Effect of Cooking on the Antinutritional Factors of Lima Beans (*Phaseolus lunatus*)

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ABSTRACT

Lima beans (Phaseolus lunatus) obtained locally were analysed for inherent antinutritional factors such as trypsin inhibitors, phytohemagglutinin, polyphenols and cyanogenic glucosides. The effect of processing (cooking) on the residual levels of these antinutritional factors, as well as crude protein and in vitro protein digestibility, was also determined.

The raw samples, cooked samples and cooking water were found to contain various levels of the antinutritional factors determined. Significant differences (P < 0.05) were observed between the raw and cooked samples for all parameters analysed.

INTRODUCTION

The phenomenal increase in population during the last 25 years has aggravated the world food crisis, and there is a high possibility that the situation could worsen as the gap between the world's protein supply and population continues to widen. Plant foods such as cereals and legumes have consistently been listed as the major potential sources of dietary protein for feeding the world of tomorrow and research efforts are being directed to this area to identify and evaluate unexploited sources.

Lima beans (*Phaseolus lunatus*) contain between 22 and 25% protein and make a significant contribution to the protein and energy requirements of many Nigerians. Their role as a source of protein is, however, affected by several factors including low protein digestibility (Aletor & Fetuga, 1984)

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flatulence (Sanchez *et al.*, 1966) and the presence of numerous antinutritional constituents, which made up the most important single factor affecting utilization. This study was carried out, therefore, to determine the effect of processing (cooking) on the level of these antinutritional factors in lima beans.

MATERIALS AND METHODS

The lima bean samples, obtained from a local market in Ogun State, were white in colour, and kidney shaped. Fifty gram portions of the beans were cooked at 60, 120 and 160 min. The resulting materials were dried in an air oven and ground in a Wiley mill. Individual bean flour and the different cooking waters were stored in screw cap bottles in the refrigerator until analysed.

Analytical procedure

Phytic acid level was determined by the method of Young and Greaves (1936), trypsin inhibitory activity was estimated by the method of Kakade *et al.* (1969) and the tannin content was measured by the modified Vanillin-Hydrochloric acid method of Price *et al.* (1978). Total polyphenols were determined by the Prussian-blue method of Price and Butler (1977), while the phytohemagglutinin activity was estimated by the method of Liener (1954). Hydrocyanic acid levels and crude protein contents were determined by standard methods (AOAC, 1980), while the in-vitro protein digestibility was determined by the method of Maga *et al.* (1975). All determinations were in duplicates on triplicate samples.

Statistical analysis

The mean values of the data were subjected to the Students t-test and oneway analysis of variance in order to compare changes in the level of antinutritional factors present in the raw and processed beans.

RESULTS AND DISCUSSION

The results of the various analyses carried out are presented in Tables 1 and 2. Trypsin inhibitor activity was observed in the raw samples with a mean value of 3.4 ± 0.6 trypsin inhibitor units (Tiu)/ml, and cooking at the

Samples	Phytic acid (mg/100 g)	Trypsin inhibitor activity (Tiu/ml)	Phytohemagglutinin activity	Hydrocyanic acid (mg/kg)	Total polyphenol (mg/g)	Tannins (mg/g)
Raw beans	234 ± 3.6	3.4 ± 0.6	+	420 ± 1.9	122 ± 1.6	0.59 ± 0.7
Cooked for 60 min	198 ± 3.6	1	+	377 ± 5.7	54.8 ± 1.5	0.31 ± 0.03
Cooked for 120 min	131 ± 3.6	l	+	217 ± 1.9	$41 \cdot 4 \pm 1 \cdot 5$	0.0195 ± 0.04
Cooked for 160 min	885 ± 3.6	ł	÷	115 ± 1.9	21.2 ± 0.6	0.11 ± 0.04
Cooking water at 60 min	25.7 ± 7.3	I	+	398 ± 1.9	30.7 ± 0.8	0.22 ± 0.008
Cooking water 120 min	$24 \cdot 2 \pm 2 \cdot 2$	1	+	377 ± 5.7	24.6 ± 1.6	0.18 ± 0.007
Cooking water 160 min	12.9 ± 3.6	ļ	÷	244 ± 1.9	15.1 ± 0.8	0.024 ± 0.003

TABLE 1

	In-vitro protein digestibility (%)	Crude protein g/100 g dry matter basis
Raw	43.0 + 1.3	23.5 ± 0.07
Cooked 60 min	51.4 ± 1.4	22.2 ± 0.07
Cooked 120 min	70.3 ± 1.4	21.2 ± 0.2
Cooked 160 min	84·6 ± 1·5	20.2 ± 0.2

 TABLE 2

 Crude Protein Content and in-vitro Digestibility of Lima beans

different times resulted in a 100% loss. It was also observed that the cooking water of the samples had no trypsin inhibitor activity.

All samples contained phytic acid at different levels and a significant difference (P < 0.05) was observed between the raw and cooked samples. The levels of phytic acid in the cooking water were not significantly different from one another (P > 0.05). The level of total polyphenols in the samples were 122, 54.8, 41.4 and 21.2 mg/g respectively (Table 1). The cooking water had levels of total polyphenols (30.7, 25.0 and 15.9 mg/g respectively), which were significantly different (P < 0.05).

All samples contained hydrocyanic acid, with the raw beans containing the highest level (420 mg/kg), while the cooked samples, at 60, 120 and 160 min, had levels of 377, 217 and 115 mg/kg respectively. A highly significant difference was observed among the samples analysed (P < 0.01). The cooking water samples also contained hydrocyanic acid levels which were significantly different at (P < 0.01). The levels of tannins in the samples were found to be 0.56, 0.31, 0.20 and 0.11 mg/g respectively (Table 1). A significant difference (P < 0.05) was observed between the raw and cooked samples at 120 and 160 min. However, the differences between the raw sample and cooked sample at 60 min, and between the 60 and 120 min cooked samples, were not significant (P > 0.05). Cooking water of the sample analysed also had high tannin levels, and a significant difference (P < 0.05) was observed (Table 1).

Although all the samples had phytohemagglutinin activity, the number of cells which were agglutinated decreased as the serial dilution was increased from 1:10 to 1:20. Similarly, agglutination was reduced with increased cooking time. The incomplete destruction of hemagglutinin activity may have been due to the high level of other antinutritional factors which interfered with its destruction.

The levels of in-vitro protein digestibility and crude protein are in Table 2. Digestibility values ranged from 43% in the raw sample to 84.6% in the fully cooked sample which were significantly different (P < 0.05). Crude protein contents on a dry matter basis were found to be 23.5, 22.2, 21.2 and

20.2 g/100 g for the raw and cooked samples respectively. A significant difference (P < 0.05) was also observed between the raw and the fully cooked samples.

Legumes have been recognised for their high protein content relative to other plants, and the results obtained in this study showed that the crude protein content of lima beans was high and capable of making a significant contribution to the nutrient intake of the consumers. All the results further showed that lima beans, though rich in protein, have some antinutritional components which could limit the nutritional quality of this legume when consumed raw.

It was observed that cooking resulted in 100% loss of trypsin inhibitor activity. This finding agrees with the observation of Osborne and Mendel (1917); Kakade *et al.* (1974) and Ologhobo (1981). Heinz-Fraenkel-Conrat *et al.* (1957) found that the activity of purified lima bean inhibitor was not affected by severe treatment with dry heat, acid, alkali, or by the action of pepsin and papain. Autoclaving and treatment in boiling water of the bean material resulted in a rapid loss of 60% of trypsin inhibitor activity. This suggests that boiling in water is the most effective way of removing the trypsin inhibitory activity of lima beans.

Although all the bean and cooking water samples showed hemagglutinin properties, the number of red blood cells which were agglutinated decreased with cooking time and an increase in the dilution of the lectin, which could be due to a decrease in the reactivity with the erythrocyte receptor cells. The decrease in both the trypsin inhibitors and phytohemagglutinin activity could explain, in part, the increase in the protein digestibility which was observed as the samples were cooked for different times.

It has been stated that cooking is a safe method for the elimination of toxicity in lima bean seeds since cooking destroys the enzyme linamarase at 72° C (Joachim & Pandittesekere, 1944), but not the glucoside. Montgomery (1965) has observed that it is yet to be proved that the glucoside itself is toxic to man, except when broken down by the enzyme. There was a threefold decrease in hydrocyanic acid content of the samples with increased cooking time. This is an indication that the process of bringing the beans to boil with cold water was effective in reducing the cyanic acid level. The cooking water contained significant amounts of hydrocyanic acid due to leaching out from the beans.

The high levels of phytic acid and polyphenol in this legume could affect the bioavailability of some nutrients present, especially the mineral elements. The decrease in the levels of these factors as a result of cooking, however, suggests that these factors may not significantly affect nutrient bioavailability with proper processing and preparation methods. It thus appears that the main problems with the lima bean is the presence of antinutritional factors which are not completely eliminated by processing (cooking). The traditional method of preparing lima bean meals, which involves discarding and changing of the cooking water many times, may reduce the level of these factors and hence increase the nutrient digestibility of this legume, while at the same time promoting its utilization in the diet.

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