as compared to the older selected variety Swarna and hybrid variety CSH-2. This may be due to differences between lysine content of those grain proteins and protein of CSH-1.

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Nutritive Value of Two Different Beans (*Phaseolus vulgaris*) Supplemented with Methionine

Miguel Hernandez-Infante, Gerardo Herrador-Peña, and Angela Sotelo-Lopez*

Two varieties of *Phaseolus vulgaris*, black and white beans, were supplemented with 0.6% methionine under different conditions: (a) the amino acid added at the beginning of the cooking process, (b) the amino acid added 30 min before the end of the cooking process, (c) the amino acid added at the end of the cooking, and (d) the samples dried with or without broth. Proximate analysis, amino acid determination, available lysine, in vitro digestibility, PER, and apparent digestibility were performed with the samples. Methionine was the limiting amino acid in the supplemented and nonsupplemented samples. The black beans had the lowest in vitro digestibility when they were dried with broth. This result correlated with the in vivo digestibility. PER and apparent digestibility were higher in both beans when they were dried without broth. The utilization of methionine was not affected by thermal treatment, which means that there is no difference if the supplementation is carried out at home or in the industrial processed beans.

Beans are the most consumed legumes in Mexico; however, their nutritive value is poor (Kakade and Evans, 1965; Liener, 1962) mainly due to the low content in sulfur amino acids (Bressani et al., 1961). The addition of methionine increases their nutritive value (Purdon and Brown, 1967), and since this amino acid is chemically synthesized at low prices, it would be convenient to focus its utilization in human nutrition as at the present it is mainly used in animal nutrition. In the present study a preestablished amount of methionine was added to two varieties of beans (Jaffẽ and Vega-Lette, 1968). The addition was done at different times of the home cooking process in order to determine whether heat affected the utilization of methionine.

Sección de Bromatología, División de Bioquímica, Subjefatura de Investigación Científica, Instituto Mexicano del Seguro Social, Apartado Postal 73-032, Mẽxcio 73, D.F., Mexico.

MATERIALS AND METHODS

Two edible beans (*Phaseolus vulgaris*) of different acceptance were studied: white bean, which has very low

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Scheme I

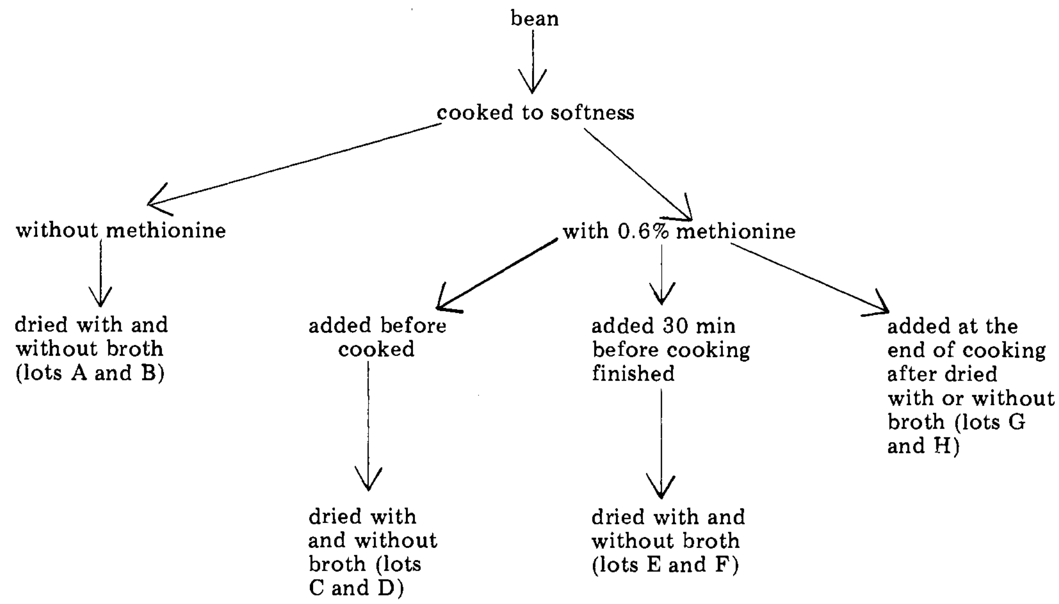

Table I. Composition of Experimental Diets

ingredients	diet groups,[a] g/100 g of diet: A, B	C, E, G	D, F, H	A, B	C, E, G	D, F, H	casein
black bean	40.16	43.86	44.84				
white bean				42.19	42.92	43.29	
sucrose	20.10	17.94	16.96	18.00	18.78	18.31	20.10
glucose	19.00	19.00	19.00	19.00	19.00	19.00	19.00
dextrin[b]	1.30						25.00
lard	7.10	7.20	7.20	7.30	7.30	7.40	8.00
corn oil	6.00	6.00	6.00	6.00	6.00	6.00	6.00
salt mix[c]	4.00	4.00	4.00	4.00	4.00	4.00	4.00
vitamin mix[d]	2.00	2.00	2.00	2.00	2.00	2.00	2.00
fiber[e]	0.34			1.51			4.41
casein (87% protein)							11.49
Protein and Energy Level in Each Diet							
N × 6.25 (g/100 g of diet)	10.0	10.0	10.0	10.0	10.0	10.0	10.0
kcal/100 g of diet (calcd)	422.1	422.6	422.0	422.0	422.3	422.8	422.4

[a] Groups A and B without methionine; C and D with 0.6% methionine added before cooking; E and F 0.6% methionine added 30 min before cooking finished; G and H with 0.6% methionine added at the end of cooking after drying. Lots A, C, E, and G were dried with broth and lots B, D, F, and H were dried without broth. [b] Productos de Maiz, S. A., México, D.F. [c] Rogers and Harper (1965). [d] See text. [e] Fiber cellulose type, Teklad test diets, Wisconsin, WI.

consumption, and black beans, whose consumption in Mexico is very high. The samples were obtained from a local market in Mexico City. Each sample was divided into eight 1-kg lots. The cooking treatment is described in Scheme I.

The DL-methionine used in this experiment was provided by ALBA-MEX Laboratories, Mexico, D.F., and was added in powder form with the other ingredients of the diet. The beans were cooked by means of boiling with water to softness (4.5 h for the black beans and 8.5 h for the white beans). After cooking, the beans were dried with or without broth as shown in Scheme I, in the oven at 60–70 °C for 24 h and then ground in an Arthur Thomas Co. mill to pass through sieve no. 20. Salt at 4% was also added in the same conditions as methionine. The chemical analyses were made in the flours and were used to prepare the diets for the protein efficiency ratio (PER) and in vivo digestibility determinations.

Methods. The proximate analysis was done according to AOAC techniques (AOAC, 1970). The amino acid determinations were performed after an acid hydrolysis using 6 N HCl at 110–115 °C for 20 h, using an amino acid analyzer Perkin-Elmer KLA-5 (Moore and Stein, 1963). Tryptophan content was measured following Lombard and Lange's technique (Lombard and Lange, 1965).

On the basis of amino acid composition, the chemical score was calculated as the percentage of the most deficient essential amino acid in the protein (Mitchell and Block, 1946), using whole egg as the reference pattern (FAO, 1970). The available lysine was measured following the Bruno and Carpenter technique (Bruno and Carpenter, 1957), and the in vitro protein digestibility was determined according to Oke and Umoh (1974). The diets used for PER determinations were isocaloric with 10% bean protein. Since the beans were supplemented with 0.6% DL-methionine, the final concentration of this amino acid in the diet was 0.26%. Diet composition and ingredients used are described in Table I. The vitamin diet fortification mixture was comprised of (g/kg) retinyl acetate (200000 units/g), 4.5; cholecalciferol (400000 units/g), 0.25; α-tocopherol, 5.0; ascorbic acid, 45.0; inositol, 5.0; choline chloride, 75.0; menadione, 2.25; p-aminobenzoic, 5.0; niacin, 4.5; riboflavin, 1.0; pyridoxine hydrochloride, 1.0; calcium pantothenate, 3.0; biotin, 20 mg; folic acid, 90 mg; vitamin B_{12}, 1.35 mg; dextrose q.s., 1 kg (Nutritional Biochemicals, Cleveland, OH).

For PER and in vivo digestibility Sprague Dawley rats, 21–23-days old and housed in individual cages, were used.

Table II. Amino Acid Composition and Chemical Score of Black and White Bean (g/16 g of N)

amino acids	black bean	white bean
Tyr	3.528 ± 0.50[a]	2.582 ± 0.20
Phe	5.812 ± 0.46	6.382 ± 0.26
Lys	7.102 ± 0.43	8.044 ± 0.57
His	1.883 ± 0.07	3.155 ± 0.26
Arg	5.503 ± 0.22	7.219 ± 0.63
Asp	13.820 ± 0.47	12.108 ± 0.96
Glu	15.308 ± 0.18	16.030 ± 1.32
Thr	4.376 ± 0.09	5.155 ± 0.46
Ser	6.301 ± 0.20	7.754 ± 0.73
Ala	4.331 ± 0.12	4.083 ± 0.30
Gly	4.337 ± 0.12	4.083 ± 0.30
Val	5.084 ± 0.14	5.902 ± 0.49
Cys	0.428 ± 0.04	0.671 ± 0.09
Met[b]	0.906 ± 0.05	0.960 ± 0.10
Met[c]	2.746 ± 0.31	3.381 ± 0.40
Ile	4.258 ± 0.11	4.855 ± 0.49
Leu	8.630 ± 0.28	12.700 ± 0.67
Trp	1.433 ± 0.06	1.200 ± 0.07

	chemical score	
	black bean	white bean
	total sulfur amino acids	total sulfur amino acids
without Met	28.51	28.89
with 0.6% Met	64.68	68.17

[a] Values represent means ± SEM of nine analyses of each sample. [b] Mean of three analysis of no supplemented beans. [c] Mean of six analyses of 0.6% methionine supplemented beans.

Table III. Remainder Methionine after the Cooking Treatment (g/16 g of Nitrogen)

lots[a]	black bean	white bean
raw	0.828	0.820
A	1.023	1.160
B	0.868	0.900
C	2.716	4.360
D	2.235	2.720
E	2.435	3.080
F	2.263	2.610
G	2.581	2.620
H	4.250	4.900

[a] The meaning of letters A–H is the same as in Table I.

The temperature of the animal house was 21 °C. Food and water intake was ad libitum. The experiment was carried out for 21 days, and food consumption and body weight were individually recorded twice a week. During the last week, the feces were collected and dried in the oven at 70 °C for nitrogen determination in order to calculate the apparent digestibility.

Variance analysis, correlation coefficient, and the Tukey multiple t test (Steel and Torrie, 1960) were used for statistical analysis. Data expressed in tables and text represent mean ± SEM.

RESULTS

Protein content was 23.1% for the black bean and 23.4% for the white bean. Carbohydrate, crude fiber, and ether extract content in both beans were similar to data previously reported (Aykroyd and Doughty, 1964; Wu Loong et al., 1966). The ashes were increased (due to the salt addition) from 3.9 to 7.2% for the black and from 3.9 to 7.6% for the white bean.

Amino acid content (Table II) was similar to that previously described by Bressani (1972).

After the addition of methionine, the total content of this amino acid increased from 0.906 ± 0.05 to 2.746 ± 0.31 g/100 g of protein in the black bean and from 0.960 ± 0.01 to 3.381 ± 0.40 g/100 g of protein in the white bean. Consequently, the chemical score increased significantly in both beans. Table III shows the methionine content after each treatment. It was found that in all the groups, except the supplemented beans at the end of cooking (lots G and H), the methionine content was higher in the beans dried with broth.

Table IV. Available Lysine Content[a] in Black and White Beans Supplemented with 0.6% Methionine and 4% Salt (g/16 g of N)

sample	black bean	white bean
raw bean	7.10	8.04
cooked beans		
lot[b]		
A	4.87	5.23
B	5.43	6.91
C	5.22	6.37
D	6.63	7.52
E	5.97	7.23
F	6.22	7.66
G	5.60	6.57
H	6.50	7.34

[a] Bruno and Carpenter's technique. [b] The meaning of letters A–H is the same as in Table I.

Table V. In Vitro Protein and in Vivo Nitrogen Digestibility of Black and White Beans under Different Treatments

	black bean digestibility, %		white bean digestibility, %	
lot	in vitro	in vivo[a]	in vitro	in vivo
A	58.4	62.8 ± 1.76 (8)[b,c]	64.3	77.0 ± 1.42 (6)
B	67.1	67.3 ± 1.06 (8)	61.5	77.1 ± 1.02 (6)
C	62.0	63.2 ± 0.75 (7)	70.4	76.4 ± 1.32 (7)
D	68.6	61.6 ± 1.66 (8)	68.6	77.7 ± 0.81 (8)
E	58.8	58.5 ± 1.29 (5)	68.9	82.4 ± 0.64 (7)
F	64.3	62.0 ± 0.95 (8)	70.9	86.1 ± 0.81 (8)
G	57.7	57.9 ± 0.98 (8)	60.2	77.7 ± 0.84 (8)
H	62.6	65.5 ± 1.59 (6)	67.9	83.0 ± 1.38 (6)

[a] Apparent digestibility. [b] Values are expressed as means ± SEM. [c] Number of rats. In some groups the number of rats was less than eight due to death.

Table IV shows the results of available lysine obtained in the samples under different treatment conditions. The highest values were found in the raw beans. It was clear that thermal treatment during cooking decreased the available lysine, and the lowest values were found in the cooked beans dried with the broth.

The in vitro digestibility was similar in both legumes; however, in vivo digestibility was better for the white bean when compared to the black bean ($P < 0.001$) (Table V).

The PER values of the beans under different treatments are shown in Table VI. PER values are consistently better for the white bean under any circumstances. However, the highest values were obtained when the beans were dried without broth and were very close to the casein values.

The variance analysis of the corrected PER of black and white beans under different treatments showed significant differences ($F = 60.982$ and 31.276, $P < 0.001$, respectively).

The Tukey t test was done in order to find the statistical discrimination between the different diets. The results are shown in the Table VI. The highest values were found in the supplemented beans and those dried without broth, at any stage at which methionine was added. Chemical score showed the highest correlation with PER ($r = 0.800$, $P < 0.001$).

DISCUSSION

Although the chemical score of the beans was remarkably improved by the addition of methionine, me-

Table VI. PER of the Black and White Beans with and without Supplementation under Different Treatment Conditions

diet lot	black bean exptl PER	corrected PER to casein at 2.50[d]	white bean exptl PER	corrected PER to casein at 2.50[d]
A	0.28 ± 0.09 (7)[a,b]	0.28[d]	0.81 ± 0.09 (6)	0.79[e]
B	0.29 ± 0.10 (7)	0.29[d]	1.40 ± 0.21 (5)	1.36[d]
C	1.88 ± 0.13 (6)	1.89[bc]	2.45 ± 0.10 (6)	2.38[ab]
D	2.10 ± 0.13 (8)	2.11[ab]	2.62 ± 0.16 (8)	2.55[a]
casein[c]	2.49 ± 0.12 (8)	2.50	2.57 ± 0.16 (7)	2.50
E	2.28 ± 0.11 (5)	2.11[ab]	2.90 ± 0.06 (6)	2.37[ab]
F	2.67 ± 0.10 (7)	2.46[a]	2.83 ± 0.16 (7)	2.32[ab]
G	1.64 ± 0.13 (7)	1.51[c]	2.32 ± 0.11 (6)	1.90[bc]
H	2.73 ± 0.08 (6)	2.52[a]	2.98 ± 0.10 (7)	2.44[a]
casein[c]	2.71 ± 0.10 (8)	2.50	3.05 ± 0.15 (8)	2.50

[a] Mean ± SEM. [b] Number of rats are indicated in parentheses. In some groups the number of rats was less than eight due to death. [c] There are four values for casein because the PER determination was done in four different experiments. [d] Values within a column not having the same superscript letter differ significantly ($P < 0.05$).

thionine continued to be the limiting amino acid. However, because of the characteristic flavor of methionine (detected even at 0.6% in the bean) it would not be desirable to increase the amount of such amino acid since it will cause a diminishing in the acceptability of the supplemented beans at least for humans.

In another experiment made in our laboratory with red beans (results not published) it was found that the PER did not change significantly in diets prepared with 0.15–1% of methionine. It suggests that the methionine content in the beans dried with broth is not significantly affecting the nutritive quality of the product.

It was expected that the cooking time used in the two beans could affect the quality of them; however, the white bean (which took more time to be cooked) gave better results than the black bean, both unsupplemented and supplemented. This was especially true in both cases if the degree of methionine defficiency was similar. This could indicate that the remaining toxics present in the cooked black bean and its lowest digestibility are responsible for this difference, as it has been previously reported (Elías et al., 1976; Sotelo et al., 1978).

The available lysine was decreased in a similar fashion in both cooked legumes, despite the fact that white beans needed the double cooking time. Apparently, the factor which most affects available lysine was the way the beans were dried since the content of the amino acid was lower in the beans dried with broth than in those dried without broth.

As Jaffé et al. (1976) have mentioned, the bean broth is a convenient food for infants because its vitamin and 10% protein content. However, if this broth is dried to a powder, its nutritive value is affected and it is reflected in the decreased PER and available lysine.

The in vitro digestibility was lower than that reported by others (Bressani et al., 1977; Gómez-Brenes et al., 1973), probably owing to differences in the method used. However, a significant linear correlation was observed between in vitro and in vivo digestibility ($r = 0.591$, $P < 0.02$). Once again, the fact that white beans were more digestible than black beans may be the consequence of the remaining toxics after the cooking process in the latter.

From the above result it is suggested that (a) the methionine utilization was not affected when it was added to the beans before cooking, (b) the addition of methionine did not improve the digestibility in these beans but did in fact improve the PER, (c) the beans dried with broth exert a negative influence upon the benefit of the supplementation. This could be due to the polyphenols content, as it was reported by Mondragón and González (1978) and also to a water-soluble undialyzable and thermal estable factor mentioned by Evans and Bauer (1978).

It would be convenient to use different drying processes or to eliminate the broth, thus reducing the time to dryness.

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