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Nutritional Status of the Protein of Corn-Soy Based Extruded Products Evaluated by Rat Bioassay

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Abstract. A rat bioassay was conducted to preclinically evaluate nutritional quality of two supplementary foods (SFs) developed based on corn and soy blends for feeding preschool children. The SFs prepared by extrusion cooking and subsequently modified to taste either sweet or salty provide 395 ± 2 kcal of energy and 20 ± 2 g protein per 100 g of food. The proximate constituents and energy contents of SFs were within the ranges prescribed for processed weaning foods and could satisfactorily meet the requirements of preschool children. Groups of male weanling rats were fed SFs for 4 weeks to evaluate the protein quality. The body weight gain of rats fed with SFs were significantly higher than those fed with skimmed milk powder (SMP) diet as control. The protein efficiency ratio and net protein utilization results of SFs were not significantly different ($p > 0.05$) from values of control group. It is inferred that these SFs were nutritionally comparable to SMP.

Key words: Bioassay, Corn-soy blends, Extrusion cooking, Food supplements, Protein quality

Introduction

Providing adequate nutrition during early childhood is of paramount importance for maintaining health. Inadequate intake of nutritionally balanced foods results in inhibition of growth. Poor growth during childhood affects a large portion of the world's population [1]. Therefore, development of nutritious supplementary foods has been suggested by FAO to combat malnutrition among children. The nutritional quality of cereals and legumes can be improved and also exploited as human foods by processing techniques such as extrusion [2].

Extrusion cooking can manufacture a wide range of plant and animal food material into a ready-to-eat form economically. Extruded corn and soy-based snacks have an improved nutritional quality [2, 3], enhanced shelf life of products, and are well accepted by children and mothers [4, 5]. Extrusion cooking not only enhances the acceptability of the product by improving its appearance, taste and texture, but also inactivates anti-nutritional factor such as trypsin inhibitors and urease activity [6] and reduces the growth of fungal pathogen fusarium [7]. Besides protein fortification, soy based supplementary foods also provide nutritional benefits due to its hypocholesterolemic and anticarcinogenic activities [8]. This has increased interest in incorporating soy as a commercial source of protein and isoflavone into foods.

Pre-extrusion addition of soy to corn products has been studied for developing supplementary foods with improved nutritional value. A few reports have shown extrusion cooking using soy-corn blends to serve as a tool to produce health foods. However, only limited studies are available on the effect of extrusion cooking on nutritional quality of foods. Noguchi et al. [9] studied the loss of available lysine during extrusion cooking of cereal and soy based mixtures at various extrusion conditions. Phillips [6] reported on inactivation of anti-nutritional factors such as trypsin inhibitors during extrusion cooking. Marsman et al. [10] studied the influence of screw configuration on the *in vivo* digestibility and protein solubility of soybean meal. The present study thus focuses on the development of ready-to-eat nutritious supplementary foods employing extrusion cooking using de-branded, de-germ corn flour and defatted soy blends. Their proximate composition and nutritional quality with respect to protein efficiency ratio (PER) and net protein utilization (NPU) were also evaluated so as to check its suitability for human consumption.

Materials and Methods

Feed Preparation for Extrusion

Corn flour (60%) and defatted soy flour (40%) were mixed (dry basis) in a Sigma mixer and water was added to achieve blend moisture content of 15%. Forty percent soy flour was found to provide a product with minimum protein content of 15%. The blend was fortified with one third of recommended daily allowance (RDA) of vitamins like vitamin-A (145.2 μg), vitamin-D (145.2 μg), vitamin-E (0.29 mg), vitamin-K (0.36 mg), thiamin (0.27 mg), riboflavin (0.32 mg), folic acid (10.9 μg), pantothenic acid (0.4 μg), vitamin-C (14.5 mg), biotin (0.12 mg), inositol (0.7 mg) and vitamin-B_{12} (0.36 μg) and minerals like iron (4.4 mg), zinc (18.7 mg), copper (0.31 mg), potassium (10.1 mg) and manganese (10.5 mg) as per the RDA for children [11]. Each food mix was blended mechanically (Sigma mixture, AMH Fabric, Peterkupper Aachan, Germany). All the vitamins and minerals were of food grade and they were purchased from Hi Media Laboratories, Mumbai, India. Defatted soy flour was obtained from

Table 1. Composition[a] of diets used for PER and NPU studies

Diet group	SFs (%)	Vegetable fat[b] (%)	Mineral mixture (%)	Vitamin mixture (%)	Corn starch (%)
Control (SMP)	30	10	4	2	54
SF (sweet)	42	10	4	2	42
SF (salty)	44	10	4	2	40
Protein free diet	Nil	10	4	2	84

[a]Dry matter basis.
[b]Groundnut oil.

Sakthi Soy Ltd., Coimbatore, India. Corn flour, sugar and common salt were purchased from the local market.

Extruders and Extrusion Cooking

A corotating intermeshing twin-screw extruder (Model ZSK 30, Werner and Pfleiderer, Ramsey, NJ, USA) with a barrel of 30.85 mm was used. The outer diameter of the extruder screws was 30.7 mm. The extruder was provided with a temperature control facility (in 5 separate zones), and has a digital display for torque (%) developed during extrusion. The length to diameter ratio of the extruder was 30.8:1. The temperature at 4 sections was maintained at 55, 70, 85, and 100 °C; and the temperature of the last section was varied between 100 and 120 °C; other details are reported elsewhere [12].

Post Extrusion Treatment

The extruded beads were coated either with sugar solution (60 ° brix) and with powdered sugar (30% of extrudate weight) or with 2% common salt and dried in a tray drier at 40 °C for 2 hour to obtain dry products (4.7 and 1.3% moisture). Sweet product was prepared in a rotary coating pan where sugar solution was poured on extrudates followed by adding powdered sugar so as all the sugar solution and powdered sugar became a part of the product. Finally each food (100 g/pack) was packed in high-density polyethylene pouches.

Proximate Composition

The fresh samples of supplements were analyzed for moisture, ash, crude protein, total fat, total carbohydrates and energy content according to AOAC methods [13]. The extruded products were ground separately using a laboratory hammer mill. The samples were sealed in polyethylene bags for subsequent use.

Bioassay

Groups of male weanling rats of Wistar-CFT strain weighing 35 ± 2 g (for PER) and 25 ± 2 g (for NPU) were obtained from the animal house facility of the institute. PER and NPU were determined by the methods of Chapman et al. [14] and Bender and Doel [15], respectively. Animals statistically grouped by randomized design were assigned to four groups of 10 animals each. They were housed individually in stainless steel cages with screen bottom and maintained at 22 ± 2 °C, 60 ± 2% RH and were exposed to 12:12 hour L:D cycle. Group 1 and 2 were fed extruded foods (sweet or salty) while group 3 and 4 were fed with control skimmed milk powder (SMP) and protein free diets, respectively. The diets were prepared at 10% level protein (N × 6.25) according to Chapman et al. [14]. The diets were kept isocaloric by adjusting the fat content about 10% by adding groundnut oil (Table 1). Diets were made adequate in other nutrients by incorporating vitamins and minerals (Sd. Fine Chemicals, Mumbai, India). The animals had access to food and water *ad libitum*. Daily food and protein intake, and weekly body weight gain were recorded (28 days for PER and 10 days for NPU) in order to determine food efficiency ratio (FER), protein efficiency ratio (PER) and net protein utilization (NPU), respectively.

Statistical Analysis

The data were subjected to analysis of variance (ANOVA) in a completely randomized design using the method of Snedecor and Cochran [16]. The probability (p) level for testing the significance was 0.05.

Results and Discussion

Proximate Composition

The nutrient components such as crude protein, total fat, total carbohydrates, total ash, water content (moisture) and

Table 2. Nutrient composition (%) of extruded supplementary foods based on corn-soy blends

Product	Moisture	Crude protein	Total fat	Total carbohydrate[a]	Total ash	Energy (kcal/100g)
SF (sweet)	4.7 ± 0.1	18.6 ± 0.8	0.4 ± 0.02	73.6 ± 2.3	2.8 ± 0.1	392.4 ± 2.2
SF (salty)	1.3 ± 0.1	23.4 ± 1.2	3.4 ± 0.3	67.9 ± 2.1	5.0 ± 0.2	397.5 ± 3.8

Note. Values are mean ± SEM of 5 independent analyses.
[a]Estimated by differential method (13).

Table 3. Growth rate, FER and PER of rats fed extruded foods

Dietary group	Food intake[a] (g)	Protein intake (g)	Body wt. gain (g)	FER	PER
Control (SMP)	277.7 ± 12.1	27.8 ± 1.2	90.6 ± 3.0	0.33 ± 0.01	3.31 ± 0.14
Extruded food (sweet)	302.4 ± 8.8_a	30.2 ± 0.9_a	102.5 ± 4.2_a	0.34 ± 0.03	3.39 ± 0.02
Extruded food (salty)	320.9 ± 7.9_a	32.1 ± 4.4_a	119.3 ± 4.4_a	0.38 ± 0.02_a	3.39 ± 0.01

Note. PER—calculated as: weight gain (g)/protein intake (g). Values are mean ± SEM of 10 animals and values in the same column with different subscripts vary significantly ($P < 0.05$) from control.
[a]Dry matter basis.

energy contents of the supplementary foods (SFs) prepared by extrusion cooking are shown in Table 2. The SFs were nutritious since the products provided more than one third of the RDA with respect to protein (20 ± 2%) and energy (395 ± 2 kcal) as recommended by World Health Organisation [17] and National Institute of Nutrition [18] for children. Further, the proximate characteristics of the SFs were within the ranges reported for weaning and supplementary foods [17] and did not differ significantly (≤0.05) between them with reference to protein, carbohydrates and energy content. However, they differed significantly (<0.05) with reference to total fat, moisture and ash content.

Bioassay

The mean body weight gain, FER and PER for control and extruded foods (sweet and salty) are shown in Table 3. Significant difference ($p < 0.05$) in food and protein intake, and body weight gain was observed between the rats fed with extruded foods, and for rats fed with SMP (control) diet. But no significant ($p > 0.05$) difference was observed in PER values between experimental and controls groups. Extruded foods promoted higher growth rate (31 and 13%) than control diet; which may be attributed to slightly higher FER values (0.34 and 0.38). Since food intake of SFs fed groups was marginally higher (7 and 13%) compared to control group, FER was also slightly higher indicating better palatability and conversion [3]. Marsman et al. [10] also reported that *in vitro* digestibility of the extruded soy meal increased significantly. Though PER of the groups fed on SFs were slightly higher than that of rats fed on control diet, however, there was no significant difference ($p > 0.05$) when compared with control group.

The higher PER values (3.39) of extruded foods was on par with the control SMP diet (3.31), and is in conformity with the standards (>2.5) laid down by the Indian Standards Institute [11] as well as the protein advisory group [19] for infant foods. Lorenz et al. [20] also reported that extrusion cooking of full fat soy flour increased the PER and body weight gain of rats. The mean NPU values of the control and extruded SFs are shown in Table 4. Though food and protein intake of rats fed with SFs were found to be higher, the NPU values of SFs (ranged from 69 to 70) were not significantly ($p > 0.05$) different from that of control value of 74. This may be due to the lower content or loss of amino acid lysine and tryptophan [21] during extrusion cooking of cereals and legumes. This makes their protein quality poorer compared to skimmed milk diet. Further, these researchers reported that the loss of available lysine during extrusion cooking was more with increased processing temperature and decreased moisture content of the feed.

In conclusion, the SFs developed during the present study showed that the products are suitable for feeding preschool children since they are nutritious with regard to quantity (nutrients component) and quality (PER and NPU) of the protein and energy content. Further, there is a potential for the extruded cereal and soy based ready-to-eat snacks to meet children's nutritional requirements.

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Table 4. Net protein utilization (NPU) of supplementary foods fed to rats

Diet group	Food intake[a] (g)	Protein intake (g)	Body wt. gain (g)	NPU
Control (SMP)	95.4 ± 1.4_a	9.5 ± 0.2_a	34.8 ± 2.4_a	74.8 ± 1.6_a
Extruded food (sweet)	123.5 ± 4.8_b	12.4 ± 0.5_b	39.4 ± 1.8_b	70.2 ± 1.9
Extruded food (salty)	118.7 ± 3.8_b	11.9 ± 0.9_c	36.5 ± 2.1_c	69.5 ± 1.5_c
Protein free diet	43.5 ± 1.5_c	Nil	(−13.0 ± 1.2_d weight loss)	Nil

Note. Values are mean ± SEM of ten animals and values in the same column with different subscripts vary significantly ($p < 0.05$) from control.
[a]Dry matter basis.

References

1. Dahiya S, Kapoor C (1994) Biological evaluation of protein quality of home processed supplementary foods for pre-school children. Food Chem 48: 183–188.
2. Faller JY, Klein BP, Faller JF (1999) Acceptability of extruded corn snacks as affected by inclusion of soy protein. J Food Sci 64: 185–188.
3. Baskaran V, Mahadevamma, Malleshi NG, Shankara R, Lokesh BR (1999) Acceptability of supplementary foods based on popped cereals and legumes suitable for rural mothers and children. Plant Foods Hum Nutr 53: 237–247.
4. Baskaran V, Mahadevamma, Malleshi NG, Jayaprakashan SG, Lokesh BR (2000) Biological evaluation for protein quality of nutritious supplementary foods based on popped cereals and legumes suitable for feeding rural mothers and children. Plant Foods Hum Nutr 56: 37–49.
5. Baskaran V, Mahadevamma, Balasubramanyan N, Malleshi NG, Lokesh BR (2000) Moisture sorption isotherms of nutritious supplementary foods prepared from cereals and legumes for feeding rural mothers and children. Eur Food Res Technol 211: 27–31.
6. Phillips RD (1989) Effect of extrusion cooking on the nutritional quality of plant proteins. In: Phillips RD, Finley JW (eds), Protein Quality and the Effect of Processing. New York: Marcel Dekker Ink, pp 219–264.
7. Castelo MM, Katta SK, Summer SS, Hanna MA, Bullerman LB (1998) Extrusion cooking reduces recoverability of fumonisin B from extruded corn grits. J Food Sci 63: 696–698.
8. Anderson JW, Johnstone BM, Newell CME (1995) Meta–analysis of the effects of soy protein intake on serum lipids. N Eng J Med 333: 276–282.
9. Noguchi A, Mosso K, Aymard C, Jeunink J, Cheftel JC (1982) Millard reaction during extrusion cooking of protein enriched biscuits. Lebens Wiss Technol 15: 105–110.
10. Marsman GLP, Gruppen N, Vanzuilichem DJ, Resink JW, Voragen AGT (1995) The influence of screw configuration on the *in vitro* digestibility and protein solubility of soy bean and rapeseed meals, J Food Eng 26: 13–28.
11. ISI (1960) Indian Standard Specification for Processed Cereal Foods. New Delhi, India: Indian Standards Institution.
12. Bhattacharya S (1997) Twin—screw extrusion of rice—green gram blend: Extrusion and extrudate characteristics. J Food Eng 32: 83–99.
13. AOAC (1998) Official Methods of Analysis, 16th edn. Arlington, MA: Association of Official Analytical Chemists.
14. Chapman DG, Castillo R, Campbell JA (1959) Evaluation of protein in foods. I. A method for the determination of the protein efficiency ratio. Can J Biochem Physiol 37: 679–686.
15. Bender AE, Doel BH (1957) Biological evaluation of proteins. New aspect. Br J Nutr 11: 140–148.
16. Senedecor GW, Cochram WG (1967) Statistical Methods. Ames, IA: Iowa state University Press.
17. WHO (1985) Energy and protein requirements. In: Technical Report Series 724. Geneva: FAO/WHO/UNU Expert Consultation, WHO.
18. National Institute of Nutrition (1992) Acceptability trails of ready to eat foods in rural ICDS centers. In: Annual Report, NIN, India, pp 1–13.
19. PAG (1976) Protein Advisory Group—Nutrition Document r. 10 Add 91/RV (WHO/FAO/UNICEF).
20. Lorenz K, Jansen GR, Harper J (1980) Nutritional stability of full fat-soy flour and corn-soy blends produced by low-cost extrusion. Cereal Food World 25: 161–162.
21. Wolf JC, Thompson DR, Ahn PC, Hegarty VJ (1974) Kinetics of available lysine losses in a soy protein isolate: Confirmation of the transition phase by protein efficiency ratio tests. J Food Sci 44: 294–295.