Short Communication

Effect of Fermentation on Antinutritive Factors in Legume Seeds

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Abstract

The effectiveness of the solid state fermentation of legume seeds in hemagglutinating activity (HA) has been investigated. A previous study showed that the HA also higher in raw and unfermented stages and in this present investigation solid state fermentation destroyed HA fully after 24 hours fermentation and enhances nutritive value of this selected legumes.

Introduction

Legume seeds are an important part of the human diet in South East Asia, the Near East and parts of East Africa[1,2]. The popularity of legume based fermented foods is due to desirable changes including texture & organoleptic characteristics[3], enhancement of keeping quality[4], partial or complete elimination of antinutritional factors or natural toxins[5] increased nutritional value[6] and reduced indigestion. The hemagglutinins of legume species inhibit growth and even cause the death of experimental animals when injected at low concentration.[7] The inhibitors of digestive enzymes are common constituents of legumes and may reduce protein digestibility, depress growth and cause pancreatic hypertrophy[8] While phytohemagglutinins of soybean have limited nutritional effects on man[9], the hemagglutinins of Phaseolus species will inhibit growth and even cause death of experimental animals when feed as raw extracts[10].

Many of the antinutritional factors in legumes e.g. Phytic acid (table-1) can be eliminated or inactivated to a larger degree by appropriate heating and processing. Wet milling and processing techniques during concentration the protein and isolation are known to be effective in the detoxification of seed materials. Therefore, to investigation the effect of solid state fermentation by mold *Rhizopus oligosporus* on hemagglutinins in *Dolichos lablab* (Seem), *Cajanus cajan* (Arhar) and *Cicer* arietinum (chola).

Bangladesh Journal of Nutrition, Vol 12, Nos. 1 & 2, December 1999. Institute of Nutrition and Food Science, University of Dhaka, Bangladesh.

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Materials and Methods

Sample of Seem, Arhar and Chola seeds were collected from Dhaka, Bangladesh and stored at room temperature until all tests could be made. The seeds 50g portions of each group were soaked in water in the proportion of 1:3. Then the seeds were dehulled by hand and skins were separated and the dhal were washed twice with distilled water. Tempeh was prepared according to modifying method[11,13] and kept in the incubator at 37°C. After 24 hours fermentation the tempehs were kept in the refrigerator. The fermented tempeh and unfermented seeds were freeze dried and grinded into powder. The powder of each group were packed into polyethylene bags and kept in the refrigerator for further experiment.

Hemagglutinating activity (HA)

For hemagglutinin analysis, a 4% red cell solution in saline was prepared[12,13] using chicken red blood cell 10% saline extracts (0.90% Nacl) of each groups were prepared and several dilutions were then prepared and 1.0 ml of each group (Sample) was mixed with 4.0 ml blood cell solution, incubated in a 37°C water bath for one hour and observed for agglutination as compared to a control. The lowest dilution to show no signs of agglutination and the result was calculated by the formula.

$$HU/g=(Db \times S)/V$$

Where Db = dilution factors, S=ml original extracts/g and V=volume of extracts in test tube (1 ml).

Results and Discussion

The hemagglutinating activity (HA) found in Seem, Arhar & Chola were shown in table-2 was similar to values obtained by Liener (1955) but there are no comparable data to confirm the low HA obtained for fermented Seem, Arhar & Chola. Tannous and Ullah[13] found that hemagglutinins and trypsin inhibitors in legume seeds can be destroyed almost completely by autoclaving the seeds for 20 minutes. Reduction of HA by Mosleudding and Tannous was similar[14] to our results.

Table 1. Phytic acid (PA) content of selected legumes

Materials	Name	% of Phytic Acid (PA)
Dolichos lablab	Seem	1.56
Cajanus cajan	Arhar	1.47
Cicer arietnum	Chola	1.46

The decrease in the HA by heating & soaking of Seem, Arhar & Chola were 88, 100 & 100% respectively at unfermented period and 100% decreased by 24 hours fermentation. The results in general indicated that there were some effects on the HA factors of the legumes after fermentation by *Rhizopus oligosporus* (NRRL 2710).

Table 2. Effect of fermentation on humagglutinating activity (HA) of legumes*

Materials	Hemagglutinating activity (HA) HU/g		
	Raw sample	Unfermented Sample	Fermented Sample
Dolichos lablab	405	50	0
% decrease	–	88	100
Cajanus cajan	200	0	0
% decrease	–	100	100
Cicer arietnum	120	0	0
% decrease	–	100	100

*Each value represents average of replicate determination

Moreover, Marquardt et al[15] and Murata[16] reported much values of HA in faba bean, field pea, mung bean and cow pea flour was obtained in their study. Some of the differences in values among laboratories could be due to difficulty in avoiding agitation of the sedimented red blood cells during the photometic measurement of the density of the unsedimented layer of red cells in the Liener procedure. In addition, the soybean and faba bean hemagglutinins[9] have no apparent toxic effects on animals as compared to the growth inhibiting effects and death attributed to hemagglutinins from *Phaseolus vulgaris*[10].

Finding a simple and low cost processing method is not only reduces some of the toxic factors in legumes, but also enhance the nutritive value of the substances. This method is specially true for the people who depend on legumes as a protein source. Solid state fermentation is such a suitable process.

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