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Effect of Germination on the Nutritional and Anti-Nutritional Composition of Soy Bean (*Glycine max*)

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ABSTRACT

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Soybean (*Glycine max*) is a superior plant food known as miracle golden bean for its treasure trove of economical and preminent protein than the high-priced meat protein. Soybean is not only known for its nutritional components but also provides significant health related benefits. This study was planned to analyze the effect of processing of soybean on its nutritional composition. Raw seeds with and without seed coats and germinated forms of three soybean varieties were analyzed for its nutritional and anti-nutritional components. Significant changes were observed during processing of the soybean seeds. Seeds when germinated contained 40.05-46.46 percent protein in SL 525, SL 744 and SL 958 soybean varieties and increase in amino acid namely lysine (5.70mg/100g), methionine (0.61mg/100g) and tryptophan (1.11mg/100g) was recorded highest in SL 958. Minerals like potassium, calcium, magnesium and phosphorus were reported to increase significantly during germination. With germination, there was an increase in *in vitro* protein digestibility with decrease in anti-nutritional factors like trypsin inhibitor and total phenols.

Introduction

Soybean (*Glycine max*) is considered as an important grain legume that belongs to species of legume which is categorized under the family 'Leguminosae' with sub family 'Papilionaceae' native to East and South-East Asia. It is commonly produced and consumed in tropical and subtropical regions of the world. Soybean is mostly found in Southeast Asia. It is a widely grown valuable crop in the central part of India where the bulk is utilized

for producing oil or for animal feed, while considerably small usage for human or direct consumption which is increasing steadily (Singh, 2016). It is superior to all other plant food because it has a good balance of all essential amino acids and contains proper amount of methionine. Soybean is therefore, not only known for its nutritional components but also provides significant health related benefits. Soybean is known as miracle golden bean. Soybean protein is economical and preminent than the high-priced meat protein

and so it is considered as the best source of protein in a vegetarian diet. Considering its nutritional attributes, mainly its high protein and fat content provided with the significant potential for cost-effective improvement of daily diets, helps in substantial reduction of protein-energy malnutrition (Goel *et al.*, 2018). It contains good amount of essential macro nutrients for regulating good nutrition which includes protein (40%), carbohydrates (18%) and fat (18%). The moisture content is 9 percent and fibre is 10 percent with other micro nutrients (5%) like folic acid, calcium, potassium and iron (Singh 2009).

Protein content is 30-45 percent with a good source of all indispensable amino acids with a digestibility value of 91.41 percent. The protein content is six times more than the rice grains and four times higher than the wheat. (Serrem *et al.*, 2011).

Germination is done to boost the nutritive value and to inactivate the anti-nutritional components or the undesirable substances namely trypsin inhibitors, phytic acid, oligosaccharides and components with lipoxygenase activity that is present in it (Ramadan 2012). It is an effective and value-added technological approach in order to enhance the nutritional quality of soybeans in Asian countries due to its digestibility (Cho *et al.*, 2009).

Germination is done in order to enhance its nutritional value which results in enriched soy flour in various nutrients, increasing its utilization and versatility as compared to the non-germinated flour. During germination, the protein content of soy flour was 6.90 percent higher with increase in essential amino acid content of mainly lysine, valine and threonine by 8.53, 8.50 and 7.93 percent. In addition, there was an increase in the iron and vitamin B2 content by 14.29 percent and 8.80 percent respectively as compared to the non-

germinated soy flour. Germinated soy flour contained 77.06 μ g/100g vitamin A, 0.29 μ g/100g vitamin B, and 0.32 μ g/100 g vitamin B₂ (Rahman *et al.*, 2016).

So, considering the properties of soybean, the present study was planned with the objective to determine the effect of processing on the nutritional composition of soybean.

Materials and Methods

Procurement and processing of soybean varieties

The sample of soybean seeds of three varieties- SL 525, SL 744 and SL 958 were procured from Department of Plant Breeding and Genetics, College of Agriculture, Punjab Agricultural University, Ludhiana. Soybean seeds were sorted based on size, colour and then thoroughly washed with water to remove any residues from the seed surface along with dirt or any other foreign material. After surface cleaning, the seeds were soaked in warm distilled water for 16-18 hours. The germination process was induced such that the seeds were placed (wrapped tightly) in sterilized wet cotton muslin cloth at room temperature in dark conditions, away from direct sunlight. The seeds were left to germinate for about 24 hours. The cloth was kept moist to support the germination process. After germination, the grains were boiled for 1 hour to reduce antinutritional factors. The seeds were later dried at 60°C up to 10 percent moisture content using mechanical dryer. The dried soybean seeds were de-vegetated and grinded to make germinated soybean flour. The flour was stored and packed in air tight plastic bags for further analysis.

Proximate analysis

Flour of raw soybean seeds with and without seed coat and of geminated soybean seeds

were analyzed for their proximate composition namely moisture, crude protein, crude fibre, crude fat and total ash by (AOAC 2000) method. The total available carbohydrate was calculated by difference of total contents from 100.

Amino acid content

The content of amino acid present in soybean (raw and germinated) were analyzed such that, tryptophan was estimated with procedure given by Concon (1975), methionine by Horn *et al.*, (1946), cysteine by Liddell and Saville (1959) and available lysine by Carpenter (1960) modified by Booth (1971).

Mineral content

Minerals like potassium, calcium, phosphorus, magnesium and zinc were estimated by atomic absorption spectrophotometer.

***In vitro* protein digestibility**

In vitro protein digestibility of soybean samples was estimated by method used by Akeson and Stachman (1964).

Anti-nutritional components

Anti-nutritional components like trypsin inhibitors and total phenols were analyzed. (Roy and Rao, 1971).

Statistical analysis

Samples for all the determinations were carried out in triplicates and results were expressed as mean + standard deviation. The data was analyzed with the help of statistical tools. To test the significant difference between raw and germinated samples, ANOVA was applied using SPSS16.0 software. The significant difference was carried out at 5% level of significance.

Results and Discussion

Proximate composition

The proximate composition of different soybeans varieties in the form of germinated (G), non-germinated without seed coat (NGSC) and non-germinated with seed coat (NGWSC) were analyzed on their dry weight basis and have been presented in Table 1. The proximate content of different varieties showed significant variability with respect to moisture, protein, fibre, ash content and carbohydrates. The moisture content was highest in NGSC with 4.73 percent and lowest in G with 1.74 percent of SL 744 variety. The percent crude protein content of G was highest in SL 958 i.e. 46.46 percent. No significant difference was observed among the NGWSC varieties, lowest protein was recorded in SL 525 with 36.05 percent Warle *et al.*, (2015) studied the effect of germination on nutritional quality of soybean. Germination of grains increased the moisture content upto 10 percent to 11 percent from 1 percent, total sugars from 3.55 percent to 5.6 percent, protein content also increased from 29.09 percent to 34.99 percent Torres *et al.*, (2006) reported that soybeans contain about 35-40 percent of the protein on a dry weight basis. Crude protein increased with increase in days germination when compared to raw control group. The protein content was increased to 6.90 percent in germinated soy flour. This finding was similar to the other study done by (Ugwuona *et al.*, 2012) and (Nwosu *et al.*, 2014).

The fibre content of NGSC was recorded highest in SL 958 i.e.18.24 percent and lowest observed in NGWSC of SL 744 (7.28%). Martino *et al.*, (2011) presented the chemical composition and iron bioavailability based on rats when provided them soybean flour with and without hulls. It was found that soybean hull flour contained 37 percent more total dietary fibre and a higher content of iron than

that of the soybean flour without hull. The crude fat content was significantly highest in SL 958 of NGSC soybean and lowest in SL 525 of G seeds with 20.32 and 8.31 percent fat respectively. Ghavidel and Prakash (2007) reported that there was a significant decrease in the fat content of some legume seeds after the seeds were germinated. Osman (2007) conducted a study in which there was a significant decrease in the fat content of germinated seeds with time period of 3 days supporting the process with use of fat as energy.

The percentage of total ash of NGSC was highest in SL 525 with 5.66 percent and lowest observed in G of SL 744 with 3.51 percent. Ari *et al.*, (2012) reported that the ash content of thermal treated soybean varied from 4.27 to 4.46 per cent. The carbohydrate content of Germinated SL 525 was reported highest with 35.91 g whereas lowest observed carbohydrate content was found in NGSC of SL 958 with 13.91 g.

Warle *et al.*, (2015) reported that the carbohydrate content of soybean was significantly decreased after soaking, germination and heating treatments i.e. 22.10 to 17.90 percent as observed in other two varieties i.e. SL 744 and SL 958. Furthermore, the fat content was reduced from 24 percent to 10 percent and ash content from 4.95 percent to 4.59 percent. The energy content was low in G seeds of SL 525 i.e. 378 Kcal whereas high of SL 958 in NGWSC (419 Kcal).

Effects of traditional processing methods on the nutritional and anti-nutritional properties of soybean were analysed by Pele *et al.*, (2016). Soybeans were subjected to various handling strategies which were mainly sun drying and processing. Sample A was considered as the controlled sample where the soybean seeds were subjected to sun for drying and then milled. For sample B, soybean

was drenched for 24 hours then it was sun dried and processed. For test C, it was almost similar to sample B but it was soaked for 12 hours instead of 24 hours which was later dehulled, sun dried and further subjected to processing. For the last sample D, the soybeans were sprouted for 120 hours, then they were sun dried and later processed. Proximate composition of soybean revealed that protein content of the samples increased from 23.98 percent to 28.44 percent with a noteworthy increment in sample B, sample C and sample D.

The fat content increased from 20.51 percent to 26.20 percent while crude fibre varied from 4.68 percent to 6.58 percent. The moisture content extended from 7.23 percent to 10.92 percent had demonstrated a critical lessening in the control.

Amino acids

The data on the amino acid content of germinated and raw seeds is presented in Table 2. Lysine content of G seeds of SL 958 reported to be the highest with 5.70 mg and lowest was observed 4.63mg in SL 744 of NGWSC treated seeds. Highest cysteine content of SL 744 in G treated seeds was 0.15 mg whereas no significant difference was observed in NGWSC and NGSC treated seeds. The methionine content of SL 958 was reported to be highest in G seeds with 0.61 mg and lowest in NGWSC of SL 525 with 0.22 mg. G seeds of SL 958 recorded highest tryptophan content of 1.11 mg and lowest in SL 744 of NGSC with 0.40 mg. Hallen *et al.*, (2004) reported that amino acids and available vitamins were increased during germination. Rahman *et al.*, (2016) studied essential amino acids content including threonine (3.13%), lysine (2.29%), valine (0.51%), glutamic acid (1.28%), isoleucine (1.49%) and methionine (1.28%) were found higher in germinated soy flour as compared to raw soy flour.

Table.1 Proximate composition of raw and germinated soybean (on dry weight basis)

Proximate Composition	SL 525			SL 744			SL 958		
	Non- Germinated		Germinated	Non-Germinated		Germinated	Non-Germinated		Germinated
	With seed coat	Without seed coat		With seed coat	Without seed coat		With seed coat	Without seed coat	
Moisture (%)	3.76 ^{ab} ±0.03	3.39 ^{ab} ±0.18	2.04 ^{aA} ±0.03	4.73 ^{bb} ±0.24	4.22 ^{bb} ±0.11	1.74 ^{aA} ±0.08	4.34 ^{bb} ±0.06	4.29 ^{bb} ±0.21	2.32 ^{aA} ±0.74
Crude protein (%)	38.59 ^{aA} ±1.11	36.05 ^{aA} ±2.47	40.05 ^{aA} ±0.99	40.25 ^{abA} ±0.74	38.59 ^{aA} ±2.1	42.50 ^{aA} ±0.74	42.53 ^{baB} ±0.74	40.51 ^{aA} ±1.11	46.46 ^{bb} ±2.85
Crude fat (%)	17.98 ^{aC} ±0.23	12.96 ^{ab} ±0.08	8.31 ^{aA} ±0.33	18.50 ^{bc} ±0.06	13.66 ^{bb} ±0.21	9.50 ^{ba} ±0.5	20.32 ^{cC} ±0.21	15.54 ^{cb} ±0.17	10.78 ^{aA} ±0.12
Ash (%)	5.66 ^{bb} ±0.11	5.60 ^{ab} ±0.06	3.60 ^{aA} ±0.11	5.65 ^{bb} ±0.21	5.60 ^{a0B} ±0.23	3.51 ^{aA} ±0.07	5.00 ^{ab} ±0.14	5.35 ^{ab} ±0.21	3.80 ^{aA} ±0.14
Crude fibre (%)	16.40 ^{bc} ±0.12	8.25 ^{ba} ±0.01	12.13 ^{ab} ±0.01	15.62 ^{ac} ±0.2	7.28 ^{aA} ±0.01	12.35 ^{ab} ±0.01	18.24 ^{cC} ±0.12	9.24 ^{aA} ±0.01	13.37 ^{bb} ±0.03
Carbohydrates (g)	21.37 ^{aA} ±20.25	29.36 ^{ab} ±3.13	35.91 ^{aA} ±0.52	19.98 ^{ba} ±20.96	34.87 ^{bc} ±0.7	32.24 ^{bc} ±0.21	13.91 ^{aA} ±6.68	37.14 ^{cC} ±24.83	25.6 ^{ab} ±3.63
Energy (Kcal)	401 ^{ab} ±1.35	409 ^{aA} ±2.41	378 ^{aC} ±11.91	407 ^{bb} ±0.98	416 ^{ba} ±2.66	383 ^b ±7.45	408 ^{bb} ±1.27	419 ^{ba} ±0.94	385 ^{bb} ±9.88

Values are expressed as Mean ± SD

Means with different notation (a, b & c) indicates significant difference at 5% level of significance between varieties.

Means with different notation (A, B & C) indicates significant difference at 5% level of significance between treatments

Table.2 Amino acid content of raw and germinated soybean varieties (mg/100g protein on dry weight basis)

Amino acid	SL 525			SL 744			SL 958		
	Non-Germinated		Germinated	Non-Germinated		Germinated	Non-Germinated		Germinated
	With seed coat	Without seed coat		With seed coat	Without seed coat		With seed coat	Without seed coat	
Lysine (mg/100g)	4.87 ^{aA} ±0.06	4.81 ^{abA} ±0.12	5.32 ^{ab} ±0.11	4.87 ^{ab} ±0.01	4.63 ^{aA} ±0.08	5.10 ^{aC} ±0.04	5.17 ^{ba} ±0.01	5.08 ^{ba} ±0.02	5.70 ^{bb} ±0.05
Cysteine (mg/100g)	0.06 ^{aA} ±0	0.10 ^{ab} ±0.01	0.11 ^{ab} ±0	0.05 ^{aA} ±0	0.10 ^{ab} ±0	0.15 ^{cC} ±0	0.05 ^{aA} ±0.01	0.11 ^{ab} ±0	0.13 ^{bc} ±0
Methionine (mg/100g)	0.34 ^{ab} ±0.01	0.22 ^{aA} ±0	0.34 ^{ab} ±0.04	0.46 ^{aA} ±0	0.36 ^{ba} ±0.05	0.50 ^{bb} ±0.04	0.40 ^{ba} ±0	0.49 ^{cb} ±0	0.61 ^{cC} ±0
Tryptophan (mg/100g)	0.43 ^{aA} ±0.04	0.70 ^{bb} ±0.02	0.91 ^{bc} ±0.04	0.40 ^{aA} ±0.02	0.41 ^{aA} ±0.01	0.60 ^{ab} ±0.01	0.51 ^{aA} ±0.05	0.80 ^{cb} ±0.02	1.11 ^{cC} ±0.03

Values are expressed as Mean ± SD

Means with different notation (a, b & c) indicates significant difference at 5% level of significance between varieties.

Means with different notation (A, B & C) indicates significant difference at 5% level of significance between treatments

Table.3 Mineral content of raw and germinated soybean varieties (mg/100g on dry weight basis)

Mineral content	SL 525			SL 744			SL 958		
	Non-Germinated		Germinated	Non-Germinated		Germinated	Non-Germinated		Germinated
	With seed coat	Without seed coat		With seed coat	Without seed coat		With seed coat	Without seed coat	
Calcium (mg/100g)	145.70 ^{ba} ±0.78	169.18 ^{ba} ±20.96	331.81 ^{bb} ±2.87	115.56 ^{aA} ±4.07	120.28 ^{abA} ±17.38	379.11 ^{bb} ±54.25	114.09 ^{aA} ±11.1	109.24 ^{aA} ±7.38	282.99 ^{aA} ±109.97
Phosphorus (mg/100g)	611.45 ^{aA} ±14.85	609.70 ^{aA} ±18.03	626.45 ^{aA} ±9.9	618.70 ^{aA} ±9.55	633.70 ^{aA} ±43.49	646.70 ^{aA} ±26.52	623.20 ^{aAB} ±20.86	585.95 ^{aA} ±14.85	649.78 ^{ab} ±27.57
Magnesium (mg/100g)	150.55 ^{ba} ±2.63	170.96 ^{ba} ±18.76	236.83 ^{ab} ±2.94	115.84 ^{aA} ±4.62	124.22 ^{aA} ±11.21	264.83 ^{ab} ±25.41	120.88 ^{aA} ±8.99	118.22 ^{aA} ±2.96	224.34 ^{aA} ±89.0
Potassium (mg/100g)	59.37 ^{aA} ±1.66	59.22 ^{ba} ±1.38	81.09 ^{ab} ±0.28	59.24 ^{aA} ±3.04	56.82 ^{abA} ±1.94	78.04 ^{ab} ±5.09	53.99 ^{aA} ±2.83	52.49 ^{aA} ±0.07	82.42 ^{ab} ±5.83
Zinc (mg/100g)	3.48 ^{aA} ±0	3.27 ^{aA} ±0.35	3.13 ^{aA} ±0.11	4.21 ^{bb} ±0.23	3.79 ^{aAB} ±0.17	3.58 ^{ba} ±0.03	5.17 ^{cb} ±0.19	4.63 ^{ba} ±0.16	5.21 ^{cb} ±0.07

Values are expressed as Mean ± SD

Means with different notation (a, b & c) indicates significant difference at 5% level of significance between varieties.

Means with different notation (A, B & C) indicates significant difference at 5% level of significance between treatments

Table.4 *In vitro* protein digestibility of raw and germinated soybean varieties

<i>In-vitro</i> nutrient digestibility	SL 525			SL 744			SL 958		
	Non-Germinated		Germinated	Non-Germinated		Germinated	Non-Germinated		Germinated
	With seed coat	Without seed coat		With seed coat	Without seed coat		With seed coat	Without seed coat	
<i>In-vitro</i> protein digestibility (%)	79.79 ^{aA} ±2.19	79.84 ^{aA} ±0.35	80.04 ^{aA} ±0.19	81.53 ^{ba} ±0.58	84.74 ^{ca} ±2.44	84.91 ^{ba} ±1.27	83.38 ^{cb} ±1.69	71.93 ^{ba} ±0.45	86.61 ^{cc} ±1.55

Values are expressed as Mean ± SD

Means with different notation (a, b & c) indicates significant difference at 5% level of significance between varieties.

Means with different notation (A, B & C) indicates significant difference at 5% level of significance between treatments

Table.5 Anti-nutritional factors of raw and germinated soybean varieties

Anti-nutritional factors	SL 525			SL 744			SL 958		
	Non-Germinated		Germinated	Non-Germinated		Germinated	Non-Germinated		Germinated
	With seed coat	Without seed coat		With seed coat	Without seed coat		With seed coat	Without seed coat	
Total phenol (mg/100g)	224.46 ^{ab} ±4.07	225.18 ^{ab} ±1.02	85.61 ^{aA} ±3.05	185.61 ^{bb} ±0.1	182.01 ^{bb} ±1.02	89.21 ^{aA} ±6.1	169.06 ^{ab} ±3.05	161.87 ^{ab} ±1.02	80.58 ^{aA} ±8.14
Trypsin Inhibitor (TIU/g of protein)	14.21 ^{cb} ±0.02	14.51 ^{bc} ±0.01	11.49 ^{aA} ±0.02	13.75 ^{bb} ±0.01	14.10 ^{ac} ±0.02	11.94 ^{ba} ±0.03	13.47 ^{ab} ±0.01	14.15 ^{ac} ±0.01	12.54 ^{ca} ±0.04

Values are expressed as Mean ± SD

Means with different notation (a, b & c) indicates significant difference at 5% level of significance between varieties.

Means with different notation (A, B & C) indicates significant difference at 5% level of significance between treatments.

Germination is highly recommended cost-effective for increasing the bioactive compounds and amino acid content in soybean seeds. The value-added bioactive compounds can be resulted due to the biochemical changes observed in the soybean seeds. It was also noted that the amino acid, free and total phenolic compounds and isoflavone content intensified by germination. The amino acid content was prominently influenced by these compounds. Considering the high amino acid content and with the potential health benefits; in order to obtain functional food, germination can be an appropriate process with its beneficial effects in chronic degenerative diseases, such as cancer and obesity. (Guzman *et al.*, 2017)

Mineral content

Soybean seeds with different varieties were given different treatments which were analyzed for potassium, calcium, phosphorus, magnesium and zinc as depicted in Table 3. The significant increase in mineral content was observed with germination. Highest and lowest content of potassium was reported in SL 958 with different treatments where G reported highest content of 82.42 mg and 52.49 mg reported to be the lowest in NGWSC treated soybean. Calcium of SL 744 in G (379.11 mg) was highest and lowest in NGWSC of SL 958 with 109.24 mg. In G, SL 958 obtained highest phosphorus content of 649.78 mg whereas lowest in NGWSC with 585.95 mg. The magnesium content in SL 744 was reported highest in 264.83 mg of G seeds and lowest in NGSC with 115.84 mg. No significant difference was observed in highest content of zinc in G and NGSC of SL 958. Rahman *et al.*, (2016) reported that germinated soy flour contained 368mg/100gm calcium, 496mg/100gm phosphorus and 16.45 mg/100gm iron. Luo *et al.*, (2013) studied that germination reduces the availability of zinc from faba bean and soybean. There was an increase in the

availability of calcium from faba bean, soybean, and rice with availability of manganese from soybean (14.4%) and rice (43%). Thus, germination could be an appropriate food-based strategy to increase the iron content with other minerals from food grains.

***In vitro* protein digestibility**

In vitro protein digestibility of germinated soybean seeds (Table 4) increased significantly from raw seeds such that the germinated seeds in SL 958 reported the highest *in vitro* protein digestibility i.e. 86.61 percent as compared to NGWSC which reported the lowest with 71.93 percent. A significant increase was observed in the *in vitro* protein digestibility of white beans, black beans and pigeon beans after germination (Sangronis and Machado, 2007). Urbano *et al.*, (2005) reported that *in-vitro* protein digestibility was slightly decreased with longer germination periods compared with the raw pea flour diet (2.3% and 7.9%) in G3 DNL (three day germination) and G6 DNL (six day germination) respectively and between germination periods no significant difference result was found.

Anti-nutritional components

The anti-nutritional factors like trypsin inhibitors and total phenol content (Table 5) decreased with germination, in SL 525 lowest trypsin inhibitor content was reported i.e. 11.49 TIU per g of protein whereas in NGWSC treated seeds obtained the highest content of 14.51 TIU per g of protein. Total phenol content in G of SL 958 was lowest whereas NGWSC of SL 525 was highest with 80.58 and 225.18 mg respectively. Effects of traditional processing methods on the nutritional and anti-nutritional properties of soybean were analysed by Pele *et al.*, (2016). Soybeans were subjected to various handling strategies which were mainly sun drying and

processing. Sample A was considered as the controlled sample where the soybean seeds were subjected to sun for drying and then milled. For sample B, soybean was drenched for 24 hours then it was sun dried and processed. For test C, it was almost similar to sample B but it was soaked for 12 hours instead of 24 hours which was later de-hulled, sun dried and further subjected to processing. For the last sample D, the soybeans were sprouted for 120 hours, then they were sun dried and later processed. The anti-nutritional properties of the test samples demonstrated that phytic acid content ranged between 5.45 percent to 8.05 percent which essentially diminish in test B, C and D. The tannin content extended from 19.23 percent to 25.23 mg/100g while the protease inhibitor ran from 4.91 to 7.09 percent with a noteworthy lessening in test B, C and D. In the end, it was recorded that there was increase in the nutritional properties of soybeans because of traditional processing methods. Also, traditional processing method remarkably lessened the anti-nutritional properties of soybeans which also led to maximizing the bioavailability of its nutrients.

Xu and Chang (2008) reported that 73.4% of the phenolic antioxidant compounds are found in the seed coat part of the soybean seeds. Kayembe and Rensburg (2013) reported that germination, soaking and de-hulling of the seeds may lower the trypsin inhibitor levels. However, there were no changes in *in vitro* protein digestibility (IVPD). (Rahman *et al.*, 2016) observed that raw soy flour contains some anti-nutritional factors which were removed by germination process. Also, germinated flour can be stored up to six months at ambient temperature and was found to be microbiologically safe for human consumption. The germination resulted in nutritionally enriched soy flour to that of non-germinated soy flour and could be used as raw material or ingredient for making bakery products.

Vagadia *et al.*, (2017) reviewed the inactivation methods of trypsin inhibitor in soybean. Soybeans contain a variety of bioactive anti-nutritional compounds including protease trypsin inhibitors, phytic acid and isoflavones that gives undesirable physiological effects and hinder its nutritional quality. The processing methods for inactivation of soybean trypsin inhibitor are physical treatments and conventional treatments. In physical treatments, under the influence of high temperatures or mechanical stress the chemical structure is changed. Therefore, most treatments involve heating the soybean samples for a certain amount of time. The heat energy helps in breaking down the physical structure of the molecule which further makes it easy for the digestion of enzyme. In conventional treatments, the thermal inactivation of trypsin inhibitors depends mainly on the time period, temperature with the water content in the sample. Some of the common methods used to eliminate the trypsin inhibitor are forced conventional drying and roasting. Furthermore, quite a few studies have observed that soaking the seed for 12-14 hours alone cannot manage the trypsin inhibitor activity of the soybean seed but if the soaked seed is cooked it results in 82 percent of its activity. Lastly, the studies have illustrated that germination of soybean seeds decrease the trypsin inhibitor activity and increases the *in-vitro* digestibility of soybean protein.

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