Protein Digestibility-Corrected Amino Acid Scores for Bean and Bean−Rice Infant Weaning Food Products

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Vegetable proteins are an integral part of infant weaning diets in Latin America. Protein quality in plant-based products, however, is constrained by amino acid composition and intrinsically present antinutritional factors. The goal of this study was to improve bean protein quality by utilizing fermentation and germination processing. The objectives were to determine if protein quality, as measured by Food and Agricultural Organization (FAO) approved True Protein Digestibility (TPD) and Protein Digestibility-Corrected Amino Acid Scores (PDCAAS), of formulated bean-based weaning products could be improved upon fermentation and germination and if protein quality could be further improved when processed beans were combined with cooked rice. Results showed that the highest TPD and PDCAAS values were obtained for cooked germinated beans combined with rice. The TPD values for products ranged from 80 to 91%, and the PDCAAS values were 0.38−0.51. There was no significant increase ($P < 0.05$) of either TPD or PDCAAS values upon fermentation. Germination increased TPD of cooked bean products; this increase was not, however, accompanied by an increase in PDCAAS. When combined with rice, the PDCAAS values for all bean products improved significantly, thus supporting the concept of cereal−legume complementation. In conclusion, this study showed the range of PDCAAS in processed black bean and bean−rice infant weaning food products. The potential for incorporation of these products into the diets of weaning age Latin American children would, however, be confirmed only after validation with growth or metabolic balance studies in human infants.

Keywords: *Black bean; rice; proteins; digestibility; fermentation; germination*

INTRODUCTION

Plant-based foods help meet the protein nutritional needs of various segments of the population, including infants and preschool children (*1*, *2*). Cereals provide more than 60% of energy and 50% of global human protein needs (*3*). Legumes also are good sources of energy, protein, and minerals and are useful sources of B vitamins (*3*). The dry bean (*Phaseolus vulgaris*) is the most widely produced and consumed legume in the world (*4*).

Common dry beans are the major protein source in the diets of people in South and Central America (*5*). Weaning-age infants in these countries are most vulnerable to protein malnutrition because plant foods have lower protein digestibility than animal-based foods (*6*, *7*). Protein malnutrition can lead to increased risk for infections, impaired physical growth, and decreased cognitive development in young children (*8*).

There are limitations to the use of dry beans as a protein source. The efficiency of utilization of proteins from beans can be compromised by the intrinsic presence of antinutritional factors such as tannins, polyphenols, and trypsin inhibitors (*9*). Black beans are rich in protein; however, they are limiting in sulfur amino acids such as cysteine and methionine (*10*).

To augment the protein quality of bean-based weaning foods and to overcome the problem of antinutritional factors, strategies have been proposed, including cereal−legume protein complementation (*11*) and the use of indigenous food-processing technologies such as fermentation (*12*) and germination (*13*).

The concept of cereal−legume complementation (e.g., rice−bean) has been applied to develop infant weaning foods with augmented protein quality (*14*, *15*). Milk proteins (rich in lysine), for instance, have been added to cereals (low in lysine) (*16*). Similarly, researchers have shown that cooked bean and cooked rice, blended in the right proportion, could replace bean broth and thus provide a balanced mixture of amino acids (*17*).

Indigenous food-processing technologies such as fermentation and germination have been used to increase protein quality. These foods have many positive attributes: favorable texture, organoleptic quality (*18*), reduced bulk (*19*), enhanced shelf life (*20*), partial or complete elimination of antinutritional factors (*21*), reduced cooking time (*22*), and improved nutritional value (*23*). The most widely used and approved method to evaluate protein quality of processed plant foods and specifically of infant weaning foods is the Protein Digestibility-Corrected Amino Acid Score (PDCAAS) (*4*, *24*−*27*). The PDCAAS provides information about the complementation potential of protein sources such as beans and rice. The U.S. FDA recognizes that the

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Table 1. Test Diet Composition[a,b]

ingredient	CB	CFB	CFCB	CGB	CB-CR	CFB-CR	CFCB-CR	CGB-CR	protein free
product[c]	35.6	34.9	35.0	34.2	48.0	50.6	51.8	51.0	0
vitamin mix[d]	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
mineral mix[d]	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5
choline bitartrate[d]	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
cellulose[d]	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
corn oil[e]	8.6	8.6	8.6	8.6	8.6	8.6	8.6	8.6	8.6
cornstarch[d]	46.1	46.8	46.7	47.4	33.7	31.1	29.9	30.66	81.7

[a] CB, cooked beans; CFB, cooked fermented beans; CFCB, cooked fermented cooked beans; CGB, cooked germinated beans; CB-CR, cooked beans−cooked rice; CFB-CR, cooked fermented beans−cooked rice; CFCB-CR, cooked fermented cooked beans−cooked rice; CGB-CR, cooked germinated beans−cooked rice. [b] Expressed as grams per 100 g. [c] Protein source. [d] Dyets, Inc. Bethlehem, PA. [e] Purchased locally.

PDCAAS method is based on human amino acid requirements and is therefore recommended for evaluating the protein quality of foods intended for human consumption (*27*). The True Digestibility of Protein (TDP) scores corrected for amino acid take into account three critical parameters of protein quality evaluation (*25*): the food protein's essential amino acid profile, its digestibility, and its ability to supply the essential amino acids in the amounts required by humans.

The current study was conducted to determine the protein quality, as measured by PDCAAS, of bean- and bean−rice-based products processed by either microbial fermentation or germination.

MATERIALS AND METHODS

Common black beans (*Phaseolus vulgaris* var. Talamanca) were provided by Dr. George Hosfield, Department of Crop and Soil Science, Michigan State University, East Lansing, MI. Extra fancy long-grain rice marketed by North Arkansas Wholesale Co. Inc., (Bentonville, AR), was purchased locally (Lafayette, IN).

Fermentation. Inoculation and fermentation procedures were conducted using the method of Rodriguez-Burger et al. (*28*) as described by Kannan et al. (*29*). Products were drum-dried (Bufloval model 175 Br5, Buffalo, NY) or combined with rice and then drum-dried.

Germination. Germination of beans was done using the same protocol as described in Kannan et al. (*29*). A half-portion of sample was drum-dried and filtered through a 35 mesh sieve. The remaining portion was combined with cooked rice and drum-dried under the following conditions: 250 °C surface temperature, 0.01 in. distance between drums, 0.8 rpm, and filtering through a 35 mesh sieve.

Analytical Methods. All products were analyzed in triplicate for moisture and fat using AOAC Methods 925.09 and 920.39C, respectively (*30*). Protein content of beans and rice was determined measuring the nitrogen content of duplicate samples with a conversion factor of 6.25, using Kjeldahl Method 960.52 (*30*). Amino acids (including sulfur-containing amino acids) were analyzed by CN Laboratories (Courtland, MN) using AOAC Official Procedure 994.12 as described in Kannan et al. (*29*).

Formulation of Bean and Bean—Rice Products. Products were formulated using the protocol described by Kannan et al. (*29*).

Formulation of Test and Control Diets. Diets were formulated as per Food and Agricultural Organization protocol (*4*). The composition of the experimental diets is given in Table 1. Test diets provided 10% protein from the bean products and 5% fat by weight. A protein-free diet served as a control. Test and control diets contained equal amounts of vitamin mix, mineral mix, choline bitartrate, cellulose, and corn oil.

Rat Feeding Protocol. Approval of the Purdue University Committee on Use and Care of Animals in Research was obtained for this study. Animals were maintained in accordance with the *Guide for the Care and Use of Laboratory Animals* (NIH Publication 86-23). Male weanling Sprague−Dawley rats (mean weight = 42.3 ± 3.8 g) were housed in individual cages at 18−26 °C and 40−70% relative humidity. Rats were fed standardized chow for an acclimation period of 2 days, and then 54 rats were divided into 9 blocks of 6 rats, with the mean weight of each block within 4 g. Water was provided ad libitum. A 15 g portion of the protein-free diet (control) or one of the test diets was fed to the rats for a 4-day preliminary period and a 5-day balance period. Rats were weighed every other day during this 11-day study. During each of the 5-day balance periods, feces and spilled food were collected from each rat and carefully separated. At the end of the 5-day balance period, spilled food was air-dried for 3 days. To determine the total food/nitrogen intake for the 5-day balance period, weights of uneaten and spilled food were deducted from total food supplied. Feces collected were dried overnight in a vacuum oven at 100 °C, weighed, ground, and analyzed for nitrogen (*30*).

Determination of PDCAAS. PDCAAS was measured following the FAO/WHO/UN protocol (*4*). For each rat, nitrogen excreted in the feces was subtracted from the amount ingested and TPD was expressed as a percentage of nitrogen intake:

$$\mathrm{TPD} = \frac{I - (F - F_k)}{I} \times 100$$

I = intake nitrogen, F = fecal nitrogen, and F_k = metabolic or endogenous fecal nitrogen. TPD values were calculated for each product.

Amino acid ratios (milligrams of an essential amino acid in 1.0 g of test product protein per milligram of the same amino acid in 1.0 g of reference protein) for nine essential amino acids plus tyrosine and cystine were calculated using the 1991 FAO/WHO/UN amino acid requirements for infants. The amino acid score is based on the amount of the most limiting amino acid. The lowest amino acid ratio provides the amino acid score for that product. Bean products were limiting in cysteine and methionine. The most limiting amino acid for the bean products with rice was tryptophan. The PDCAAS of the bean-only test products were calculated by multiplying the TPD by the cysteine and methionine amino acid score. The PDCAAS of bean−rice test products were calculated by multiplying the TPD by the tryptophan amino acid score. These scores were then expressed as a decimal.

Statistical Analysis. Data were analyzed using SAS (*31*) and tested for normality. Mean TPD values and PDCAAS for all products were computed using PROC MEANS and tested for significant differences between test groups using Student−Newman−Keuls (SNK) test ($P < 0.05$).

RESULTS

Table 2 presents the TPD (expressed as a percentage) and PDCAAS (expressed as a decimal) for the tested bean products. A wide range in TPD and PDCAAS values was observed for the products: TPD values ranged from 80 to 91% and PDCAAS between 0.38 and 0.51. CGB-CR had the highest TPD (91%) as well as the highest PDCAAS (0.51). CB, CFB, and CFCB had the lowest TPD (80%) and the lowest PDCAAS (0.38).

Table 2. True Protein Digestibility (TPD) and Protein Digestibility-Corrected Amino Acid Scores (PDCAAS) of Formulated Products[a−c]

test diet	TPD (%)	PDCAAS
CB	80.94 ± 4.4[a]	0.38 ± 0.02[a]
CFB	80.31 ± 3.8[a]	0.38 ± 0.18[a]
CFCB	80.30 ± 4.0[a]	0.38 ± 0.02[a]
CGB	86.80 ± 3.2[b,c]	0.39 ± 0.03[a]
CB-CR	84.30 ± 3.8[b]	0.47 ± 0.02[b]
CFB-CR	84.60 ± 3.3[b]	0.47 ± 0.02[b]
CFCB-CR	81.34 ± 3.7[a]	0.46 ± 0.02[b]
CGB-CR	91.00 ± 3.7[c]	0.51 ± 0.02[c]

[a] $n = 6$ rats per diet group; values are expressed as mean ± standard deviation. [b] CB, cooked beans; CFB, cooked fermented beans; CFCB, cooked fermented cooked beans; CGB, cooked germinated beans; CB-CR, cooked beans−cooked rice; CFB-CR, cooked fermented beans−cooked rice; CFCB-CR, cooked fermented beans−cooked rice; CGB-CR, cooked germinated beans−cooked rice. [c] Different superscripts within columns denote significant differences at $P \leq 0.05$ SNK.

Table 3 compares the FAO/WHO recommended essential amino acid patterns for infants with the essential amino acid composition of experimental diets used in this study. All of the cooked bean products that were not combined with cooked rice are colimiting in methionine plus cysteine and in tryptophan. The bean products that are combined with rice are colimiting in isoleucine and tryptophan and are marginally limiting in lysine and valine. The methionine plus cysteine pattern is increased in diets containing rice.

DISCUSSION

The TPD measured in this study for CB is consistent with findings reported for a similar bean product by Sarwar (*32*) and is 18 and 8% higher than that reported by Hernandez et al. (*11*) and Sarwar and McDonough (*33*), respectively. It is encouraging to note that TPD values for CFB and CFCB (both 80.3%) are similar to those reported for autoclaved pinto beans (*34*), canned kidney beans, and canned lentils (*32*) and are only somewhat higher than that reported for pinto beans (72−76%) (*35*). PDCAAS for CFB and CFCB duplicate the score reported for wheat gluten (0.25−0.37) (*32*).

Results of the current study show no improvement in PDCAAS when bean−rice products were fermented. This finding is similar to reports that fermentation did not improve the protein quality of whole white and whole black bean−rice diets (*36*). In contrast to these findings, Goyal (*37*) showed an improvement in digestibility for fermented rice−soy flour blend products. Boralkar and Reddy (*38*) and Goyal (*37*) attributed the increase in protein digestibility to an increase in proteinase activity in fermented soybeans and in fermented rice−soy flour blended products. Khalifa and Zinay (*21*) attributed the increase in the protein nutritional value of sorghum to a decrease in the tannin content of the globulin fractions, following fermentation.

Germinating the beans raised the TPD from 81 to 87% ($P < 0.05$). This increase is especially notable because it brought the TPD of CGB close to that of milk-based infant formulas (*34*). This finding agrees with Oyeleke et al. (*39*), who found that germination improves digestibility in legumes such as mung beans and garbanzo beans. However, the current study did not show an effect of germination of beans on PDCAAS. The lack of effect on PDCAAS is probably related to the similarity in the amino acid contents of cooked beans and germinated beans (Table 3). Another reason is likely to be related to the stringent amino acid scores for infants that were used in calculating the PDCAAS in the evaluation of the test products.

Nielsen (*40*) attributed the improvement in digestibility upon germination to the modification of storage proteins. Germination reportedly causes protein mobilization with the help of proteases, leading to the formation of polypeptides, oligopeptides, and free amino acids, thus facilitating an improvement in protein digestibility. It has been suggested that germination can lead to lower levels of trypsin inhibitors, tannins, phytate (*41*), and lectins (*42*) and can decrease phytohemagglutinating activity. For example, germination reduced the tannin content in mungbeans by 23−36% after a 48 h germination (*43*). It is likely that the germination time used in the current study (25 h) might not have allowed for sufficient degradation of antinutritional factors, including trypsin inhibitors, tannins, and phytate.

Digestibility increased when cooked beans and cooked fermented beans were combined with rice; this was accompanied by a marked increase in PDCAAS, from 0.38 to 0.48 ($P < 0.05$). The PDCAAS value for CB-CR observed in the current study is higher than that of

Table 3. Recommended Essential Amino Acid Patterns for Infants and Essential Amino Acids in Experimental Infant Food Products[a−c]

essential amino acid	recommended essential amino acid pattern for infants (<1 year), mean (range)[a]	CB[b,c]	CFB[b,c]	CFCB[b,c]	CGB[b,c]	CB-CR[b,c]	CFB-CR[b,c]	CFCB-CR[b,c]	CGB-CR[b,c]
histidine	26 (18−36)	29.86	31.35	31.35	31.34	28.51	26.30	26.63	28.21
isoleucine	46 (41−53)	46.72	49.90	49.90	47.44	26.98	26.84	26.84	27.49
leucine	93 (83−107)	83.45	84.60	84.60	81.54	84.53	85.63	85.63	85.88
lysine	66 (53−76)	73.83	69.13	69.13	78.10	54.64	54.78	54.78	54.97
methionine + cysteine	42 (29−60)	19.72	19.62	19.62	18.70	33.88	34.68	34.68	35.01
phenylalanine + tyrosine	72 (68−118)	91.43	95.30	95.30	89.22	94.68	97.72	97.72	96.63
threonine	43 (40−45)	43.3	42.37	42.37	40.03	39.38	40.23	40.23	38.55
trytophan	17 (16−17)	11.4	11.78	11.78	11.00	11.96	12.03	12.03	11.86
valine	55 (44−77)	51.46	51.46	53.66	49.41	56.04	56.62	56.62	55.93
total									
including histidine	460 (408−588)	451.1	457.7	457.71	446.8	430.6	434.8	434.8	434.5
minus histidine	434 (390−552)	421.3	426.3	426.3	415.4	402.0	408.8	408.5	406.3

[a] Food and Agricultural Organization/World Health Organization.[4] [b] Essential amino acid is expressed as milligrams per gram of protein. [c] CB, cooked beans; CFB, cooked fermented beans; CFCB, cooked fermented cooked beans; CGB, cooked germinated beans; CB-CR, cooked beans−cooked rice; CFB-CR, cooked fermented beans−cooked rice; CFCB-CR, cooked fermented beans−cooked rice; CGB-CR, cooked germinated beans−cooked rice.

breakfast cereals based on rice−wheat gluten and whole wheat and is lower than that of rolled oats (*33*). This increase in digestibility and PDCAAS was expected because previous research has shown that protein complementation in the appropriate proportion leads to two proteins of higher PDCAAS than either of the two individual proteins (*16*). TPD values measured in bean and bean−rice products are similar to those reported for South and Central American bean and maize−bean diets (*44*). Except for CGB-CR, TPD was slightly lower than what has been reported for similar North American diets (88−94%) and higher than that of Asian Indian plant protein diets. Digestibilities of the bean−rice products in this study were 14% higher than that reported for bean and corn combined together (*11*). This probably is a reflection of the protein quality of rice versus corn as has been suggested by Sarwar et al. (*34*), who found that adding rapeseed protein to wheat flour increased digestibility from 41 to 67%, whereas adding casein increases digestibility of the same product dramatically, to 91%. The PDCAAS value for CGB-CR (0.51) was similar to values reported for rice−soybean, wheat flour−soy protein, wheat flour−pea flour (*32*), peanut products (*33*), and milk−wheat (*11*).

PDCAAS values for the experimental products fall within the range reported by the Protein Advisory Group (*4*). PDCAAS values were higher in products combined with rice, relative to those without rice, thus confirming the hypothesized benefit of cereal−legume complementation.

Methodological differences between the current study and other similar research published in the literature merit discussion. It is well-known that the reference amino acid score influences the PDCAAS for a product. In evaluating the protein quality of the weaning food products in the present study, we utilized the 1991 FAO/WHO Consultation amino acid scoring patterns recommended for infants, which is based on human milk amino acid composition (milligrams per gram of protein). It has been suggested by Young and Pellett (*26*) that adoption of the pattern for preschool children might overestimate the value of a protein for weaning age infants. Other researchers, including Sarwar et al. (*34*), have evaluated the TPDs of their bean products on the basis of the amino acid scoring pattern used for preschool children. Also, in the present study, TPD and PDCAAS were determined at the 10% protein level compared to the 8% protein in the study by Sarwar et al. (*34*).

As has been reported by Rodriguez-Burger et al. (*28*), in vitro protein digestibilities of CB and CFB used in this study were 88 and 86%, respectively, compared to 81 and 80% obtained for the same products in the current study with the in vivo rat model. This is consistent with a report by Eggum et al. (*45*), who have shown a similar discrepancy between results of in vitro and in vivo protein digestibility of legume products. These discrepancies in in vitro versus in vivo digestibility measurements may be partly explained by strong bacterial growth in the lower gut of animals when certain legumes are consumed (*4*).

Results reported here are based on an animal model. The rat is a good model for predicting protein digestibility (*34*) and especially for screening a great number of products such as in this study; however, confirmation of these results with metabolic balance studies in infants is needed.

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LITERATURE CITED

(1) Chau, C. F.; Cheung, C. K.; Wong, Y. S. Chemical composition of three underutilized legume seeds grown in China. *Food Chem*. **1998**, *61* (4), 505−509.
(2) Simango, C. Potential use of traditional fermented foods for weaning in Zimbabwe. *Soc. Sci. Med*. **1997**, *7*, 1065−1068.
(3) Moussa, W. A.; Tadros, M. D.; Mekhael, K.; Darwish, A. E. H.; Shakir, A. H.; El-Rehim, E. A. A. Some simple methods of home processing and their implication with weaning foods. *Nahrung* **1992**, *36* (1), 26−33.
(4) Food and Agricultural Organization and World Health Organization. *Protein Quality Evaluation*; Report of a Joint FAO/WHO Expert Consultation, Food and Agricultural Organization of the United Nations: Rome, Italy, 1991; pp 26−28.
(5) Bressani, R. Grain quality of common beans. *Food Rev. Int*. **1993**, *9* (2), 237−297.
(6) Huffman, S. L.; Martin, L. H. First feedings: Optimal feeding of infants and toddlers. *Nutr. Res*. **1994**, *14*, 127−159.
(7) Underwood, B. A. Weaning practices in deprived environments: the weaning dilemma. In *Feeding the Normal Infant*; Supplement to *Pediatrics*; American Academy of Pediatrics: 1995; pp 49−57.
(8) Scrimshaw, N. S. Effect of infection on nutritional status. *Proc. Natl. Sci. Council* **1992**, *16*, 46−64.
(9) Hughes, J. S.; Acevedo, E.; Bressani, R.; Swanson, B. G. Effects of dietary fiber and tannins on protein utilization in dry beans (*Phaseolus vulgaris*). *Food Res Int*. **1996**, *29* (34), 331−338.
(10) Wu, W.; Williams, W. P.; Grimes, L. W. True protein digestibility and digestibility-corrected amino acid scores of red kidney beans (*Phaseolus vulgaris* L). *J. Agric. Food Chem*. **1995**, *43*, 1295−1298.
(11) Hernandez, M.; Montalvo, I.; Sousa, V.; Sotelo, A. The protein efficiency ratios of 30:70 mixtures of animal: vegetable protein are similar or higher than those of the animal foods alone. *J. Nutr*. **1995**, 574−581.
(12) Sarkar, P. K.; Jones, L. J.; Craven, G. S.; Somerset, S. M.; Palmer, C. Amino acid profiles of kinema, a soybean fermented food. *Food Chem*. **1997**, *59*, 69−75.
(13) Donangelo, C. M.; Trugo, L. C.; Trugo, M. F.; Eggum, B. O. Effect of germination of legume seeds on chemical composition and on protein and energy utilization in rats. *Food Chem*. **1995**, *53*, 23−27.
(14) Prinyawiwatkul, W.; Beuchat, L. R.; McWatters, K. H.; Phillips, R. D. Functional properties of cowpea (*Vigna unguiculata*) flour as affected by soaking, boiling, and fungal fermentation. *J. Agric. Food Chem*. **1997**, *45*, 480−486.
(15) Nnanna, I. A.; Phillips, R. D. Protein and starch digestibility and flatulence potential of germinated cowpeas (*Vigna unguiculata*). *J. Food Sci*. **1990**, *55*, 151−153.
(16) Codex Alimentarius Commission Document CX/VP 82/8. *The Utilization of Vegetable Protein Products in Foods and Labeling Considerations*; Food and Agricultural Organization: Rome, Italy, 1982; pp 2−3.
(17) Wondimu, A.; Malleshi, N. G. Development of weaning foods based on malted, popped, and roller-dried barley and chickpea. *Food Nutr. Bull*. **1996**, *17*, 169−174.
(18) Trugo, L. C.; Farah, A.; Trugo, N. M. F. Germination and debittering lupin seeds reduce α-galactosides and intestinal carbohydrate fermentation in humans. *J. Food Sci*. **1993**, *58*, 627−630.
(19) Bhutta, Z. A.; Hendricks, K. M. Nutritional management of persistent diarrhea in childhood: A perspective from the developing world. *J. Pediatr. Gastroenterol. Nutr*. **1996**, *22*, 17−37.

(20) Hesseltime, C. W. The future of fermented foods. *Nutr. Rev*. **1983**, *41*, 293−301.
(21) Khalifa, A. O.; Zinay, A. H. Effect of fermentation on protein fractions and tannin content of low- and high-tannin cultivars of sorghum. *Food Chem*. **1994**, *49*, 265−269.
(22) Reddy, N. R.; Piersen, M. D.; Salumkhe, D. K. Introduction. In *Legume Based Fermented Foods*; CRC Press: Boca Raton, FL, 1986; pp 1−3.
(23) Steinkraus, K. H. Indonesian tempe and related fermentations. In *Handbook of Indigenous Fermented Foods*, 2nd ed.; Steinkraus, K. H., Ed.; Dekker: New York, 1996; Chapter 1, p 7.
(24) Dewey, K. G.; Beaton, G.; Fjeld, C.; Lonnerdal, B.; Reeds, P. Protein requirements of infants and children. *Eur. J. Clin. Nutr*. **1996**, S119−S150.
(25) Henley, E. C.; Kester, J. M. Protein quality evaluation by protein digestibility corrected amino acid scoring. *Food Technol*. **1994**, 74−77.
(26) Young, V.; Pellett, P. L. Protein evaluation, amino acid scoring and the Food and Drug Administration's Proposed Food Labeling Regulations. *J. Nutr*. **1991**, *121*, 145−155.
(27) FDA. Food Labeling; general Provisions; Nutrition labeling; Label format. Nutrient Content Claims; Health Claims; Ingredient Labeling, State and local requirements and exemptions, final rules. *Fed. Regist*. **1993**, *5893*, 2101−2106.
(28) Rodriguez-Burger, A.; Mason, A.; Nielsen, S. S. Use of fermented black beans combined with rice to develop a nutritious weaning food. *J. Agric. Food Chem*. **1998**, *46*, 4806−4813.
(29) Kannan, S.; Nielsen, S. S.; Rodriguez-Burger, A. P.; Mason, A. C. Iron and zinc bioavailability in rats fed intrinsically labeled bean and bean−rice infant weaning food products. *J. Agric. Food Chem*. **2001**, *46*, 5063−5069.
(30) AOAC. *Official Methods of Analysis*, 16th ed.; Association of Official Analytical Chemists: Gaithersburg, MD, 1995.
(31) SAS Institute. *SAS System for Statistical Analysis*; Cary, NC, 1990.
(32) Sarwar, G. Digestibility of protein and bioavailability of amino acids in foods: effects on protein quality assessment. *World Rev. Nutr. Diet*. **1987**, *54*, 26−70.
(33) Sarwar, G.; McDonough, F. E. Evaluation of protein digestibility corrected amino acid score method for assessing protein quality of foods. *J Assoc. Off. Anal. Chem*. **1990**, *73* (3), 347−356.
(34) Sarwar, G.; Peace, R. W.; Botting, H. G.; Brole, D. Relationship between amino acid scores and protein quality indices based on rat growth. *Plant Foods Hum. Nutr*. **1989**, *39*, 33−44.
(35) McDonough, F. E.; Steinke, F. H.; Sarwar, G.; Eggum, B. O.; Bressani, R.; Huth, P. J.; Barbeau, W.; Mitchell, W. E.; Mitchell, G. V.; Phillips, J. G. In vivo rat assay for true protein digestibility: Collaborative study. *J. Assoc. Off. Anal. Chem*. **1990**, *73* (5), 801−805.
(36) Joseph, E.; Swanson, B. G. Protein quality of "Idli" fermented steam cakes prepared from beans (*Phaseolus vulgaris*) and rice. *Nutr. Res*. **1994**, *14* (4), 553−568.
(37) Goyal, R. Nutritional improvement and utilization of the cereal-legume blends through fermentation. M.Sc. thesis, Haryana Agricultural University, Hissar, India, 1991.
(38) Boralkar, M.; Reddy, N. S. Effect of roasting, germination and fermentation on the digestibility of starch and protein present in soybean. *Nutr. Rep. Int*. **1985**, *3*, 833−836.
(39) Oyeleke, O. A.; Morton, I. D.; Bender, A. E. The use of cowpeas (*Vigna unguiculata*) in improving a popular Nigerian weaning food. *Br. J. Nutr*. **1985**, *54*, 343−347.
(40) Nielsen, S. S. Digestibility of legume proteins. *Food Technol*. **1991**, 112−118.
(41) Gupta, C.; Sehgal, S. Development, acceptability and nutritional value of weaning mixtures. *Plant Foods Hum. Nutr*. **1991**, *41*, 107−116.
(42) Smets, G.; Van Driessche, E.; Beckman, S. Ultrastructural localization of pea lectin in the embryo and cotyledon during development by the colloidal-gold method. In *Lectins*; de Gruyter: Berlin, Germany, 1985; Vol. IV, p 453.
(43) Barrogo, C. F.; Laurena, A. C.; Mendoza, E. M. T. Polyphenols of mungbean *(Vigna radiata(L)* Wilceck): Determination and removal. *J. Agric. Food Chem*. **1985**, *33*, 1006−1009.
(44) Hopkins, D. T. *Protein Quality in Humans: Assessment and in Vitro Estimation*; Bodwell, C. E., Adkins, J. S., Hopkins, D. T., Eds.; AVI Publishing: Westport, CT, 1981; pp 169−193.
(45) Eggum, B. O.; Hansen, I.; Larsen, T. Protein quality and digestible energy of selected foods determined in balance trials with rats. *Plant Foods Hum. Nutr*. **1989**, *39*, 13−21.

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