MAXIMIZATION CARROT MINERALS PRESERVE AND ANTIOXIDANT CAPACITY BY FOLIAR APPLICATION OF AQUEOUS BARLEY SPROUTS EXTRACT, TREHALOSE, AND CALCIUM

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ABSTRACT
This research was implemented at vegetable field of the College of Agricultural Engineering Sciences - University of Baghdad during fall seasons, 2018 and 2019. The experiment was conducted using factorial within Randomized Complete Block Design arrangement with three factors and replicates (3X3X2). The aqueous barley sprouts extract (B0, B1) (0, 100 g.L^{-1}) represented the first factor. Trehalose (T0, T1, T2) (0, 50, 75 mmole.L^{-1}) represented the second factor. Calcium (C0, C1, C2) (0, 1, 2 ml.L^{-1}) represented the third factor. The research objectives are assessing the impact of the mentioned factors and their interactions on carrot plant minerals and antioxidant accumulation. Results showed the effectiveness of three ways interaction treatment B1T2C1 in producing significant increases in phosphorus (0.302, 0.311%) total carotenoids (15.83, 15.93 mg.100^{-1}g), β carotene (7.9, 7.967 mg.100^{-1}g) and DPPH (97.94, 98.41%) in roots for both seasons respectively.in compare with control treatment B0T0C0.

Keywords: sugars; total carotenoids; β carotene; DPPH; phosphorus; core/cortex Proportion
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تعاظم مخزون العناصر ومضادات الاكسدة في الجزر بالرش بالمستخلص المائي لمستنبتات الشعير والتريهالوز والكالسيوم

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المستخلص
نفذت تجربة حقلية في حقول كلية علوم الهندسة الزراعية/جامعة بغداد للموسمين خريفي 2018 وخريفي 2019. طُبقت التجربة باستعمال تصميم القطاعات الكاملة المعشاة حسب ترتيب التجارب العاملية وبثلاث عوامل وثلاث مكررات (2X3X3)، مَثَّل الرش بالمستخلص المائي للحبوب المستنبتة لنبات الشعير (0، 100 غم حبوب جافة.لتر$^{-1}$) العامل الاول والذي رُمز له (B0 وB1)، اما العامل الثاني فتضمن الرش بسكر التريهالوز (0، 50 ، 75 ملي مول.لتر$^{-1}$) والذي رمز له (T_0 وT_1وT_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_1وC_2)، تتمثل أهداف البحث في اختبار تأثير العوامل المذكورة وتداخلاتها في تراكم العناصر ومضادات الأكسدة لنبات الجزر. اظهرت النتائج التفوق المعنوي لمعاملة التداخل الثلاثي B1T2C1 في زيادة النسبة المئوية للفسفور (0.302، 0.311%) والكاروتينات الكلية (15.83، 15.93 ملغم . 100غم$^{-1}$) والبيتا كاروتين (7.9، 7.967 ملغم . 100غم$^{-1}$) والنسبة المئوية لكسح جذر DPPH (97.94، 98.41%) في الجذور مقارنة بمعاملة القياس B0T0C0.

الكلمات المفتاحية: سكريات، كاروتينات كلية, بيتا كاروتين, DPPH, فسفور، نسبة الخشب الى اللحاء
مستل من اطروحة دكتوراه للباحث الاول

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INTRODUCTION

Health food awareness has risen in recent years. That resulted from aggravation of health problems such as diabetes, heart disease, high blood pressure, cancers, autoimmune, and gastrointestinal diseases since of applying wrong diets for several decades (18). That led to the concept of therapeutic nutrition that emphasizes the daily consumption of vegetable crops, especially those of higher antioxidant content, which are preferred by the consumer so they could be included in the daily diet of the individuals (4; 14; 15). Carrot that belongs to Apiaceae one of the most important vegetables that shifted people perception about healthy food as does not taste good. Unlike most of root vegetables and spices, carrots combine the sweet favorable flavor, crunchy texture, and vibrant color with huge health benefits. They classified as the maximum 10 produced vegetables, which there're intake have maximized 3 folds for the last 10 years in compare to the 20th century statistics (11). Notably, carrots occupied the second most consumed vegetable after potato (30). In a comparison study based on nutrients density score done by preventing chronic disease center (9); carrots preceded radish, tomato, turnip, leek and sweet potato according to their distinctive nutritional profile such as having good amounts of Carotenoids, which considered the most prominent characteristic of carrot, from which its name derived. Carotenoids are fat-soluble phytonutrients that have anti-oxidant, anti-mutagenic and anti-cancer impact. As a result, they help in cells homeostasis. Since this effect works for one time, it is recommended to eat carrots in a regular basis. In addition to that, carrots have sufficient amount of electrolytes, B vitamins, and fibers (26). Thus, production of carrots has become a fertile ground for scientific research to keep pace with the latest developments in authentic medicinal centers as well as people demands for carrots in the global market. Using sprouted seeds has elevated interest in the last 20 years. Because sprouting considerably increases nutrients bioavailability, i.e. breaking down proteins to amino acids, starch to sugars, and fats to fatty acids (23). Moreover, they have sharp increase in gibberellins and rapid decrease in ABA (31). Hence, their aqueous extracts could be sprayed on plant. Trehalose is non-reduced disaccharide that forms from combining two glucose molecules (17). It has multi tasks in plant such as, metabolizing carbon, protecting plant from abiotic stress (7) and signaling (6; 30). In addition, it has proven that trehalose is anti-oxidant molecule that eradicates free radicals (12). Shafiq et al (29) demonstrated that spraying trehalose (50 mmole.L^{-1}) increased the anti-oxidant capacity under drought conditions of radish. Aldesuquy and Ghanem (1) noticed that foliar fertilizing with trehalose (1.5 mmole) on wheat significantly support the enzymatic and non-enzymatic defense system. Calcium is a structural element since it a component in cell walls. Moreover, it stabilizes plasma membrane, regulates osmotic relationships, and responds to the signals as a second messenger for many cellular and hormonal cues (6; 31). Lee et al (21) observed that carrot plant foliar feeding by calcium nitrate 2% improved storage characteristics. Hussein (16) showed that spraying a fertilizer having 5% CaO (6 ml.L^{-1}) on cauliflower plant increased the total yield. Saaseea and Al-a'amry (27) reported that foliar spraying by calcium (1000 mg.L^{-1}) remarkably enhanced tuber weight of potato plant. Consequently, we need to uncover the role of barley aqueous sprouted Grains, trehalose, and Calcium on P, Ca tissue preserve, and antioxidant potency in carrot plant.

MATERIALS AND METHODS

This experiment was conducted during two fall seasons (2018 and 2019) 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 both seasons. The seeds of carrot were sowed on lines on terraces in 15/September for both seasons. The field was under drip irrigation system. 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 planting (2). Thinning carried out after 30 days from planting seeds for both seasons. The spacing between one plant and another for both seasons was 0.05 m. The seeds of carrot var. Nantes were purchased from Modesto

Seed Company (California, U.S.A.) for the first season. The seeds of the fall season 2019 produced locally in the spring season 2019 from the stecklings of the fall season 2018. The seeds were planted in a plant density 1,000,000 plants.ha^{-1}. The entire plots harvested after 115 days of the planting day for the1st season and 85 days for the 2nd season. The experiment was implemented as factorial arrangement (2X3X3) within randomized complete block design 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 first spraying was after 10 days from thinning. The second spraying was after 15 days from the first spraying. The third spraying was after 15 days from the second spraying. 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. The Study traits were Determination of the following; calcium in leaves and root (%) (28), phosphorus in leaves and root (%) (25), carotenoid in roots (mg.100^{-1}g FW) (13), beta-carotene in roots (mg.100^{-1}g FW) (24), DPPH radical assay (%) in roots (19), Core/cortex proportion (%) in roots. The collected data analyzed using analyses of variance and the means were compared according to L.S.D. test under 5% probability.

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

character	Values	
	Fall 2018	Fall 2019
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	

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 ($\mu g\ L^{-1}$)	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

RESULTS AND DISCUSSIONS

Tissue nutrients status

It is evident from Table 3 A that there is a significant effects of the three ways interaction treatments in the percentages of P and Ca in leaves and roots of carrot for both seasons. B1T2C1 treatment produced the highest percentage of phosphorus in leaves (0.423, 0.436%) and roots (0.302, 0.311%) for both seasons respectively, compared to the lowest percentages in B0T0C2. Also, table 4 A showed that B1T2C2 significantly influenced the percentage of calcium in leaves (2.932, 2.933%) and roots (1.298, 1.311%) for both seasons respectively, compared to the lowest percentages produced by B0T0C0 for both seasons.The results of Table 3 B indicated that there were significant differences in two ways interaction between the aqueous extract of barley sprouts and trihalose. (B1T2) produced the highest percentage of phosphorus in leaves (0.347, 0.362%) and roots (0.248, 0.258%) for both seasons respectively. However, B1T2 increased Calcium percent in roots in season 2018 only (1.361%). With reference to the interaction between aqueous germinated barley grains extract and calcium (Table 3B); B1C1 showed the highest percent of P in both tested tissues (leaves, roots) for both seasons than the control plants. B1C2 produced the highest percentage of calcium in carrot roots than the control plants for fall 2018 season only (1.463%). The results of the two ways interaction between calcium and trehalose (Table 3B) showed the consistent differences between treatments, T2C1 treatment increased P percentage in leaves (0.343, 0.356%) and roots (0.245, 0.253%) for both seasons respectively compared to the lowest percentages in T0C0.

Table 3 A. Leaf and root tissues P, and Ca (%) of carrot plant after treatment for three ways interaction of fall 2018 and fall 2019 seasons

traits	P (leaves)		P (roots)		Ca (leaves)		Ca (roots)	
treatments	2018	2019	2018	2019	2018	2019	2018	2019
B0T0C0	0.205	0.212	0.137	0.141	2.033	2.004	1.003	1.000
B0T0C1	0.209	0.215	0.139	0.143	2.367	2.383	1.171	1.158
B0T0C2	0.205	0.211	0.136	0.140	2.467	2.390	1.400	1.406
B0T1C0	0.215	0.229	0.143	0.152	2.310	2.295	1.152	1.200
B0T1C1	0.236	0.249	0.168	0.178	2.667	2.717	1.232	1.226
B0T1C2	0.236	0.243	0.168	0.173	2.667	2.797	1.409	1.394
B0T2C0	0.223	0.253	0.148	0.168	2.267	2.300	1.185	1.183
B0T2C1	0.263	0.276	0.187	0.197	2.687	2.757	1.268	1.265
B0T2C2	0.236	0.250	0.168	0.178	2.700	2.793	1.421	1.422
B1T0C0	0.216	0.236	0.143	0.157	2.233	2.215	1.140	1.178
B1T0C1	0.223	0.236	0.148	0.157	2.433	2.430	1.217	1.223
B1T0C2	0.226	0.240	0.150	0.157	2.500	2.502	1.410	1.410
B1T1C0	0.271	0.278	0.180	0.185	2.367	2.333	1.134	1.256
B1T1C1	0.370	0.383	0.263	0.273	2.867	2.866	1.361	1.355
B1T1C2	0.326	0.350	0.232	0.249	2.933	2.934	1.479	1.479
B1T2C0	0.256	0.270	0.183	0.192	2.333	2.370	1.286	1.287
B1T2C1	0.423	0.436	0.302	0.311	2.932	2.933	1.298	1.311
B1T2C2	0.363	0.380	0.259	0.271	2.933	3.033	1.499	1.493
LSD 0.05	0.034	0.037	0.023	0.026	0.135	0.131	0.041	0.070

The percentage of calcium increased in elevated concentration of sprayed materials. The highest percentage of calcium in leave and roots tissues was found in T2C2 (2.817, 2.913%) and roots (1.460, 1.457%) treatment compared to the lowest percentage in T0C0 treatment. The results of the statistical analysis of the individual factors showed the significant superiority of B1 in p and Ca percentages in leaves and roots (Table 3C) for both seasons compared to the lowest percentages in B0. Plants that received foliar trehalose application (75 mmol/l^{-1}) showed higher p and Ca percentages in leaves and root tissues for both seasons, while the lowest percentages were found in T0, spraying calcium (1 ml.l^{-1}) was produced the highest P percent in leaves for both seasons, compared to the lowest rates in C0 treatment. C2 produced the highest percentage of calcium in leaves and roots tissues for both seasons, compared to the lowest percentages in C0.

Table 3 B. Leaf and root tissues P, and Ca (%) of carrot plant after treatment for two ways interaction of fall 2018 and fall 2019 seasons

treatments	P (leaves)		P (roots)		Ca (leaves)		Ca (roots)	
	2018	2019	2018	2019	2018	2019	2018	2019
				TXB				
B0T0	0.206	0.213	0.137	0.141	2.289	2.259	1.191	1.188
B0T1	0.229	0.240	0.160	0.167	2.548	2.603	1.258	1.273
B0T2	0.241	0.260	0.168	0.181	2.551	2.640	1.291	1.290
B1T0	0.221	0.237	0.147	0.158	2.389	2.383	1.256	1.270
B1T1	0.322	0.337	0.225	0.235	2.722	2.711	1.363	1.364
B1T2	0.347	0.362	0.248	0.258	2.733	2.756	1.361	1.363
LSD 0.05	0.019	0.021	0.013	0.015	N.S.	N.S.	0.023	N.S.
				CXB				
B0C0	0.214	0.231	0.143	0.154	2.203	2.223	1.107	1.128
B0C1	0.236	0.247	0.165	0.172	2.596	2.619	1.224	1.216
B0C2	0.226	0.235	0.157	0.164	2.611	2.660	1.410	1.407
B1C0	0.248	0.261	0.169	0.178	2.311	2.283	1.226	1.240
B1C1	0.338	0.352	0.238	0.247	2.744	2.744	1.292	1.296
B1C2	0.305	0.323	0.214	0.226	2.789	2.822	1.463	1.461
LSD 0.05	0.019	0.021	0.013	0.015	N.S.	N.S.	0.023	N.S.
				CXT				
T0C0	0.211	0.224	0.140	0.149	2.133	2.110	1.072	1.089
T0C1	0.216	0.226	0.143	0.150	2.400	2.408	1.194	1.190
T0C2	0.215	0.225	0.143	0.150	2.483	2.445	1.405	1.408
T1C0	0.243	0.253	0.162	0.168	2.338	2.314	1.192	1.228
T1C1	0.303	0.316	0.216	0.225	2.767	2.792	1.296	1.291
T1C2	0.281	0.296	0.200	0.211	2.800	2.865	1.444	1.436
T2C0	0.240	0.261	0.165	0.180	2.300	2.335	1.235	1.235
T2C1	0.343	0.356	0.245	0.253	2.810	2.845	1.283	1.288
T2C2	0.300	0.315	0.214	0.224	2.817	2.913	1.460	1.457
LSD 0.05	0.024	0.026	0.016	0.018	0.095	0.092	0.029	0.041

Table 3 C. Leaf and root tissues P, and Ca (%) of carrot plant after treatment for individual factors of fall 2018 and fall 2019 Seasons

treatments	P (leaves)		P (roots)		Ca (leaves)		Ca (roots)	
	2018	2019	2018	2019	2018	2019	2018	2019
				B				
B0	0.225	0.238	0.163	0.155	2.463	2.501	1.247	1.252
B1	0.297	0.312	0.217	0.207	2.615	2.617	1.327	1.318
LSD 0.05	0.011	0.012	0.008	0.008	0.045	0.043	0.013	0.023
				T				
T0	0.214	0.225	0.149	0.142	2.339	2.321	1.223	1.229
T1	0.276	0.288	0.201	0.192	2.635	2.657	1.311	1.318
T2	0.294	0.311	0.219	0.208	2.642	2.698	1.326	1.327
LSD 0.05	0.014	0.015	0.010	0.009	0.055	0.053	0.016	0.028
				C				
C0	0.231	0.246	0.166	0.156	2.257	2.253	1.166	1.184
C1	0.287	0.299	0.210	0.201	2.659	2.682	1.258	1.256
C2	0.265	0.279	0.195	0.186	2.700	2.741	1.436	1.434
LSD 0.05	0.014	0.015	0.010	0.009	0.055	0.053	0.016	0.028

Antioxidant traits performance in roots (carrots): The experimented seasons shared the significant increases in carotenoid content, Beta carotene, and DPPH. The most increases was observed in B1T2C1 for both seasons (15.83, 15.93 mg.100^{-1}g), (7.9, 7.967 mg.100^{-1}g) (97.94, 98.41%) (Table 4A) respectively. Two ways interaction between T2 and B1 tended to consistently promoting antioxidant performance in both seasons over the control (13.14, 13.23 mg.100^{-1}g), (6.484, 6.556 mg.100^{-1}g) (81.24, 81.60%) (Table 4A) respectively (Table 4B). The results in the same table also showed the superiority of B1C1 by producing the highest concentration of total carotenoids (12.89, 13.16 mg.100^{-1}g), beta-carotene (6.294, 6.461 mg.100^{-1}g) and reduced DPPH (78.56, 78.90%) in the roots for both seasons compared to the lowest numbers that found in B1C2 for both seasons. Exogenous application of calcium and trehalose significantly enhanced antioxidant traits (Table 4b). T2C1 showed the highest numbers in the mentioned traits (13.65, 13.73 mg.100^{-1}g), (6.717, 6.867 mg.100^{-1}g) (85.34, 85.64%) for both seasons respectively, compared to the lowest numbers that were in T0C2 for both seasons respectively (Table 4B). Statistical analysis of the individual factors (Table 4C) reveals the significant results of B1 treated plants by producing the highest concentration of total carotenoids, beta-carotene and reduced DPPH in roots over B0 for both seasons. Application of (T2) had a significant effect of total carotenoids, beta-carotene and reduced DPPH in the roots for both seasons over T0 for both seasons (Table 4C). As for calcium factor, C1-treated plants achieved the highest concentration of in the mentioned traits for both seasons compared to the lowest numbers in C2 for both seasons (Table 4C).

Table 4A. Total carotenoid, beta carotene (mg.100^{-1}g FW), reduced DPPH (%),and core/cortex (%) of carrot plant after treatment for three ways interaction of fall 2018 and fall 2019 seasons

treatments	total carotenoid		beta carotene		Reduced DPPH		core/cortex	
	2018	2019	2018	2019	2018	2019	2018	2019
B0T0C0	9.603	9.663	4.033	4.137	54.42	54.86	32.73	31.64
B0T0C1	9.597	9.707	4.833	4.783	59.16	59.39	37.16	36.34
B0T0C2	9.067	9.000	4.267	4.283	52.11	52.25	37.34	36.45
B0T1C0	10.67	10.96	5.200	5.317	64.05	64.18	29.79	28.94
B0T1C1	9.967	10.20	5.100	5.233	65.44	66.33	33.58	32.3
B0T1C2	9.500	10.66	4.633	4.533	58.78	57.33	36.37	35.62
B0T2C0	12.00	12.26	5.867	5.933	71.48	72.01	27.03	25.45
B0T2C1	11.46	11.53	5.333	5.517	72.73	72.87	32.31	31.43
B0T2C2	10.00	9.93	4.833	4.733	60.03	60.63	36.07	35.08
B1T0C0	10.33	10.73	5.050	5.217	66.41	66.48	31.24	30.17
B1T0C1	10.16	10.43	4.933	5.050	60.04	60.30	34.21	33.23
B1T0C2	9.530	9.367	4.517	4.517	59.07	59.94	40.87	40.03
B1T1C0	13.07	13.73	6.350	6.267	81.93	82.56	26.59	25.78
B1T1C1	12.66	13.13	6.050	6.367	77.71	77.96	32.09	31.04
B1T1C2	9.600	9.600	4.467	4.483	60.46	60.52	47.13	46.25
B1T2C0	13.60	13.70	6.617	6.783	82.56	82.43	28.76	27.32
B1T2C1	15.83	15.93	7.900	7.967	97.94	98.41	28.62	26.54
B1T2C2	10.00	10.07	4.935	4.917	63.22	63.95	49.07	47.36
LSD 0.05	0.98	0.93	0.485	0.498	6.02	6.40	N.S.	N.S.

Core/cortex proportion

Results reveal that core/cortex proportion significantly affected by elevated concentration of calcium. In fact, it observed from individual factors that C2 treated plants had high core/cortex proportion (41.14, 40.13%) over C1 and C0. Additionally, two ways interaction with C2 exclusively increased the mentioned proportion (Tables 4B and 4C). However, three ways interaction did not show any significant differences.

Table 4 B. Total carotenoid, beta carotene (mg.100^{-1}g FW), reduced DPPH (%), and core/cortex (%) of carrot plant after treatment for two ways interaction of fall 2018 and fall 2019 seasons

treatments	total carotenoid		beta carotene		Reduced DPPH		core/cortex	
	2018	2019	2018	2019	2018	2019	2018	2019
				TXB				
B0T0	9.422	9.447	4.378	4.401	55.23	55.50	35.74	34.81
B0T1	10.01	10.27	4.978	5.028	62.72	62.62	33.25	32.29
B0T2	11.15	11.24	5.411	5.478	68.08	68.33	31.80	30.66
B1T0	10.01	10.18	4.833	4.928	61.84	62.24	35.44	34.47
B1T1	11.78	12.15	5.622	5.706	73.36	73.69	35.27	34.35
B1T2	13.14	13.23	6.484	6.556	81.24	81.60	35.48	33.74
LSD 0.05	0.57	0.54	0.279	0.287	3.48	3.70	N.S.	N.S.
				CXB				
B0C0	10.76	10.45	5.033	5.129	63.32	63.51	29.85	28.68
B0C1	10.34	10.98	5.156	5.261	65.78	66.20	34.35	33.36
B0C2	9.522	9.533	4.578	4.517	56.97	56.74	36.59	35.72
B1C0	12.33	12.72	6.006	6.089	76.97	77.16	28.86	27.75
B1C1	12.89	13.16	6.294	6.461	78.56	78.90	31.64	30.27
B1C2	9.711	9.681	4.639	4.639	60.92	61.47	45.69	44.54
LSD 0.05	0.57	0.54	0.279	0.287	3.48	3.70	3.87	3.951
				CXT				
T0C0	9.970	10.22	4.542	4.677	60.42	60.67	31.98	30.90
T0C1	9.882	10.03	4.883	4.917	59.60	59.85	35.68	34.79
T0C2	9.298	9.183	4.392	4.400	55.59	56.09	39.10	38.24
T1C0	11.87	12.35	5.775	5.792	72.99	73.37	28.19	27.36
T1C1	11.31	11.66	5.575	5.800	71.57	72.15	32.84	31.67
T1C2	9.550	9.633	4.550	4.508	59.62	58.93	41.75	40.93
T2C0	12.80	12.98	6.242	6.358	77.02	76.95	27.89	26.39
T2C1	13.65	13.73	6.717	6.867	85.34	85.64	30.47	27.99
T2C2	10.00	10.00	4.884	4.825	61.62	62.29	42.57	37.22
LSD 0.05	0.70	0.66	0.342	0.352	4.26	4.53	4.735	4.84

Table 4 C. Total carotenoid, beta carotene (mg.100^{-1}g FW), reduced DPPH (%), and core/cortex (%) of carrot plant after treatment for individual factors of fall 2018 and fall 2019 seasons

treatments	total carotenoid		beta carotene		Reduced DPPH		core/cortex	
	2018	2019	2018	2019	2018	2019	2018	2019
				B				
B0	10.20	10.32	4.922	4.969	62.02	62.15	33.60	32.58
B1	11.64	11.85	5.646	5.730	72.15	72.51	35.40	34.19
LSD 0.05	0.33	0.31	0.161	0.166	2.01	2.131	N.S.	N.S.
				T				
T0	9.717	9.813	4.606	4.664	58.54	58.87	35.59	34.64
T1	10.91	11.21	5.300	5.367	68.06	68.15	34.26	33.32
T2	12.15	12.24	5.947	6.017	74.66	74.69	33.64	32.20
LSD 0.05	0.40	0.38	0.197	0.203	2.46	2.61	N.S.	N.S.
				B				
C0	11.54	11.81	5.519	5.609	70.14	70.33	29.36	27.22
C1	11.61	11.85	5.725	5.861	72.17	72.55	32.99	31.81
C2	9.617	9.607	4.609	4.578	58.94	59.10	41.14	40.13
LSD 0.05	0.40	0.38	0.197	0.203	2.46	2.61	2.73	2.79

Exogenous application of sprouted barley aqueous extract, trehalose, and calcium was efficiently utilized by increasing the percentage of P and Ca of carrot plant tissues. The reason behind this could be due to the effect of each factor in triggering different activation of different cellular process. Sprouted barley aqua extract has bioavailable and easily absorbed form of phosphorous (in the form of soluble phosphate) (Table 2), which led to the ease of absorption by leaves of and then significantly increased its percentage. In addition to that; trehalose spray may have a contribution to phosphorous

increase due to its role in preserving phosphorous-rich cell membranes (5). Calcium role in the mentioned increase could be resulted from its work as a second messenger in the CBL-CIPK network that involved in the signal pathway of phosphorous (20).The significant impact of T2C1 in P percentage and T2C2 in Ca percentage may be due to the synergetic effect of trehalose and calcium in preserving cell membranes. In fact, trehalose contributes in cells membranes stability and homeostasis by forming hydrogen bonds between its hydroxyl groups and hydrogen bonds of membranes phospholipids (5). Calcium maintains plasma membranes by increasing its activity by constantly opening or closing Ca^{+2} channels in response to a specific signal, which leads to better functioning of membranes. This means better control over the biological processes in the plant, better diagnosis and reception of signals and stimuli from outside the cell, and better connection between cells and their biological pathways (6; 31). As a result, better efficiency and homeostasis of the plant tissues and improving the growth and development of the plant. The significant influence of all individual treatments in P and Ca percentages is due to their impact on plant metabolism from different angles, which leads to improving plant fitness and increasing its growth, development and responses to external signals and enhances its absorption and assimilation of the elements, i. e. The richness of aqueous extract of barley sprouts in gibberellin and bioavailable nutrients (Table 2) and the effect of trehalose in increasing photosynthesis rates and plant biomass (22). Finally, calcium effect in building plant structure as a major component of plant cell walls as well as being a cellular and osmotic regulator and co-enzymatic for several biological processes in plants (6; 10; 31). The emphasis on increasing the content of antioxidants in fruits and vegetables has elevated in recent years. In fact that was shown in many research papers (2; 4; 29). The significant superiority of B1T2C1 by producing the highest antioxidant indicators for both seasons could be interpreted by the synergetic effect of the sprayed substances on the plants of the mentioned treatment. carotenoids share the same precursor with gibberellin, (geranylgeranyl diphosphate. GGPP) (8), as a result, spraying the aqueous extract of barley sprouts resulted in supplying the plant in sufficient amount of gibberellin (Table 2) and that substitute for its synthesis by the plant and focusing on carotenoids biosynthesis. In addition to that; Trihalose is antioxidant and a free radicals scavenger, making it unnecessary to oxidize carotenoids and as a result preserving their amounts in roots. Calcium may indirectly contribute in carotenoids biosynthesis by through its function as a cellular and osmotic regulator of plant cells. In fact that influencing the permeability of cell membranes. Thus, it achieved the homeostasis of the plant cells function, which created optimum conditions for the of carotenoids biosynthesis in chromoplasts. as all of what mentioned resulted increase the percentage of scavenging DPPH .The significant impact of B1T2 in in total carotenoids and beta-carotene increase is due to the role of each of the aqueous extract of barley sprouts and trehalose in phosphorous increase in roots (Table 3B). Phosphorous involved in all intermediates and some of enzymes of carotenoids and beta-carotene biosynthesis (8), and that may cause increase in their concentration and then an increase in the percentage of scavenging DPPH. B1C1 and T2C1 significant results might be due to phosphorus increase in their roots (Table 3B), in addition to having low proportion of core/cortex in the mentioned treatments plants (Table 4B), as is known that carotenoids and their derivatives are more concentrated in cortex. The reason for the superiority of plants that individually treated with trehalose and aqueous barley grains extract over the control in all antioxidant indicators is due to their effective effect on plant metabolism (6; 31), by providing a good environment inside cells to increase the biosynthesis of carotenoids and their derivatives. The opposite effect of elevated level of calcium is due to its effect on increasing the proportion of core/cortex. As it known, high core/cortex proportion is marketabley unfavorable. Our findings indicate that calcium increase in roots is closely correlated with high core/cortex proportion (Tables 3B, 3C, 4B, and 4C). In fact, calcium accumulates in xylem since it

doesn't translocate across phloem. After that, calcium combines with lignins and pectins in xylem cell walls causing xylem walls thickening (6) which in turn producing increase in xylem diameter (core). In conclusion; two seasons of field and laboratory studies authenticate that aqueous extract of barley sprouts, trehalose, and lowered level of calcium had a profound role in enhancing nutrients profile and increasing antioxidant traits in carrots. The elevated level of calcium largely influenced core/cortex percentage and that in turn negatively impacted total carotenoids, beta carotene and reduced DPPH%, which suggests that spraying the mentioned substances with taking in account a moderate dose of calcium on carrots maximizes their nutritional value.

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