Effect of some antioxidant treatments on physical and chemical characters of jerusalem artichoke tubers under cold storage conditions

Abdullah M.A.A. ¹* and A.M. Mounir ²

¹Vegetable Handling Department. Horticulture Research Institute, Agriculture Research Center, Giza, Egypt.

²Natural Products Research Department. National Center for Radiation Research and Technology, Atomic Energy Authority, Cairo, Egypt.

Abstract: Jerusalem artichoke is one of the non-traditional crops of the family Asteraceae. Its tubers are considered one of the richest vegetable crop in sugars especially inulin. Jerusalem artichoke faces some problems during storage such as tubers browning. The aim of this study was to enhance tubers quality and storability by soaking treatments for 5 min with 3% ascorbic acid, 3% citric acid, 1% calcium chloride or water, which served as control, before storage. The results clearly indicated that 3% ascorbic acid reduced the weight loss and decay. Also, it maintained better tubers appearance as well as higher contents of carbohydrates, total soluble solids, protein and inulin compared to other treatments. The observed effects of ascorbic acid on the tubers quality and storability could be due to its effect on Polyphenol oxidase, whereas its activity was inhibited by ascorbic acid.

Key words: Jerusalem artichoke, ascorbic acid, citric acid, calcium chloride.

Introduction

Jerusalem artichoke, girasol and sunchoke are the same synonym of (Helianthus tuberosus L.), belonging to the family Asteraceae. It is originated in North America but did not specify exactly its home land (Ben Chekroun et al., 1994). Some countries like Mexico, United States, and China produced it on a commercial scale (Kays and Nottingham, 2008), as its tubers are rich in inulin content (14-19%), non-digestible oligosaccharides. The tubers play an important role in lowering sugar blood, and enhancing the availability of minerals (Niness, 1999).

The main factor which complicates the storage of tubers is the highly evaporation of tubers through thin peel and the browning of tubers peel (Saengthobpinit and Sajjaanantakul, 2005). There are many antioxidants compound that could be used to prolong the storage ability of different fruits, and act the role of antioxidant agents. Ascorbic acid, citric acid and calcium chloride are using as additives to store tubers. Ascorbic acid plays an important role in reducing the activity of polyphenol oxidase enzyme that reduces the browning of fruits (McGhie et al., 2005). It has been used in different concentrations from 0.5 to 4% (w/v). The mechanism of ascorbic acid in inhibiting the activity of polyphenol oxidase could be summarized in its capability in the reduction of O-quinones which produced by PPO-catalyzed oxidation of polyphenols, back to dihydroxy polyphenols (Ozoglu and Bayindirli, 2002).
Citric acid is an important organic acid in plants, plays an important role in some physiological processes in plant such as the respiration. Also it has an inhibitory effect on the activity of polyphenol oxidase enzyme through its effect as a phenolase copper (Cu)-chelating agent and prohibition fruits browning and extending their shelf life (Jiang et al., 1999).

On the other hand, one of the most important macro elements is Calcium, which plays a considerable role in plant growth and fruit development. It connects with the carboxyl groups located at the backbone of pectin homogalacturonan, as hypothesized by the model of egg box (Braccini and Pérez, 2001). The presence of calcium increases fruit firmness strengthens plant and fruit cell wall, protects fruits against the degradations of enzymes and inhibits their action (White and Broadley, 2003). The presence of calcium delays fruit softening and enhances the storage ability of fruits, increases their shelf life and decreases the physiological disorders like internal or external fruit browning (Martín-Diana et al., 2007). In addition, calcium retards fruit ripening and senescence (Lester and Grusak, 2004; Mahajan and Dhatt, 2004; Singh et al., 2007).

Thus the aim of this study was to evaluate the effect of ascorbic acid, citric acid and calcium chloride on enhancing the storage ability and preservation of Jerusalem artichoke tubers, as well as decreasing the undesirable chemical changes in tubers under cold storage conditions.

2. Materials and methods

This study was carried out at Horticulture Research Institute, Agriculture Research Center, Giza governorate during the two successive seasons of 2016-2017 and 2017-2018. Jerusalem artichoke, Helianthus tuberosus L. cv. Fuseau, plants were grown in the experimental farm of the National Center of Radiation Research and Technology, Nasr City, Cairo, Egypt. Tubers were harvested on the 30th of December in both seasons, then transported immediately to the Vegetable Handling Department and kept overnight at 5°C with 90-95% relative humidity. The following morning, tubers were carefully selected, free of visual damage or defects, washed initially with water, then air dried. Tubers were divided into four groups dipping in the solution of 3% Ascorbic Acid for 5 minutes, 3% Citric Acid for 5 minutes, 1% Calcium chloride (CaCl₂) for 5 minutes and tap water for 5 minutes which served as control.

Jerusalem artichoke tubers were placed in plastic bags weighting 250 g then in carton boxes for each treatment and arranged in a complete randomized design consisting of three replicates and stored at 5°C and 90-95% relative humidity for 100 days. The treatments were examined immediately after harvest and every twenty days intervals for the following parameter.

2.1. Weight loss percentage: it was estimated according to the following equation: Weight loss% = [(Initial weight - weight of tubers at sampling date)/Initial weight of fruits] x 100.

2.2. General appearance: it was determined as score system of excellent > 9, good > 7 to 8.9, fair > 5 to 6.9, poor > 3 to 4.9, and unassailable > 2.9. The scale depends on morphological defects such as shriveling, fresh appearance and color change of tubers. Tubers rating (5) or below considered unmarketable (Watada and Morris, 1996; Jimenez et al., 1998).

2.3. Decay

Decay was determined as score system of 1= none, 2= slight, 3= moderate, 4= moderately severe, 5= severe. This depends on decay percentage on fruits (Watada and Morris, 1996; Jimenez et al., 1998).

2.4. Percentage of total soluble solids (T.S.S)

It was determined in Jerusalem artichoke tubers juice sample by digital refract meter of
model Abbe Leica according to the method described by (A.O.A.C., 2012).

### 2.5. Total carbohydrates content (g/100gm D.W)

After freezing, the tissues were ground to a powder for carbohydrate extraction. The petals (0.1 g) were extracted with 5 ml HCL (2.5 N) and boiled at 100 °C in a water bath for 2 h. After cooling to room temperature, the extracts were centrifuged at 4500 g for 15 min at 20 °C to remove contaminants and the supernatant was removed. Briefly, 1 ml of sample was added to test tube with 500 μL 5% phenol, then 2.5 ml concentrated sulfuric acid was added. The reaction of carbohydrate with phenol and sulfuric acid in aqueous solution gives a brown color and generates heat. The reaction mixture was allowed to cool to room temperature for 20-25 minutes, shaken, and the absorbance was measured at 490 nm (hexoses) and 480 nm (pentoses) in a spectrophotometer against a blank cell. The sugar concentration was obtained by referring to the standard graph. The assay for this standard glucose (Merck) graph was carried out by adding phenol and sulfuric acid to a standard glucose solution. Total carbohydrates were expressed in mg/0.1g fresh weight. (Dubois et al., 1956).

### 2.6. Crude protein percentage

Crude protein percentage was determined by microkjeldhl method as described by (A.O.A.C., 2012).

### 2.7. Inulin content (mg/g D. W.)

Inulin content tubers were longitudinally sliced into thin pieces at the middle part of the tubers. Fifty grams of sliced tuber was soaked in absolute ethanol at 4 °C for 24 hands the samples were stored at -20 °C until analyzed. The samples were oven dried at60 °C for 10 hours. To extract inulin, 2 g of dried sample was mixed with distilled water at 80 °C for 20 minutes. The solution was cooled to room temperature and filtered through a 0.45 μm membrane filter. The extracts (500 μl) were pipette into 25 ml volumetric flasks containing 3% HCl and diluted to 25 ml with water. The mixtures were then heated at 80 °C in a water-bath for 45 minutes. After cooling, the solutions were stored in plastic bottles before being analyzed by spectrophotometer. Inulin content was determined according to the method mentioned by (Saengkanuk et al., 2011).

### 2.8. Polyphenol oxidase (PPO)

It was extracted by homogenizing tubers samples with 5 fold of their weight sodium phosphate buffer (0.1 m, pH 6.5) containing 30 mM sodium ascorbate and 0.4 mM sucrose at 25°C. The homogenate fruit was centrifuged at 10000 g for 15 min. Supernatant was collected and stored at 4°C. Catechol was dissolved in the phosphate buffer (10 mM) then a volume of 3 mL was mixed with 1.0 enzyme extract. The increment of absorption of 495 nm was spectrophotometrically recorded. The increase in absorbance of 0.01 per minute at 495 nm at the specified condition was defined as one unit of PPO activity. The results were expressed as IU per mg protein (Dogan et al., 2002).

### Statistical analysis

All data were subjected to statistical analysis according to the procedures reported by Snedecor and Cochran (1982) and means were compared by Duncan’s Multiple Range Tests (Duncan, 1955).

### 3. Results and discussions

#### 3.1. Weight loss percentage

Data presented in Table (1) showed the effect of different storage periods, antioxidant treatments and their interaction on weight loss percentage of Jerusalem artichoke tubers. It was observed that the lowest weight loss percentage was at 20 days after storage then a relatively decrease in weight loss percentage was remarked through the increment of storage period till reaching its maximum depression at 100 days of storage.
in both seasons. Similar result was obtained by (Danilčenko et al., 2008; Attia and Alian 2011; Rashed et al., 2018) who declared that the increase of storage period was accompanied by an increase in weight loss percentage of Jerusalem artichoke tubers. The increase in weight loss percent might be related to the thin crust of tubers which facilitate the water loss through the transpiration and the amount of dry matter through respiration (Wills et al., 1981).

As for the effect of different antioxidant treatments on weight loss percentage, data show that the lowest weight loss percentage was obtained when tubers were treated with 3% ascorbic acid. On the contrary the highest weight loss percentage was found in untreated tubers. This result agrees with that obtained by Kasim et al. (2015) on fresh-cut carrot. This finding might be attributed to the effect of ascorbic acid on diminishing the respiration rate of tubers and increasing its cell capacity of scavenging ROS (Lin et al., 2007).

Regarding the interaction between storage periods, different antioxidant treatments on weight loss percentage of tubers, data revealed that tubers treated with 3% ascorbic acid stored for 20 days scored the lowest weight loss percentage followed by tubers treated with 3% citric acid than 1% calcium chloride and stored for the same period in both seasons. The depression of weight loss percentage during different storage period scored its lowest decrease when tubers were treated with 3% ascorbic acid till 100 days of storage in both seasons.

3.2. Decay (score)

As presented in Table (1), data show that tubers stored till twenty days scored the lowest decay, then the increase in storage period was accompanied by an increase in decay in both seasons. This result is on the same line of previous findings (El- Sharkawy et al., 2003; Kader, 2011; Attia and Alian, 2011) showed that the prolongation of cold storage period of Jerusalem artichoke tubers was related with an increase in unmarketable percentage and damaged tubers. The decay of tubers during storage reaches its maximum percentage throughout the increase of storage period as a result of high respiration rate, microbiological load, the activity of different enzymes and biochemical changes in tubers during storage period (Rashed et al., 2018).

Concerning the effect of different treatments on decay score, the obtained results indicate that tubers treated with 3% ascorbic acid gave significantly the lowest decay. This finding is on the same line of Ouzounidou et al. (2012) who returned the effect of ascorbic acid on reducing the decay to its effect on reducing pathogenic effect and inhibiting the enzymatic browning.

Respecting the interaction between storage periods and different treatments, data showed that there was no significant difference among antioxidant treatments during forty days of storage, and then it was observed that treated tubers with 3% ascorbic acid scored lower decay than other treatments during sixty days and eighty days of storage in both seasons.

3.3. General appearance (score)

Data in Table (1) indicated that tubers stored till twenty days maintained good appearance, and then a gradual deterioration was observed till the end of storage period. This finding is in accordance with that obtained by (El- Sharkawy et al., 2003; Kader, 2011; Attia and Alian 2011) who concluded that the increase of storage period of Jerusalem artichoke under 2°C was linked by a reduction in general appearance. The decline in general appearance might be related to several factors that increase damages and decay of tubers such as accelerating ripening and senescence, respiration rate subsequently water loss, which lead to fruit shriveling and affect tubers general appearance (Sams, 1999).

As for the effect of different antioxidant treatments and their effect on tubers general appearance, data revealed that 3% ascorbic
acid scored significant result and gave better general appearance than other treatments. This result might be related to its effect on decreasing weight loss percentage, diminishing microbial load and the activity of enzymes, like polyphenol oxidase, which was reflected on the general appearance of tubers (Lin et al., 2007).

Regarding the effect of different antioxidant treatments on tubers general appearance during storage period, it was observed that there was no significant difference among different treatment through forty days of storage, then a significant difference was observed in the rest of storage period, which explained that treating tubers with 3% ascorbic acid or 3% citric acid maintained their general appearance during sixty days of storage in both seasons and 3% of ascorbic acid alone till eighty days of storage in the first and the second seasons, respectively.

3.4. Percentage of total soluble solids (T.S.S)

Data presented in Table (2) show that the increment in storage period of Jerusalem artichoke tubers was accompanied by a decrease in total soluble solids (%) which reaches its maximum depression at the end of storage period. This result agrees with that obtained by (Danilcenko et al., 2008; Rashed et al., 2018) on Jerusalem artichoke tubers. Most of the biochemical changers are reduced under low temperature, except Jerusalem artichoke tubers metabolism that could continue even under cold storage conditions (Saengthobpinit and Sajjanaantakul, 2005) which led to the decrease in total soluble solids as a result of the consumption of carbohydrates through the respiration. Concerning the effect of different antioxidant treatments and their effect on total soluble solids, data revealed that there was a notable difference among different treatments. Tubers treated with 3% ascorbic acid had the highest content of total soluble solids (2.96%), while untreated tubers have the lowest percentage of total soluble solids (2.82%). Same result was obtained by Kasim and Kasim (2016) and Kumhar et al., (2014) who declared that the high concentration of ascorbic acid increase the percentage of total soluble solids of ready to use carrot shreds and custard apple (Annonasquamosa L.) pulp respectively.

Regarding the interaction between storage periods and different antioxidant treatments, it was noticed that all treatments in the beginning of storage period gave the highest total soluble solids in both seasons. On the other hand, twenty days of storage period, data show that both 3% ascorbic acid and 3% citric gave higher total soluble solids than other treatments generally in both seasons. This result might be referred to the effect of both ascorbic acid and citric acid in reducing respiration rate causing a decrease in metabolic activities and preserve total soluble solids during storage period. Same result was obtained by (Li et al., 2014).

3.5. Total Carbohydrates content (g/100gm D.W)

As presented in Table (2), the obtained results show that the prolongation of storage period led to a gradual reduction in carbohydrates content which reached its maximum extent at the end of storage period in both seasons.

This finding is on the same line of Ghoneem et al. (2016) who was found that the degradation rate of carbohydrates in Jerusalem artichoke tubers extend with the increase of storage period.

Concerning the effect of different antioxidant treatments and their impact on carbohydrates content, data showed that tubers treated with 3% ascorbic acid had higher content of carbohydrates than the other treatments. This result might be referred to the effect of ascorbic acid in delaying the respiration rate
Table 1. Effect of citric acid, ascorbic acid and calcium chloride on weight loss % and decay as well as general appearance of Jerusalem artichoke (*Helianthus tuberosus* L.) tubers under cold storage conditions.

| Characters | First season |   |   |   |   | Mean |   |   |   |   |   | Mean |   |   |   |   |   | Mean |
|-----------|--------------|---|---|---|---|------|---|---|---|---|---|------|---|---|---|---|---|------|
| Treatments | Days after storage | 0 | 20 | 40 | 60 | 80 | 100 | Mean | 0 | 20 | 40 | 60 | 80 | 100 | Mean |
| Weight loss % | C. | 4.62p | 14.13l | 27.23f | 42.14b | 56.81a | 28.98A | - | 5.107p | 15.04l | 27.913f | 43.04b | 57.41a | 28.98A | 2.23E | 7.37D | 18.63C | 28.82B | 37.59A | - | 2.59E | 8.20D | 19.28C | 29.63B | 38.60A |
| C.A3% | - | 1.52s | 5.05o | 16.23k | 24.50h | 31.26d | 15.71C | - | 1.53s | 6.22o | 16.913k | 25.13h | 32.64d | 16.49C | 1.00E | 1.00E | 1.41D | 1.83C | 2.58B | 3.41A | 1.00D | 1.00D | 1.50C | 2.08B | 2.5B | 3.58A |
| A.A3% | - | 0.73t | 3.80q | 12.56m | 21.71i | 28.15e | 13.39D | - | 0.92t | 4.32q | 13.11m | 22.52i | 28.91e | 13.95D | 1.00E | 1.00E | 1.41D | 1.83C | 2.58B | 3.41A | 1.00D | 1.00D | 1.50C | 2.08B | 2.5B | 3.58A |
| CaCl2 1% | - | 4.62p | 14.13l | 27.22f | 42.13b | 56.81a | 17.62B | - | 2.843r | 7.23n | 19.183j | 27.81g | 35.44c | 18.5B | 1.00E | 1.00E | 1.41D | 1.83C | 2.58B | 3.41A | 1.00D | 1.00D | 1.50C | 2.08B | 2.5B | 3.58A |
| Decay | C. | 1.00g | 1.00g | 2.00d-g | 2.66c-e | 3.66a-c | 4.33a | 2.44A | 1.00f | 1.00f | 2.00d-f | 2.66b-d | 3.33a-c | 4.33a | 2.38A |
| C.A3% | 1.00g | 1.00g | 1.33fg | 1.66e-g | 2.00d-g | 3.00b-d | 1.66C | 1.00f | 1.00f | 1.33ef | 2.00d-f | 2.33c-e | 3.66ab | 1.88B |
| A.A3% | 1.00g | 1.00g | 1.00g | 1.00g | 1.66e-g | 2.33d-f | 1.33D | 1.00f | 1.00f | 1.00f | 1.33ef | 1.66d-f | 2.66b-d | 1.44C |
| CaCl2 1% | 1.00g | 1.00g | 1.33fg | 2.00d-g | 3.00b-d | 4.00ab | 2.05B | 1.00f | 1.00f | 1.66d-f | 2.33c-e | 2.66b-d | 3.66ab | 2.05AB |
| General appearance | C. | 9.00a | 9.00a | 7.00a-d | 5.66c-e | 3.66a-g | 2.33g | 6.11D | 9.00a | 9.00a | 7.00a-c | 5.66c-e | 4.33d-f | 2.33f | 6.22C |
| C.A3% | 9.00a | 9.00a | 8.33ab | 7.66a-c | 7.00a-d | 5.00d-f | 7.66B | 9.00a | 9.00a | 8.33ab | 7.00a-c | 6.33b-d | 3.66ef | 7.22B |
| A.A3% | 9.00a | 9.00a | 9.00a | 9.00a | 7.66a-c | 6.33b-d | 8.33A | 9.00a | 9.00a | 9.00a | 8.33ab | 7.66a-c | 5.66c-e | 8.11A |
| CaCl2 1% | 9.00a | 9.00a | 8.33ab | 7.00a-d | 5.00d-f | 3.00g | 6.88C | 9.00a | 9.00a | 7.66a-c | 6.33b-d | 5.66c-e | 3.66ef | 6.88BC |
| Mean | 9.00A | 9.00A | 8.16B | 7.33C | 5.83D | 4.16E | 9.00A | 9.00A | 8.00B | 6.83C | 6.00C | 3.83D |

Means followed by different letters are significantly different at P ≤ 0.05 level; Duncan´s multiple range test. C: Control, C.A: Citric acid, A.A: Ascorbic acid, CaCl2: Calcium chloride.
3.6. Crude protein percentage

The effect of different storage period and several antioxidant treatments as well as their interaction effect on crude protein percentage is presented in Table (2). A notable reduction in crude protein percentage was observed during storage period which reached its maximum run-down at the end of storage period. Same result was obtained by Ghoneem et al. (2016) who found that the increase in storage period was related by a decrease in protein percentage of Jerusalem artichoke tubers.

As for the effect of different antioxidant treatments, it was found that tubers treated with 3% ascorbic acid scored the highest percentage of crude protein.

Regarding the interaction between storage period and different antioxidant treatments and their impact on protein percentage, data revealed that there was no significant difference between different treatments during the first twenty days. While it was remarked that 3% ascorbic acid gave higher protein percentage than the other treatments in both seasons.

3.7. Inulin content (mg/g D. W.)

The effect of storage periods, different antioxidant treatments and their interaction on inulin content in Jerusalem artichoke tubers is shown in Table (3) it is clear that there was a reversible relation between storage period and inulin content, Whereas the highest content of inulin was observed at the beginning of storage period while the lowest content was observed at the end of storage period. These results are in agreement with Cabezas et al. (2002) on Helianthus tuberosus (Jerusalem artichoke) and Cichorium intybus tubers and Attia and Alian, (2011); Ghoneem et al. (2016) and Rashed et al. (2018) on Jerusalem artichoke tubers. This result might be related to the continues metabolism which continued in tuber even under low temperature and finally led to the breakdown of inulin into short chain through the partial enzymatic hydrolysis that degrades it into lower DP frictions, sucrose, glucose and fructose (Rubel et al., 2014).

As for the effect of different antioxidant treatments, the obtained results show that tubers treated with 3% ascorbic acid had higher content of inulin than the other treatments. This result may be related to the effect of ascorbic acid in reducing the respiration rate (Lin et al., 2007) which was reflected on diminishing the breakdown of inulin into short chain.

Respecting the effect of the interaction between storage period and different antioxidant treatments, data show that tubers treated with 3% ascorbic acid during the first twenty days gave higher inulin content than other treatments in both seasons.

3.8. Polyphenol oxidase activity

As presented in Table (3), the increase in storage period was related to the increase in polyphenol oxidase acidity. This result is in agreement with that obtained by El-Awady et al. (2015) and Abdullah et al. (2017) on Jerusalem artichoke tubers.

Concerning different antioxidant treatments and their effect on polyphenol oxidase activity, it could be concluded that treated tubers with 3% ascorbic acid showed the lowest polyphenol oxidase activity. This result might be related to the important role of ascorbic acid through the direct or indirect scavenger of AOS in plant cell (Smirnoff and
Table 2. Effect of citric acid, ascorbic acid and calcium chloride on T.S.S.% , carbohydrates content (g/100gm D.W) and total crude protein% of Jerusalem artichoke (*Helianthis tuberosus* L.) tubers under cold storage conditions.

| Characters | First season | Second season | Days after storage | Days after storage |
|------------|--------------|---------------|--------------------|--------------------|
|            | Treatments   |               | 0                  | 20                 | 40 | 60 | 80 | 100 | Mean | 0   | 20 | 40 | 60 | 80 | 100 | Mean |
| T.S.S.%    | C            | 3.18a         | 3.10b-d            | 3.01ef             | 2.71hi             | 2.50f | 2.4m | 2.82D | 3.2a       | 3.10ab | 2.86de | 2.64gh | 2.50jk | 2.46k | 2.79C |
|            | C.A3%        | 3.18a         | 3.13a-c            | 3.07c-e            | 2.82g              | 2.64ij| 2.54kl| 2.90B | 3.2a       | 3.13a | 2.94cd | 2.8ef  | 2.64g-i | 2.5jk | 2.86B |
|            | A.A3%        | 3.18a         | 3.15ab             | 3.1b-d             | 2.96f              | 2.77gh| 2.62j | 2.96A | 3.2a       | 3.17a | 3.00bc | 2.82d-f| 2.74fg | 2.64g-i | 2.93A |
|            | Cacl2 1%     | 3.18a         | 3.1b-d             | 3.06de             | 2.81de             | 2.61jk| 2.51l | 2.88C | 3.2a       | 3.1ab | 2.9c-e | 2.74fg | 2.61h-j | 2.52i-k | 2.84B |
| Mean       | 3.18A        | 3.12B         | 3.06C              | 2.83D              | 2.63E              | 2.52F |        |       | 3.2A       | 3.12B | 2.92C  | 2.75D  | 2.62E  | 2.53F  |
| Carbohydrates | Control   | 50.22a      | 47.35e             | 43.74i             | 40.16m             | 36.54r| 32.72t| 41.79D | 49.3a      | 46.82e | 43.04j | 39.72n | 36.03s | 32.04u | 41.16D |
|            | C.A 3%       | 50.22a       | 48.63c             | 45.04g             | 42.64j             | 39.34o| 36.73q| 43.76B | 49.3a      | 48.12c | 44.63g | 42.03k | 38.85p | 36.12r | 43.17B |
|            | A.A 3%       | 50.22a       | 49.33b             | 46.23f             | 44.44h             | 42.03k| 39.83n| 45.34A | 49.3a      | 48.81b | 45.73f | 43.83j | 41.41l | 39.24o | 44.72A |
|            | Cacl2 1%     | 50.22a       | 48.04d             | 44.53h             | 41.82l             | 38.13p| 35.33s| 43.01C | 49.3a      | 47.52d | 44.03h | 41.15m | 37.53q | 34.70t | 42.37C |
| Mean       | 50.22A       | 48.34B       | 44.88C             | 42.26D             | 39.01E             | 36.15F|        |       | 49.3A      | 47.82B | 44.35C | 41.68D | 38.45E | 35.52F | |
| Crude protein% | Control   | 4.08a       | 3.63c              | 3.20e              | 2.73h              | 2.31j | 1.83l | 2.96D | 4.19a      | 3.71d | 3.33g  | 2.86jk | 2.42m  | 1.96o  | 3.08D |
|            | C.A 3%       | 4.08a        | 3.74b              | 3.44d              | 3.04f              | 2.71h | 2.24j | 3.21B | 4.19a      | 3.81c | 3.64e  | 3.25h  | 2.92j  | 2.37m  | 3.35B |
|            | A.A 3%       | 4.08a        | 3.81b              | 3.52d              | 3.22e              | 2.91g | 2.51i | 3.34A | 4.19a      | 3.98b | 3.61e  | 3.35g  | 3.07i  | 2.63l  | 3.47A |
|            | Cacl2 1%     | 4.08a        | 3.76b              | 3.46d              | 2.95g              | 2.71h | 2.11k | 3.18C | 4.19a      | 3.80c | 3.50f  | 3.01i  | 2.83k  | 2.21l  | 3.25C |
| Mean       | 4.08A        | 3.73B        | 3.40C              | 2.99D              | 2.66E              | 2.17F |        |       | 4.19A      | 3.82B | 3.51C  | 3.12D  | 2.81E  | 2.29F  |

Means followed by different letters are significantly different at P ≤ 0.05 level; Duncan’s multiple range test. C: Control, C.A: Citric acid, A.A: Ascorbic acid, CaCl2: Calcium chloride.
Table 3. Effect of citric acid, ascorbic acid and calcium chloride on inulin content (mg/g D. W.) and polyphenol oxidase (IU per mg protein) of Jerusalem artichoke (*Helianthus tuberosus* L.) tubers under cold storage conditions.

| Characters | Treatments | First season Days after storage | Second season Days after storage | Mean |
|------------|------------|---------------------------------|---------------------------------|------|
|            |            | 0 | 20 | 40 | 60 | 80 | 100 | Mean | 0 | 20 | 40 | 60 | 80 | 100 | Mean |
| inulin     | C.         | 29.53a | 24.63f | 20.81i | 16.81n | 12.72r | 8.94s | 18.91D | 28.71a | 25.17c | 21.23h | 17.03l | 12.92q | 8.94s | 18.91D |
|            | C.A3%      | 29.53a | 26.72c | 23.22g | 20.71i | 17.6m | 14.33p | 22.02B | 28.71a | 25.13c | 22.33f | 19.42j | 16.93m | 13.72p | 21.04B |
|            | A.A3%      | 29.53a | 27.15b | 25.40e | 23.16g | 20.41j | 17.91l | 23.92A | 28.71a | 26.03b | 25.14g | 22.91k | 19.74o | 16.51r | 23.17A |
|            | CaCl2 1%   | 29.53a | 25.81d | 22.52h | 19.82k | 16.08k | 13.62q | 21.23C | 28.71a | 24.61d | 21.52g | 18.84k | 15.36o | 12.73r | 20.29C |
| Mean       | 29.53A | 26.08B | 22.99C | 20.13D | 16.70E | 13.70F | 28.71A | 25.233B | 22.55C | 19.55D | 16.23E | 13.02F | 28.71A | 25.233B | 22.55C |
| ppo        | C.         | 61.80t | 66.23o | 71.05j | 77.16f | 84.05b | 91.24a | 75.25A | 63.14m | 68.02h-k | 73.41f | 79.06c-e | 86.23b | 93.92a | 77.30A |
|            | C.A 3%     | 61.80t | 63.93q | 66.42n | 70.24k | 75.04h | 79.41d | 69.47C | 63.14m | 65.24k-m | 68.61h-j | 72.21fg | 77.53e | 81.21cd | 71.32B |
|            | A.A 3%     | 61.80t | 62.71s | 65.73p | 69.66l | 74.41i | 78.72e | 68.84D | 63.14m | 64.63lm | 67.04i-l | 71.15f-h | 73.81f | 80.14c-e | 69.99C |
|            | CaCl2 1%   | 61.80t | 63.51r | 67.02m | 71.15j | 75.92g | 80.23c | 69.94B | 63.14m | 65.95j-m | 69.52g-i | 73.44f | 78.03de | 82.31c | 72.07B |
| Mean       | 61.80F | 64.09E | 67.56D | 72.05C | 77.36B | 82.40A | 63.14F | 65.96E | 69.65D | 73.96C | 78.90B | 84.39A | 61.80F | 64.09E | 67.56D |

Means followed by different letters are significantly different at $P \leq 0.05$ level; Duncan´s multiple range test. C: Control, C.A: Citric acid, A.A: Ascorbic acid, CaCl2: Calcium chloride.
Wheeler, 2000) and preserving the relative stability of the mono hydroascorbate radical. Also ascorbic acid is considered one of the important antioxidant agents, because it is one of two considerable soluble antioxidants in chloroplast, where it creates a condition of equilibrium between antioxidants and AOS production (homeostasis) of sensitive plant tissue (Foyer and Noctor, 2000), conserve α-Tocopherol and repair it from α-Tocopheroxyradical (Munné-Bosch and Alegre, 2002).

Respecting the interaction between storage period and different antioxidant treatments on polyphenol oxidase activity, the obtained results showed that all treatments at the beginning of storage period gave the lowest polyphenol oxidase activity followed by treated tubers with 3% ascorbic acid, 3% citric acid and 1% calcium chloride respectively during the first twenty days in both seasons.

Conclusion

Our results have demonstrated that the best treatment which maintains tubers of Jerusalem artichoke under cold storage conditions is the dipping in 3% ascorbic acid which preserved the physical and chemical properties of tubers.

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