

# Protein digestibility and protein quality of common beans (*Phaseolus vulgaris*) fed alone and with maize, in adult humans using a short-term nitrogen balance assay<sup>1-3</sup>

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**ABSTRACT** Various experiments using young human adults were carried out to measure the protein digestibility of common beans, fed alone and in different forms and fed together with maize as tortillas. In all, 36 young men were used. Common bean protein digestibility was significantly lower than that observed from meat, and similar to the digestibility of a textured vegetable protein soy protein product. It was also observed that forms of consumption may affect protein digestibility of beans. Using a short-term multiple intake protein quality assay method, it was found that feeding beans alone, N equilibrium was reached with an intake of 114 mg N/kg/day, while when fed with maize in a 87:13 ratio, N equilibrium was reached with 98 mg N/kg/day. This was attributed to both a complementary protein effect and an increase in protein digestibility. *Am. J. Clin. Nutr.* 34: 1893-1898, 1981.

**KEY WORDS** Bean protein digestibility, bean protein quality

## Introduction

Maize or rice in combination with beans constitute the basic staples of the Central American Diet. Data collected from nutritional surveys show that black beans are consumed in Guatemala and El Salvador, while the red-colored seed is preferred in the rest of the countries (1). The amount consumed in all cases is relatively low compared to the cereals. The protein intake from such diets is, therefore, low in amount and quality. Emphasis is then made on the advantage of increasing bean consumption in order to improve the nutritional value of the diet. The contribution of lysine and tryptophan from the beans to the limited amount existing in the cereal should considerably improve the nutritional value of the diet, as it has been reported by several authors (2).

Protein digestibility of common beans may be a limiting nutrition factor when increasing the amount consumed. Most of the data reported in the literature on the digestibility of beans (*Phaseolus vulgaris*) protein have been obtained from animal studies. Data on humans are relatively scarce. It is the purpose of this paper to present and discuss the find-

ings from a series of experiments with humans designed to measure nitrogen digestibility of various bean samples fed alone or in mixtures with maize. The report also includes the evaluation of the protein quality, in young human subjects, of black beans fed alone and in a 87:13 mixture with maize as tortilla, evaluated by the nitrogen balance index short-term assay (3).

## Materials and methods

Nitrogen balances were run using a total of 36 young men 23 to 35 yr of age. Most of them had participated at one time in similar studies as members of the diet squad of INCAP. They were given a physical checkup and a

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clinical analysis of blood, urine, and feces to assure their good physiological condition.

Common black and red beans (*P. vulgaris*) purchased in the market were used. Cooked beans were prepared in batches so that they could be eaten in a week's time. The proportion 1:3 beans:water for overnight soaking (16 to 18 h) was used. This was followed by cooking in the retort at 16 lb pressure for 30 min. It was necessary, in some cases, to add water to cover the grain before placing the 3-gallon containers into the cooker. The cooked beans were salted to taste and used whole in one experiment. In a second experiment a lot of beans was passed through a pulper finisher using a fine sieve at low speed to remove part of the skin. This was considered as sieved beans or "colados". In another case, the beans were ground wet using a Waring Blendor and these were called ground beans or "molidos". These three preparations represent the forms in which beans are usually consumed. Individual portions were weighed out for each meal for every subject. After preparation and portion weigh out, the portions were frozen until the time of feeding. Fried beans were prepared with 20 g of cottonseed oil and 10 g of chopped onion for seasoning.

A large lot of tortillas made from white maize by the common household method was also prepared and divided in portions to be fed with the beans, 3 times a day.

Digestibility tests were run with 2 days adaptation and 3 days of quantitative fecal collection. On the other hand, the methodology for the protein quality evaluation of beans fed alone or in an 87:13 weight ratio with maize was the same as for the nitrogen balance index short method described in earlier publications (3). Carmine and charcoal markers were used to separate the fecal samples between periods. All subjects also received a complete multivitamin and mineral capsule.

## Results

### Experiment I

Fifteen men participated in this study. They were fed 0.7 and 0.8 g protein/kg/day from common black or red beans or meat for the digestibility test, with a basal diet made of foods low in protein and high in calories as described in Table 1. Therefore, beans or meat provided over 95% of the protein intake. Energy intake was adjusted to 45 kcal/kg/day with a constant intake of water. The results are shown in Table 2. Apparent digestibility was similar for the black or red bean diets but significantly lower when compared to ground meat ( $p < 0.05$ ).

For comparative purposes, results on the protein digestibility of ground meat and a textured vegetable protein (TVP) soy product are included (4). The value for meat in the present study was slightly higher when compared to the value reported previously. These differences may be due to nitrogen intake.

TABLE 1  
Ingredient composition of basal diet

	g
Instant coffee	3
Low protein bread	300
Apple marmalade	30
Sugar	25
Margarine	30
Vegetable oil	60
Starch cookies	300
Cornstarch soup	480
Fresh fruit (apple or pineapple)	200
Soft drink	1 bottle
Artificial flavored drink	2 glasses
Vitamin and mineral supplement*	1 capsule
Sugar candies	To meet individual
Artificial flavored drink	caloric needs

\* UNICAP T. Upjohn Company, S.A., Guatemala City, Guatemala, C.A.

When the intake was 129.8 mg, the digestibility was 82.1%, while the same group of subjects with an intake of 188.2 gave digestibility of 86%. These results confirm other protein digestibility analyses in humans in that lower intakes give lower values (4, 5). No difference was found in the digestibility between the TVP and the beans even though the intake was significantly less for the textured soybean (4). Common beans have been called the "poor man's meat" from the economical point of view; however, they seem to be poorer because of their lower nutritional value which starts with a lower protein digestibility.

### Experiment II

It has been observed that when beans are consumed whole, a good number of them are excreted without the effect of the digestive process. This observation, and the fact that rural populations consume up to 32% of the diet as whole beans, led the investigators to search if digestibility would be affected by the form of preparation for consumption. If whole beans pass through the gut and are excreted whole without breaking the bean skin, it is then assumed that the digestibility would be lower than when compared to beans consumed ground or sieved. A nitrogen balance test with nine subjects participating was planned using a Latin square design. The

TABLE 2

Nitrogen digestibility of beans (*P. vulgaris*), meat, and a texturized soybean product fed to adult humans as the main protein sources

Diet	n	Intake	Fecal excretion	% Apparent digestibility	% True digestibility
		mg/kg/day	mg/kg/day		
Ground black beans	5	173.6 ± 3.3	39.6 ± 4.1	77.0 ± 2.4	91.4 ± 4.5
Ground red beans	5	175.1 ± 3.8	41.3 ± 4.8	76.6 ± 2.4	90.0 ± 2.7
Ground meat	5	188.2 ± 1.0	26.2 ± 2.2	86.0 ± 1.2	101.9 ± 1.6
Ground meat*	5	129.8 ± 1.4	23.2 ± 3.4	82.1 ± 2.7	97.4 ± 1.8
Texturized soybean*	5	131.4 ± 1.2	29.4 ± 4.2	77.7 ± 3.1	93.7 ± 3.7

\* Navarrete et al. (4).

TABLE 3

Nitrogen digestibility of black beans as the main source of protein fed to adult men

Form of ingestion of beans	Subjects	Intake	Fecal excretion	Digestibility	
				Apparent* %	True %
		mg/kg/day	mg/kg/day		
Whole	8	127.4 ± 0.7	57.9 ± 5.6	54.6 ± 4.1	75.9 ± 3.4
Ground	9	125.4 ± 1.0	53.1 ± 5.2	57.8 ± 3.9	81.2 ± 4.6
Sieved	9	123.2 ± 0.9	63.5 ± 4.8	48.4 ± 4.8	71.9 ± 4.6
Nitrogen-free diet	9		30.0 ± 2.4		

$$* \% \text{ Apparent digestibility} = \frac{\text{N intake} - \text{fecal N}}{\text{N intake}} \times 100.$$

$$\% \text{ True digestibility} = \frac{\text{N intake} - (\text{fecal N} - \text{metabolic fecal N})}{\text{N intake}} \times 100$$

subjects were fed black whole beans, ground or sieved, for 5 days added to the basal diet described in Table 1. During the last 3 days of each 5-day feeding period, intake and fecal data were collected to measure nitrogen digestibility. A protein level of 0.65 g/kg/day was fed with 45 to 50 cal to maintain body weight. At the end of the bean feeding period, all subjects received a low protein diet as used in other experiments (3, 5) to determine endogenous losses and in order to calculate true digestibility.

The results are shown in Table 3. Nitrogen digestibility of whole, ground and sieved black beans was obtained as follows: 54.6, 57.8, and 48.4%, respectively. Fecal excretion was higher in the group fed the sieved beans (63.5 mg/kg/day) while the group fed the ground beans excreted 53.1 mg/kg/day and the group fed whole beans, 57.9 mg/kg/day. Thus protein digestibilities of whole and ground beans were statistically alike, but different from that found for sieved beans. Nitrogen digestibility of black beans was significantly different ( $p < 0.01$ ) in this group when compared to the previous experiment.

Protein intake must have influenced the results, as they were 173.6 mg in the first experiment and 125.4 in the second. True digestibilities were calculated using an endogenous fecal excretion value of 30.4 mg N/kg/day. As expected, values for all preparations increased.

### Experiment III

A third experiment was carried out to measure the protein quality of ground black beans and of the 87:13 mixture by weight of maize tortilla:black beans fed with the basal diet described in Table 1. Twelve young adult men participated in a balance study using the short nitrogen balance index method as described in other publications (3-5). The subjects received a diet low in nitrogen (22.0 mg/kg/day) for 3 days to determine endogenous losses during the last 2 days, followed by increases every 2 days in protein intake of 0.2, 0.4, and 0.6 g protein/kg/day. Energy was kept constant, as indicated previously. Six of the men received their protein from black ground beans, while the other six received a mixture of 73.25% protein from tor-



tilla and 26.75% protein from beans, equivalent to the relation 87:13 on a raw weight basis. Apparent nitrogen digestibility shown in Table 4 for the black beans and the mixtures of tortilla and black beans was similar when having a protein intake of 0.6 g/kg/day. These data were obtained from the last 2 days of the trial, when all the subjects received that intake. Regression equations between nitrogen intake and nitrogen balance were calculated for all the subjects, and the values for each subject are shown in Tables 5 and 6. The average equation was  $Y = -62.69 + 0.54 X$  for the black beans, and  $Y = -86.96 + 0.88 X$  for the tortilla:black beans, with  $r$  values of 0.75 and 0.89, respec-

tively. It was found that the protein quality of the 87:13 tortilla:bean mixture was higher than that of the beans alone. When the subjects were fed cooked beans alone, all six subjects were in negative nitrogen balance at the 0.6 g p/kg/day level of intake; however, all subjects on maize and beans were in positive nitrogen balance at the same level of protein intake.

### Discussion

The low protein digestibility of common beans and other food legumes in comparison with food proteins of animal origin has been recognized by various workers; however, no

TABLE 4  
Nitrogen digestibility of black beans and of tortilla: black beans fed to adult men

Diet	Subjects	Intake mg/kg/day	Fecal excretion mg/kg/day	Digestibility	
				Apparent %	True %
Black beans	6	115.6 ± 0.9	45.8 ± 2.6	60.0 ± 2.2	74.9 ± 2.9
Tortilla:black beans	6	127.6 ± 0.5	42.0 ± 6.1	61.0 ± 9.0	86.7 ± 2.7
Nitrogen-free diet			20.9		

TABLE 5  
Regression equations between nitrogen intake and nitrogen balance when feeding cooked black beans\*

Subject	NR = a + b (NI)	r	SE-B
1	NR = - 39.73 + 0.31 (NI)	0.56	0.32
2	NR = -100.97 + 0.85 (NI)	0.93	0.23
3	NR = - 50.68 + 0.46 (NI)	0.71	0.31
4	NR = - 85.98 + 0.80 (NI)	0.83	0.36
5	NR = - 35.99 + 0.30 (NI)	0.99	0.61
6	NR = - 63.15 + 0.62 (NI)	0.96	0.12
Average	NR = - 62.69 + 0.54 (NI)	0.75	0.10
Average	NR = - 57.4 + 0.81 (NA)	0.82	0.12

\* NR, nitrogen retained; NI, nitrogen intake; NA, nitrogen absorbed.

TABLE 6  
Regression equation between nitrogen intake and nitrogen balance when feeding corn:black beans in a 87:13 weight ratio\*

Subject	NR = a + b (NI)	r	SE-B
1	NR = - 89.47 + 0.94 (NI)	0.96	0.19
2	NR = -105.91 + 0.88 (NI)	0.82	0.43
3	NR = - 75.68 + 0.74 (NI)	0.86	0.30
4	NR = - 83.54 + 0.90 (NI)	0.96	0.18
5	NR = - 88.42 + 0.90 (NI)	0.94	0.22
6	NR = - 80.58 + 0.94 (NI)	0.99	0.08
Average	NR = - 86.96 + 0.88 (NI)	0.89	0.09
Average	NR = - 63.82 + 0.95 (NA)	0.92	0.08

\* NR, nitrogen retained; NI, nitrogen intake; NA, nitrogen absorbed.

explanation has been found, although various possibilities have been suggested (6). Most of the studies have been conducted with experimental animals, and only a few with human subjects. The results presented clearly indicate that beans, as tested in adult humans, also have a low digestibility. Although it is recognized that the differences in protein digestibility of beans as compared to meat (Table 2) may have some limitations due to the relatively short fecal collection period (3 days), these differences still persist even when corrected for endogenous fecal loss. In the present investigation the individual's own endogenous nitrogen excretion value obtained at the 3rd day on the basal diet was used for calculating true protein digestibilities. These values varied from 21.0 to 30.0 mg N/kg body weight/day, and are significantly higher than the value reported at the end of 14 days on a nitrogen-free diet containing about 10 mg/kg/day (7). If this value is used, the differences in true protein digestibility between beans and meat are larger. In general, very little attention has been given to protein digestibility as such, even though it represents the first biological process affecting protein quality evaluation and estimation of protein allowances. One reason may be due to the fact that most workers deal mainly with animal protein which, unless heat damaged, always has a high digestibility.

Analysis of data derived from human assays relating protein intake to protein digestibility has shown that at the lower levels of protein intake, digestibility of the protein is lower than at levels close to, and slightly above, the nitrogen equilibrium point (4). This is an important consideration in the evaluation of protein by multiple point assays, and further studies are needed to establish if such relationship really exists.

The results of the present report are of interest, since the high protein quality observed when beans were fed with maize is due, to some degree, to an improved nitrogen digestibility and to the protein complementary effect, which has been well documented, between bean and maize proteins (2). Nitrogen equilibrium as an average with the mixture of maize and beans was 98.1 mg N/kg/day, equivalent to 0.6 g protein/kg body weight/day, as compared to 114.2 mg N/kg/day for beans fed alone.

In a recent study, Calloway and Kretsch (8) reported a protein digestibility of 69% for subjects fed a Guatemalan diet based on beans and cereal grains (maize, rice, and wheat). This result compared well with the value of the present report of 61%, although there was a difference in protein intake, between the two studies, of 0.875 g for the first study and 0.61 g for the second study. One further difference is that in the present study fecal calculations represent short-term observations.

The large fecal output of nitrogen found in the present study and those of Calloway and Kretsch (8) and Graham et al. (9) is probably due to the poor digestibility of beans. The data suggest that lower protein intake values for nitrogen equilibrium could be possible if the protein digestibility of beans was increased, even with present levels of sulfur containing amino acids in beans. Therefore, it would appear to be worth investigating the reasons for the low protein digestibility of common beans. In one study conducted with dogs (10) it was found that a sodium hydroxide soluble protein fraction extracted from cooked beans had a lower digestibility than the sodium hydroxide insoluble fraction. Similar results have been shown recently with adult humans. Therefore, studies are being conducted to learn more about such a fraction. The effects of particle size on digestibility as conducted in this study were surprising, since it was expected for sieved beans to have a higher digestibility than whole beans. This had led to studies designed to learn if polyphenolic compounds present in beans (11) are responsible for their low digestibility. ■

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