EFFECT EXTRACTION METHODS OF RICE BRAN PROTEIN ON THEIR FUNCTIONAL PROPERTIES
Abd El-Hady, Sahar R. and Badiaa A. Bessar
Food Tech. Dept., Fac. of Agric., Kafr El-Sheikh Univ., Egypt

ABSTRACT

This investigation was carried out to study the functional properties and amino acids compositions of rice bran protein isolate (RBPI) and rice bran protein concentrate (RBPC). The results indicate that (RBPI) had better water binding capacity, protein content and yield as compared to (RBPC), while oil binding capacity lower in (RBPI) than (RBPC). Nitrogen solubilities of (RBPI) were 52.3, 8.4, 63.5, 79.1, 83.7 and 81.5% at pH 2, 4, 6, 8, 10 and 12, respectively. RBPI had better foaming and emulsifying properties than (RBPC). While, RBPI had similar foaming properties in comparison to egg white, but emulsifying properties of RBPI were lower than those of bovine serum albumin. The results of amino acid analysis showed that the relative content of cystine and methionine in RBPC were lower than that of RBPI and RBP, while leucine and phenylalanine were higher in RBPC. The computed protein efficiency ratio (C-PER) and biological value (B V) of RBPI were higher than those of RBPC and value of all samples were lower than that of standard casein protein. Obtained results indicate that the RBPC and RBPI have high qualities of both nutritional and functional properties and could be competitive proteins ingredients in food products.

INTRODUCTION

Rice bran, by-product which obtained from rice milling process, contains a number of valuable components such as, proteins, carbohydrates and other phytochemicals that exhibit health benefits (Khuwijitjaru et al., 2007).

Rice bran is considered a good source of hypoallergenic proteins, and, rice bran protein may serve as a suitable ingredient for infant food formulations (Burks and Helm, 1994).

The most commonly used solvent to extract proteins from rice bran is alkali. High alkaline conditions could cause undesirable side reactions and potential toxicity such as lysinoalanine, thus losing the nutritive values of protein (Shih, 2003).

Carbohydrases (cellulase, pectinase, hemicellulase and viscoenzyme L) have been used to improve the extractability of plant proteins (Ansharullah and Chesterman 1997). The carbohydrases were considered to be macerating which disintegrate the cell wall tissue, and thus facilitate subsequent nitrogen extraction (Ansharullah and Chesterman 1997 and Shih and Daigle, 2000).

Understanding basic physicochemical and functional properties of a protein is very important for its application in food products (Gorinstein et al., 1996).
Determination of the functional properties of proteins is desirable for the utilization of any new proteinaceous material. The functionality of proteins depends in part upon the size and structure of the proteins and in part on their interactions with other food components like carbohydrates and fats, and is influenced by the processing conditions including the method of drying (Prakash and Narasinga Rao, 1986). The hypoallergenic property and the high nutritive quality could make rice protein concentrate or isolate a competitive protein ingredient in the food ingredients market (Chrastil, 1992).

The present work was carried out to prepare protein concentrate and protein isolate from defatted rice bran, which consider a by product of rice milling. Also some functional properties of rice bran protein concentrate, rice bran protein isolate, and amino acids content of these samples were studied.

**MATERIALS AND METHODS**

**Materials:**
Rice bran was purchased from the Rice Research and Training Center (RRTC), Sakha, Kafr El-sheikh Governorate, Egypt.

**Methods:**

1-**Defatting of rice brane:**
Defatting was prepared by mixing bran with hexane (1 to 20) at ambient temperature for 20 hours. Defatted samples were stored at -20°C until use. Protein content of the defatted bran was 14.2% (Kjeldahl method, conversion factor of 5.95).

2-**Preparation of protein concentrate (RBPC):**
Preparation of protein concentrate from defatted rice bran was done by using the procedure of Kumagai et al. (2006). Defatted bran was suspended in distilled water (1: 10). The pH of the slurry was set at 9.0 using NaOH solution (4 M), continuously stirred for 1 h and centrifuged (12600 xg, 15 min.). The supernatant protein solution was adjusted to pH 4.5 using HCl acid (4 M), stirred for 30 min. and left undisturbed for cold precipitation overnight (4°C). The supernatant was carefully siphoned off and precipitated protein was washed 3-4 times with distilled water. The protein slurry was neutralized to pH 7.0 and lyophilized. The resultant product was termed as rice bran protein concentrate (RBPC).

3-**Preparation of protein isolate:**
Preliminary trials were conducted to optimize conditions to extract rice bran protein with maximum protein content and yield in the presence of a combination of phytase and xylanase using the method described by Shih (2003). The final procedure for preparing of rice bran protein is given in Figure (1).
Defatted rice bran (10 grams) + deionized water (75 ml)

Adjust pH (5.0)

Add phytase (4,000 PU) and xylanase (2,400 GXU)

Incubate at 55°C for 2 hr

Inactivate enzymes (adjusting pH 10.0)

Centrifuge (10,000 rpm, 30 mn)

Supernatant

Adjust pH (4.0)

Centrifuge (8,000 rpm, 10 min)

Neutralize reside (pH 7.0)

Freeze

Freeze dry and store at 5°C

Figure 1: Procedure for the preparation of protein isolate from defatted rice bran.

Cake preparation:
The cake was processed at home according to Hanneman (1984). The ingredients used to make various cake blends are in table (1).

Table (1) : The blends of cake processed using RBPI and RBPC as substitution for eggs.

<table>
<thead>
<tr>
<th>Ingredients (gm)</th>
<th>Control</th>
<th>RBPI a</th>
<th>RBPI b</th>
<th>RBPI c</th>
<th>RBPC A</th>
<th>RBPC b</th>
<th>RBPC c</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat flour (72%)</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>RBPI</td>
<td>-</td>
<td>10</td>
<td>20</td>
<td>30</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>RBPC</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>10</td>
<td>20</td>
<td>30</td>
</tr>
<tr>
<td>Margarine</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
</tr>
<tr>
<td>Sugar</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
<td>66.5</td>
</tr>
<tr>
<td>Eggs</td>
<td>100</td>
<td>90</td>
<td>80</td>
<td>70</td>
<td>90</td>
<td>80</td>
<td>70</td>
</tr>
<tr>
<td>Milk</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Baking power</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
</tr>
</tbody>
</table>

RBPI : rice bran protein isolate  
RBPC : rice bran protein concentrate

Analytical methods:

1-Protein content and yield determination:

The protein contents of RBPC and RBPI were determined by the kjeldahl method (AOAC, 1990). The value of 5.95 was used as a protein conversion factor. Protein yields were calculated as yield (%)

\[
\text{Weight of RBPI(g) x protein content(\%) of RBPI} = \frac{\text{weight of DRB (g) x protein content(\%) of DRB}}{100}
\]

2-Water and fat absorption:

Water and fat absorption of protein were determined by Prakash and Ramanatham (1995). Sample (0.5 g) was taken and mixed with 3 ml of distilled water or refined palm oil. The slurry was centrifuged at 750 rpm for 15 min. The pellet was drained for 30 min. and the gain in weight per unit weight was reported as water or oil absorption capacity (g/g), respectively.

3-Bulk density:

A known weight of the protein concentrate was added to graduated measuring cylinder. The cylinder was gently tapped and volume occupied by the sample was determined. Bulk density was reported as weight per unit volume (g/ml) (Wang and Kinsella, 1976).

4-Solubility of proteins:

Protein solubility (PS) was determined by the method of Bera and Mukherjee (1989). Samples (20 mg each) were dispersed in 2 ml of deionized (DI) water. The pH was adjusted to 2.0, 4.0, 6.0, 8.0, 10.0 and 12.0 using either 1.0, 0.1 N HCl or NaOH. Samples were shaken at 250 rpm for 30 min. at room temperature (25°C) and then centrifuged at 4000 rpm for 30 min. Nitrogen contents of the supernatants (NS) were determined by the kjeldahl method, and percent protein solubility was calculated as follows:
$PS\% = \frac{\text{protein in the supernatant (mg)}}{\text{Total protein a 100 mg sample}} \times 100$

5-Foaming capacity and stability:
Foaming capacity (FC) of proteins was determined by measuring the volume of foams immediately after the introduction of air (90 cm$^3$/min) for 15 min. into 5 mL of 0.2% protein solution in 0.05 M phosphate buffer (pH 7.4) in a glass tube (2.4 x 30 cm).

Foaming stability (FS) was calculated from the following equation:
$$FS = V_0 \left( \frac{\Delta t}{\Delta V} \right)$$
where $\Delta V$ is the change in the volume of foam (V), occurring during the time interval, $\Delta t$ (30 min), and $V_0$ is the volume of foam at zero (0) time (Kato et al., 1989).

6-Emulsifying activity and emulsion stability.
Emulsifying activity (EA) and emulsion stability (ES) of protein were determined by the turbidimetric method of Pearce and Kinsella (1978). A 1% of protein solution was adjusted to pH 7.0. A 2 ml of palm oil was added into the protein solution and homogenized in a mechanical homogenizer at a setting of 6 for 1 min to produce the emulsion. The 50 µL portions of emulsion were pipetted at 0 and 10 min after homogenizing and mixed with 5 ml of 0.1% sodium dodylsulfate. Absorbance of emulsions was measured at 500 nm. The absorbance was measured immediately after emulsion formation and expressed as emulsifying activity of protein. Emulsion stability index was determined as follows:
$$ES = T_0 \left( \frac{\Delta t}{\Delta T} \right)$$
where $\Delta T$ is the change in turbidity, $T_0$ occurring during the time interval $\Delta t$.

7-Amino acid composition:
Amino acids of studied rice bran protein (RBP), rice bran protein isolate (RBPI), rice bran protein concentrate (RBPC) and casein were subjected to acid hydrolysis using 6 N HCl and few drop of mercaptoethanol. The hydrolyzate was recovered by removing the acid by evaporating in a rotary evaporator. Amino acids were estimated in the hydrolyzate using amino acid analyzer (Beckman amino acid analyzer, Model 119 CL) as described by Sadasivam and Manickam (1992) method. Tryptophan was determined colourimetrically after subjecting to alkaline hydrolysis as outlined by Miller (1967). Chemical score of indispensable amino acids was calculated using the equations of Pellet and Young (1980). Computation procedure for protein efficiency ratio (C-PER) of different studied samples was calculated as described by Alsmeyer et al. (1974) using following equation:
$$C\text{-PER} = -1.816 +0.435 \text{ (Methionine)} + 0.780 \text{ (leucine)} + 0.211 \text{ (histidine)} - 0.944 \text{ (tyrosine)}.$$
Biological values were calculated using the following equation reported by Farag et al. (1996):
$$\text{Biological value (B.V.)} = 49.9 + 10.53 \text{ C-PER},$$
where $C\text{-PER} = \text{computed protein efficiency ratio}$. 

Statistical analysis:
The data were statistically analyzed using the analysis of variance and the means were further tested using the least significant difference test as outlined by Steel and Torrie (1980).

RESULTS AND DISCUSSION

Protein content and yield of RBPI and RBPC and their functional properties:

Protein content and yield according to extraction methods were compared (Table 2). The highest protein content (88.8%) and yield (70.66%) were obtained with RBPI comparing to those of RBPC. These results demonstrate that the effectiveness of the combination of phytase and xylanase in releasing protein from rice bran. A combination of phytase and xylanase seemed to have advantages of releasing and increasing the extractability of proteins bound to cellular components, minerals and/or phytase. Therefore, released proteins can be solubilized, separated, and obtained in the form of an isolate. The aforementioned results coincide with those obtained by Grossman et al. (1980), Ansharullah et al. (1997), and Sogi et al. (2002).

Table (2): Protein content, protein yield, water absorption, oil absorption and bulk density of rice bran protein isolate (RBPI) and rice bran protein concentrate (RBPC).

<table>
<thead>
<tr>
<th>Samples</th>
<th>Protein content (%)</th>
<th>Protein yield (%)</th>
<th>Water absorption (g/g)</th>
<th>Oil absorption (g/g)</th>
<th>Bulk density (g/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBPI</td>
<td>88.8 b</td>
<td>70.66 b</td>
<td>2.4 b</td>
<td>3.2 b</td>
<td>0.21 a</td>
</tr>
<tr>
<td>RBPC</td>
<td>67.26 a</td>
<td>31.26 a</td>
<td>2.1 a</td>
<td>2.8 a</td>
<td>0.22 a</td>
</tr>
</tbody>
</table>

Each value was an average of three determinations. Values followed by the same letter in column are not significantly different at P < 0.05

In relation to water absorption capacity, data in Table (2) indicate that RBPI had low water binding capacity of 2.1% as compared with that of RBP(2.4). Water absorption might be related to nitrogen solubility index (NSI) since RBPC with least NSI had also low water solubility. Furthermore, RBPC and RBPI possess good water absorption capacity and can be used for products requiring high water retention. These results are in the line with those of Knorr and Betschart (1983), Prakash and Ramantham (1995). In this relation, Aletor et al. (2002) reported that high water absorption of protein helps to reduce moisture loss in packed bakery goods. Also, it is required to maintain freshness and moist mouth feel of baked foods. Water absorption capacity values ranging from 1.49 to 4.72 (g/g) are considered critical in viscous foods such as soups and gravies.

In respect of Oil absorption capacity, RBPI had the highest value (3.2%) and RBPC had the lowest one (2.8%) (Table 2). These results are in accordance with those obtained by Gandhi et al. (2000) and Aletore et al. (2002), who reported that RBPC had higher oil absorption capacity as compared to casein, various leaf protein concentrates and soy protein
isolates. High oil absorption is essential in the formulation of food systems like sausages, cake batters, mayonnaise and salad dressings.

Concerning with bulk density which consider as an important parameter that determines the packaging requirement of product. Bulk density signifies the behavior of a product in dry mixtures. Also it varies with the fineness of particles. High bulk density is disadvantageous for the formulation of weaning foods, where low bulk density is required (Onimawo and Egbekun, 1998). RBPC and RBPI had similar bulk density of 0.22 and 0.21 (g/ml), respectively (Table 2).

A similar observation of lowered bulk density was found by Venkatesh and Prakash (1993) for freeze-dried sunflower proteins. RBPC had very low bulk density as compared to casein (0.89 g/ml) and tomato seed protein concentrates (Sogi et al., 2002).

**Protein solubility (PS):**

The protein solubility profiles at different pH values(2.0, 4.0, 6.0, 8.0, 10.0 and 12.0) for RBPC and RBPI are shown in Table (3).

### Table (3): The solubility profiles of rice bran protein isolate (RBPI) and rice-bran protein concentrate (RBPC) at different pH values.

<table>
<thead>
<tr>
<th>pH</th>
<th>Samples</th>
<th>Protein solubility %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>RBPI</td>
</tr>
<tr>
<td>2</td>
<td></td>
<td>52.3 b</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td>8.4 a</td>
</tr>
<tr>
<td>6</td>
<td></td>
<td>63.5 c</td>
</tr>
<tr>
<td>8</td>
<td></td>
<td>79.1 d</td>
</tr>
<tr>
<td>10</td>
<td></td>
<td>83.7 f</td>
</tr>
<tr>
<td>12</td>
<td></td>
<td>81.5 e</td>
</tr>
</tbody>
</table>

Each value was an average of three determinations

Values followed by the same letter in column are not significantly different at P < 0.05

The solubility of RBPI had minimum values at pH 4.0. It was increased below pH 4.0 and above pH 6.0. Above pH 8.0, the solubility continued to increase but at a slower rate. Maximum (PS) of RBPI was observed at pH 10.0. However, the PS of RBPI at all used pH values was higher than that of corresponding values of RBPC extracted by alkali. This might be due to the change in ionic and/or other surface properties of rice bran proteins due to the removal of cell wall components after phytase and xylanase treatments. This solubility pattern is in agreement with those obtained by Gnanasambandam and Hettiarachchy (1995) and Hamada (2000). They mentioned high nitrogen solubility is required for protein concentrates or isolates to be used as functional ingredients in many foods including beverages, dressings, coffee whiteness, whipped toppings, confections etc.

**Foaming capacity and foam stability:**

Foaming activity is a measurement of ability of liquid to form foam after aeration. Protein can help forming the foam because of their surface active property. The foaming capacity (FC) and foam stability (FS) of egg white, RBPI and RBPC are presented in Table (4). Egg albumin protein is the most frequently used standard for foaming comparisons among proteins.
because of its good foaming properties (Symers, 1980). The egg white had relatively high values of FC and FS (20.1 ml and 121.1 min, respectively) than RBPI and RBPC. The FC and FS of RBPC were lower than those of RBPI.

Table (4): Foaming properties of, rice bran protein isolate (RBPI) and rice bran protein concentrate (RBPC) comparing to egg white.

<table>
<thead>
<tr>
<th>Samples</th>
<th>FC (ml)</th>
<th>FS (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Egg white</td>
<td>20.1 c</td>
<td>121.1 c</td>
</tr>
<tr>
<td>RBPI</td>
<td>17.6 b</td>
<td>105.0 b</td>
</tr>
<tr>
<td>RBPC</td>
<td>16.1a</td>
<td>101.20 a</td>
</tr>
</tbody>
</table>

Each value was an average of three determinations
Values followed by the same letter in column are not significantly different at P < 0.05
FC: Foaming capacity
FS: Foaming stability

It has been shown that molecular properties of proteins required for good FC and good FS are different (Cheftel et al., 1985). The formation of protein-based foams involves the diffusion of soluble proteins toward the air-water interface and rapid conformational change and rearrangement at the interface: the FS requires formation of a thick, cohesive, and viscoelastic film around each gas bubble (Damodran, 1994 and Tang et al., 2003). The good foaming capacity of RBPI, which is similar to that of egg white, might suggest fewer secondary and tertiary structure(s) in the RBPI molecules. RBPI released from rice bran hydrolyzed by phytase and xylanase might have more flexible random-coiled structure. These proteins might be more flexible due to a loss of complex secondary or tertiary structure which is due to the loss of phytate, mineral, and cellular components. Foaming capacity has been reported to be favored when proteins have more flexible random coiled structure (Halling, 1981 and Damodaran, 1990). However, the observed lower FS of RBPI might be due to the lack of formation of a thick, cohesive, and viscoelastic film around gas bubbles that prevented the foams from collapsing (Halling, 1981 and Damodaran, 1990).

Emulsifying activity (EA) and emulsion stability (ES):

The results of (EA) and (ES) of RBPI, RBPC and bovine serum albumin (BSA) estimated using corn oil are presented in Table (5). BSA is a good emulsifier. Therefore, it is the most frequently used as standard for comparing the effectiveness of emulsifying properties of protein. No significant differences of EA and ES were observed between RBPI and RBPC values (P < 0.05). Emulsifying properties of BSA were significantly higher than those of RBPI and RBPC (P < 0.05). Surface hydrophobicity is an important factor in determining the emulsifying properties (Petrucelli and Anon, 1995, Phillips et al., 1994, and WU, 2001).

Table (5): Emulsifying properties of rice bran protein isolate (RBPI) and rice bran protein concentrate (RBPC) compared to bovine serum albumin (BSA),
When compared with that of BSA, the lower emulsifying capacities of RBPI and RBPC might be due to its lower hydrophobicity value than that of BSA. The low hydrophobicity of RBPI would not facilitate the interaction between proteins and oils, resulting in a decrease of emulsifying properties (Halling, 1981 and Phillips et al., 1994).

**Amino acid content composition:**

The amino acids composition of rice bran protein (RBP), rice bran protein isolate (RBPI), rice bran protein concentrate (RBPC) and casein are shown in Table (6). Since casein based formula has been successfully used as the primary source of nutrition for infants (due to their good amino acid composition), the amino acid composition of casein was also used for comparison. RBP was found to have higher levels of isoleucine, methionine, cysteine, lysine and glycine in comparison to those amino acids of RBPI and RBPC.

**Table (6): Amino acids composition of RBP, RBPI, RBPC compared with casein (g/16 g N).**

<table>
<thead>
<tr>
<th>Amino acids</th>
<th>RBP</th>
<th>RBPI</th>
<th>RBPC</th>
<th>Casein</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Indispensable amino acids</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leucine</td>
<td>7.69</td>
<td>803</td>
<td>8.20</td>
<td>8.6</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>3.51</td>
<td>3.39</td>
<td>3.45</td>
<td>4.95</td>
</tr>
<tr>
<td>Valine</td>
<td>6.16</td>
<td>6.05</td>
<td>6.16</td>
<td>6.0</td>
</tr>
<tr>
<td>Methionine</td>
<td>2.18</td>
<td>1.91</td>
<td>1.65</td>
<td>2.5</td>
</tr>
<tr>
<td>Cystine</td>
<td>2.52</td>
<td>2.31</td>
<td>1.60</td>
<td>0.04</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>5.16</td>
<td>5.45</td>
<td>5.65</td>
<td>4.5</td>
</tr>
<tr>
<td>Tyrosine</td>
<td>3.62</td>
<td>3.72</td>
<td>3.77</td>
<td>-</td>
</tr>
<tr>
<td>Lysine</td>
<td>4.48</td>
<td>4.46</td>
<td>4.45</td>
<td>7.1</td>
</tr>
<tr>
<td>Threonine</td>
<td>4.53</td>
<td>4.73</td>
<td>4.55</td>
<td>3.7</td>
</tr>
<tr>
<td>Tryptophan</td>
<td>1.28</td>
<td>1.31</td>
<td>1.29</td>
<td>1.4</td>
</tr>
<tr>
<td><strong>Dispensable amino acids</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Histidine</td>
<td>3.16</td>
<td>3.41</td>
<td>3.35</td>
<td>3.0</td>
</tr>
<tr>
<td>Arginine</td>
<td>8.82</td>
<td>9.10</td>
<td>9.05</td>
<td>3.3</td>
</tr>
<tr>
<td>Serine</td>
<td>5.51</td>
<td>5.75</td>
<td>5.85</td>
<td>4.6</td>
</tr>
<tr>
<td>Alanine</td>
<td>6.79</td>
<td>6.80</td>
<td>6.77</td>
<td>2.7</td>
</tr>
<tr>
<td>Glutamic acid</td>
<td>14.30</td>
<td>14.21</td>
<td>14.32</td>
<td>18.0</td>
</tr>
<tr>
<td>Aspartic acid</td>
<td>9.32</td>
<td>9.35</td>
<td>9.45</td>
<td>6.3</td>
</tr>
<tr>
<td>Glycine</td>
<td>5.81</td>
<td>5.75</td>
<td>5.73</td>
<td>1.6</td>
</tr>
<tr>
<td>Proline</td>
<td>4.39</td>
<td>4.40</td>
<td>4.41</td>
<td>5.5</td>
</tr>
</tbody>
</table>

In comparison to casein, RBPI and RBPC had similar or higher levels in valine, cystine, phenylalanine, threonine, histidine, arginine, serine, alanine, aspartic and glycine. These results coincide with those of Chrastil (1992).
The relative contents of cystine and methionine in RBPC were lower than those of RBPI and RBP, while leucine and phenylalanine were higher in RBPC. During preparation of rice protein, a sulfur smell was detected during the steps from neutralization to drying, only with RBPC. It was reported that mild alkali treatment of a sulfur rich protein solution resulted in degradation of cystine and H₂S occurred when the solution was acidified (Nashef et al., 1977 and Florence 1980). Thus, the sulfur compounds appeared to have eliminated from rice protein in the alkaline solution.

**Amino acid scores (A.A.S.):**

The amino acid scores can be considered as an imperfect indicator of protein quality, but it still is the best one based on amino acid composition (Pellett and Young, 1980).

The amino acid scores of the indispensable amino acid of samples are given in Table (7). The results revealed that, the lowest amino acids score were found in RBPC. First limiting amino acid score was recorded for lysine. The results of FAO,(1993) support these results.

Table (7): *Chemical scoring of the indispensable amino acids of samples.*

<table>
<thead>
<tr>
<th>Indispensable amino acids</th>
<th>RBP</th>
<th>RBPI</th>
<th>RBPC</th>
<th>FAO/WHO pattern g/16 g N</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leucine</td>
<td>109.86</td>
<td>114.71</td>
<td>117.14</td>
<td>7.0</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>87.75</td>
<td>84.75</td>
<td>86.25</td>
<td>4.0</td>
</tr>
<tr>
<td>Lysine*</td>
<td>81.45</td>
<td>81.09</td>
<td>80.9</td>
<td>5.5</td>
</tr>
<tr>
<td>Methionine + cystine</td>
<td>134.29</td>
<td>120.57</td>
<td>92.86</td>
<td>3.5</td>
</tr>
<tr>
<td>Phenylalanine + tyrosine</td>
<td>146.33</td>
<td>152.83</td>
<td>157</td>
<td>6.0</td>
</tr>
<tr>
<td>Threonine</td>
<td>113.25</td>
<td>118.25</td>
<td>113.75</td>
<td>4.0</td>
</tr>
<tr>
<td>Valine</td>
<td>123.2</td>
<td>121</td>
<td>123.2</td>
<td>5.0</td>
</tr>
<tr>
<td>Tryptophan</td>
<td>128</td>
<td>131</td>
<td>129</td>
<td>1.0</td>
</tr>
</tbody>
</table>

*C-chemical scoring was calculated as a percentage of FAO/WHO (1973) recommended amino acids.

The computed protein efficiency ratio (C-PER):  

The computed protein efficiency ratio (C-PER) of different samples were lower than that of standard casein protein (PER = 2.50) as given in Table (8). The lower (C-PER) value was recorded in RBP, while the highest one was recorded in RBPI.

Table (8): Computed protein efficiency ratio (C-PER) and biological value (B.V) of samples.

<table>
<thead>
<tr>
<th>Samples</th>
<th>C-PER</th>
<th>B.V.</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBP</td>
<td>2.37</td>
<td>74.86</td>
</tr>
<tr>
<td>RBPI</td>
<td>2.49</td>
<td>76.08</td>
</tr>
<tr>
<td>RBPC</td>
<td>2.40</td>
<td>75.20</td>
</tr>
<tr>
<td>Casein*</td>
<td>2.50</td>
<td>76.23</td>
</tr>
</tbody>
</table>

*C-PER: Computed protein efficiency ratio  
B.V.: Biological value  
*C-PER and B.V. of casein according to (FAO/WHO pattern, 1973).
The biological values of protein of samples are also shown in Table (8). Biological values are very useful parameter for evaluating the effect of processing on food protein quality (Abd Alla, 1981). The results revealed that, RBPI had high biological value than other samples. These results may be related to the (C-PER) which was higher in RBPI than other samples.

Organoleptic evaluation of the cake Prepared using RBPI and RBPC:

The Organoleptic evaluation of the cake prepared using different percentages of RBPI and RBPC, as replacement for egg was performance and the obtained results were recorded in Table (9). The results indicate that the cake made without RBPI and RBPC(control) gave highest scores for all characteristics followed by that made using 10%of RBPI either from RBPC. The cake prepared using 30% RBPC as replacement for egg gave the lowest scores for the tested characteristics. It could be observed that the scores of the cake Prepared with RBPC were lower than those of RBPI. This may be related to variation of functional properties. The data in this table show also that the scores for all characteristics of cakes decreased with increasing the ratio of RBPI and RBPC replacement.

Table (9) : Organoleptic evaluation of the cake Prepared by using different replacement of egg by RBPI and RBPC

<table>
<thead>
<tr>
<th>Cake samples</th>
<th>Color</th>
<th>Taste</th>
<th>Aroma</th>
<th>Texture</th>
<th>Sponginess</th>
<th>Overall acceptability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>9.1 c</td>
<td>9.5 e</td>
<td>8.9 c</td>
<td>9.0 c</td>
<td>9.0 c</td>
<td>9.1 e</td>
</tr>
<tr>
<td>RBPI 10%</td>
<td>9.0 c</td>
<td>9.0 d</td>
<td>8.8 c</td>
<td>8.7 b</td>
<td>8.8 c</td>
<td>8.86 d</td>
</tr>
<tr>
<td>RBPI 20%</td>
<td>9.0 c</td>
<td>8.5 c</td>
<td>8.8 c</td>
<td>8.5 b</td>
<td>8.5 b</td>
<td>8.66 c</td>
</tr>
<tr>
<td>RBPI 30%</td>
<td>8.6 b</td>
<td>8.3 c</td>
<td>8.7 bc</td>
<td>8.1 a</td>
<td>8.3 ab</td>
<td>8.4 b</td>
</tr>
<tr>
<td>RBPC 10%</td>
<td>9.0 c</td>
<td>8.5 c</td>
<td>8.8 c</td>
<td>8.6 b</td>
<td>8.5 b</td>
<td>8.68 c</td>
</tr>
<tr>
<td>RBPC 20%</td>
<td>8.5 b</td>
<td>7.6 b</td>
<td>8.5 b</td>
<td>8.6 b</td>
<td>8.2 a</td>
<td>8.34 b</td>
</tr>
<tr>
<td>RBPC 30%</td>
<td>7.8 a</td>
<td>7.3 a</td>
<td>8.2 a</td>
<td>8.0 a</td>
<td>8.1 a</td>
<td>7.86 a</td>
</tr>
</tbody>
</table>

Values followed by the same letter in column are not significantly different at P < 0.05 Control with out any replacement of egg.

Conclusion

RBPI had better water binding capacity, protein content and yield as compared to RBPC. The foaming properties of RBPI were similar to those of egg white. But emulsifying properties of RBPI were lower than that of BSA. The obtained results indicate that defatted bran can be effectively used for preparing protein isolate which suitably used as an ingredient in foaming type products. The result of amino acid analysis show that the profile of amino acids of RBPC and RBPI were similar or higher levels of casein.

REFERENCES


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Tأثير طرق استخلاص بروتينات رجيع الأرز على الخواص الوظيفية لها

سحر رمضان عبدالهادى و بديعة عبدالرحمن بيصار
قسم تكنولوجيا الأغذية ـ كلية الزراعة ـ جامعة كفرالشيخ

أجريت هذه الدراسة بهدف دراسة تأثير طرق الاستخلاص على الخواص الوظيفية لبروتينات رجيع الأرز، حيث تم دراسة الخواص الوظيفية ومحفوظ الأحماض الأمينية للبروتين المنزول والبوتروتين المركز الناتج من رجيع الأرز وكان مختص النتائج كما يلي:
• البروتين المنزول من رجيع الأرز كان هو الأفضل من حيث سعة إنتاج المبيض ومحفوظ من البروتين والنتائج النهائي للبروتين مقارنة بالبوتروتين المركز الناتج من رجيع الأرز بينما كانت سعة البروتين المنزول لامتصاص الزيت أقل مقارنة بالبوتروتين المركز من رجيع الأرز.
• كانت نسبaNية البروتين المنزول من رجيع الأرز هي 67% - 76% - 67% - 83%. pH 6.7 - 7.8 - 8.4 - 7.2.
• وظَف أيضا أن البروتين المنزول من رجيع الأرز كان هو الأفضل في خصائص الرغوة والاستحلاب بالمقارنة بالبوتروتين المركز من رجيع الأرز. وكانت خصائص الرغوة للبروتين المنزول من رجيع الأرز متشابهة مع زلال البنيا. ولكن خصائص الاستحلاب كانت أقل في البروتين المنزول من رجيع الأرز بالمقارنة بالبوتروتين المركز.
• نتائج الأحماض الأمينية أشارت إلى أن نسبة الأحماض الأمينية السستين والميثونين في مركز بروتين الرجيع كانت أقل منها في كل من البروتين المنزول من رجيع الأرز وكذلك بروتين الرجيع بينما كانت نسبة الأحماض الأمينية الليوسين والفينالين الأثنا عب في الأقل في مركز بروتين الرجيع.

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تفوقت القيمة الكيميائية للأحماض الأمينية الأساسية في البروتين المعزول عن البروتين المركز، وظهرت النتائج أن هناك زيادة في كفاءة الاستفادة من البروتين وكذلك القيمة الحيوية للبروتين المعزول المستخلص من رجيع الأرز المنزوع الدهن عن البروتين المركز، ومما سبق نجد أن البروتينات المعزولة أو المركز الناتجة من رجيع الأرز عالية الجودة الغذائية والوظيفية، وذلك يمكن أن تعد هذه البروتينات منافسة لمضادات جيدة لمنتجات بعض الأغذية.