235.3	2437	545.3	2030
$B_1T_0C_1$	1363.3	2283	1268.0	2312	742.7	2007
$B_1T_0C_2$	1160.0	2250	1149.3	2457	547.7	2038
$B_1T_1C_0$	1333.3	2033	1292.0	2208	733.0	1985
$B_1T_1C_1$	1556.0	2230	1441.7	2358	833.0	1993
$B_1T_1C_2$	1205.0	2173	964.00	2208	615.7	2012
$B_1T_2C_0$	1400.0	2050	1380.0	2198	796.3	2025
$B_1T_2C_1$	1628.3	2308	1620.3	2386	874.7	2081
$B_1T_2C_2$	1101.7	2117	947.00	2170	650.3	2016
$LSD_{0.05}$	96.76	N.S.	66.59	N.S.	23.92	N.S.

Table 4A. Germination speed (G_P)(%) and germination percent (G_S) (%) of primary, secondary, tertiary umbels of carrot seeds after treatment for two ways interaction

Treatments	Primary umbels		Secondary umbels		Tertiary umbels	
	G_S	G_P	G_S	G_P	G_S	G_p
			B			
B_0	68.26	79.00	62.96	77.07	49.56	65.37
B_1	80.48	86.19	78.04	84.07	63.44	72.89
LSD $_{0.05}$	3.337	1.380	3.018	1.177	1.819	1.385
			T			
T_0	67.78	76.44	58.94	74.56	48.11	63.00
T_1	77.61	85.22	75.61	83.17	60.44	71.78
T_2	77.72	86.11	76.94	84.00	60.94	72.61
LSD $_{0.05}$	4.087	1.690	3.697	1.442	2.227	1.696
			C			
C_0	77.56	83.17	71.72	81.44	57.28	70.33
C_1	84.11	89.16	82.28	87.44	67.28	76.83
C_2	61.44	75.44	57.50	72.83	44.94	60.22
LSD $_{0.05}$	4.087	1.690	3.697	1.442	2.227	1.696

Results of table (3 C) reveal that there is a significant effect of third order interaction treatments in embryo length in different umbels order. $B_1T_2C_1$ treatment produced the longest embryos (1628.3, 1620.3, and 874.7 µm) respectively, compared to the lowest percentages in $B_0T_2C_2$. However; endosperm length didn't reach to significant level.

Germination speed (GP) (%) and germination percent (GS) (%) of primary, secondary, tertiary umbels:

The results of the statistical analysis of the individual factors show the significant superiority of B_1 in germination speed and germination percent of primary, secondary, tertiary umbels of carrot seeds (80.48, 86.19%); (78.04, 84.07%); (63.44, 72.89%) respectively compared to the lowest percentages in B_0. Application of T2 treatment reveals significant results in the mentioned traits (77.72, 86.11%); (76.94, 84 %); (60.94, 72.61%) respectively over T_0, spraying calcium (1 ml.l^{-1}) (C_1) was effective in increasing germination speed and percent (84.11, 89.16%); (82.28, 87.44 %); (67.28, 76.83%) respectively compared to the lowest percentages in C_2 (Table 4A).

Table 4B. Table 3 A. Germination speed (G_P)(%) and germination percent (G_S) (%) of primary, secondary, tertiary umbels of carrot seeds after treatment for two ways interaction

treatments	Primary umbels		Secondary umbels		Tertiary umbels	
	G_S	G_P	G_S	G_p	G_s	G_p
			BXT			
B_0T_0	59.11	71.56	44.78	69.89	37.33	59.00
B_0T_1	74.12	82.11	73.78	80.11	55.78	68.22
B_0T_2	71.56	83.33	70.33	81.22	55.56	68.89
B_1T_0	76.44	81.34	73.11	79.22	58.89	67.00
B_1T_1	81.11	88.33	80.11	86.22	65.11	75.33
B_1T_2	83.89	88.89	80.89	86.78	66.33	76.33
$LSD_{0.05}$	5.78	2.389	5.23	2.04	3.15	N.S.
			BXC			
B_0C_0	73.67	79.33	64.78	78.00	50.78	67.56
B_0C_1	80.78	85.78	78.00	83.89	62.89	72.33
B_0C_2	50.33	71.89	46.11	69.33	35.00	56.22
B_1C_0	81.44	87.00	78.67	84.89	63.78	73.11
B_1C_1	87.43	92.56	86.56	91.00	71.67	81.33
B_1C_2	72.56	79.00	68.89	76.33	54.89	64.22
$LSD_{0.05}$	5.78	N.S.	5.23	N.S.	3.15	N.S.
			TXC			
T_0C_0	71.17	75.17	52.67	72.17	43.00	60.83
T_0C_1	74.83	83.18	71.67	81.83	58.50	71.33
T_0C_2	57.33	71.00	52.50	69.67	42.83	56.83
T_1C_0	75.50	86.17	78.33	85.00	60.67	73.83
T_1C_1	88.17	91.50	87.33	90.00	70.33	79.33
T_1C_2	69.17	78.00	65.50	74.50	50.33	62.17
T_2C_0	86.00	88.17	84.17	87.17	68.17	76.33
T_2C_1	89.33	92.83	88.17	90.50	73.00	79.83
T_2C_2	57.83	77.33	54.50	74.33	43.67	61.67
$LSD_{0.05}$	7.08	2.927	6.40	2.50	3.86	2.98

The results of Table (4 B) indicate that there were significant differences in second order interaction between the aqueous extract of barley sprouts and trihalose. (B_1T_2) produced the highest percentages of GP and GS for the seeds of primary (83.89, 88.89%), secondary (80.89, 86.78%), tertiary umbels (G_S only) (66.33,) respectively. With reference to the interaction between aqueous germinated barley grains extract and calcium (Table 4B); B_1C_1 shows the highest percent of GP and GS for the seeds of primary (83.89, 88.89%), secondary (80.89, 86.78%), tertiary umbels (G_S only) (66.33) respectively. The results of the second order interaction between calcium and trehalose (Table 4B) shows the consistent differences between treatments, T_2C_1 treatment increased GP and GS for the seeds of primary (89.33, 92.83%), secondary (88.17, 90.5%), tertiary umbels (73, 79.83%) respectively compared to the lowest percentages in T_0C_2.

Table 4 C. Germination speed (G_P) (%) and germination percent (G_S) (%) of primary, secondary, tertiary umbels of carrot seeds after treatment for three ways interaction

traits	Primary umbels		Secondary umbels		Tertiary umbels	
treatments	G_S	G_P	G_S	G_P	G_S	G_P
$B_0T_0C_0$	60.67	69.00	35.67	67.67	30.00	58.67
$B_0T_0C_1$	71.00	81.00	65.00	79.00	53.33	68.68
$B_0T_0C_2$	40.67	64.67	33.67	63.00	28.67	49.67
$B_0T_1C_0$	71.00	83.33	75.00	82.00	54.67	70.00
$B_0T_1C_1$	84.67	87.67	83.00	86.00	65.00	75.00
$B_0T_1C_2$	67.67	75.33	63.00	72.00	47.67	59.67
$B_0T_2C_0$	85.33	85.67	83.67	84.33	67.67	74.00
$B_0T_2C_1$	86.67	88.68	85.67	86.33	70.33	73.33
$B_0T_2C_2$	42.68	75.65	41.67	73.00	28.67	59.33
$B_1T_0C_0$	76.67	81.33	69.67	76.67	56.00	63.00
$B_1T_0C_1$	78.66	85.34	78.33	84.67	63.67	74.00
$B_1T_0C_2$	68.00	77.33	69.00	76.33	57.00	64.00
$B_1T_1C_0$	81.00	89.00	81.67	88.00	66.67	77.67
$B_1T_1C_1$	91.67	95.33	90.67	93.67	75.67	83.67
$B_1T_1C_2$	70.66	80.67	68.00	77.00	53.00	64.66
$B_1T_2C_0$	86.68	90.68	84.67	90.00	68.67	78.67
$B_1T_2C_1$	92.00	97.00	90.67	94.67	75.68	86.33
$B_1T_2C_2$	73.00	79.00	67.33	75.68	54.67	64.00
LSD $_{0.05}$	10.01	4.14	9.06	3.531	5.46	4.154

It is evident from Table (3 C) that there is a significant effect of the third order interaction treatments in the percentages of GP and GS for the seeds of all umbels orders. $B_1T_2C_1$ treatment produced the highest percentages **(**92, 97%); (90.67, 94.67%); (75.68, 86.33%) respectively, compared to the lowest percentages in $B_0T_0C_2$.

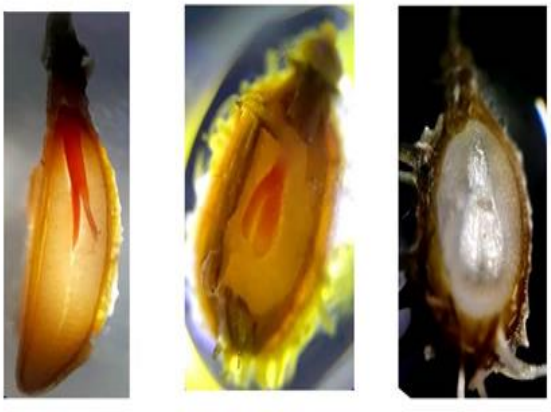

Figure 2. variable embryos length in second order umbels as affected by study treatments

Exogenous application of sprouted barley aqueous extract, trehalose, and calcium was efficiently utilized by increasing embryo length. The elevating impact of barley sprouts aqueous extract, trehalose, and somehow calcium, and in the process of embryogenesis and fruit development, which led to the growth of the embryo and its increase in its length compared to the endosperm, achieving several treatments first- and second-order, the required maturity rate (length of the embryo = two-thirds of the length of the endosperm), and overcoming the problem of ruidementary or immature embryos (figure 2) (25). and then obtain regular germination and emergence, as several studies have confirmed the work of the trehalose precursor in embryos maturation in the Arabidopsis plant through its contribution to completing the embryogenesis stages of the process to the fullest extent (9), therefore Trehalose metabolism is important in regulating food reserves, as T_6P is essential for utilization of carbohydrates (22). Moreover; Calcium regulates the growth of developing fruit in carrots after fertilization) due to its direct action in forming cell walls and maintaining the structure and permeability of cell membranes. These results agree with (18, 19). the effect of sprouted barley aqueous extract is came from the bioavailable nutrients that had (Table 2), which had a role in fruit development and seeds, such as nitrogen, phosphorus, and potassium (7, 14), as well as

the effective action of gibberellin in regulating the secretion of starch-degrading enzymes in the endosperm and supplying the embryo with nutrition (24).

REFERENCES

1. Ali, N. S., H. S. Rahi, and A. A. Shackir, 2014. Soil Fertitity. Arabic Community Publisher for Publication and Distribution 1st ed , pp: 307.

2. Ali, Q., M. Ashraf, and F. Anwar. 2012. Trehalose-induced changes in seed oil composition and antioxidant potential of maize grown under drought stress. J Am Oil Chem Soc 89:1485–1493.

3. Al-Khafaji, A. M. H. H. and K. D. H. Al-Jubouri. 2022. Enhancing growth and production of carrot plant by spraying aqueous barley sprouts extract, trehalose, and calcium. Journal of Kerbala for Agricultural Sciences, 9(4): 134-144. https://doi.org/10.59658/jkas.v9i4.1069

4. Al-Khafaji, A. M. H. H. and K. D. H. Al-Jubouri. 2022. Influence of aqueous extract of barley sprouts, trehalose, and calcium on growth, quality and yield of carrot. Iraqi Journal of Agricultural Sciences,53(1):133-140. https://doi.org/10.36103/ijas.v53i1.1517.

5. Al-Khafaji, A. M. H. H. and K. D. H. Al-Jubouri. 2022. Maximization carrot minerals preserve and antioxidant capacity by foliar application of aqueous barley sprouts extract, trehalose, and calcium. Iraqi Journal of Agricultural Sciences, 53(1):122-132. https://doi.org/10.36103/ijas.v53i1.1515.

6. Al-Rubaie; A. H. S. and K. D. H. Al-Jubouri. 2023. Effect of tocopherol, trehalose and soil improvement in water productivity and industrial potatoes under water stress. Iraqi Journal of Agricultural Sciences, 54(4):979-995. https://doi.org/10.36103/ijas.v54i4.1787

7. Bhatla, S. C., and M. A.Lal. 2019. Plant Physiology, Development and Metabolism. Springer Nature, Library of Congress, pp: 1237.

8. Burek, M, S. Waskiewicz, L. Wandzik, and K. Kaminska. 2015. Trehalose – properties, biosynthesis and applications. Chemik, 69(8): 469-476.

10. Eastmond P J, A. J. van Dijken, M. Spielman, A. Kerr, and A. F. Tissier. 2002. Trehalose-6-phosphate synthase 1, which catalyses the first step in trehalose synthesis, is essential for Arabidopsis embryo maturation. Plant J. 29:225–35.

11. El Habbasha S.F., and F. M. Ibrahim, 2015. Calcium: physiological function, deficiency and absorption. International Journal of Chem. Tech Research, 8(12): 196-202.

12. Feofilova E. P., A. I. Usov, I. S. Mysyakina, and G. A. Kochkina. 2014. Trehalose: chemical structure, biological functions, and practical application. Microbiology 2014, 83, 184–194. https://doi.org/10.1134/S0026261714020064

13. Ge, L., H. Tian, and S. Russell. 2007. Calcium function and distribution during fertilization in angiosperms. American Journal of Botany. 94(6): 1046–1060.

14. Iturriaga, G., R. Suárez, and B. N. Franco. 2009. Trehalose metabolism: from osmoprotection to signaling. Int. J. Mol. Sci., 10, 3793-3810.

15. Kennedy, E. L. T., A. K. Jacobs, and S. J. Roy. 2015. The role of the CBL–CIPK calcium signalling network in regulating ion transport in response to abiotic stress. Plant Growth Regul , 76, 3–12. https://doi.org/10.1007/s10725-015-0034-1

16. Lima, C. B., A. C. Boaventura, and T. T. Villela. 2018 Comparing procedures for performing tetrazolium test on carrot seeds. Horticultura Brasileira 36:240-245.

17. Lin, Y., J. Zhang, W. Gao, Y. Chen, H. Li, D. W. Lawlor, M. J. Paul, and W. Pan. 2017. Exogenous trehalose improves growth under limiting nitrogen through upregulation of nitrogen metabolism. BMC Plant Biology, 17(247): 1-13. https://doi.org/10.1186/s12870-017-1207-z

18. Marton, M., and Z. Mandoki. 2010. The role of sprouts in human nutrition a review. Acta Univ. Sapientiae, Alimentaria, (3) 81-117.

19. Mohammed, R. R. and B. H. Majeed. 2023. Effect of moringa leaves extract, calcium, and potassium silicate on the growth, yield, and quality of strawberry fruits. Iraqi Journal of Agricultural Sciences,54(6):1703-1715. https://doi.org/10.36103/ijas.v54i6.1869.

20. Mohammed, R. R. and B. H., Majeed, 2024. Response of strawberry growth, yield and marketable fruit quality to spraying with

moringa leaf extract, calcium and potassium silicate. Iraqi Journal of Agricultural Sciences, 55(1):440-452.
https://doi.org/10.36103/yf9f0c65
21. Palumbo, F., A. Vannozzi, and G. Barcaccia, 2021. Impact of genomic and transcriptomic resources on apiaceae crop breeding strategies. International Journal of Molecular Sciences, 22(18), 9713. https://doi.org/10.3390/ijms22189713
22. Rubatzky, V. E., C. F. Quiros and P. W. Simon. 1999. Carrots and Related Vegetables Umbelliferae. CABI Publishing. pp:294.
23. Schluepmann H, T. Pellny, A. van Dijken, S. Smeekens, and M. Paul.2003. Trehalose 6-phosphate is indispensable for carbohydrate utilization and growth in Arabidopsis thaliana. Proc. Natl. Acad. Sci. USA 100:6849–54.
24. Simon, P., M. Lorizzo, D. Grzebelus, and R. Baranski. 2019. The Carrot Genome. Springer Nature Switzerland AG 1st ed. pp: 372.
25. Taiz, L., E. Zeiger, I. M. Moller, and A. Murphy. 2014. Plant Physiology and Development. 6th ed, Sinauer Assotiates, Inc., Publishers Sunderland, Massachusetts. pp, 761.
26. Tanja M. H., J. R. Hewitt and H. Nonogaki. 2003. Endo-β-mannanase activity is associated with the completion of embryogenesis in imbibed carrot (*Daucus carota* L.) seeds. Seed Science Research, 13, pp 219-227.