Effects of Freezing and an Alginate Coating on the Shelf Life of Fresh *Pistacia Vera*

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Abstract

Introduction: Despite nutritional and economic importance of pistachios, there are still many problems facing their production and marketing. Changes made in the quality of fresh pistachios during storage could reduce their shelf life. Thus, they should be consumed in a short period of time if not stored under proper conditions.

Materials and Methods: This study was conducted using treatments of freezing (for 0, 1.5, and 3 months at -20 °C) and an alginate-based edible coating (at concentrations of 0, 0.5, and 1%) to increase durability of fresh pistachios in a completely randomized design. The measured parameters included moisture reduction, the peroxide value, and the phenolic content.

Results: Results of the current research showed that freezing temperatures (up to 1.5 months) reduced the peroxide value of fresh pistachios. The use of an alginate-based edible coating (especially at the concentration of 1%) helped maintain some parameters of fresh pistachios, such as the moisture content and peroxide value.

Conclusions: Quality changes in fresh agricultural products during storage are mostly caused by enzyme activities and metabolic alterations. The use of chemical compounds is not welcomed by consumers due to their detrimental effects on the environment and human health. The freezing temperature could extend the shelf life of fresh products by decreasing their respiration rate. The other new technology employs edible coatings, such as alginates, to create a barrier on the surface of fruits to prevent water loss and gas exchange (oxygen and carbon dioxide).

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1. Introduction

The name of pistachios (*Pistacia vera* L.) is closely associated with Iran. Despite significant nutritional and economic values of pistachios, there are still many problems facing their production, marketing, and sales. This agricultural product is mainly supplied as dried fruit, while it is possible to offer and sell the product fresh [1]. Durability of fresh pistachios would generally decrease with an increase in the temperature and the storage time. Nuts with a good quality and intact hulls could be stored for 48 h or more under ambient conditions with no increase in the staining of the shell. However, pistachios with hulls of a poor quality would be damaged after only 8 h at 40°C, 24 h at 30°C, and 40 h at 25°C [2].

Quality changes are mostly related to enzyme activities and metabolic alterations [3, 4] that cause moisture loss, decrease fruit firmness, cause fruit discoloration, and increase fungal and bacterial activities in the fruit [5, 6]. Postharvest investigation of fresh pistachios in Akbari and Ohadi cultivars indicates that nuts' weight loss and microbial activities increase during storage, while firmness of hulls and kernels decreases [7]. Besides, upon an increase in the storage duration, the content of fatty acids, carbohydrates, and color characteristics of fresh pistachios decrease [8].

Today, new methods and technologies have considered the extending of the shelf life of fresh horticultural products by decreasing their respiration rate. Applying chemical compounds to agricultural products is not accepted by consumers as they could be harmful to humans and the environment. Storing fresh pistachios with and without hulls as well as placing dried pistachio nuts inside paper envelopes at temperatures 4 and 12°C indicate that an increase in the storage duration and temperature could stain shells, damage kernels, and reduce the shelf life [9]. These treatments increase acidity and peroxide values as well as water loss, but decrease the carbohydrate content, taste index, and fruit firmness, while the amount of proteins does not change. Accordingly, keeping in-hull fresh pistachios at 4°C would increase their shelf life for up to 45 days.

The use of edible coatings or films is a new technology employed to overcome lipid oxidation and microbial deterioration. An edible film is a membrane formed by interactions between different molecules, which comes from nature and edible materials, such as polysaccharides, proteins, etc. In fact, the function of edible coatings is to create a barrier against some materials, such as water and gases (oxygen and carbon dioxide), to preserve food compounds, transfer additives (colors, flavorings, antioxidants, antimicrobials, etc.), restrict growth of microorganisms on the product surface, and provide mechanical protection for products [10-13]. Among constituents of edible coatings, hydrocolloids (carbohydrates and proteins) are the best barriers to oxygen and water due to the

formation of a regular structure of hydrogen networks [14-16]. Alginates are biological polymers with unique colloidal properties, which form strong gels and insoluble polymers after reacting with cations, such as calcium and sodium [17]. In fact, they are polysaccharides derived from brown algae (the Phaeophyceae), which are employed in the food industry increasingly for their gel forming properties [14, 18, 19].

In this research, effects of the freezing temperature (-20°C) and different concentrations (0, 0.5 and 1%) of alginate-based edible coatings are studied on the shelf life of fresh pistachios.

2. Materials and Methods

2.1. Sample preparation

Fresh *P. vera* nuts of the Akbari cultivar were harvested from pistachio trees at station no. 2, Pistachio Research Center, Rafsanjan, which were immediately transferred to the laboratory, with their cluster axis divided in parts. Next, fresh, ripe, healthy, and uniform pistachios were separated from unripe, damaged, and cracked ones. The samples were coated by immersion in sodium alginate for 3 min at different concentrations (0, 0.5, and 1%) (Sigma Aldrich Co.) and were dried for 1 min at the laboratory temperature. To form a gel, coated fruits were soaked in 2% $CaCl_2$ for 2 min. In addition, about 400-450 g of the prepared samples were packed in polyethylene containers and stored at -20°C for 0, 1.5, and 3 months. Next, different parameters, such as the moisture content (weight loss), peroxide value, and total phenolic content were measured.

2.2. Moisture content

Fresh pistachios were weighed at initial and final points of the specified freezing period, and moisture reduction was calculated using the following equation [20]:

$$\text{Moisture reduction } (\%) = \frac{IW - FW}{IW} \times 100$$

Where IW and FW are initial and final weights (g) of pistachio fruits, respectively, at any specified time.

2.3. Peroxide value

For the purpose of lipid extraction, unshelled pistachio nuts were ground in a laboratory mill. The oil was extracted using a solvent of n-hexane and the Soxhlet extraction system. Next, the extract was evaporated by a rotary evaporator [21]. Besides, iodometric titration was used to measure the peroxide value [22]. Five g of the extracted oil was mixed with the solvent consisted of acetic acid and chloroform at a ratio of 3:2. The mixture was then placed in the dark for 1 min. Next, 25 ml of distilled water and a few drops of a 1% starch solution were added as the indicator. Titration continued using sodium thiosulfate solution, 0.1 N [3], until blue color of the solution disappeared. The peroxide value was calculated as meq kg^{-1} using the following formula:

$$\text{Peroxide value} = \frac{1000 \times \text{normalization of ST} \times \text{the used amount of ST (ml)}}{\text{sample weight (g)}}$$

2.4. Total phenolics

The Folin-Ciocalteu method was used to determine the concentration of total phenolics [23]. As much as 0.5 g of pistachio kernels was ground with 3 ml of 85% methanol to produce a homogeneous mixture. Next, the mixture was centrifuged at 10,000 rpm for 15 min. Besides, 300 µl of the supernatant was mixed with1200 µl of 7% sodium carbonate and 1500 µl of the 10-fold-diluted Folin-Ciocalteu reagent. The final mixture was shacked for 1.5 h in the dark, and its absorbance was recorded at 765 nm. Measurements were carried out based on the standard curve of gallic acid. Besides, total phenolic compounds were considered as the equivalent amount of gallic acid (mg) per dry matter (g) [24].

2.5. Statistical analysis:

This research was conducted in a completely randomized design with 3 replications. Besides, statistical analyses were performed by SPSS 16.0 using the one-way analysis of variance (ANOVA). Next, the means were compared through the Duncan's test ($P< 0.05$).

3. Results

3.1. Moisture content

Effects of the freezing duration and the concentration of the alginate edible coating on the moisture reduction content of fresh pistachios have been shown in Fig. 1. As the results show, the increase in the freezing duration from 0 to 1.5 and 3 months in fresh pistachios coated with 0% alginate has reduced the moisture content (Fig. 1).

In fresh fruits coated with 0.5% alginate and stored for 1.5 and 3 months at the freezing temperature, moisture reduction was higher than in samples which were not frozen (control) (Fig. 1).

The moisture reduction in pistachios with the 1% alginate coating stored for 1.5 months at the freezing temperature was significantly higher than in the unfrozen samples (Fig. 1). However, there was no significant difference in the moisture reduction between pistachios frozen in 1.5 and 3 months with the 1% alginate coating (Fig. 1). Besides, Fig. 1 shows that the moisture reduction in pistachios frozen in 1.5 and 3 months coated with 1% alginate was lower than in those covered with 0% alginate (the control). In addition, there was no significant differences between 1 and 0.5% coatings in these samples.

3.2. Peroxide value

Fig. 2 shows effects of the freezing duration and the concentration of the alginate coating on the peroxide value of fresh pistachios. As the results indicate, the peroxide value enhanced significantly by the increase in the freezing duration from 0 and 1.5 months to 3 month. This result was repeated at alginate concentrations of 0 and 0.5% as well.

In pistachios coated with 1% alginate, the freezing duration had no significant effect on the peroxide value (Fig. 2). However, peroxide values of pistachios frozen for 3 months were significantly lower at the 1% alginate concentration than at 0 and 0.5% concentrations (Fig. 2).

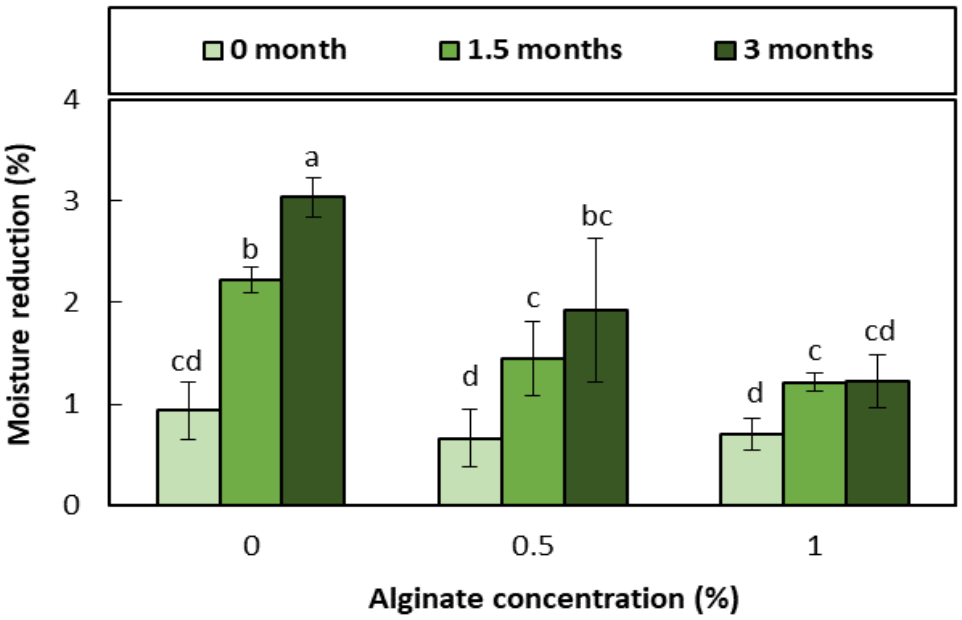

Fig. 1- Effects of the freezing duration (0, 1.5, and 3 months) and the concentration of the alginate edible coating (0, 0.5, and 1 %) on moisture reduction in fresh pistachios; data are shown as means ± SD (n = 3), and the letters above the bars show significant differences between mean values (Duncan test, $P < 0.05$).

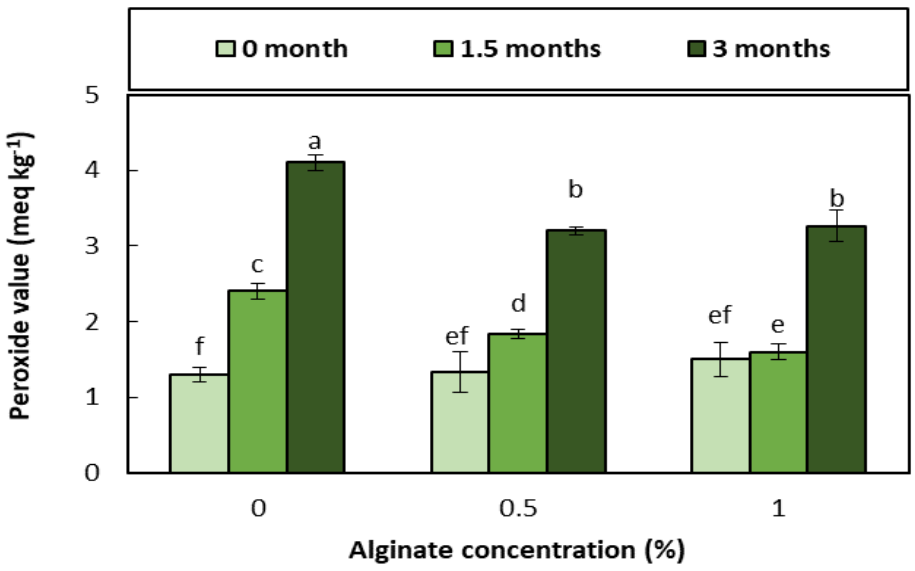

Fig. 2- Effects of the freezing duration (0, 1.5, and 3 months) and the concentration of the alginate edible coating (0, 0.5, and 1 %) on the peroxide value in fresh pistachios; data are shown as means± SD (n = 3), and the letters above the bars show significant differences between mean values (Duncan test, $P < 0.05$).

3.3. Total phenolics

Fig. 3 shows effects of different freezing durations (0, 1.5, and 3 months) as well as alginate coating concentrations (0, 0.5, and 1%) on the total phelolic content of fresh pistachios. As the results show, the freezing of fresh pistachios (for 1.5 and 3 months) reduced the phenolic content. This trend was almost the same in fruits coated at 0, 0.5, and 1% alginate concentrations (Fig. 3).

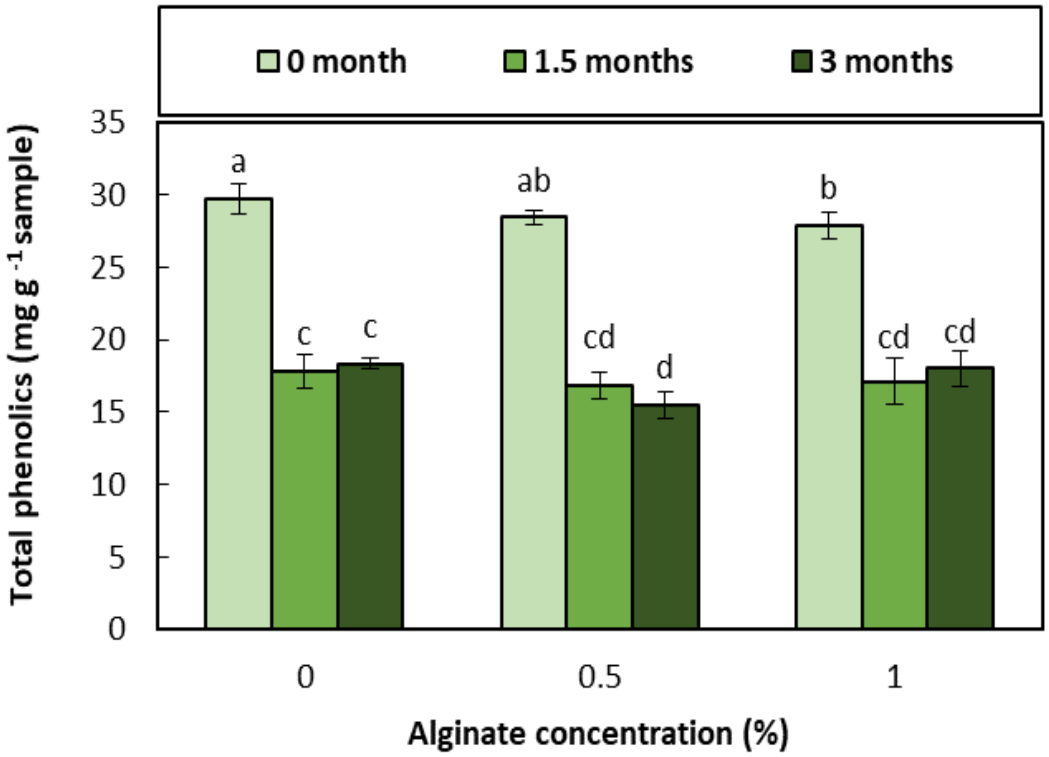

Fig. 3- Effects of the freezing duration (0, 1.5, and 3 months) and the concentration of the alginate edible coating (0, 0.5, and 1 %) on total phenolic compounds in fresh pistachios; data are shown as means± SD (n = 3), and the letters above the bars show significant differences between mean values (Duncan test, P<0.05).

4. Conclusions

4.1. Moisture content

The results of this research indicated that freezing fresh pistachios could reduce their moisture content. However, the alginate edible coating (especially at a 1% concentration) prevented moisture reduction during storage. In fact, edible coatings prevent exchange of water and gases (as oxygen and carbon dioxide) by creating a uniform and thin layer on the product surface [10, 19, 25]. To preserve kiwifruit, Xu et al used an edible coating prepared from soybean stearic acid, protein isolates, and pullulan [26].

Their results showed that edible coatings extended the shelf life of kiwifruit by about 3 times. In another research, Sipahi et al studied effectiveness of an alginate-based antimicrobial edible film in preserving fresh cut watermelons [27]. In this research, a coating was prepared from sodium alginate (0.5, 1, 2 g $100g^{-1}$), beta-cyclodextrin, microencapsulated trans-cinnamaldehyde, pectin, and calcium lactate using the layer-by-layer technique. The coated samples were stored for 15 days at 4°C, which were highly welcomed by consumers except at the 2 g $100g^{-1}$ alginate concentration. Both alginate coatings of 1 and 2 g $100g^{-1}$ extended durability of fresh-cut fruits from 7 (control) to 12-15 days [27].

4.2. Peroxide value

Pistachios are a type of nut of a high nutritional value with 89.1% of their total fatty acids being unsaturated and 30% of which being polyunsaturated. Therefore, the high lipid content and unsaturated fatty acids existing in pistachios make them very susceptible to decay and fungal deterioration [27]. The results of the current research indicated that freezing pistachio products for 1.5 months decreased the kernels' peroxide value. It was also established that pistachios with an alginate coating (especially at the 1% concentration) had a lower peroxide value.

In a study, walnut kernels with an edible coating composed of a protein isolate of soybean, catechin, and carboxymethylcellulose had a lower peroxide value than the control samples after 21 days [28]. In fact, soy protein-based coatings are proper carriers of antioxidant agents and effective preservative methods for extending shelf life as well as improving durability of oxidation sensitive nuts.

In another research, effects of different concentrations of edible coatings composed of β-glucan and chitosan on quality stability of dried peanuts were investigated [29]. Peanuts coated with both β-glucan and chitosan showed the least lipid oxidation values and the highest level of polyunsaturated fatty acids after four months of storage.

4.3. Total phenolics

In the current study, the content of total phenolics decreased during storage at the freezing temperature. However, the edible coating of alginate had no significant effect on the concentration of phenolics. Reque et al [30] studied antioxidant properties both

in blueberry juice (refrigerated at 4°C for 10 days) and fruit (frozen at -18°C for 6 months). The anthocyanin content decreased significantly in the frozen fruit (59%) and refrigerated juice (83%). It was also shown that the antioxidant status was kept constant during cold storage of the whole juice and fruit. However, the anthocyanins were degraded probably by oxidants and/or strong reactions with other phenolic compounds. In another research, effects of chitosan 1% and alginate 3% together with the extract of olive leaves were studied on the quality of sweet cherries [31].

The results showed that the ripening process and the increase in anthocyanin were delayed using coatings, especially those coatings composed of chitosan enriched with the olive leaf extract. In the end, it could be stated that edible coatings decrease evaporation and respiration rates by providing a barrier that maintains the quality of products, thereby increasing their shelf life. In addition, the olive leaf extract helps maintain fruit quality as a source of antioxidant and antimicrobial compounds.

5. Conclusions

In general, fresh pistachios' appearance and chemical properties, such as the moisture content, peroxide value, and phenolic compounds would be changed during the storage period. The results of this study showed that freezing (up to 45 days) and an alginate-based edible coating (especially at the concentration of 1%) helped maintain appearance and biochemical properties of fresh pistachios to some extent.

Conflict of Interest

The authors declare no conflict of interest.

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