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Effect of Pre-Treatments on the Proximate Composition of Pumpkin Flour

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Abstract: *Pre-treatments are required to preserve the quality of the product. Studies were carried out to optimize the different pre-treatments for preparation of pumpkin flour. The Pre-treatments included steam blanching, hot water blanching, NaCl blanching, KMS, Citric acid and a combination of blanching followed by sulphitation. Untreated samples served as control. Drying of the samples was performed on a tray drier at 55℃ for 8 hrs. The dried samples were pulverized and sieved through a standard mesh. Quality characteristics of pumpkin flour viz. moisture, total carotene, colour, ash, crude fibre, protein, minerals like iron and phosphorus, NEB as affected by pre-treatments were studied. Storage study was also carried out for a period of six months by packing in MPE pouches. Changes in carotene, colour and NEB were studied during storage period at room temperature. Pre-treatments did not improve the drying rate of pumpkin flour, but influenced the colour of the final product. The pre-treatment of blanching combined with sulphitation was selected as the best process for retention of carotene and other quality parameters during storage.*

Keywords: *Pre-treatments, Quality, Pumpkin flour*

1. INTRODUCTION

Pumpkin belongs to the genus Cucurbita of the family *Cucurbitaceae.* It includes squash and cucumbers and is extensively grown in tropical and sub-tropical countries. The most common types of pumpkin worldwide are *Curcurbita pepo*, *Curcurbita maxima* and *Curcurbita Moschata* [1]. Pumpkin can be found in different shapes, sizes and colours. Pumpkins are sweet when fully mature with yellow or orange flesh and are rich in carotene, vitamins, minerals and dietary fibre [2]. Carotenoids, the natural plant pigments responsible for the orange colour of pumpkin, are intensely investigated because of their health promoting effects. Carotenoids are a primary source of vitamin A for most of the people living in developing countries [3] where vitamin A deficiency is still common [4]. Pumpkins are poor in total solids [5] and are high in β-carotene, which gives its yellow or orange color [6]. Murkovic *et al*. [7] reported that three species of pumpkin namely, *C. pepo, C. maxima and C. moschata* consisted of β-carotene (0.06-7.4 mg/100g)α-carotene (0-7.5 mg/100g) and lutein (0-17 mg/100g). Similarly, Amotz and Fishler [8] reported that pumpkin consisted of both β-carotene and lycopene. The β-carotene content of pumpkin varies from 1.6 to 45.6 mg/100 g [9] and 2.8 to 3.4 mg/100 g [10]. Indian cultivars of pumpkin have 132 to 527 mg/100 g (on dry weight basis) of β-carotene content [11].

Pumpkins are consumed in a variety of ways such as fresh or cooked, as well as being stored frozen or canned [12]. Rheological and thermal characteristics of pumpkin puree have been studied by Dutta *et al.* [13]. Pumpkin puree is an intermediate product that is mainly used for the manufacture of jams, jelly, sweets, beverages and other products. Fernandez *et al.* [14] developed pumpkin flakes as a source of carotene for infant foods. Pumpkin is also known for the properties of its seeds that are rich in fat, protein, thiamin, niacin and various minerals [15]. Much research has been carried out investigating the pumpkin seed as a valuable source of oils and proteins. Stevenson *et al*. [16] showed that pumpkin seed oil has high oxidative stability and is suitable for food and industrial applications, as well as high unsaturation and tocopherol content that could potentially improve the nutrition of human diets.

Pumpkin can be processed into flour which has a longer shelf-life and is used because of its highly-desirable flavour, sweetness and deep yellow-orange colour. Pumpkin powder has been reported 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 [17]. Studies by Pongjanta *et al.* [18] revealed that pumpkin powder produced by juice extraction and cabinet drying and then ground with pin mill and sifted through an 80 mesh sieve was cheap to produce, of good quality and easy to use as a β-carotene supplement in food products. Pongjanta *et al.* [19, 20] indicated that the use of 10-20% pumpkin powder substituted for wheat flour in Thai desserts improved the yellow color and β-carotene content. Lee *et al*. [21] found that noodles made from pumpkin flour were attractive with yellow colour and were popular in terms of appearance, taste, texture and acceptability. Krokida *et al.* [22] have studied the influence of parameters of the drying agent and size of samples on the dehydration kinetics of different vegetables, including pumpkin.

Fruits and vegetables are usually pre-treated for extending the shelf life, preservation of flavour and colour, minimization of nutrients loss, elimination of enzyme activity etc. Pumpkin, like most vegetables, is a perishable food. Therefore it becomes necessary to use methods that allow preserving its properties. One of the most commonly used methods is drying, which is considered the oldest and the most important method of food preservation [23]. The two most commonly employed methods for drying are conventional hot air drying and freeze drying [24]. Nowadays, to dry agricultural products such as vegetables, the most used process is hot air drying [23]. Air drying is generally favoured due to its low operating costs, and also because less drying time is required in comparison to freeze drying. In conventional air drying, high temperatures are employed that adversely affect the texture, colour, flavour, and nutritional value of the products in question [24]. However, this method allows obtaining a uniform, hygienic and attractive dried product [25]. One way of producing dried products of good quality is to use pre-treatments, which is able to improve product quality [26-29]. Also, pre-treatments help in the inhibition of enzymatic browning. Use of pre-treatments such as blanching and sulfiting before drying results in minimum quality degradation. Sulfiting protects the product against non-enzymatic browning during dehydration [30]. In general, various pre-treatments prior to drying like blanching, chemical treatments viz. sodium metabisulphite, citric acid are suggested for obtaining better quality characteristics of vegetables. The present study was undertaken with the objective to optimize different pre-treatments to obtain good quality pumpkin powder and β-carotene retention during storage.

2. MATERIALS AND METHODS

2.1 Chemical Reagents

Chemicals and solvents used in the study were of analytical and laboratory grade and were procured from SD Fine-Chem Ltd. (Mumbai, India).

2.2 Sample Preparation

Pumpkins (*Cucurbita maxima*) were procured from local market in Hyderabad, Telengana. They were cleaned, peeled and the seeds were separated manually. The pumpkin was then washed and cut into slices of 5 mm thickness using a vegetable slicer.

2.3 Preparation of Pumpkin Flour (PF)

The pumpkin slices were subjected to different pre-treatments (Table 1). In each of the pre-treatment (T_1 to T_7), around 10 kg pumpkin slices (Tray load: 1kg/tray) were taken. After the pre-treatment, the samples were dried in a tray drier (Chemida India, Bombay, Model No.12-969) till constant weight. In case of control, the slices were dried without any pre-treatment. Dried slices were pulverized and then sieved through a standard 72 BSS mesh. Pumpkin flour was packed in MPE pouches for carrying out further analysis. All the samples were dried at 55 ± 2°C till constant weight.

2.4 Analysis of Fresh pumpkin and Pre-treated Pumpkin Flour

Fresh pumpkin and its flour were analyzed for various proximate parameters. Moisture, crude fibre, starch, ash and minerals like phosphorus and iron were analyzed as described by Ranganna [31].

2.5 Colour Measurement

Pumpkin flour was subjected to colour measurement [32]. The change of colour was measured and compared using Hunter Colorimeter (Model No. USVIS1417, Hunter Associates Laboratory, USA). Among the three colour coordinates, namely L*, a* and b*, "L*" represents the lightness index, "a*" represents red-green, while "b*" represents yellow-blue colour components. The measurement of L*, a* & b* values of colour was carried out in triplicate and the average values were reported.

2.6 Estimation of Total carotenoids

The estimation of total carotenoids was done after extraction of the sample with acetone and purification with hexane and distilled water. The sample (1.0 g) was extracted with acetone until the residue became colourless. The extracts were transferred to a separating funnel followed by addition of hexane and water. The Hexane extract was collected and its volume adjusted. The solution was then filtered with anhydrous sodium sulphate and read on a spectrophotometer at 452 nm against hexane as blank.

2.7 Estimation of Starch

Starch in the sample was estimated by acid hydrolysis method. To the sample about 100 ml of 95% alcohol was added and centrifuged till the precipitate settled at the bottom. After filtering the residue was washed with about 50% alcohol. The residue was then transferred to a 500ml conical flask with about 200ml of water followed by addition of conc. H_2SO_4. A funnel was placed in the neck of the flask to prevent evaporation, and heated in a boiling water bath for 2.5 hours, cooled, neutralized with a definite volume of water and determined the reducing sugars.

2.8 Estimation of Phosphorus and Iron

Phosphorus and iron was analyzed by colorimetric method according to Ranganna [31]. To the ash solution, molybdate reagent was added and mixed followed by addition of aminonaphthosulphonic acid solution with constant mixing and making upto volume. Similarly a blank was prepared by using water instead of the sample. It was allowed to stand for 10 minute and measured the colour at 650 nm setting the blank at 100% transmission.

The iron was determined by converting the iron to ferric form using oxidising agents like potassium persulphate or hydrogen peroxide and treating thereafter with potassium thiocyanate to form the red ferric thiocyanate which was measured colorimetrically at 480nm setting the blank at 100% transmission.

2.9 Non-Enzymatic Browning

The sample (1 g) was soaked overnight in 20 ml ethanol (90%) and NEB was expressed as optical density of the ethanol extract at 420 nm using a spectrophotometer [33].

2.10 Moisture sorption studies

The studies on Moisture content-Relative Humidity relationship of products were carried out at 27 $\pm$ 1°C by exposing known weight of the samples in duplicate to different relative humidity conditions ranging between 11- 92% in air tight desiccators using appropriate saturated salt solutions [34]. The samples were weighed periodically till they attained constant weight or showed signs of visible mould growth whichever was earlier. The equilibrated moisture content (EMC) at different relative humidity was calculated. The effect of different pre-treatments on sensory attributes was also assessed.

2.11 Storage studies

The pumpkin flour samples were packed in MPE pouches and stored at room temperature (65% RH at 27°C). Samples were withdrawn after 3 and 6 months to analyze the changes in total carotene degradation and Non-enzymatic browning during storage period.

3. RESULTS AND DISCUSSION

The different pre-treatments for pumpkin are presented in Table 1. The fresh pumpkin contained 92.52 % moisture and 1.07 mg/100 g of β-carotene. The crude fibre constituted about 0.60 %. The mineral matter, represented by total ash content was 0.68% in fresh pumpkin (Table 2). The proximate parameters of fresh pumpkin in the present study are almost in line with those reported by See *et al.* [35] as 92.24 % moisture, 0.56% crude fibre and 0.76% ash.

3.1 Effect of pre-treatments on the quality characteristics of pumpkin flour

Moisture content of the dried product is an indicator of efficiency of dehydration. It is directly related to drying method, drying time and conditions of storage. The effects of different pre-treatments on quality characteristics of pumpkin flour are presented in Table 3. The moisture content of the different pre-treated pumpkin flour ranged between 6.40-13.8%. The moisture content among different pre-treatments varied with maximum moisture recorded in T_5 sample. In the present study, pre-treated flour samples retained higher moisture content compared to control flour sample which could be due to the moisture imbibed during the pre-treatment stage. The protein content of pumpkin flour was in the range of 5.17 to 9.54% for the pre-treated samples on dry weight basis (Table 3). Temperature significantly affected the protein content of pumpkin flour. From the table it can be observed that blanching has significantly influenced the protein content. The cold pre-treated samples (T_1, T_2 and T6) had higher protein values as compared to the blanched (heat pre-treatment) samples. Similarly, Van Hal [36] observed that the heat processing treatments have a negative influence on protein quantity and quality depending on the period and the intensity of heat exposure in sweet potato. Also, blanching considerably reduced the quantity of phosphorous and iron. This could be due to the leaching of nutrient during hot water blanching. However, the influence was less on ash and crude fiber. Pumpkin flour had a lighter colour (higher L value), less red colour (low a value) and increased yellowness (higher b value) than fresh pumpkin (Table 3).The discoloration of the fresh pumpkin was prevented by subjecting the slices to different pre-treatments before hot air drying. The T_7 sample showed higher b* values as compared to other pre-treated samples which could be due to the combined effect of blanching and sulphiting. However, blanching or sulphitation treatments of the samples individually showed lower or higher L* values. The higher L* value of sulphited pumpkin powder may be due to the bleaching effect of the sulphite treatment. Blanching of pumpkin protected the total carotenoids during dehydration. SO_2 gave further protection of total carotenoids of the blanched pumpkin. Among the different pre-treatments, the total carotene content ranged between 2.8-10.35 mg/100 with higher amount of total carotene recorded in T_7 samples which might be due to the combined application of blanching with potassium metabisulphite (KMS). Retention of total carotene using pre-treatments like blanching with addition of additives like potassium metabisulphite, Citric acid (CA) etc. has also been reported by various workers. It was observed that the use of chemical pre-treatments improved the amount of total carotene in pumpkin flour. But blanching and sulphiting when used together showed a most favourable effect on total carotenoid stability. Although good amounts of total carotenoids are present in pumpkin flour, it decreases over time with processing in both raw form and during heat treatment. Also the total carotenoid content in different pre-treated pumpkin flour varied greatly which could be due to the type of processing. Blanching treatment slightly affected starch content. The blanched flours had lower starch content than non-blanched flours. In general, blanching of pumpkin decreased protein, ash, fiber and starch content due to leaching out during blanching. A similar observation was made in the preparation of sweet potato flour. The fat, protein, ash, and crude fibre content in the blanched sweet potato flour reduced [37].

3.2 Moisture sorption isotherm of pumpkin powder

Moisture sorption isotherms (Fig 1) were drawn to evaluate the hygroscopicity of the different pre-treated pumpkin flour. The control and the pre-treated pumpkin flour samples had moisture content in the range of 6.40 to 13.8% equilibrated to a relative humidity of 28 to 51% respectively. This indicated that the products dried below their respective ERH and absorbed moisture above their ERH. The moisture sorption isotherm of all the pumpkin flour samples (T_1 – T_7) was almost similar and sigmoid indicating the typical of low fat starch rich products. The sharp increase in moisture above 32% RH in the samples (T_1, T_2, T_3, T_4, and T_5) indicated deterioration of the product above 32% RH. However, the samples (T_6 and T_7) equilibrated at 44% RH. The treatment T_7 (Blanching + 0.2% KMS) attained highest score for colour (8.0) and overall acceptability (7.5) followed by T_6 (0.2% KMS) in comparison to all the other samples (T_1 – T_5) which scored low for colour. The additive potassium metabisulphite (KMS) inhibited the browning reaction by binding with the carbonyl group of reducing sugar and other compounds to retard the browning process [38] resulting in bright yellow colour of the product. This is an important data to predict the shelf life of products for the deteriorations caused by moisture and also indicate the requirement of a high moisture barrier at Relative Humidity (RH) range 32-56% for different pumpkin powder variants.

3.3 Storage Studies

In general, pumpkin flour had poor carotene stability unless processed carefully and immediately placed in sealed packages and kept under proper storage conditions. The main cause of carotene degradation during processing and storage could be oxidation [36]. Total carotenoids in pumpkin flour were influenced by pre-treatments. All samples showed a progressive loss of carotene throughout the storage period with a different rate of degradation and colour changes. It was seen that sample T_7 showed a good effect on the retention of carotene as compared to the other samples. Results also showed that this combination could delay significantly the changes in carotene degradation and subsequently more protective effect in comparison to other pre-treatments (Fig 2). Blanching and sulphiting were effective in reducing the loss of carotenoids in T_7 sample which could be due to the reactions of sulfites with carbonyl groups, reducing sugars and disulfite bonds in proteins and enzymes [39].

Non-enzymatic browning values were used as an index of discoloration of pumpkin powder. During storage, the orange-yellow colour of pumpkin gradually changed to brown. This phenomenon which is known as non-enzymatic browning produces dark pigments and destroys the natural colour of the products [40]. Browning was observed in all the pre-treated samples with significant variations depending on the pre-treatment. Lesser browning effect was observed in T_7 samples during storage which might be due to the fact that blanching and other additives like KMS might have checked the non-enzymatic browning as observed in Fig 3. Similarly, Arya *et al.* [41] indicated that metabisulfite reduces the formation of browning compounds during dehydration and subsequent storage.

4. CONCLUSION

The results have shown that blanching of pumpkin slices followed by pre-treatment with KMS (T_7) before drying gave higher colour stability. Blanching prior to dehydration improves the retention of β-carotene during storage of the pumpkin flour, probably due to inactivation of enzymes and sulphite treatment has an enhancing effect. Browning during storage of pumpkin flour was less prominent in blanched + sulphited samples than only blanched or only sulphited samples. Processing of pumpkin into flour improves the shelf life and makes it easier to incorporate it into food products. This could lead to efficient and profitable utilization of pumpkin thereby ensuring reduced postharvest losses. Pumpkin flour can act as an important source of β-carotene.

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Table 1: *Different Types of pre-treatments*

Samples	*Pre-treatment*
T_1	Control
T_2	Dipping in 0.1% Citric acid (CA) for 15 min
T_3	Hot water blanching at 95 for 3 min
T_4	Steam blanching for 5 min
T_5	Blanching at 95 in 1% NaCl for 3 min
T_6	Dipping in 0.2 % Potassium meta bisulphite (KMS) for 45 min
T_7	Hot water blanching for 2 min followed by dipping in Potassium meta bisulphite (KMS) for 45 min

Table 2: *Proximate composition of Fresh Pumpkin*

Composition	*Fresh Pumpkin*
Moisture (%)	92.52 ± 0.01
Protein (%)	1.25 ± 0.02
Ash (%)	0.40 ± 0.04
Crude Fibre (%)	0.60 ± 0.03
Total Carotene (mg/100g)	1.07 ± 0.02
Colour	L* 61.95± 0.00 a* 12.19±0.01 b* 39.11±0.01

#Average of duplicate analysis ± SD

Table 3: *Effect of Pre-treatments on the proximate parameters of pumpkin flour*

Composition	*Pumpkin Flour (0 days)*						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Moisture (%)	6.40 ± 0.05	7.38 ± 0.01	12.78 ± 0.02	12.62 ± 0.01	13.8 ± 0.05	11.44 ± 0.01	10.99 ±0.01
Protein(%)	8.51± 0.01	7.25±0.01	5.17±0.02	6.16±0.01	6.68± 0.01	9.54±0.01	5.45± 0.02
Ash(%)	6.52± 0.02	5.70±0.15	4.02±0.01	6.61±0.02	6.59± 0.03	6.04±0.01	6.54± 0.01
Crude Fiber (%)	6.58± 0.02	6.41±0.03	6.9±0.2	7.04±0.02	7.5±0.05	8.36±0.01	12.011±0.02
Minerals (mg/100g) Phosphorus Iron	241.977±0.02 22.54 ±0.05	317.545±0.02 16.01 ±0.03	177.449±0.06 5.078± 0.00	167.514±0.01 10.075 ±0.01	28.35 ±0.05 11.629±0.00	269.451±0.02 18.61 ±0.04	142.988±0.04 21.794±0.01
Total carotene (mg/100g)	2.816 ±0.01	5.492± 0.03	9.196± 0.01	10.35± 0.01	2.17± 0.01	7.581± 0.00	17.769±0.00
Starch (%)	30.16 ±0.05	40.77± 0.01	19.8±0.01	23.7±0.05	22.68 ±0.02	30.22± 0.03	32.14 ±0.04
Colour	L* 81.60 ±0.03 a* 3.90± 0.00 b* 27.12 ±0.01	L* 80.51± 0.01 a* 5.79±0.01 b* 28.93± 0.00	L* 77.06± 0.01 a* 4.72±0.00 b* 27.12± 0.01	L* 74.20± 0.1 a* 3.21±0.02 b* 26.81± 0.02	L* 76.50 ±0.00 a* 2.95± 0.01 b* 25.45 ±0.00	L* 82.37± 0.00 a* 6.44±0.01 b* 36.61± 0.01	L* 83.27 ±0.01 a* 3.82± 0.02 b* 39.71 ±0.01
SO_2 (mg/kg)	-	-	-	-	-		1279.14±0.03

#Average of triplicate analysis ± SD

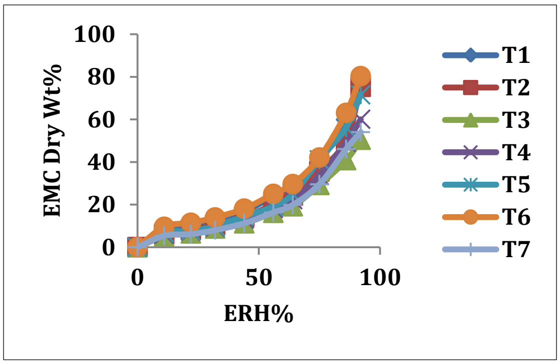

Chart 1: *Equilibrium Relative Humidity for pre-treated pumpkin powder*

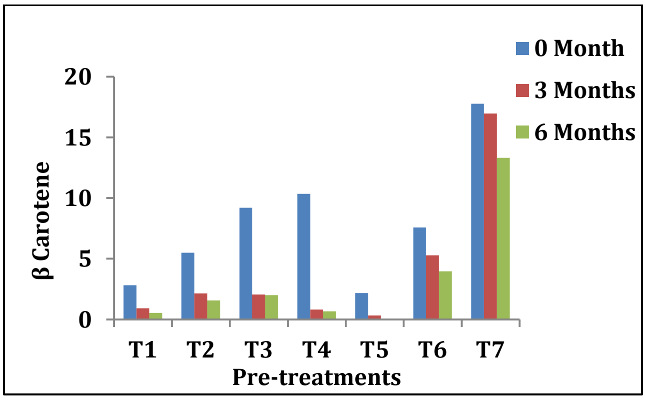

Chart 2: *Effect of Pre-treatments (As mentioned in Table 1) on Total carotene during storage*

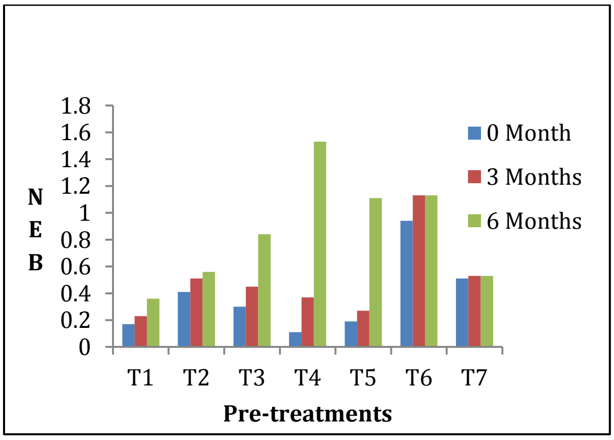

Chart 3: *Effect of Pre-treatments on NEB during storage.*

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Development of viable processing methodologies to arrest the post-harvest losses in selected fruits, vegetables and tubers and development of novel and extruded products in combination with minor cereals, millets and pulses.

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