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Biochemistry

Influence of Storage Temperature on Chemical and Microbial Quality of Carotene Rich Pumpkin Powder

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Abstract

Attempts were made to store the carotene rich pumpkin powder at 30 ± 2°C & 62±5% RH and 7 ± 1°C & 80±5% RH in aluminum laminated flexible pouch for the period of 180 days. The shelf life was determined on the basis chemical and microbiological analysis of stored powder at an interval of every 15 days. As the storage period was increased from 0 to 180 days, the carotene retention was found to decrease. More carotene could be retained (82.95%) when pumpkin powder was stored at 7°C as compared to 52.28% retention at 30°C when the stored for 180 days. Standard plate count of pumpkin powder stored at 7°C was found to be only 440 cfu / g at the end of 180 days of storage. Similarly, yeast and mold count observed at the end of 180 days storage was only 7 cfu/g. Coliform was found to be absent throughout storage. Pumpkin powder was found to be more stable upto only 75 days, if stored at 30°C. However, it can be safely stored for almost 180 days, if stored at 7°C.

Highlights

- Pumpkin powder was found to be rich in carotene content (42.2 mg/100g) which is precursor of vit A.
- Storage temperature and period had a significant effect on carotene retention in the pumpkin powder.
- 82.95% carotene retained in the pumpkin powder when stored at 7 + 10C for 180 days without microbiological spoilage.

Keywords: Carotene, pumpkin powder, SPC, yeast and mold, coliform, storage, temperature.

Pumpkin is extensively cultivated throughout the world and its preservation is of commercial importance. Pumpkin is traditionally consumed in the form of freshly boiled, steamed or as a processed food items such as soup or curry. In many countries pumpkin is one of the main vegetable products. It has been used traditionally as medicine in many countries such as China, Yugoslavia, Argentina, India, Mexico, Brazil and America (Jia *et al.*, 2003, Adolfo and Michael, 2005). It is characterized by taste, colour and flavor. It also provides several vitamins, minerals and other valuable nutrients. Pumpkin is high in β-carotene, vitamins, minerals, pectin and dietary fiber (Bhaskarachary *et al.*, 2008 and Djutin, 1991). Among carotenoids, β-carotene is most widely used in food industry, it provides uniform natural colour, enhances colour, imparts yellow colour and adds to vit A activity. (Gayathri and Prakash, 2003; Lee, 1983). Extent of deficiency of vit A in the diets of male and female is 80 and 84%, respectively and is the highest among the deficiency of other micronutrient i.e. iron (62%), riboflavin (50%), vit C (30%) (NNMB, 2002).Consumption of foods containing carotene helps in prevention of eye disorders, cancer and skin diseases (Bendich, 1989). Incorporation of β-carotene rich foods in diets is the best measure to improve vit A nutrition of individuals to overcome the problems and diseases caused by vit A deficiency (Chandrashekhar and Kowsalya, 2002; Siems *et al.* 2005). Pumpkin powder is used because of its highly-desirable flavour, sweetness and deep yellow-orange colour. It has been reported to be used to supplement cereal flours in bakery products, for soups, sauces, instant noodle and spice as well as a natural colouring agent in pasta and flour mixes. To enhance the shelf life of pumpkin powder and to make it easily available to the consumers, a technology was developed for the manufacture of carotene rich pumpkin powder (Kulkarni and Joshi, 2013). Like all other products the quality of pumpkin powder also deteriorates during storage. The changes in chemical composition and microbiological quality of pumpkin powder during storage are of utmost importance from commercial point of view, as they are most important attributes that govern the consumer acceptability and storage life (Khamrui and Pal, 2003). The effect of storage temperatures on the chemical and microbiological quality changes of pumpkin powder packed in aluminum laminated metalized flexible pouches are reported in this paper.

Materials and Methods

Raw Material

Seeds of popularly grown pumpkin (*Cucurbita maxima*) variety MPH-1 in Anand region were procured from the local market. These seeds were cultivated on the AAU farm. The pumpkin fruits were harvested at a typical maturity of 100 days. These fruits were stored in the laboratory under ambient conditions (27$\pm$3°C, 62$\pm$5%RH).

Chemical composition of edible portion of pumpkin fruit and powder

The fresh pumpkin fruit pulp and powder prepared from it were analyzed for moisture, total soluble solids, carbohydrate, protein, fat, crude fibre, ash, carotene, ascorbic acid, phosphorus, calcium, iron and pH value. The standard prescribed methods for analysis employed (AOAC, 1990). All chemicals used in the experiment were of analytical grade from reputed manufacturers like Qualigens, E-Merck etc

Pumpkin powder production

The peeled pumpkin fruit was converted into 10 mm size cubes and subjected for pretreatments such as blanching (temperature 94°C, time 2 min.) and sulphitation for 10 min carried out in 500 ppm solution of $K_2S_2O_3$ prior to drying. The pretreated cubes of pumpkin fruit were dried for preparation of powder using vacuum dryer. Vacuum drying of pumpkin cubes was carried out at 80^{O}C & 700 mm Hg vacuum (Kulkarni and Joshi, 2013). The Pumpkin powder was analyzed for carotene content by the method described by Jensen (1978).

Storage of Pumpkin Powder

Storage study of pumpkin powder produced using all the optimized parameters was carried out for 6 months both at ambient temperature i.e. 30$\pm$2°C & 62$\pm$5% RH and at 7$\pm$1°C & 80$\pm$5% RH. The shelf life was determined on the basis of chemical and microbiological analysis of stored powder. A 1 g of sample was filled in a small aluminum polythene laminated bag and packed air tightly. These bags were kept in the laboratory at ambient temperature and also in refrigerator maintained at 7°C in dark condition and studied for following parameters.

Total carotene content

The powder samples were analyzed both initially (0 day of storage) as well as thereafter at every 15 days interval for carotene stability. Percent carotene was estimated by the method given by Jensen (1978). Analysis was carried out in duplicate and the average values are reported.

Microbiological analysis

Initially (0 day) and thereafter at every 15 days regular interval, the samples stored at both room temperature and at 7^{O}C in duplicate were analyzed for microbiological quality. The samples were subjected to analysis for standard plate count (SPC) using plate count agar, coliform count using violet red bile agar (VRBA) and yeast and mold count using freshly prepared acidified (pH adjusted to 3.5 by sterile 10 per cent tartaric acid solution) potato dextrose agar as per the standard procedure (Ranganna, 1986). A 1 g of sample was dissolved in 99 ml sterile water to obtain 1:100 dilutions. Subsequently, 5 ml of the dilution was poured in a set of sterile petri plates in duplicate. The data obtained from the replicates during the investigation were analyzed statistically for the analysis of variance as described by Snedecor and Cochran (1994).

Results and Discussion

Chemical composition of edible portion of pumpkin fruit

The data obtained regarding the chemical composition of fresh pumpkin pulp i.e. moisture, TSS, carbohydrate, protein, fat, crude fibre, ash, carotene, ascorbic acid, calcium, phosphorous, iron content etc. are presented in Table 1. All determinations are carried out in triplicate and the average values are reported.

Table 1. Chemical composition of pumpkin pulp

Sr. No.	Parameter	Composition[a]
1	Moisture (%)	95.06±0.482
2	TSS (^{O}Bx)	3±0.00
3	Carbohydrate (%)	2.65±0.151
4	Protein (%)	1.44±0.031
5	Fat (%)	0.34±0.028
6	Crude fibre (%)	0.77±0.015
7	Ash (%)	0.51±0.027
8	Carotene (mg/100 g)	3.0±0.028
9	Vit. C (mg/100 g)	2.42±0.031
10	Calcium (%)	0.026±0.004
11	Phosphorous (%)	0.036±0.002
12	pH	6.48±0.1
13	Pottassium (%)	0.07±0.03
14	Iron (ppm)	8.21±0.17

[a] Mean ± SD

The results indicate that the pumpkin pulp had very high moisture content of 95.06% (wb), thus pumpkin fruit is grouped under the category of perishable commodity. The protein and fat were estimated to the tune of 1.44 and 0.34%, respectively, which are very less. Carbohydrate and crude fibre were found to be moderately high i.e. 2.65 and 0.77%, respectively. The content of micronutrients in the pumpkin pulp such as carotene (3 mg /100 g) was found to be very high while vitamin C and minerals such as Ca, P, Fe and K were also present in considerable amounts.

The results obtained are in good accordance with the results reported by several other investigators for pumpkin (Teotia., 1992; Pawar *et al.,*1985; Park *et al.,*1997; *Lee et al.,*2002). However, the possibility of differences in the composition of pumpkin pulp may be due to variety, stress, environment, geographical and cultivation practices. Nevertheless, there is good measure of agreement with regard to carotene content of the fruit.

Chemical composition of pumpkin powder

Pumpkin powder as produced using optimized processing, was analyzed for moisture, carbohydrate, protein, fat, crude fibre, ash, carotene, vit. C, calcium, phosphorus, potassium, iron content and water activity (aw). All determinations were carried out in triplicate and the average values are reported in Table 2.

Table 2. Chemical composition of pumpkin powder

Sr. No.	Parameters	Composition[a]
1	Moisture (% db)	3.65± 0.01
2	Carbohydrate (%)	83.19± 0.59
3	Protein (%)	6.21± 0.02
4	Fat (%)	1.61± 0.02
5	Crude fibre (%)	3.88± 0.02
6	Ash (%)	4.77± 0.05
7	Carotene (mg%)	42.2± 0.09
8	Vit. C (mg%)	0.31± 0.03
9	Calcium (%)	0.103± 0.00
10	Phosphorous (%)	0.215± 0.00
11	Potassium (%)	0.84± 0.04
12	Iron (ppm)	92.66± 13.42
13	Water activity (a_w)	0.267± 0.01

[a] Mean ± SD

The results illustrate that the pumpkin powder had a very low moisture content of 3.65% (db) with a water activity (aw) of 0.267.

The major contributor in nutritional composition of pumpkin powder is carbohydrate which was estimated to the tune of 83.19% and crude fibre was around 3.88%. The protein and fat content were 6.21 and 1.61%, respectively. The major functional constituent of pumpkin powder is carotene content which was estimated at 42.2 mg/100g and relatively vit.C was much lower to the tune of 0.31 mg/100g. In mineral matter, the ash content was 4.77% which comprised of minerals such as calcium, phosphorus, potassium and iron in the amount of 0.103, 0.215, 0.84% and 92.66 ppm, respectively.

The results obtained are in good agreement with the results reported by several other investigators for pumpkin powder (Pogjanta *et al.*, 2006; Lee *et al.*, 2002).

Effect of storage conditions on carotene degradation in pumpkin powder

Fig.1 represents the data on variations in carotene content (µg/g) in pumpkin powder after 0, 15, 30, 45, , 180 days of storage both at 7°C and 30°C. As the period of storage was extended, the carotene content in powder reduced for both the temperatures of storage. From the results it was observed that at 7°C storage, the total reduction in carotene content in the pumpkin powder was only 17.04% from 422 µg/g (0 day) to 350.08 µg/g carotene at 180 days. While at 30°C, the total reduction was 48.71% from 422 µg/g (0 day) to 216.44 µg/g (180 days). The total carotene retained after 180 days at 7°C and 30°C storage was found to be 82.95 and 51.28%, respectively. However, after 90 days of storage at 7°C and 30°C, the carotene retention was 90.90 and 79.25%, respectively. Steady loss of carotene was found through entire storage period, but the effect was found to be more prominent after 75 days of storage at 30°C storage. The more degradation of carotene was found at 30°C storage for all the samples. This might be due to the possible higher oxidation and increase in non enzymatic browning at higher storage temperature (Pawar *et al.*, 1985; Madan *et al.*, 2005). Similar kinds of results are reported by Dutta *et al*. 2005 and Suman *et al.*, 2002.

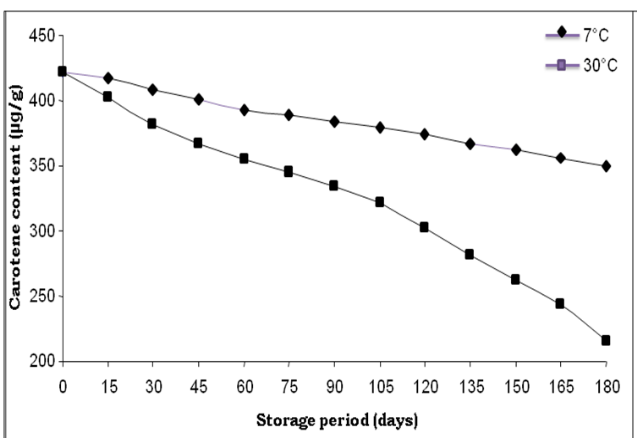

Figure 1. Changes in carotene content of pumpkin powder during storage at different temperatures

Effect of storage conditions on standard plate count (SPC)

The changes in standard plate count (cfu/g) of pumpkin powder during storage are depicted in Fig 1. The SPC of pumpkin powder showed a decreasing trend during entire course of storage at both the storage temperatures. At the end of storage period (180 days), the lowest SPC was recorded for the samples stored at 7°C as compared to those stored at 30°C. At 7°C, the initial count of 2100 cfu/g was decreased to 440 cfu/g by the end of storage. However, at 30°C, the initial count of 2100 cfu/g was reduced to only 800 cfu/g at the end of storage of 180 days. It is evident from the results mentioned in Table 2 that the water activity (a_w) of pumpkin powder was 0.267, which is well below the minimal water activity (a_w) required for the growth of microorganisms. Because of low moisture levels and lower water activity (a_w) of pumpkin powder, microorganisms that might be present were unable to grow and decreased in number during storage. It could be noted that the decrease in count at 7°C was higher as compared to 30°C; this is probably because most of the microorganisms present in pumpkin powder have their optimum temperature for growth and survival between 20 and 30°C.

The above results are in good agreement with the results reported by several other investigators (Khamrui and Pal., 2003; Adu-Gyamfi., 2006).

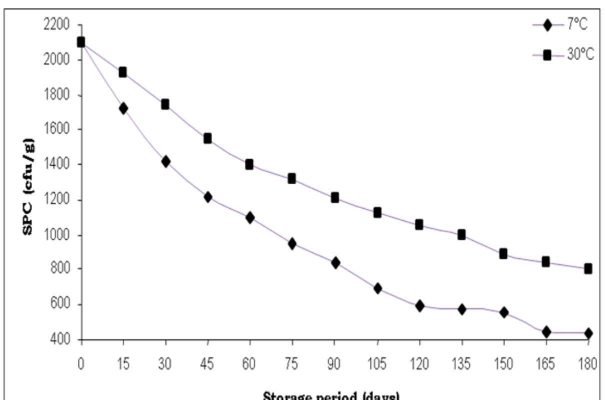

Fig : 2 Change in SPC of pumpkin powder during storage at different temperatures

Effect of storage conditions on yeast and mold count

The changes in yeast and mold count (cfu/g) in pumpkin powder during storage at different temperatures in metalized aluminium pouches are shown in Fig. 3. The yeast and mold count also showed a decreasing trend during the entire period of storage at both the storage temperatures. Lower the temperature of storage faster was the rate of decrease of yeast and mold count. For example, at 90 days of storage, the yeast and mold count at 7°C and 30°C were 13 and 25 cfu/g, respectively. But, at 180 days of storage the yeast and mold count at 7°C and 30°C were only 7 and 20 cfu/g, respectively. This was probably because of the fact that the optimum temperature for growth and survival of yeast and mold is between 20 and 30°C. Hence the rate of decrease of yeast and mold count at 30°C is lower as compared to that at 7°C storage temperature. The above results are in good agreement with the results reported by several other investigators (Khamrui and Pal., 2003; Adu- Gyamfi., 2006).

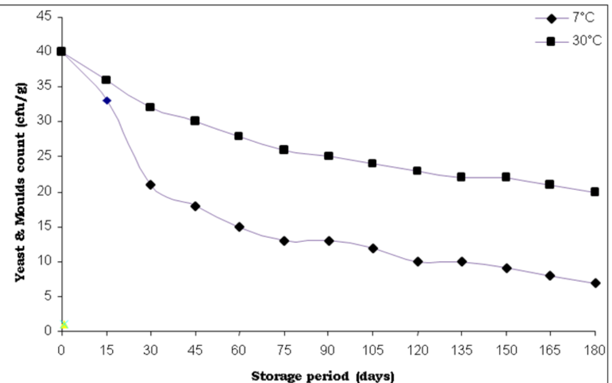

Fig : 3 Change in yeast & mould count during storage at different temperatures

Effect of storage conditions on coliform count

During the storage of pumpkin powder at different temperatures, it was found that the coliform which were absent initially remained absent throughout the period of storage (180 days). Absence of coliform during entire storage period at both the temperatures indicates that hygienic conditions were maintained during preparations and further no toxins are produced in the pumpkin powder during storage.

Statistically, temperature of storage had significant effect on SPC and yeast & mold count during the storage. However, the temperature as well as intervals of storage individually had significant ($p<0.01$) influence on the retention of carotene in pumpkin powder.

Storage stability of pumpkin powder

The results confirm that the microbial quality of the pumpkin powder remained good even after the storage for 180 days, both at the temperatures of 7°C & 30°C. However, there was gradual reduction was observed in the carotene content of the pumpkin powder during storage. Though the loss in carotene content was high at 30°C storage temperature, it was only 17% at the end of 180 days of storage at 7°C. These samples were stored in dark chamber and maintained at 80 and 62% RH, respectively.

The carotene degradation was faster when the pumpkin powder was stored at 30°C as compared to that at 7°C. The carotene content in the powder after 30 days of storage at 30°C was similar to the value after 90 days of storage at 7°C. Carotene content in powder after 75 days of storage at 30°C was similar to the value after 180 days of storage at 7°C.

Pumpkin powder was found to be more stable at 7°C temperature of storage as compared to that at 30°C. The powder can be safely stored at 7°C for almost 180 days. Alternatively, at 30°C, the pumpkin powder should not be stored for longer period than 75 days.

Conclusion

During storage of pumpkin powder, the storage temperature and period had a significant effect on carotene retention in the product. As the storage period was increased from 0 to 180 days, the carotene retention was found to decrease. More carotene could be retained (82.95%) when pumpkin powder was stored at 7°C as compared to 52. 28% retention at 30°C when the stored for 180 days. Standard plate count of powder stored at 7°C was found to be only 440 cfu / g at the end of 180 days of storage. Similarly, yeast and mold count observed at the end of 180 days storage was only 7 cfu/g. Coliform was found to be absent throughout storage. Pumpkin powder was found to be more stable upto only 75 days, if stored at 30°C. However, it can be safely stored for almost 180 days, if stored at 7°C.

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