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Research Article

PROCESSING AT GREEN CHEMISTRY CONCEPT ON PROTEIN QUALITY ENHANCEMENT OF MODERN SOYBEAN [*GLYCINE MAX* (L.) MERRILL] VARIETIES

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Abstract- The most important heat stable biologically activity trypsin inhibitor strongly prohibited the uses of soybean for protein rich food. The quantification of protein content and trypsin inhibitor of five improved varieties were carried out. The inactivation of trypsin inhibitor activity value and effects on protein content percent of autoclaved processing technology till the 10, 20, 30 minutes and sprouted of same cultivars seeds were quantified for enhancement concerned *in-vitro* protein digestibility percent. Germinated seeds, protein content increases at least 0.18 exceeded to 1.68 percent as compared to control mean. The effect on trypsin inhibitor was found decreased significantly during thermal processing and also same on sprouted seeds. The effects of processing on IVPD were found to increase significantly only more than 20 to 30 min autoclaved processing time seeds for all five varieties seeds as compared with their control seeds mean.
No soybean constituent is removed or enriched during the processing of the raw materials. Thus, all components of soybean remains in soy-powder serve both controlled and processed seeds in which green chemistry prevails and no unhygienic chemicals are used for processing. So, this is safe for consumption and hygienically processing techniques of any soy food products, soy fortified foods and dietary utilization in under malnourished community.

Keywords- Soybean, Trypsin inhibitor, Protein, Green Chemistry, IVPD

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Introduction

The phytonutrients and intuitively of modern soybean [*Glycine max* (L.) Merrill.] high yielding five cultivars were analyzed as raw comprised with different level of autoclaved thermal processing as well as cold processing sprouted seeds with their control means for the utilization as safe in the commercial food commodities. The proteins are naturally phytochemicals significant basic food content be healthful played preventive and therapeutic roles for several diseases protein calories malnutrients with potential for heart problem, cancers, obesity, osteoporosis, hypertension, menopause to females and more. Soybean is leading *kharif* season oilseed crop in India and about 43 percent of the oilseed crops and 25 per cent of the edible oil production in the country.

Soybean known as a good source of proteins in food commodities contained 38-42 percent range of protein in improved verities, moreover about 5% minerals content and rich of dietary fiber. Scientist had found the soybean oil content have majorly five types of fatty acids saturated along with unsaturated [1]. Many researcher have been announced that soy protein are highly digestible and their amino and profile is well balanced to meet the requirements for human nutrition. Processing of soybean typically by steam moist heat treatment under high pressure, improve nutritional value IVPD through conversion of its refractive or native protein more digestible denatured form. Bioactive constituent of soybean trypsin inhibitor is a major limited the uses of soybean for protein rich food and accounts for 30-50% of the growth response of inhibitors effect the pancreatic hypertrophic response of animal fed row soybean meal [2]. Soybean seeds had higher levels of trypsin inhibitor found mainly in soybean reported by [3]. The biological depressive effect on protein digestion and utilization such as protease inhibitor was observed by [4]. Hunger and malnutrition continue to be serious problem in the world, even in countries that report a surplus food grain production The United Nations' Food and Agriculture Organization provides a depiction of the number of people in the world who are under nourished [5].

Trypsin inhibitors are probably the best known and the most important of all the anti-nutritional factors known to be present in soybean. According to [6] the thermal inactivation of these inhibitors is accompanied by a marked enhancement in the nutritive quality of the soybean protein. There are several lines of research which indicate that trypsin inhibitors are only partially responsible for the poor nutritive value of raw soybeans. High level of trypsin inhibitors in a diet stimulates the pancreatic juice secretion, causes pancreatic hypertrophy and poor growth performance in animals [7,8].

An optimal thermal process inactivates deleterious enzymes, microbes and biologically active component, while maximum retention of nutrient and other quality attributes. The flavor of soybean products is major acceptable quality produced by lipoxygenase and peroxides enzymes activity, results in rancidity of oils, painty, grassy or beany off-odors. Hydroperoxides formed by lipoxygenase and arising secondary products through its decomposition or denatured protein and amino acids through formation of covalent bonds [9]. Whereas germination defined as natural biological process of all superior plants, seed comes out of its latency stage. During the sprouting process some quantitative and qualitative changes occur within the seed.

The process of germination used as an alternative technique to remove

undesirable taste, aroma and anti-nutrients of soybean. Food scientists use the life dreams a miracle golden bean due to nutritive value of soybean, as a substitute complementary protein. Soybean protein supplies sufficient amount of various kinds' of amino acids that's why announced as a complete protein.

Materials and Methods

Soybean seeds of five improved varieties *viz.* JS-20-29(V_1), JS-20-34(V_2), JS-97-52(V_3), JS-93-05(V_4) and JS-95-60(V_5) of Soybean Research Unit,(BSP), JNKVV, Jabalpur (M.P.) were used as experimental material.

Before chemical analysis of raw seeds, they were cleaned and dried in cabinet dryer on 50^{0}C for 5-6 hours for equilibrium moisture. Then they were milled using laboratory hammer mill. Each five cultivars at moist high pressure on 15 psi at 121^{0}C temperature for 10, 20 and 30 minutes was autoclaved separately and immediately drawn out and cooled it at 8-10 ^{0}C. After that dried it, again in same way while, germinated seeds were milled after dried on same and packed in Zip-lock polyethylene sample bags and stored at 4^{0}C until analysis. Analysis was carried out of each raw, autoclaved and sprouted seeds on the dry matter basis.

The protein content in sample was determined by using conventional micro-Kjeldahl digestion and distillation procedure as given in [10].For sprouting seed were kept in wet muslin cloths rolled and kept in BOD seed germinator at 25 ^{0}C and 90% humidity for 92 hours. The trypsin inhibitor activity in soybean was determined through methods given by [11]. An improved colorimetric method was used for determining anti-tryptic activity in soybean products.

According to AOAC [12] the *in-vitro* protein digestibility were recorded by calculating the difference between the amount of total nitrogen in the sample before and after digestion with pepsin enzymes, nitrogen contents was calculated by Kjeldahl method than multiplied by the factor 6.25 to obtained the crude protein.

Statistical analysis has done with RBD, Dunnett's t-test was used to identify the significant differences with confidence level at 95% of five soybean varieties at four processing level and triplicate mean of replications with their control mean [13].

Result and Discussion

Effect on protein: The results [Table-1] reveals about the effect of processing technology on protein content of five soybean varieties.

The results indicated that among the variety V_1 cooking process for 20 (T_2) to 30 (T_3) minutes and sprouted differed significantly, while V_4T_2 and V_5T_4 vary with significantly to control as raw seeds.

The confidence interval level for various varieties at different cooking processing was also given in [Table-1]. The results indicate that among the varieties differ significantly reduced while V_4T_2, V_5T_2 and V_5T_4 differ significantly with their control mean. Hence, we can say that with 95% of confidence for variety V_1 the cooking processes at 20 minutes reduced the control by at least 4.20, 3.07 and for T_4 exceeded at least 0.86 with their control mean. While in case of variety V_4 and V_5 at the cooking process V_4T_2 reduced 7.26 and V_5 reduced T_2 at least 7.76 and T_4 are 2.73 with their control mean.

We conclude that V_2 and V_3 are found to be superior rather than that of V_1, V_4 and V_5 with respect to the loss of protein content (%) among the 5 varieties of soybean. Similar work has been obtained by [14]

Table-1 *Effect of processing on Protein content (%) at different varieties of soybean*

Control / Treatments	Ȳ- Ȳ0		d value	't' value	SEm	CD%	Confidence level of treatment effect [d ± (Dunnet) Sd] at 95%		
V1T1	38.32	-38.8	-0.48	1.27	0.270	0.882	-1.634	to	0.674
V1T2	35.75	-38.8	-3.05	7.96	0.270	0.882	-4.204	to	-1.896
V1T3	36.88	-38.8	-1.92	5.01	0.270	0.882	-3.074	to	-0.766
V1T4	40.04	-38.8	1.24	3.22	0.270	0.882	0.086	to	2.394
V2T1	38.09	-37.45	0.64	0.83	0.541	1.763	-1.670	to	2.950
V2T2	35.7	-37.45	-1.75	2.27	0.541	1.763	-4.060	to	0.560
V2T3	37.3	-37.45	-0.15	0.19	0.541	1.763	-2.460	to	2.160
V2T4	39.13	-37.45	1.68	2.19	0.541	1.763	-0.630	to	3.990
V3T1	34.94	-39.92	-4.98	2.36	1.487	4.858	-11.331	to	1.371
V3T2	35.52	-39.92	-4.4	2.09	1.487	4.858	-10.751	to	1.951
V3T3	38.47	-39.92	-1.45	0.69	1.487	4.858	-7.801	to	4.901
V3T4	40.16	-39.92	0.24	0.11	1.487	4.858	-6.111	to	6.591
V4T1	40.45	-40.65	-0.2	0.27	0.698	1.739	-2.770	to	2.370
V4T2	35.96	-40.65	-4.69	6.23	0.698	1.739	-7.260	to	-2.120
V4T3	39.45	-40.65	-1.2	1.59	0.698	1.739	-3.770	to	1.370
V4T4	40.83	-40.65	0.18	0.23	0.698	1.739	-2.390	to	2.750
V5T1	41.19	-41.27	-0.08	0.07	0.698	2.282	-3.064	to	2.904
V5T2	36.49	-41.27	-4.78	4.83	0.698	2.282	-7.764	to	-1.796
V5T3	39.84	-41.27	-1.43	1.44	0.698	2.282	-4.414	to	1.554
V5T4	41.52	-41.27	0.25	8.24	0.698	2.282	-2.734	to	3.234

df = 8; tab. value at 0.05 = 3.02; Ȳ= triplicate mean treatment; Ȳ0= triplicate mean raw seed

Effect on Trypsin Inhibitor

The data [Table-2] reveals about the effect at different processing level on trypsin inhibitor (mg/g protein) each 5 varieties of soybean with their control mean. Dunnett's t-test was used to identify the significant difference of the varieties at the various cooking process levels.

The results indicated that among varieties V_1T_3, V_2T_3, V_2T_4, V_3T_3, V_4T_2, V_4T_3, V_5T_2, V_5T_3 and V_5T_4 differ significantly with their control mean.

The difference level of the varieties at different cooking level were also given in the [Table-2].The results indicated that 30 minute cooking for the variety V_1 and V_2 reduced at least 25.85 and 25.55 found almost similar with their control mean.

While in case of V_4 and V_5 the similar results were obtained and accepted 20 min. of V_5 in declining that it exceeds at least 1.81 and similarly V_4 exceeds 3.84 at 95% confidence level.

The sprouted indicated that variety V_2 and V_5 differ significantly with their control mean also indicated for V_2 and V_5 reduced at least 19.00 and 18.76, which was also similar finding for reduction in trypsin inhibitor mg/g protein of seeds. Similar findings were also reported by [15-19].

Effect on in-vitro digestibility of protein

[Table-3] and [Fig-1] reveals about the effect of processing an in-vitro digestibility of protein % of five varieties of soybean at different cooking process level.

The results indicated that among all the varieties i.e. V_1, V_2, V_3, V_4 and V_5 at cooking process level 20 and 30 minutes autoclaved differ highly significant with their control mean.

While all the varieties at cooking level 10 min autoclaved and sprouted were found to be not significant differ with their control (raw) mean.

The confidence level of among the varieties at significance cooking level was also given in the [Table-3]. It was found that among the varieties V_1 and V_2 the cooking level (T_2) 20 and (T_3) 30 min. exceed 16.35 and 15.56 at least with their control mean at 95% confidence level than V_3, V_4 and V_5. Thus, it was observed that all

the varieties at the cooking level T_2 and T_3 were found to be best than (T_1) autoclaved cooking and (T_4) hydroponically sprouted of soybean seeds

Table-2 *Effect of processing on TI (mg/g)at different varieties of soybean*

Control / Treatments	Ȳ- Ȳ0		d value	't' value	SEm	CD%	Confidence level of treatment effect [d ± (Dunnet) Sd] at 95%		
V1T1	24.29	-17.84	6.45	1.92	2.368	7.738	-3.667	to	16.567
V1T2	27.61	-17.84	9.77	2.91	2.368	7.738	-0.347	to	19.887
V1T3	2.10	-17.84	-15.74	4.69	2.368	7.738	-25.857	to	-5.623
V1T4	8.99	-17.84	-8.85	2.64	2.368	7.738	-18.967	to	1.267
V2T1	18.54	-19.50	-0.96	0.33	1.992	6.507	-8.165	to	8.849
V2T2	26.92	-19.50	7.42	2.63	1.992	6.507	-1.087	to	15.927
V2T3	2.45	-19.50	-17.05	6.04	1.992	6.507	-25.557	to	-8.543
V2T4	9.00	-19.50	-10.5	3.72	1.992	6.507	-19.007	to	-1.993
V3T1	19.81	-17.19	2.62	0.47	3.461	12.614	-13.872	to	19.112
V3T2	32.01	-17.19	14.82	2.72	3.461	12.614	-1.672	to	31.312
V3T3	2.12	-17.19	-15.07	4.75	3.461	12.614	-31.562	to	1.422
V3T4	7.867	-17.19	-9.323	1.70	3.461	12.614	-25.815	to	7.169
V4T1	23.23	-17.50	5.73	1.22	3.296	10.768	-8.346	to	19.806
V4T2	35.44	-17.50	17.94	3.84	3.296	10.768	3.864	to	32.016
V4T3	1.93	-17.50	-15.57	3.34	3.296	10.768	-29.646	to	-1.494
V4T4	8.75	-17.50	-8.75	1.87	3.296	10.768	-22.826	to	5.326
V5T1	23.35	-17.67	5.68	1.95	2.058	6.723	-3.108	to	14.468
V5T2	27.64	-17.67	9.97	3.42	2.058	6.723	1.182	to	18.758
V5T3	1.80	-17.67	-15.87	5.45	2.058	6.723	-24.658	to	-7.082
V5T4	7.69	-17.67	-9.98	3.43	2.058	6.723	-18.768	to	-1.192

df = 8; tab. value at 0.05 = 3.02; Ȳ= triplicate mean treatment; Ȳ0= triplicate mean raw seed

Table-3 *Effect of processing on IVPD (%) at different varieties of soybean*

Control / Treatments	Ȳ- Ȳ0		d value	't' value	SEm	CD%	Confidence level of treatment effect [d ± (Dunnet) Sd] at 95%		
V1T1	68.96	-64.63	4.33	2.33	1.311	5.423	-1.209	to	9.869
V1T2	86.48	-64.63	21.85	11.75	1.311	5.423	16.311	to	27.389
V1T3	85.73	-64.63	21.10	11.37	1.311	5.423	15.561	to	26.639
V1T4	62.75	-64.63	-1.88	1.01	1.311	5.423	-7.419	to	3.659
V2T1	67.67	-63.84	3.83	2.40	1.128	5.032	-0.990	to	8.650
V2T2	85.01	-63.84	21.17	13.25	1.128	5.032	16.350	to	25.990
V2T3	84.10	-63.84	20.26	12.88	1.128	5.032	15.440	to	25.080
V2T4	61.23	-63.84	-2.61	1.63	1.128	5.032	-7.430	to	2.210
V3T1	70.57	-67.57	3.00	1.62	1.844	2.153	-12.417	to	18.417
V3T2	87.82	-67.57	20.25	10.97	1.844	2.153	4.833	to	35.667
V3T3	87.11	-67.57	19.54	10.60	1.844	2.153	4.123	to	34.957
V3T4	64.46	-67.57	-3.11	1.68	1.844	2.153	-18.527	to	12.307
V4T1	69.73	-66.73	3.00	1.57	1.907	2.189	-13.480	to	19.480
V4T2	87.46	-66.73	20.73	10.86	1.907	2.189	4.250	to	37.210
V4T3	86.38	-66.73	19.65	10.29	1.907	2.189	3.170	to	36.130
V4T4	64.21	-66.73	-2.52	1.32	1.907	2.189	-19.000	to	13.960
V5T1	68.79	-65.86	2.93	1.72	1.697	2.066	-10.119	to	15.979
V5T2	87.79	-65.86	21.93	12.92	1.697	2.066	8.881	to	34.979
V5T3	85.04	-65.86	19.18	11.30	1.697	2.066	6.131	to	32.229
V5T4	63.67	-65.86	-2.19	1.29	1.697	2.066	-15.239	to	10.859

df = 8; tab. value at 0.05 = 3.02; Ȳ= triplicate mean treatment; Ȳ0= triplicate mean raw seed

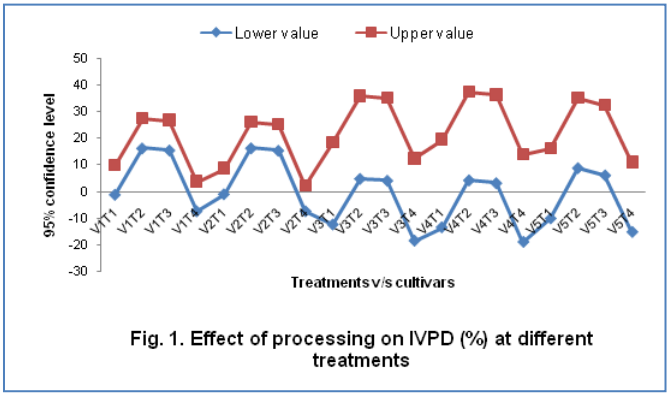

Fig. 1. Effect of processing on IVPD (%) at different treatments

Conclusion

The variety V3 was least affected to be by autoclaved and sprouted the losses of protein content. The highly significant losses of Soybean Trypsin Inhibitor were found during 30 minutes autoclaved. The *in-vitro* protein digestibility percentage increased very highly significant 20.25 to 21.93 percent during 20 minutes autoclaved cooked which was similar followed by 30 minutes autoclaved 19.18 to 21.10 percent with their control mean for each five improved varieties. The finding of the research data are directly justified to enhancement of protein quality be used for safe must be cooked for at least 20 minutes moist heat at high pressure thermal processing.

Conflict of Interest: None declared

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