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ORIGINAL ARTICLE

The Role of Perforated Packaging in Increasing the Shelf Life of Fresh Pistachios

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	ABSTRACT: Fresh pistachios do not have a long shelf life and should be consumed in a short period of time. In this
KEYWORDS	research, the effect of perforating in pistachio packaging on some quality characteristics of pistachio including
Peroxide value;	peroxide value, moisture content, mold and yeast count, and total microbial count of Ahmad Aghaei cultivar pistachio
Moisture content;	was investigated. The effect of 3 different levels of perforating including high perforating (by creating 6 holes in the
Microbial	package), medium perforating (by creating 3 holes in the package) and no perforating conditions were used in the
contamination;	packaging of pistachio samples. Then, the pistachio samples were kept at room temperature for 12 days and the quality
Packaging	characteristics were determined on the 3rd, 6th and 12th days of storage. The results showed that the pistachio sample
	with the most perforating (6 holes in the package) and the longest storage time (12 days) (S3T4) significantly had the
	highest amount of fat oxidation and peroxide value (1.03 meq kg ⁻¹). It was also observed that none of the samples
	stored for 3 days had any significant difference with the control sample (S0T0) which showed a peroxide value of 0.14
	meq kg ⁻¹ in this respect ($p < 0.05$). Pistachio samples with the highest perforating after 12 days of storage (S3T4)
	significantly had the lowest moisture content (39.92%) among other samples ($p < 0.05$), followed by samples with
	moderate perforating after 12 days of storage (S2T4) (42.74 %) and no perforating after the same time of storage
	(S1T4) (44.50 percent). The amount of mold and yeast in the sample without perforating after 12 days of storage
	(S1T4) (2566 cfu g ⁻¹) was significantly higher than the number of other samples ($p < 0.05$). Regardless of the amount
	of perforating applied in pistachio packaging, after 12 days of storage, the samples had the highest total microbial
	count and there was no statistically significant difference between them ($p < 0.05$), but these samples showed
	significant difference from other samples. The results of this research revealed that the amount of perforating applied
	in the packaging of Ahmad Aghaei cultivar pistachios has an important effect on the factors affecting the quality of
	pistachios during their short-term storage.

INTRODUCTION

Fresh pistachios do not have a long shelf life and should be consumed in a short period of time [1]. In recent years, consumption of fresh fruits and vegetables with low processing and high nutritional value has been noticed. Increasing the shelf life of fresh pistachios is a suitable solution that does not change the color of the pistachio hull and does not change its taste, smell, taste and texture. Because improper storage conditions or prolongation of the



storage period cause the skin on the fresh pistachio to crack and on the other hand, the color material is removed from the skin on the pistachio and causes the skin to become discolored and ultimately leads to a decrease in its quality. In this situation, due to the high humidity of the product and respiration of fresh pistachios, there is a risk of production and increase of aflatoxin, which threatens the health of the consumer [2-4].

In a research to evaluate the ability of aloe vera gel to reduce the quality loss of fresh pistachios after harvesting, in comparison with the natural polysaccharide chitosan, as a coating material with antifungal activity, was studied. The results showed that chitosan and aloe vera gel are potential candidates for maintaining the quality of pistachios after harvest. Their results showed that surface coating increases the resistance of fruit hull against gas permeability. Therefore, the internal atmosphere is modified and the breathing rate decreases. They concluded that any coating that reduces oil oxidation is more suitable for coating [5].

In another study, the effect of chitosan in combination with salicylic acid on some physicochemical, microbial, enzymatic and sensory characteristics of fresh pistachio fruit during storage in the refrigerator was investigated⁶. The weight loss as well as the values of peroxide value and free fatty acids of fresh pistachio fruit treated were lower than the control group at the end of the storage period. The activity of peroxidase enzymes in the treated pistachio fruit was significantly higher than the control group (p<0.05) and the lowest amount of peroxide value on day 28 was related to the treatment of chitosan and salicylic acid combination. Pistachio fruit treated with salicylic acid showed the lowest polyphenol oxidase activity. Also, the treatment of salicylic acid led to a significant superiority of the fruit color score among the samples. In addition, chitosan and salicylic acid treatments alone or in combination significantly reduced bacterial and fungal growth. They concluded that salicylic acid treatments individually and the combination of chitosan and salicylic acid can guarantee the safety and quality of fresh pistachio fruit in refrigerated storage [6].

In a research, the effect of alginate coating and Shirazi thyme on the storage quality of fresh pistachios was

investigated. The results showed that in uncoated fruits (control and thyme essential oil alone), the peroxide value increased with the increase in storage time, and its maximum value was observed in the fruits after 39 days of storage. In addition, the fruit peroxide value increased mainly by using alginate alone, and adding thyme essential oil to the edible coating formula decreased the peroxide value. After 39 days of storage, the lowest peroxide value (0.3meq kg⁻¹) was observed in the fruits coated with 1% alginate enriched with 0.3% thyme essential oil, and this treatment reduced the growth of mold and yeast. The highest peroxide value (1.5 meq kg⁻¹) was observed in fruits coated with 1.5% alginate after 39 days of storage. Samples treated with 1% alginate enriched with 0.3% or 0.5% thyme essential oil showed contamination of 1.308 and 1.544 log CFU g-1, respectively, compared to the control 3.89 log CFU g⁻¹) and other treatments showed significantly less contaminations [3].

In another study, edible coatings of gum arabic (6 and 8 %) with Shirazi thyme (0.3 and 0.5 %) as potential postharvest treatments to protect the quality of fresh pistachios at a temperature of 2 °C and a relative humidity of 85% was evaluated up to 36 days. The results showed that the combination of gum arabic 6% with thyme in concentrations (0.3 and 0.5%) reduced the color change and PPO activity of fresh pistachio compared to untreated control fruits. The fruit treated with 6% gum arabic alone and also with the addition of 0.3% thyme also had higher PAL and total phenol activity. The amount of free fatty acid and higher peroxide value were also found in fruits coated with 8% gum arabic containing 0.5% thyme. The results showed that the amount of peroxide value in all three compounds increased with time and after 36 days, the lowest amount of peroxide value was observed in gum arabic 6%, which was equal to 0.41 meq kg⁻¹ [7].

The effect of packaging fresh raw pistachios in vacuum, modified atmosphere and conventional packaging at 4°C for 30 days was investigated [8]. The results showed that the moisture content of fresh raw pistachios decreased with storage time, but no significant difference was observed between the moisture content of samples in different packaging systems during storage (p < 0.05). The highest amount of moisture was observed in vacuum packed pistachios at the end of storage. The moisture content of pistachios in the modified atmosphere after storage was lower than other packages. In this study, the microbial population increased during storage in vacuum-packed and conventionally packed samples. While the use of controlled atmosphere can significantly reduce the microbial population in fresh raw pistachios. Also, time and type of packaging showed a statistically significant difference on microbial growth.

In another study, modified atmosphere packaging was used to increase the shelf life of fresh pistachios in pistachio shells [9]. The quality of fresh shelled pistachios at 5°C and in three different gas conditions including MAP1 (10% oxygen, 20% carbon dioxide and 70% nitrogen), MAP2 (100% carbon dioxide) and ambient atmosphere for 42 days was investigated. Pistachios packed in modified atmospheric conditions showed a slight weight loss compared to the control sample. This could be due to the lower respiration rate in the modified atmosphere, which results in lower fresh product weight loss. In this study, the use of MAP can significantly slow the growth of the microbial population in fresh nuts. The results showed that the amount of microbial growth decreases with the increase of carbon dioxide concentration. Therefore, it was concluded that it can be more useful to store fresh pistachios in the shell in modified atmosphere packaging with 100% carbon dioxide and can increase the shelf life of fresh fruit in the pistachio core from 2 weeks to about 35 days [9].

In a research (2016), fresh pistachio samples of the Ohadi cultivar were exposed to ultraviolet rays and polyethylene terephthalate packaging (non-porous and porous) [10]. The results showed that ultraviolet radiation caused a significant decrease (p<0.01) in total microbial count and mold and yeast count in the irradiated samples. Also, at the end of the storage period, the microbial count in the samples exposed to ultraviolet radiation was significantly (p<0.05) lower than the control sample. Microbial population was observed less in non-porous packaging. The results of the present

study showed that ultraviolet radiation reduces microbial growth in fresh pistachio fruit and this treatment can be used as a suitable method to increase the shelf life of pistachio by reducing spoilage [10].

In a research (2021), the effect of polyethylene packaging with four perforation levels (0, 0.5, 1 and 1.5%), storage time (harvest, 30 and 60 days) and their interaction were investigated on pistachio nuts¹¹. They concluded that for a time of about 30 days, 0.5% perforation in polyethylene packaging is somehow able to preserve some qualities of fresh pistachios such as firmness, lipid content and chlorophyll content of kernels and carotenoid content and chroma of hulls. While for a longer time of about 60 days, 1 and 1.5% perforation are more effective for preservation of some properties such as firmness, and anthocyanin content of hulls and firmness, lipid content, antioxidant activity, phenolic content, and flavonoid content of kernels. Therefore, 0.5 and 1.5% perforation levels are suggested for 30- and 60-days storage periods respectively [11].

Objective

In this research, the effect of packaging of fresh pistachios with different levels of perforating on the quality and shelf life of fresh pistachios was investigated.

MATERIALS AND METHODS

The pistachio used in this research was the Ahmed Aghaei variety, which was obtained from the local market in Kerman. In order to investigate the effect of different perforating rates in pistachio packaging on the quality characteristics of pistachios, first, 3 kg of pistachios were divided into 200 gram packages and placed in disposable containers. In order to create different perforating, different number of holes in the packaging containers were used. For this purpose, 6 holes (S3) were used to apply high perforating in the packages. Also, non-perforated packaging (S1) was prepared for comparison. The samples were kept at ambient temperature for 0 days (T0), 3 days (T1), 6 days (T2), 9 days (T3) and 12 days (T4) and the following tests were performed on them.

Peroxide value

5 grams of the fat sample was weighed and poured into a 250 ml Erlenmeyer flask. 30 ml of acetic acid and chloroform solution was added to it and it was slowly rotated to mix the solution and the fat inside the Erlenmeyer flask. 0.5 ml of saturated potassium iodide was added to it. The Erlenmeyer was closed and mixed for a full 60 seconds without drawing air into the solution. Then the Erlenmeyer lid was removed and 30 ml of freshly boiled and cooled distilled water was added to it. Immediately titrate the released iodine with a few milliliters of 0.1 normal sodium thiosulfate solution until the color of the solution changes from orange yellow to pale yellow. By adding 0.5 milliliters of starch solution, a purple color is created, which continues with the titration. It becomes colorless. As soon as the color of the solution disappeared, the titration was stopped. The amount of peroxide (PV) was calculated in milliequivalents of active oxygen per kilogram using the following formula:

$$\frac{(V-V_0).\,C_{thio}.\,F\times 1000}{m}$$

The parameters of this equation are as follows:

V: the volume of the normal 0.01 sodium thiosulfate standard solution used for the test in milliliters;

 V_0 : the volume of normal 0.01 sodium thiosulfate standard solution used for the control in milliliters;

 C_{thio} : Approximate concentration of normal 0.01 sodium thiosulfate standard solution in terms of moles per liter; m: test mass in grams

F: Solubility factor of sodium thiosulfate 0.01.

Moisture content

two grams of laboratory sand was poured into a container and placed in a greenhouse at a temperature of 70 °C for 2 hours. Then we put the container inside the desiccator to reach the laboratory temperature. Then, we weighed it with a laboratory scale. We weighed 5 grams of the ground sample and spread it completely inside the container. We recorded the weight of the sample and placed it in a greenhouse at a temperature of 70 degrees Celsius for 6 hours. After drying, we removed the container from the greenhouse and cooled it in a desiccator until it reached the laboratory temperature. Then we weighed the container along with the sample and put it in the following formula and calculated the percentage of goodness. It was done according to the national standard of Iran number 672. After preparing the sample and doing the necessary steps, the humidity was determined from the following formula:

Moisture percentage by weight $= \frac{M_1 - M_2}{M_1 - M_0} \times 100$ which in this equation:

M1: weight of container with lid and sample before drying M2: weight of container with lid and sample after drying M0: the weight of the empty container with lid.

Total count of microorganisms (mold and yeast)

Using a sterile pipette, we transferred 1 ml of the sample (the dilution prepared from the 10^{-1} sample) into the sterile plate. Then 12 to 15 ml of plate count agar culture medium with a temperature of 44 to 47 °C was poured into each plate. The interval between preparing the initial suspension and adding the culture medium should not be more than 45 minutes. We carefully mixed the plates with the culture medium by rotating motion and placed them on a horizontal and cool surface until they solidified completely. In cases where the product probably had microorganisms whose colonies grew excessively on the surface of the culture medium, after the plates were completely solidified, in about 4 ml of the culture medium, the top layer or plate count agar at a temperature of 44 to 47°C was poured on the surface of the inoculated culture medium, then it was placed horizontally to cool and solidify. The prepared plates were placed at 30°C for 72 hours. The total counting of microbes was done based on the national standard of Iran No. 8923 Microbiology of food and animal feed - test preparation - initial suspension and decimal dilutions for microbiology tests.

Statistical analysis

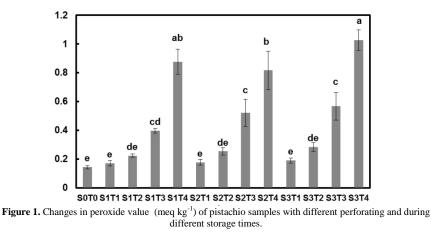
In order to statistically analyze the results of a completely

random statistical design in the form of a factorial test and mean comparison and data analysis was done with Minitab 20 software.

RESULTS

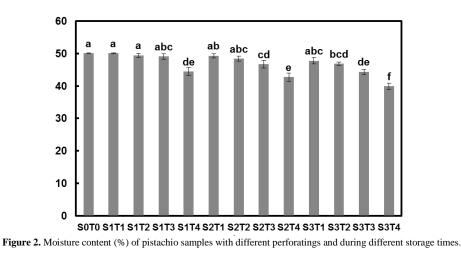
Peroxide value

Peroxide value is one of the quality parameters that are often determined during production, storage and marketing. The peroxide value shows the degree of oxidation in the substance and measures the total amount of peroxides as the primary product of oil oxidation. The pistachio sample with the most perforating (6 holes in the package) and the longest storage time (12 days) (S3T4) significantly had the highest amount of fat oxidation and peroxide number (Figure 1) (1.03 meq kg⁻¹). After this sample, pistachio samples with moderate perforating (3 holes in the package) (S2T4) and without perforating (S1T4) showed the highest amount of peroxide value after 12 days of storage. It was also observed that none of the samples stored for 3 days had any significant difference with the control sample (S0T0) which showed a peroxide number of 0.14 (meq kg⁻¹) in this respect (p < 0.05).



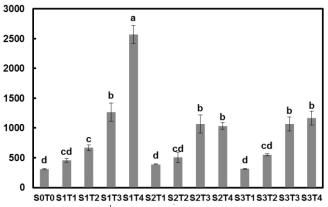
Moisture content

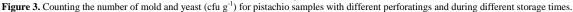
Pistachio samples stored in non-ventilated conditions until the 9th day of storage have no significant difference in terms of moisture content with the control sample (Figure 2) (p < 0.05), but on the 12th day this difference becomes significant (p < 0.05). For samples with moderate perforating up to day 6 and for samples with high perforating up to day 3 of storage, no significant difference was observed with control samples (p < 0.05). Pistachio samples with the highest perforating after 12 days of storage (S3T4) significantly had the lowest moisture content (39.92%) among other samples (p < 0.05), followed by samples with Average perforating after 12 days of storage (S2T4) (42.74 %) and no perforating after 12 days of storage (S1T4) (44.50 %) were located.



Counting mold and yeast

As it is clear from the results (Figure 3), the amount of mold and yeast in the non-ventilated sample after 12 days of storage (S1T4) (2566 cfu) was significantly and with a significant difference higher than that of the other samples. For samples with moderate perforating (S2) and high perforating (S3), there was no significant difference between the 9th and 12th days of storage in terms of mold and yeast counts. Also, the results showed that the number of mold and yeast in the pistachio samples, regardless of the amount of perforating applied to them, did not differ significantly from the control sample 3 days after storage (p < 0.05).





Total microbial count

Regardless of the amount of perforating applied in pistachio packaging, the samples had the highest total microbial count after 12 days of storage and there was no statistically significant difference between them (p < 0.05) (Figure 4), but this sample were significantly different from other samples stored in 3 and 6 days (p < 0.05). In general,

the amount of microbial population increased with the increase of storage time, but there was no statistically significant difference between the storage time of 3 and 6 days for all samples (p < 0.05). Also, these samples did not show any significant difference with the control sample (p < 0.05).

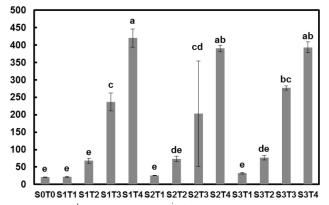


Figure 4. Total microbial count (cfu g⁻¹) for pistachio samples with different conditions and during different storage times.

DISCUSSION

From the results obtained for the peroxide value of pistachio samples, it can be concluded that the pistachio storage time is a more important factor in increasing the peroxide value, although the amount of perforating also had an almost direct effect on the peroxide value. Peroxide values obtained in this research for pistachio variety Ahmad Aghaei for packaging without perforating in the range of 0.17-0.88, for packaging with medium perforating in the range of 0.18-0.82 and for packaging with high perforating in the range. It was 0 meq kg⁻¹ on 0.19-1.03. The Previous studies have shown that chitosan and aloe vera gel are potential candidates for maintaining pistachio quality and reducing fat oxidation after harvest [12]. The lowest peroxide value of the coated pistachios was related to the chitosan coating of 0.05%, which was reported as 0.3 meq kg⁻¹. After that, 50% aloe vera gel and 0.5% chitosan were covered respectively. These results showed that the surface coating increases the resistance of the fruit skin against gas permeability, so the internal atmosphere is modified and the respiration rate decreases. They concluded that any coating that reduces oil oxidation might be more suitable for coating. Increasing the storage time of fresh pistachios decreases its quality Treatments such as freezing (especially for 1.5 months) can help preserve some characteristics of fresh pistachios, including peroxide value [12]. The use of edible alginate coating, especially at a concentration of 1%, preserved moisture and also reduced peroxide value (especially during the 3-month freezing period) in fresh pistachios. The results have shown that the combination of chitosan with salicylic acid in the coating of pistachio nuts causes a decrease in the peroxide value during 28 days of pistachio storage at refrigerator temperature (4 degrees Celsius) [6]. The results of the previous research showed that the amount of peroxide value increased in uncovered pistachios with the increase in storage time, and its maximum value was observed in pistachios stored at the maximum time studied (39 days) [7]. In addition, the peroxide values of pistachios mainly increased with alginate coating alone with increasing storage time, while the addition of thyme essential oil to the alginate coating composition decreased the peroxide value. After 39 days of storage, the lowest peroxide value (0.3 meq kg⁻¹) was observed in fruits coated with 1% alginate enriched with 0.3% thyme essential oil. This value obtained for the peroxide value of the coated pistachio is much lower than the values obtained during 12 days after storage for the pistachio samples in all the perforating conditions tested in the present research.

Moisture is one of the important quality factors of dried fruit quality. It is clear that as the pistachio storage time increases, the moisture content of the samples decreases more. The reason for this is that there is more opportunity for moisture loss. Higher perforating and more air circulation on pistachios can also increase the rate of evaporation of moisture and in this sense help to further moisture reduce. Of course, it should be noted that the relative humidity difference between the air and the pistachio plays a decisive role in the moisture loss rate, and the lower the relative humidity of the air (the higher humidity difference), the faster the pistachio's moisture loss in the vicinity of the air. To show the rate of moisture loss due to the difference between the relative humidity of the air and the moisture content of the pistachio itself, we can refer to the results of a previous similar study [13]. This study reported that if pistachios are roasted at 150 degrees Celsius, their moisture content decreases to about 1%, and storing these pistachios for a month increases their moisture content to about 1.2% due to the absorption of moisture from the surrounding air. The rate of moisture transfer between the food item and the surrounding atmosphere is reduced by completely covering the food item with a film or edible coating. The previous study [14] have shown that the coating of pistachio kernels with chitosan causes the moisture content of pistachio samples to remain almost unchanged during 6 months of storage and at the same initial moisture level, while for uncoated samples, changes in moisture content were observed during storage. The results have shown that the use of alginate coating, especially at a concentration of 1%, preserves the moisture of pistachio samples [12]. Edible coatings create a barrier between air and produce that reduces water loss and limits gas exchange, which in turn slows respiration and fruit moisture loss.

The results obtained in this research showed that the amount of perforating does not have much effect on the amount of mold and yeast, but the storage time of pistachios has a significant effect on the amount of this population. Almost the same result was obtained regarding total microbial counts for pistachio samples stored in different perforating conditions during 12 days of storage. The research results⁷ have shown that the use of plant extracts that have an antibacterial role, such as thyme extract, can play an effective role in reducing the population of mold and yeast in pistachios during storage.

CONCLUSIONS

Pistachios are one of Iran's valuable, nutritious, energyrich, export products, which are mainly dried and used for snacks due to storage problems. A very small amount of it is consumed fresh at the time of harvest. Fresh pistachios have a short shelf life, and after harvesting, they cannot be kept healthy for a long time, because the color of the outer soft hull gradually darkens and it suffers weight loss and fungal contamination. In this research, the effect of packaging of fresh pistachios with different levels of perforating on the quality and shelf life of fresh pistachios was investigated. The results revealed that the amount of perforating applied in the packaging of Ahmad Aghaei cultivar pistachios has an important effect on the factors affecting the quality of pistachios during their short-term storage.

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Conflict of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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