

The influence of freezing conditions on the organoleptic attributes of Iranian leafy vegetable foods

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ABSTRACT

The edible leafy vegetables contain nutritional ingredients that are necessary for human health and it is important that nutrients protection be monitored during processing and storage. The aim of this study was to study some organoleptic attributes of a very popular Iranian meal named Coco-Sabzi, which was prepared with a mixture of edible grinded leafy vegetable pre-stored at different frozen conditions. So, by sensorial evaluation we can conclude about nutritional loss of products. The mixture of five edible grinded leafy vegetables including *Allium ampeloprasum* var. *porrum*, *Lepidium sativum*, *M. spicata* (*M. sativa*), *Ocimum basilicum* and *Allium porrum* were stored at -9, -12 and -18°C for 120, 150 and 180 days. The organoleptic attribute of the prepared Coco-Sabzi was compared with the above three different time-temperature combinations during the frozen storage period. Results indicated that the best colors were observed at -18, -12 and -9°C, respectively. Taste and overall acceptability at -18°C after 120 and 150 days and also at -12°C after 180 days ranked 1st ($P < 0.05$). Data analysis showed that the color, taste and overall acceptability of samples were not statistically different at three different time-temperature combinations during the frozen storage period. As a result, organoleptic attribute during six months of frozen storage was affected by freezing temperature, not by frozen storage period.

Keywords: Sensory evaluation; Leafy vegetables; Freezing conditions; Iranian meal; Storage

INTRODUCTION

Considering the development of societies and special interest in ready-to-use food in the developed/developing countries, consumption of vegetables has increased. Nowadays, consumers are aware of the need to consume a variety of fresh vegetables for getting the most nutritional value and complete antioxidant support [1]; however, many people do not have the opportunity to eat fresh vegetables every day and they frequently use frozen vegetables or cooked vegetables [2]. Thus, maintenance and improvement of postharvest quality of fresh vegetables is becoming increasingly important [3]. For preserving any type of food products, the retention of nutritional components is a critical point; for this reason, freezing is probably the least destructive method for long-time storage if properly applied [4]. But, as frozen storage can induce chemical, microbial and organoleptic changes during time, it is important to study the quality factors

thoroughly. Chemical and microbial changes are generally intrinsic factors that can affect final products directly or indirectly whereas organoleptic parameters are classified as extrinsic factors/quality. Loss of quality factors like organoleptic attributes including color, flavor and overall acceptability can reduce the acceptances of products by consumers. The quality of frozen products and consumer acceptability can be enhanced by optimizing the processing conditions such as freezing temperature and storage time condition [4].

It is well known that one of the destructive sources during frozen storage time which affects color and taste is enzymatic activity. This reaction can be reduced by controlling the time-temperature combination during freezing [5]. Since vegetables have a great share in daily diets, quality losses of their nutritional content should be minimized. Furthermore, consumption of leafy vegetables is very popular in Asian countries, especially in Iran, and these

leafy vegetables are very susceptible to different deteriorations during cold storage. Considering these factors, the present study is undertaken in order to develop the best time-temperature combination during the frozen storage of leafy vegetable mixtures which are subjected to minimal processing and to study their storage stability.

MATERIALS AND METHODS

Ready-to-use and edible grinded leafy vegetable mixtures (a typical and popular mixture for preparing Coco-Sabzi, an Iranian meal mainly made of leafy vegetables and eggs) were stored at -9 , -12 and -18°C for 120, 150 and 180 days and, subsequently, Coco-Sabzi was prepared. Their organoleptic attributes after each freezing time-temperature combination were compared using observation methods and questionnaire completion [6]. The study population comprised of packaged ready-to-use fresh leafy vegetable mixtures prepared for making Coco-Sabzi and included *Allium ampeloprasum* var. *porrum*, *Lepidium sativum*, *M. spicata* (*M. sativa*), *Ocimum basilicum* and *Allium porrum*. These five popular vegetables are commonly used in Iranian meals and are widely available in bulk and in comminuted package in supermarkets and groceries.

Sample preparation

Allowing for three replicates, a total of 90 packages of edible grinded leafy vegetable mixtures were selected from stores at three different regions of Tehran using the cluster sampling method [6]. Sampling of each package was done in a completely randomized method [7].

Preparation of leafy vegetable mixtures was carried out using fresh ready-to-use leafy vegetables on the day of production. Each package of leafy vegetable mixtures was assigned a four-digit alphanumeric code indicating the type of leafy vegetables, freezing temperature, frozen storage period and replication frequency. The samples were put in the CoolSelect Zone and freezer compartments of a Samsung RL44QEUS refrigerator/freezer (Seoul, South Korea). For temperature monitoring, a JR-1001 temperature data logger (ACR Systems Inc., Surrey, BC, Canada) with a functional ability in the range of -40 to 85°C was placed in each freezer. The sensors recorded the temperature every 10 min and the data were extracted using TrendReader Express

2.22 software (ACR Systems Inc., Surrey, BC, Canada) every 15 days. At each stage of sensory evaluation, there were three packages of ready-to-use vegetable mixtures for each temperature [6].

Coco-Sabzi preparation

The content of each package of pre-frozen edible leafy vegetable mixture (380 ± 20 g) was mixed with 1.5 g salt and 3 whole eggs and was stirred for 30 s. The mixture was fried in 4 tablespoons of vegetable oil in a large pan with 28 cm diameter for 40 min (20 min each side).

Microbiological analysis The total count of viable micro-organisms (TVC) was determined on a plate count agar according to ISO 4833 [8] using the pour plate method. The plates were incubated at 30°C for 72 h. Coliforms (including faecal coliforms) were enumerated in lauryl sulfate tryptose broth according to ISO 4831 [9] using the most probable number (MPN) technique. The plates were incubated at 37°C for 48 h. *Escherichia coli* was enumerated according to ISO 7251 [10] using the MPN technique. The plates were incubated at 37 and 44°C for 48 h, respectively. *Salmonella* was enumerated according to ISO 6579 [11].

Parasite larvae were looked for and counted in saturated saline solution using the flotation method [12]. The microbiological analyses were conducted on 180th day of storage at all three temperatures. The results (\log_{10} colony-forming units (CFU) g^{-1} for TVC, coliforms, *E. coli* and *Salmonella*; number g^{-1} for parasites) were presented as the average of duplicate measurements of three replicates for each freezing conditions. All media were purchased from Merck (Darmstadt, Germany) or Sigma-Aldrich (Munich, Germany).

Sensory analysis

Assessors

Fifty untrained assessors were collected and sensory evaluation carried out using the ranking method. Assessors were selected on the base of interest and understanding of evaluation method. They consumed Coco-Sabzi previously and were informed how to complete the sensory sheets. Each time, six persons separately evaluated the samples in six special chambers at the same time. Evaluation was carried out under white and yellow fluorescent lights (similar to daylight) and ambient temperature (25°C). Order of sample presentation to each assessor was different. Each assessor was requested to wash his/her mouth with mineral water (20°C)

and, if necessary, use salt-free biscuit before testing and after each evaluation. Among 50 assessors, 10 were eliminated and 40 persons were remained until the end of storage period.

Sensory procedure

Prepared Coco-Sabzi, sliced in 7 cm length and 3 cm width pieces, was placed randomly on a disposable white microwavable plate with no color or odor. Each plate had one of the codes of A, B or C. Before evaluation, the plate along with the prepared Coco-Sabzi was warmed using a microwave oven with the power of 700 W for 20 s. Evaluation was such that rank 1 belonged to the best quality and rank 3 belonged to the least one/quality.

All of variables including microwave power, waming time, Coco-Sabzi preparation and

cooking procedure remained constant. The treatments were manufactured with the same raw materials. All assessments were conducted on the day of Coco-Sabzi preparation.

Statistical analysis

Microbial data were subjected to one-way analysis of variance (ANOVA) using SPSS 12.1 statistical software (SPSS Inc., Chicago, IL, USA). Data from sensory evaluation were analyzed by the Kruskal–Wallis H non-parametric test .The Mann–Whitney U test was used to determine the statistically significant differences among the means. A 95% ($P < 0.05$) significance level was considered in all comparisons [13].

Table 1. Microbial characteristics of prepared Coco-Sabzi made of ground frozen leafy vegetable mixtures after 180 days of storage at different freezing temperatures

Freezing Temperature (°C)	T.V.C (log ₁₀ CFU g ⁻¹)	Coliforms (log ₁₀ CFU g ⁻¹)	E.coli (log ₁₀ CFU g ⁻¹)	Salmonella (log ₁₀ CFU g ⁻¹)	Parasite Larvae (Number g ⁻¹)
-9	3.0	ND	ND	ND	ND
-12	3.8	1.7	ND	ND	ND
-18	3.7	ND	ND	ND	ND

Values are mean of three replicates for each treatment. ND, not detected.

Table 2. Ranking of organoleptic attributes for the prepared Coco-Sabzi of ground frozen leafy vegetable mixtures after 120 days of storage at different freezing temperatures

Freezing Temperature (°C)	Color ^{A,B,C}	Taste	Overall Acceptability
-9	2.41±0.82 ^a	2.44±0.75 ^a	2.45±0.76 ^a
-12	2.00±0.65 ^b	1.87±0.73 ^a	1.92±0.75 ^a
-18	1.54±0.76 ^c	1.67±0.77 ^b	1.63±0.75 ^b

^A Values are mean ± standard deviation of 40 replicates for each treatment.

^B Means with different letters in the same column are significantly different ($P < 0.05$).

^C For each attribute the lowest value denotes highest quality and the highest value denotes lowest quality.

Table 3. Ranking of organoleptic attributes for the prepared Coco-Sabzi of ground frozen leafy vegetable mixtures after 150 days of storage at different freezing temperatures

Freezing temperature (°C)	Color ^{A,B,C}	Taste	Overall Acceptability
-9	2.54±0.72 ^a	2.08±0.77 ^a	2.26±0.68 ^a
-12	1.92±0.58 ^b	2.21±0.73 ^b	2.08±0.77 ^b
-18	1.54±0.82 ^c	1.72±0.89 ^b	1.67±0.90 ^b

^A Values are mean ± standard deviation of 40 replicates for each treatment.

^B Means with different letters in the same column are significantly different ($P < 0.05$).

^C For each attribute the lowest value denotes highest quality and the highest value denotes lowest quality.

Table 4. Ranking of organoleptic attributes for the prepared Coco-Sabzi of ground frozen leafy vegetable mixtures after 180 days of storage at different freezing temperatures

Freezing Temperature (°C)	Color ^{A,B,C}	Taste	Overall Acceptability
-9	2.71±0.61 ^a	2.34±0.85 ^a	2.37±0.82 ^a
-12	1.95±0.57 ^b	1.79±0.70 ^a	1.79±0.66 ^a
-18	1.34±0.63 ^c	1.87±0.81 ^b	1.84±0.86 ^b

^A Values are mean ± standard deviation of 40 replicates for each treatment.

^B Means with different letters in the same column are significantly different ($P < 0.05$).

^C For each attribute the lowest value denotes highest quality and the highest value denotes lowest quality.

RESULTS AND DISCUSSION

Microbiological evaluation

Microbiological characteristics of prepared Coco-Sabzi made of edible grinded frozen leafy vegetable mixtures are presented in Table 1. TVC of microorganisms was lower than the permitted limit and far from the critical value. Enumeration of Coliforms, *Escherichia coli*, *Salmonella* and Parasite larvae are indicative of adequate hygienic conditions.

Sensory evaluation for determination optimum freezing temperature

Effect of freezing temperature after 120 days

The effect of freezing temperature of the edible grinded leafy vegetable mixture at -9, -12 and -18°C after 120 days (Table 2) on the organoleptic attributes of prepared Coco-Sabzi indicated that color rank from 1 to 3 belonged to -18, -12 and -9°C, respectively. The color of samples stored at these temperatures had significant differences to each other ($P<0.05$). Ranks 1 to 3 of taste evaluation belonged to -18, -12 and -9°C, respectively. The results showed the significant difference ($P<0.05$) at -18°C from two other temperatures; but, sample tastes at -12°C and -9°C had no significant differences from each other. For overall acceptability, ranks 1 to 3 were allotted to -18, -12 and -9°C, respectively. The overall acceptability of samples at -12 and -9°C had no significant differences from each other; but, both of them had significant differences ($P<0.05$) from the overall acceptability at -18°C.

Effect of freezing temperature after 150 days

The effect of freezing temperature of the edible grinded leafy vegetable mixtures at -9, -12 and -18°C after 150 days (Table 3) on the organoleptic attributes of prepared Coco-Sabzi showed that the color ranks from 1 to 3 was significantly ($P<0.05$) related to -18, -12 and -9°C, respectively. Taste ranks of 1 to 3 belonged to -18, -9 and -12°C, respectively. The results showed that taste at -9°C had significant differences ($P<0.05$) from that at -12 and -18°C whereas taste at -12 and -18°C showed no significant differences from each other. Overall acceptability ranks from 1 to 3 belonged to -18, -12 and -9°C, respectively. The overall acceptability at -9°C had significant differences ($P<0.05$) from the overall acceptability at -12 and -18°C; but, the two last temperatures (-12 and -18°C) had no significant difference from each other.

Effect of freezing temperature after 180 days

The effect of freezing temperature of the edible grinded leafy vegetable mixtures at -9, -12 and -18°C after 180 days (Table 4) on the organoleptic attributes of prepared Coco-Sabzi indicated that color ranks from 1 to 3 were significantly ($P<0.05$) related to -18, -12 and -9°C, respectively. Taste ranks from 1 to 3 were related to -12, -18 and -9°C, respectively. The results showed that taste at -12 and -9°C had no significant difference from each other; but, both of them showed significant differences ($P<0.05$) from the taste at -18°C. Ranks of overall acceptability from 1 to 3 were related to -12, -18 and -9°C, respectively. The overall acceptability at -12 and -9°C had no significant difference from each other; but, they had a significant difference ($P<0.05$) from the overall acceptability at -18°C.

Discussion

The freezing condition does not significantly destroy the microorganisms that may be present in vegetables [4]. Thus, the microbial results are due to the hygienic processing and packaging of the vegetables mixture. However, comparison with other research is difficult owing to differences in raw materials and preparation procedures.

It is conceivable that compounds related to green color are more resistant to freezing than those related to yellow/red [2] although this phenomenon could not be observed in prolonged storage period. According to Baardseth [14], no off-flavor and off-odor were observed for the frozen leek during 15 months of storage at -30°C; but, the color changed from green to yellow-green. Also, no significant differences of organoleptic attributes were observed between storage at -20°C and -30°C [14]. In another study, during 9 months of freezing, the level of chlorophyll did not significantly reduce in frozen products, except for non-blanched and blanched parsley leaves stored at -20°C compared to -30°C [15]. In some of leafy vegetables which contain thiol structures in their functional groups (such as spinach), the high initial vitamin C content has a direct correlation with the sulfur content. In this leafy vegetables due to an enzyme-catalyzed reaction in which reduced glutathione (a sulfur-containing tripeptide) reduces dehydroascorbic acid to ascorbic acid in chloroplasts, a good overall acceptability could be observed.

Enzymes such as lipase, peroxidase and catalase contribute to worsening features of organoleptic attributes and their activity declines in prolonged freezing times (after 6 months). In the present study, at -9°C , the overall acceptability had a worst value in comparison with two other temperatures (-12°C and -18°C) that may be due to the higher rate of reduction reactions catalyzed by enzymatic activity at this temperature which is in accordance with the findings of Lisiewska and Kmiecik [16] and Giannakourou and Taoukis [17].

CONCLUSION

Analysis of sensory evaluation data showed that the best storage temperatures of edible

grinded leafy vegetable mixtures are -18 , -12 and -9°C , respectively. Length of storage period (180 days) does not have any effect on organoleptic parameters. However, organoleptic parameters of Coco-Sabzi mixtures stored for six months are affected by freezing temperature, not freezing period.

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