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Functional Properties of Flour, Protein Concentrate and Protein Isolate from Dolichos Lablab Bean [*Dolichos lablab purpureus* (L.) Sweet]

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Abstract. Protein solubility, fat and water absorption capacity, emulsion and foaming capacities and stabilities of Dolichos lablab flour, protein concentrate and protein isolate were determined. Protein solubility at different pH revealed that Dolichos lablab bean protein had minimum solubility at pH 4.0. The protein isolate showed better solubility than bean flour and protein concentrate below and above pH 4.0. Protein isolate had the highest oil absorption capacity, but lowest water absorption capacity among the samples studied. Emulsion capacity of the Dolichos lablab proteins was fair, with high stability. Although the protein isolate exhibited excellent foaming capacity, it had less foam stability in comparison to the bean flour and protein concentrates.

Keywords: Dolichos lablab, protein solubility, functional properties.

Introduction

Legumes are important source of proteins, carbohydrates, dietary fiber and minerals that are consumed worldwide. Although many species have been extensively studied and commercially promoted, several potential legumes growing in tropical and subtropical areas are still little known. These potential legumes might be of great importance in many developing countries, where there is a pressing need for good source of high energy and good protein quality. The Dolichos lablab bean (*Dolichos lablab purpureus* (L.) Sweet) is an indigenous legume of South and East Asia. The plant is drought tolerant and grows well with limited moisture. As in other legumes, the main nutritional advantage of the bean lies in the mature seed which contains (21.5 - 24.9%) protein, (0.8%) fat and

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(60.1%) carbohydrates [1, pp. 102-106, 275-278; 2, pp. 273-276, 290-294]. The amino acid composition of the Dolichos lablab bean is similar to that of soybean protein, with sulfur containing amino acid (methionine and cystine) being the limiting amino acid [1]. Most of the previous studies carried out on the Dolichos lablab bean were limited to proximate analysis [1; 2], mineral content [3], antinutritional factors [4; 5] protein quality [6] and animal feeding [7; 8]. However, there is no information on functional properties of the Dolichos lablab bean proteins; such information is needed for the potential use of the protein in food and for the comparative studies with other legume proteins. The purpose of this study is, therefore, to determine protein solubility, water and oil absorption and functional properties of Dolichos lablab flour, protein concentrate and protein isolate.

Materials and Methods

Materials

Dolichos lablab beans (*Dolichos lablab purpureus* (L.) Sweet) were obtained from the Agriculture Experiment station, College of Agriculture, King Saud University, Saudi Arabia. Beans were cleaned manually and ground in hammer mill to pass through a 30mesh screen size (bean flour).

Preparation of protein isolate and protein concentrate

For the preparation of protein concentrate the method of Mattil [9] was employed. The bean flour was mixed with (70%) ethanol (1:10 wt/vol). After stirring for 30 min, the mixture was centrifuged at 10,000 rpm for 20 min. The precipitate was air dried in the laboratory, then ground.

Protein isolate was prepared by extracting bean flour with water (1:10) and adjusting the pH of suspension to pH 10. After stirring for 30 min, the mixture was centrifuged at 10,000 rpm for 20 min. The supernatant was brought to its isoelectric point (IP) by adding 1N HCl. The suspension was centrifuged for 20 min. The precipitate protein was dissolved in water and the pH was adjusted to 7. The sample was then freeze-dried.

Protein determination

Crude protein (N \times 6.25) of bean flour, protein concentrate and protein isolate were determined in duplicate with micro – Kjeldahl method AACC [10]. Solubilized protein was determined by Lowry *et al.* [11].

Protein solubility

Protein solubility was performed according to the method described by Sathe [12]. Bean flour, protein concentrate and protein isolate were dissolved in 0.1 N NaOH (1:10 wt/vol) and stirred for 30 min. The extract was centrifuged at 12600 g for 20 min and supernatant was diluted with 9-volume water. The pH of aliquots (20 ml) was adjusted to the desired value with 1N HCl or 1N NaOH. The suspensions were centrifuged at 12600

g for 15 min. Analysis was performed in duplicate and the means were reported.

Water and oil absorption

For oil and water absorption determination, the method of Beuchat [13] was followed. Duplicate samples of 1 g were individually weighed into a 25 ml centrifuge tube, mixed with 10 ml water or oil for 30 sec. The samples were then allowed to stand at

room temperature for 30 min, centrifuged at $5000 \times g$ for 30 min, and the volume of supernatant noted in a 10 ml graduated measuring cylinder. Results are expressed as gram of corn oil or water absorbed per gram of samples.

Emulsion capacity and stability

The emulsion capacity and stability of the samples were determined by method of Yasumatsu et al [14]. Duplicates of each sample were homogenized with 50 ml of water for 30 sec. A polytron homogenizer was used at 10,000 r/min. Pure corn oil (25 ml) was added to each sample and homogenized for 90 sec. The emulsion obtained was divided evenly in four centrifuges, then centrifuged at $1100 \times g$ for 5 min. The emulsion activity was determined by dividing the volume of emulsified layer by the volume emulsion before centrifugation and expressing the result as a percentage of the added oil. Emulsion stability was determined by method of Naczk *et al.* [15], using the material prepared for measurement of emulsifying activity. The mixture was heated at 85 c for 15 min and cooled. After cooling to room temperature, the mixture was centrifuged as described above. Emulsion stability was expressed as the percentage emulsifying activity remaining after the heating.

Foaming capacity and foam stability

Foaming capacity and stability was determined as described by Lin *et al.* [16]. Duplicate samples of the bean flour, protein concentrate and protein isolated were used individually to prepare (3%) (w/v) solution in distilled water using low speed mixing for 30 sec and high speed whipping for 6 min in electric blender. The mixture was then transferred immediately into a 250 ml measuring cylinder and the foam volume was recorded. Foam capacity was expressed as percentage of the original volume of the liquid. Foam stability was expressed as the volume of the foam remaining after a 0.5, 10, 20, 40, 60 and 120 min.

Protein solubility

Results and Discussion

The results on protein solubility profile of Dolichos lablab bean flour, protein concentrate and protein isolate are shown in Fig 1. The samples exhibited lowest protein solubility at pH 4, indicating the acidic nature of the proteins. The protein's solubility increased below pH 3 and above pH 6. Alkaline pH was more effective in solubilizing proteins than acidic pH. Protein isolate showed lowest protein profile, minimum solubility (3.2%) among the three at pH 4.0. In contrast at pH below 3 and above 6, the protein isolate was markedly more soluble than bean flour and the protein concentrate.



Fig. 1. Solubility profile of the flour, protein concentrate and protein isolate of lablab proteins.

Similar protein solubility profile was observed on the flour, protein concentrate and protein isolate of different types of legumes. Narayana and Rao [17] and Sathe et al [18] independently reported that winged bean flour and protein concentrate had low protein solubility at pH 4.0. Sosulski and McCurdy [19] showed also that field pea and faba bean flour, protein fraction and protein isolate had minimum protein solubility at pH 4 to 5.0 and highly soluble below and above this pH. Such pattern were also reported on lupin flour and concentrate [20, 21] and tepary bean flour [22]. This result was similar to those reported on Dolichos lablab protein concentrate from Chinese variety [23]. Because of their high solubility, the bean flour and protein isolate could be used in food system where this advantage is needed.

Oil and water absorption

The water and oil absorption capacity of Dolichos lablab bean protein is shown in Table.1. The protein isolate showed lower water absorption capacity than the bean flour and protein isolate. Similar trends were observed for great northern bean. Al-ban and winged bean [17; 24; 25]. The value reported here for bean flour was in agreement with data reported for the great northern bean flour [24], and Lupin flour [21] but higher than those reported for faba bean, field bean and tepary bean flour [19: 22]. The oil absorption for the bean flour, protein concentrate and protein isolate were 1.54 ± 0.06 , 2.03 ± 0.03 and 3.38 ± 0.06 glg respectively. The oil absorption by bean flour in the present investigation was comparable to that of winged bean flour [17] and great northern bean flour [24]. The protein isolate showed higher oil absorption than bean flour and protein concentrate, suggesting that this fraction have more hydrophobic residues exposed which can bind to the hydrocarbon side chain of the oil. The protein isolate was found to be an excellent fat binder indicating that it may be used as meat binder or in bakery.

Sample	Protein content %	Water absorbed g/g	Oil absorbed g/g
Bean flour	22.7 ± 0.18	1.56 ± 0.09	1.54 ± 0.05
Protein concentrate	28.17 ± 0.9	1.70 ± 0.04	1.60 ± 0.02
Protein isolate	82 ± 0.18	1.24 ± 0.06	3.38 ± 0.06

Table 1. Protein concentration, water and oil absorption capacity of Dolichos lablab proteins

Emulsion properties

The emulsion capacity and emulsion stability of the Dolichos lablab bean flour, protein concentrates and protein isolates are presented in Table 2. The differences between samples in oil emulsification were not great. Similar characteristics were noticed on soybean, faba bean and field pea flour, protein concentrate and protein isolate [19]. The emulsifying capacity of bean flour and protein isolate were in agreement with those reported for winged bean flour, protein isolate and soy bean isolate [26] but was lower than those for great northern bean [24], field bean, faba bean flour and protein isolate [19]. As noted by Crenwelge et al [27] the comparison of emulsion of various materials is difficult and difference in the results may occur because of protein source, protein concentration and protedure used. Emulsion stability data of the Dolichos lablab proteins

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showed that the emulsions formed were fairly stable, since 97.8 to 99% of emulsifying activity were retained after heating at 80°C for 15 min. Similar results were reported for Dolichos lablab and soy bean concentrate [23]. The high emulsion stability of the Dolichos lablab proteins may be due to the globular nature of the major protein of the bean protein [21]. They may be of use in meat, ice cream and baking mixture, where the emulsion stability is desired.

Sample	Emulsion capacity g oil / g sample	Emulsion stability %	Foaming capacity %
Bean flour	12.63 ± 0.66	98	72 ± 1.0
Protein concentrate	12.10 ± 0.53	97.8	30.5 ± 0.5
Protein isolate	13.25 ± 0.50	99	110 ± 0.0

Table 2. Emulsion capacity and stability and foaming capacity of Dolichos lablab proteins

Foaming capacity and stability

Foam capacity and stability are important in food preparation such as bakery and dairy products. As shown in Table 3, the protein isolate indicated the highest foaming capacity compared to the bean flour and protein concentrate. Foaming capacity of both protein isolate and bean flour was higher than those reported for great northern bean [24] and field pea, faba bean and soy bean flour and isolate [19]. The Dolichos lablab protein concentrate showed similar foaming capacity to that of winged bean protein concentrate [18], but were lower than those reported for protein concentrate from great nothern bean [24]. Foaming stability was determined by measuring the decrease of foam as function of time. As shown in Table 3, foams from protein concentrate was more stable than those obtained from protein isolate and the flour. The protein isolate showed high foaming capacity but lower foam stability. The foam stability of the bean flour was higher than those reported for field pea bean and faba bean flour [19], but has lower foaming stability than winged bean flour and lupin flour [21; 26]. The foaming stability of the protein isolate was lower than that reported for soybean isolate and field pea isolate but higher than of faba bean isolate [19]. These differences may be due to the difference in protein nature or protein concentration used.

In conclusion, the present study reveals that Dolichos lablab proteins, had good functional properties. Protein isolate showed high protein solubility at acid and alkaline pH ranges and a low solubility at pH 4.0. The protein isolate also exhibited excellent oil absorption but poor water absorption, indicating its potential use in bakery or as meat binder. The Dolichos lablab proteins exhibited high emulsion stability but fair emulsion capacities. The foaming capacity was particularly high in protein isolate in comparison to the flour and protein isolate. The protein isolate showed promising functional properties; these require further studies on their potential use as food ingredient.

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Time (min)	Bean flour	Protein concentrate	Protein isolate
0.5	72.0 ± 1	30.5 ± 0.5	110 ± 0.0
5	62 ± 0	28.5 ± 0.5	83.5 ± 0.5
10	60 ± 0	28.0 ± 0.0	81.5 ± 1.5
20	58.5 ± 0.5	27.5 ± 0.5	72.5 ± 0.5
30	57.8 ± 0.5	27.0 ± 0.0	72.0 ± 0.5
60	56 ± 7.00	26.5 ± 0.5	65.5 ± 0.5
90	51.0 ± 1.0	25.0 ± 0.0	50.0 ± 0.00
120	49.0 ± 0.0	25 ± 0.0	10.0 ± 0.0

 Table 3. Foam stability of suspension of Dolichos lablab proteins at different times of standing

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الوظائف الحيوية للدقيق والمركز البروتيني والبروتينات المفصولة بواسطة الأس الهيدروجيني لبذرة الكشرنجيج

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(قدم للنشر في ٢٠/٢/١١؛ وقبل للنشر في ١٤٢٠/٦/١٧هـــ)

ملخص : تم في هذه الدراسة تقدير ذوبانية البروتين وسعة امتصاص الزيت والماء وسعة وثباتية الاستحلاب والرغوة للدقيق والبروتين المركز والبروتين المفصول لبذرة الكشر نجيج. بينت نتائج الذوبان أن بروتينات البذرة أقسسل ذوبانساً عنسد الرقسم الهيدروجيني ٤ (pH 4.0) كما بينت أن البروتين المفصول بواسطة الأس الهيدروجيني له قابلية ذوبان أفضل من دقيق البسذرة والبروتين المركز عند pH أقل وأكبر من ٤ . وأظهرت النتائج أيضاً أن البروتين المفصول بواسطة الأس مع معام معام والرغوة للدقيسي المستحلاب والرغوة للدقيمي ق امتصاص أعلى للزيت وسعة امتصاص أدن للماء مقارنة بالعينات الأخرى.

وأظهرت دراسة الخواص الاستحلابية أن بروتينات بذرة الكشرنجيج لها سعة استحلاب مقبولة ودرجة ثباتية عاليــــة. أوضحت النتائج أن البروتينات المفصولة بواسطة الأس الهيدروجيني لها سعة عالية على تكوين الرغوة ولكن درجة ثباقما كانت أدن. وتبين أن البروتين المفصول بواسطة الأس الهيدروجيني له سعة عالية على تكوين الرغوة ولكن درجة ثباته كـــانت أقـل بالمقارنة مع الدقيق والمركز البروتيني.