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## EFFECT OF PROCESSING ON NUTRACEUTICAL PROFILE AND AMINO ACID CONTENT ON PUMPKIN (*CUCURBITA PEPO* L.) SEEDS

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### ABSTRACT

A well-known, nutrient-rich, multipurpose food, pumpkin has recently provided scientists with new avenues for research. In addition to proteins, carbohydrates, polyunsaturated fatty acids, monounsaturated fatty acids, amino acids, minerals, and a variety of phytochemicals, the pumpkin fruit, comprising the flesh, seed, and peel, is a rich source of primary and secondary metabolites. Raw pumpkin seeds may have a compromised flavor, color, as well as digestibility. Therefore, the objective of the present study is to assess the influence of roasting (160°C for 10 min) and germination on the nutritional profile, mineral content and amino acid content of pumpkin seeds. Our results indicated that fibre and protein content was increased in the processing methods i.e., roasting and germination. In the meantime, the content of minerals changed after roasting and germination and revealed significant increase at ( $p < 0.05$ ) in potassium and phosphorus and after germination calcium, magnesium and iron were also increased. Results obtained from processed pumpkin seeds further revealed that the essential and non-essential amino-acid content of germinated pumpkin seeds were increased when compared to raw and roasted pumpkin seeds. Processed pumpkin seeds had significant mineral content and excellent amino acid content. It can be concluded that subjecting pumpkin seeds to various processing methods enhanced its nutritional value and opened up potential uses for them in the food sector.

**Keywords :** Pumpkin seeds, roasting, germination, nutraceutical profile, amino acids

### Introduction

Scientists are becoming more interested in pumpkin due to its nutritional profile. It is a nutritious and cost-effective product from the Cucurbitaceae family. Cucurbita consists of 12 species, the most economically important of which is *Cucurbita pepo* L. and its varieties, which are cultivated worldwide due to their cost-effectiveness and green properties (Dotto and Chacha, 2020). Due to the rich nutraceutical and therapeutic potential of pumpkin, it is becoming more and more recognised as a functional food and has been extensively utilized in the healthcare industry recently. Even though some areas consider pumpkin seeds as snacks or as a protein supplement, they are commonly discarded after the oil gets extracted (Wang *et al.*, 2017). Pumpkin seeds contain carotenoids, vitamin, phytosterols, cucurbitacin, squalene and phenolic compounds in addition to carbohydrates, protein, and other essential nutrients (Potocnik *et al.*, 2018). Pumpkin seeds have been shown to have anti-helminthic, anti-hypercholesterolemic, and anticancer actions, as well as the ability to prevent prostate cancer and urinary problems, hypertension, and diabetes (Aziz *et al.*, 2018). Traditional processing techniques like germination, fermentation and roasting not only change physical characteristics of food materials but also improve the nutritional content, add value and help in extending the

shelf life in addition to sensory quality of a variety of foods (Flores *et al.*, 2010). Roasting is a cooking method that can use direct or indirect heat sources. When the heat treatment is applied, it induces the development of the desirable color and flavor with unique taste and has a significant effect on a product's nutritional value, mineral and amino acid content. Furthermore, the heat generated during roasting reduces microbial contamination and proliferation, as well as inactivates enzymes that can contribute to deterioration of food (Akintade *et al.*, 2019). Germination of seed improves its nutritional quality and the bioavailability of constituents present in the seeds. It is an economical method that can enhance the quality and quantity of bioactive compounds that causes an important change in nutritional value of the products and sensory characteristics (Shemi, 2013). This process hydrolyzes macronutrients that are inside the grain, such as starch, fat, and proteins, into new cell components, resulting in sensory and nutritional advancements (Gan *et al.*, 2017). It was reported that germination resulted in an appreciable reduction in the factors responsible for flatulence, thus increasing the intake and utilization of available protein and carbohydrate. Thus, it decreases and eliminates the anti-nutritional factors present in the legumes and seeds (Kajihaua *et al.*, 2014; Singh *et al.*, 2016).

In order to enhance the utilization of *Cucurbita pepo* L. seeds, it is essential to determine how traditional processing of food can affect the usefulness of the nutrients in the food system. Therefore, understanding the effects of germination and roasting on the nutraceutical and amino acid profile of *Cucurbita pepo* L. seeds that will boost the food system and promote value addition.

## Materials and Methods

### Materials

Pumpkin (*Cucurbita pepo* L.) seeds used for this study were procured from Krishi Vigyan Kendra, Banasthali Vidyapith. The laboratory received mature fruits for additional processing, and all of the reagents utilised were of analytical grade (Sigma Aldrich Research Laboratories Pvt. Ltd., Mumbai (Maharashtra, India).

### Sorting and cleaning of pumpkin seed

A sharp knife was used to harvest and slice open mature pumpkin fruits. The seeds were washed, and manually decorticated, left to drain in a strainer, and then dried in a hot air quick drying oven at 50° C for 12 hours. The pumpkin seeds with fine quality were packaged and kept in an ambient environment in an HDPE white plastic pouch until further use (Akintade *et al.*, 2019).

### Roasting of pumpkin seeds

Fresh pumpkin seeds were boiled with salt for 10 min, air dried and roasted in an earthen pot (160°C for 10 min) with coarses and (which was properly washed and sun-dried before use). The roasted pumpkin seed was allowed to cool and then stored in an HDPE white plastic pouch for further analyses (Abdulrahman *et al.*, 2014).

### Germination of Pumpkin Seeds

Fresh pumpkin seeds were soaked in water for 12 h after which the water was drained, the steeped pumpkin seeds were spread on a moistened jute bag, covered and left for 3 days with regular wetting and turning of seeds. After sprouting, the seeds were air dried and stored in an HDPE white plastic pouch and labelled (Badau *et al.*, 2005).

### Proximate analysis of processed and raw Pumpkin seeds

Proximate content of processed and raw pumpkin seeds were determined according to the Association of Official Analytical Chemists (AOAC, 2016).

### Mineral content of processed and raw pumpkin seeds

The minerals Na and K were determined from the resulting solution using AOAC methods as described by James (1995). Emission flame photometer, while Ca, Mg, Zn, P, Cu, Fe and, Mn were determined using atomic absorption spectrophotometer (Model A A-6200, Shimadzu, Corp., Kyoto, Japan) using standard methods.

### Determination of amino acid profile of processed and raw Pumpkin seeds

Amino acid content of raw, roasted and germinated pumpkin seeds were determined following the method of (Ismail *et al.*, 2010) using the PTH amino acid analyzer (model 120A).

## Statistical analysis

All the results were obtained in triplicate, subjected to analysis of variance ANOVA and the New Duncan Multiple Range Test (NDMRT) was used to distinguish the means. All samples' means and standard deviations were computed and compared by Version 22.0 of the SPSS for Windows programme to analyse the results.

## Results and Discussion

### Proximate composition of raw, roasted and germinated pumpkin seeds

Table 1 revealed the proximate composition of raw, roasted and germinated pumpkin seeds. Raw pumpkin seeds had the significantly highest moisture content 6.78g/100g, while that of germinated and roasted was the lowest 5.49g/100g and 6.31g/100g respectively which may be valuable in view of the sample's shelf-life. (Devi *et al.*, 2018) reported the moisture content of raw pumpkin seeds was 5.53g/100g. The protein content of germinated pumpkin seeds was insignificantly increased after the germination process (32.75g/100g) while the protein value of raw pumpkin seeds was 32.38g/100g. Results of protein for pumpkin seeds estimated during this study were similar with results reported by (Bhat and Bhat, 2013). Plant foods that provide about 12g/100g of their calorific value from protein are considered good sources of protein (Langyan *et al.*, 2022). Fats are essential in diets as they increase the palatability of foods by absorbing and retaining their flavours and help in the transport of nutritionally essential fat-soluble vitamins (Reid *et al.*, 2017). As shown in Table 1, fat content of raw and roasted pumpkin seeds were 45.65g/100g and 40.31g/100g respectively, as the significant reduction in fat content was observed during the roasting process when compared to germinated seeds 43.14g/100g. It may be suggested that shelf life of pumpkin seed flour will be prolonged as the rate of rancidity will be slow and also contribute to the low energy of the samples (Sade, 2009). Germinated pumpkin seeds were found to possess the highest fibre content of 7.99g/100g, followed by the roasted 6.83g/100g and raw seeds 5.89g/100g. (Kindki, 2017) investigated that fiber content in whole pumpkin seeds was 18.54g/100g on a dry basis which coincides that the presence of dietary fibers in pumpkin seeds reduces sugar levels. Ash content of raw, germinated and roasted pumpkin seeds were 4.78, 3.86 and 4.06g/100g respectively, processing significantly reduced the ash content of pumpkin seeds which is parallel to observations of (Ohtsubo *et al.* 2005). The results revealed that carbohydrate content was significantly increased at (p<0.05) level after roasting (6.65g/100g) of pumpkin seeds when compared to raw and germinated seeds 4.21 and 4.44g/100g respectively, Similar results were reported by (Olawepo *et al.*, 2014) who recorded an increase in carbohydrate content after roasting pumpkin seeds. The energy content of pumpkin seeds were significantly decreased and recorded to be 488.97, 534.66 and 562.13 Kcal/100g in roasted, germinated and raw seeds respectively. These seeds are usually discarded by people and therefore they do not make use of the nutrients inherent in the seeds. Results obtained from this research are in agreement with previous reports which suggested that processing techniques such as germination and roasting improved the nutritional quality of food products (Akeem *et al.*, 2019).

### Mineral composition of raw, roasted and germinated pumpkin seeds

Table 2 represents the mineral content of raw and processed pumpkin seeds that comprises a substantial amount of minerals which were significantly increased at ( $p < 0.05$ ) level. It was found that processing methods, especially germination showed immense retention while increases were noticed in sodium, phosphorus, iron, potassium and magnesium whereas roasting showed increase in zinc and manganese content. The mineral content after processing showed the potassium and phosphorus having the highest value of 135.13 g/100g and 58.78 g/100g after germination followed by 110.12 g/100g and 54.33 g/100g after roasting when compared to raw pumpkin seeds. Increase in mineral content during germination could be as a result of activities of microorganism and biosynthesis during processing (Ijarotimi, 2012). (Wang *et al.* 2010) reported similar results that the roasting increased the potassium 110.12%, zinc 0.36% and phosphorus 54.33% content in pumpkin seeds significantly.

### Amino acids content of raw, roasted and germinated pumpkin seeds

Table 3 depicts different essential and non-essential amino acid content of raw and processed pumpkin seeds. Pumpkin seeds were recorded to be high in crude protein, roughly 35%, and this translates to a significant and different amount of amino acids (Jafari *et al.*, 2012). Amino acids play important roles both as building units of proteins and as intermediates in metabolism. It was observed that germination and roasting improved some of the amino acids present in pumpkin seeds. The essential and non-essential amino-acid content of germinated pumpkin seeds were 34.91 g/100g and 47.67 g/100g respectively, whereas the roasted pumpkin seeds recorded 25.51 g/100g and 40.45 g/100g of essential and non-essential amino acids when compared to raw pumpkin seeds i.e. 31.76 g/100g and 49.61 g/100g respectively. Thus, there was significant increase at ( $p < 0.05$ ) level in essential amino acid content after germination and decrease in both essential and non-essential amino acids after roasting. (Ijarotimi, 2012) observed similar effects that germination enhanced some of the amino acids present in pumpkin seeds while there were reductions also in some amino acid values of roasted seeds as compared to raw pumpkin seeds content.

**Table 1:** Proximate composition (g/100g) of raw, roasted and germinated pumpkin *Cucurbita pepo* L. seeds on dry weight basis

Samples of pumpkin seeds (g/100g)	Raw	Roasted	Germinated
Ash	4.78±0.49 <sup>a</sup>	4.06±0.26 <sup>b</sup>	3.86±0.83 <sup>c</sup>
Fat	45.65±1.11 <sup>a</sup>	40.31±1.98 <sup>c</sup>	43.14±0.67 <sup>b</sup>
Fibre	5.89±0.20 <sup>c</sup>	6.83±0.14 <sup>b</sup>	7.99±0.28 <sup>a</sup>
Protein	32.38±0.54 <sup>c</sup>	32.77±1.54 <sup>b</sup>	32.75±1.44 <sup>a</sup>
Moisture	6.78±0.39 <sup>a</sup>	6.31±0.12 <sup>b</sup>	5.49±0.18 <sup>c</sup>
Carbohydrate	4.21±0.06 <sup>b</sup>	6.65±0.02 <sup>a</sup>	4.44±0.09 <sup>b</sup>
Energy (Kcal/100g)	562.13±0.03 <sup>a</sup>	488.97±0.19 <sup>b</sup>	534.66±0.11 <sup>a</sup>

Means of triplicate determinations ± S.D with different superscripts on the same row are significantly different

**Table 2:** Mineral composition (g/100g) of raw, roasted and germinated pumpkin *Cucurbita pepo* L. seeds

Samples of pumpkin seeds (g/100g)	Raw	Roasted	Germinated
Sodium	40.42±0.05 <sup>b</sup>	38.98±0.02 <sup>c</sup>	52.22±0.01 <sup>a</sup>
Potassium	98.76±0.50 <sup>c</sup>	110.12±0.11 <sup>b</sup>	135.13±0.32 <sup>a</sup>
Calcium	4.65±0.11 <sup>a</sup>	2.78±0.04 <sup>c</sup>	4.01±0.19 <sup>b</sup>
Magnesium	1.48±0.04 <sup>b</sup>	1.05±0.03 <sup>c</sup>	2.45±0.02 <sup>a</sup>
Zinc	0.29±0.02 <sup>b</sup>	0.36±0.01 <sup>a</sup>	0.19±0.01 <sup>c</sup>
Phosphorous	47.49±0.06 <sup>c</sup>	54.33±0.01 <sup>b</sup>	58.78±0.04 <sup>a</sup>
Copper	0.49±0.01 <sup>a</sup>	0.08±0.03 <sup>b</sup>	0.43±0.02 <sup>a</sup>
Iron	2.79±0.05 <sup>b</sup>	1.24±0.11 <sup>c</sup>	3.60±0.01 <sup>a</sup>
Manganese	0.01±0.00 <sup>a</sup>	0.04±0.02 <sup>a</sup>	0.00±0.00 <sup>a</sup>

Means of triplicate determinations ± S.D with different superscripts on the same row are significantly different at  $p < 0.05$ .

**Table 3 :** Amino acid content (g/100g) of raw, roasted and germinated pumpkin *Cucurbita pepo* L. seeds

Samples of pumpkin seeds (g/100g)	Raw	Roasted	Germinated
Lysine	4.36±0.03 <sup>b</sup>	4.12±0.15 <sup>c</sup>	4.99±0.07 <sup>a</sup>
Histidine	2.49 ±0.01 <sup>a</sup>	2.12±0.04 <sup>b</sup>	2.51±0.05 <sup>a</sup>
Glutamic	12.88±0.01 <sup>b</sup>	10.45±0.15 <sup>c</sup>	11.44±0.14 <sup>a</sup>
Aspartic	9.11±0.09	7.44±0.09 <sup>b</sup>	8.46±0.08 <sup>b</sup>
Arginine	10.45±0.07 <sup>b</sup>	7.66±0.08 <sup>c</sup>	11.23±0.31 <sup>a</sup>
Glycine	2.36±0.04	2.20±0.01 <sup>b</sup>	3.52±0.04 <sup>a</sup>
Proline	3.13±0.03 <sup>b</sup>	2.25±0.05 <sup>c</sup>	3.96±0.02 <sup>a</sup>
Alanine	4.99±0.03 <sup>a</sup>	4.87±1.00 <sup>a</sup>	3.41±0.05 <sup>b</sup>
Valine	4.52±0.05 <sup>b</sup>	3.45±1.00 <sup>c</sup>	4.78±0.05 <sup>a</sup>
Cystine	0.89±0.05 <sup>a</sup>	0.47±0.00 <sup>c</sup>	0.68±0.00 <sup>b</sup>
Methionine	2.45±0.01 <sup>b</sup>	2.20±0.05 <sup>c</sup>	3.19±0.06 <sup>a</sup>
Leucine	5.25±0.29 <sup>b</sup>	4.24±1.00 <sup>c</sup>	5.34±0.02 <sup>a</sup>
Isoleucine	3.58±0.04 <sup>b</sup>	2.47±0.06 <sup>c</sup>	4.54±1.00 <sup>a</sup>
Phenylalanine	3.89±0.05 <sup>b</sup>	3.15±0.04 <sup>c</sup>	4.25±0.02 <sup>a</sup>
Tryptophan	2.03±0.02 <sup>a</sup>	1.54±0.01 <sup>c</sup>	1.87±0.08 <sup>b</sup>
Threonine	3.19±0.08 <sup>b</sup>	2.22±0.04 <sup>c</sup>	3.44±1.00 <sup>a</sup>
Serine	3.61±0.04 <sup>b</sup>	3.55±1.00 <sup>b</sup>	3.99±0.04 <sup>a</sup>
Tyrosine	2.19±0.08 <sup>b</sup>	1.56±0.02 <sup>c</sup>	3.39±0.06 <sup>a</sup>

Means of triplicate determinations ± S.D. with different superscripts on the same row are significantly different at  $p < 0.05$ .

### Conclusion

In conclusion, processing pumpkin seeds significantly improved the nutritional profile and mineral bioavailability. Fibre and protein content in pumpkin seeds increased after germination, whereas carbohydrate content was found to be high during the roasting process when compared to raw pumpkin seeds. Minerals such as sodium, potassium, and phosphorus were increased in both germination and roasting processing methods. The study found that the essential amino acid content of germinated pumpkin seeds responded well with the reference standard i.e. raw pumpkin seeds, while non-essential amino acid content increased after roasting. It is fair to conclude that pumpkin seeds, which are typically discarded as vegetable waste, can be efficaciously used for value addition through simple processing methods such as roasting and germination, and that they possess functional attributes which render them advantageous for product development.

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### Conflict of Interest

None

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