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Yeast (*Saccharomyces cerevisiae*) Protein Concentrate: Preparation, Chemical Composition, and Nutritional and Functional Properties

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The main objective of the present study was to compare the composition and functional and nutritional properties of whole yeast cells (WY) from an ethanol distillery with those of a phosphorylated protein concentrate (PPC) prepared from the same cells. Comparisons were also made of PPC with texturized soy protein (TSP) and soy protein isolate (SPI), both acquired in the local market. Yeast (*Saccharomyces cerevisiae*) is a rich source of protein, soluble fiber, and some minerals. Saturated fatty acids predominated over monounsaturated and polyunsaturated in both WY and PPC. The functional properties of PPC were similar to those of SPI and TSP. Both soy products and PPC replaced 20 or 40% chuck roll protein without affecting the emulsion properties of the meat products. Amino acid scoring was high for both WY and PPC; digestibility was higher (90%) for PPC and lower (68%) for WY. The protein nutritive value of PPC did not differ from that of casein and was significantly higher than that for WY.

KEYWORDS: Yeast; Saccharomyces cerevisiae; protein; functional properties; nutritive value

INTRODUCTION

Brazil is one of the major producers of ethanol from sugar cane, with a production of 12 billion liters in the fiscal year 1999/2000 (1). Experiments done in Brazilian distilleries have shown that 20-30% (w/w) of the yeast biomass can be withdrawn from the fermentation tanks, after each cycle of fermentation, without any drop in the alcohol yield in the following cycle (2, 3).

This operation yields an annual production of \sim 250000 metric tons of dried biomass (3), which is still underutilized in the country. A significant additional amount of yeast biomass is produced in breweries as a byproduct.

According to Halász and Lásztity (4), yeast is used in the fermentation industries for the production of alcohol and wine and in bakeries for the fermentation of bread dough, among other bakery products. Inactivated yeast and yeast derivatives have been used as nutritive complements and as food ingredients for the formulation of a variety of industrial food products (4–9).

The nutritive value and functional properties of whole yeast cells, mechanically ruptured cells, and protein concentrates of *Saccharomyces cerevisiae* originated as a byproduct in brewing industries have been reported (10-13).

In this work the main objective was to develop a pilot scale preparation method for a modified yeast protein concentrate, from distillery yeast, and to study its composition and nutritional properties related to whole yeast cells and also compare its functional properties to those of commercial texturized soy protein (TSP) and soy protein isolate (SPI).

MATERIALS AND METHODS

Material of Study. The yeast biomass (*S. cerevisiae*) was provided by an ethanol distillery near Campinas, in the form of a suspension of 20% cells (w/v). The suspension was diluted (1:1 v/v) with water and centrifuged using a plate centrifuge (Alfa Laval BRPX 20739 S60) to obtain a biomass slurry and a supernatant. The biomass slurry was spray-dried (Niro Atomizer CB3 104D) at 180 ± 5 °C at the entrance and 80 ± 5 °C at the exit of the chamber.

TSP Maxten R-100 and SPI Samproy MP-90 were commercial products provided by Ceval Alimentos Co., Paraná, Brazil.

Analytical grade reagents used were from Merck, Sigma, Pharmacia, and Pierce. Sodium trimetaphosphate (STMP) was of food grade, supplied by Solutia (Augusta, GA). All of the ingredientes used for preparing rat diets were of food grade purchased from local suppliers.

Phosphorylated Yeast Protein Concentrate (PPC). The isolation of a PPC involved mechanical rupturing of cell walls, centrifugation to remove cell debris, phosphorylation at alkaline pH with STMP, and precipitation of the phosphorylated protein at acidic pH (*14*).

Rupturing of the cell walls was performed in a 10% cell suspension (w/v) adjusted to pH 9.5, with 2 N NaOH solution forced by a peristaltic pump through a mill (Dyno Mill DDL-PILOT mill) at 2400 rpm, at a flow rate of 4.8 L h⁻¹. For the operation 70% of the mill chamber was filled with 0.6–0.9 mm glass spheres. During operation the cell

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suspension was kept at 25 $^{\circ}$ C by circulating a mixture of water/ethylene glycol through a double-jacket chamber.

The ruptured cell suspension was centrifuged (5300g, 15 min), and the soluble fraction was adjusted to pH 11.0 with 2 N NaOH solution and treated with food grade STMP at a concentration of 4% (w/w/ total solids) at 35 °C for 3 h. The phosphorylated protein was acidified (3 N HCl) to pH 3.2 and centrifuged. The precipitate was washed twice with acidified distilled water, diluted in distilled water, adjusted to pH 7.0 with NaOH, and then freeze-dried.

Proximate Percent Composition. Water content, ashes, and crude protein (N \times 5.8) were determined according to the AOAC (*15*) procedures. Total lipids were determined according to the Bligh and Dyer method (*16*). Soluble and insoluble fibers were quantified by treating the sample first with proteolytic enzymes (pepsin/pancreatin) to digest sample protein, followed by filtration to retain the insoluble fiber and precipitation of the soluble fiber from the filtrate with ethanol. Both the precipitate (soluble fiber) and the material retained in the filter (insoluble fiber) were quantified after drying in an oven, to constant weight, according to AOAC procedure 985.29 (*15*). Mineral composition was determined in a plasma spectrometer (ICP 2000 BAIRD simultaneous version) with an argon flame detector. Quantification was done by using a pure standard mixture of known concentration. Preparation of the samples for analysis was done according to the method of Angelucci and Mantovani (*17*) and IMO Industries Inc. (*18*).

Nucleic Acid (RNA). RNA was extracted with a 0.5 M solution of perchloric acid at 37 °C for 2 h. RNA content was determined colorimetrically with orcinol reagent (19).

Amino Acid Determination. Amino acids were determined in an acidic hydrolysate (6 N HCl, 110 °C, 22 h) essentially according to the procedure of Spackman et al. (20) using a Dionex D-300 analyzer with cation exchange column and postcolumn ninhydrin reaction, using a Pierce standard amino acids mixture for quantification. Tryptophan was quantified in a Pronase (40 °C, 24 h) hydrolysate by the reaction with 4-dimethylaminobenzaldehyde (DAB), according to the method of Spies (21).

Fatty Acid Determination. Fatty acid composition was determined by gas-liquid chromatography after acidic interesterification with methanol, according to the procedure of Firestone (22).

Viscosity. Viscosity was determined in an RVA apparatus (Rapid Visco Analyzer, Newport Scientific). The tests were run at 160 rpm at two sample concentrations (6 and 15%, w/v). Measurements were made at 25, 33, 60, 70, and 80 °C during 2 min. Temperature increasing time in each step was 1 min. After reaching 80 °C, the products were cooled to 30 °C. For each temperature the experiment was done twice.

Solubility. Protein solubility of the concentrates was determined according to the method of Morr et al. (23). Samples were suspended (1% w/v) in distilled water, agitated at 25 °C for 1 h, and then centrifuged (10000g, 5 °C, 15 min). The supernatant protein was determined by using the micro-Kjeldhal method (14), and percent solubility was calculated on the basis of total nitrogen in the original sample.

Water-Holding Capacity (WHC). WHC was determined according to the method of Regenstein (24). Aqueous sample dispersions were adjusted to the desired pH with 0.1 N HCl or NaOH solution, and the final volume was adjusted to give 1% protein concentration. After agitation at 25 °C for 1 h, 30 mL aliquots were centrifuged (30000*g*, 5 °C, 15 min). The supernatant protein was determined (*14*), and the precipitated protein was calculated by difference from the total protein in the original sample. WHC was estimated as grams of water retained per 100 g of precipitated protein.

Emulsifying Capacity (EC). These analyses were performed according to the procedure of De Kanterewicz et al. (25) using a homogeneizer (Ultra-Turrax T-25, Junkel & Junkel). Protein dispersion (1% w/v) and soy oil were combined at various volume ratios, maintaining 50 mL of total volume to find the proximity of the emulsion breaking point. To the nearest breaking point ratio, 1 mL volumes were added, consecutively, until the true emulsion breaking point was reached. The emulsion was kept under stirring (9500 revolutions min⁻¹) in an ice bath during the whole operation to avoid sample heating.

Emulsifying capacity was also performed in mixtures of meat (chuck roll) with yeast or soy protein. Minced chuck roll (25 g) was

Table 1. Percent Composition (Dry Matter Basis) of Whole Yeast Cells (WY) and Phosphorylated Yeast Protein Concentrate (PPC)^a

component (g/100 g of dm)	WY	PPC
protein (N \times 5.8)	39.6	62.6
total lipids	0.5	8.5
ashes	4.6	13.2
total fiber	31.4	6.0
insoluble fiber	1.1	nd
soluble fiber	30.3	nd
RNA	9.0	10.4
others	14.9	0.0

^a Results are the average of two determinations; nd, not determined.

homogeneized in 100 g of 1 M NaCl solution for 2 min in a Sorvall-Omni mixer, and the emulsifying capacity was determined as above (25).

Emulsion Stability (EE). EE was determined according to the Acton and Safle procedure (26). Moisture content was determined in 10 mL aliquots pipeted from the bottom of the emulsion container immediately after preparation and after 24 h of standing at 37 ± 2 °C. Emulsion stability was determined with the expression EE (%) = $100 - U_{24h}/$ $100 - U_{bp} \times 100$, where U_{24h} is the moisture content of the aliquot after 24 h of standing and U_{bp} the moisture content immediately after emulsion preparation.

Essential Amino Acid Score (EAE). The EAE represents the smallest ratio of the most limiting essential amino acid in the protein under study with regard to the same amino acid of a reference standard. In this work the FAO/WHO (27) reference was used.

Protein Digestibility Corrected Amino Acid Scoring (PDCAAS). This index is calculated by multiplying the EAE by the true protein digestibility (TD), normally expressed in percentage (28).

Rat Assay. The basic experimental diet was that recommended by the American Institute of Nutrition, AIN-93 (29), modified with respect to protein type and content. Diets with 10% protein provided by casein (CAS), whole yeast cells (WY), phosphorylated yeast protein concentrate (PPC), and a protein-free diet (PFD) were prepared. Thirty-two male rats of the Wistar strain, specific pathogen-free (SPF), 21-25days old, were used. The rats were distributed into four groups following a randomized block design, located in individual cages receiving water and diet ad libitum. The temperature of the experimental room was automatically controlled at 22 ± 2 °C with alternating dark/light periods of 12 h. Diet consumption and body weight gain were recorded for the calculation of true digestibility (28), PDCAAS, net protein ratio (NPR), and protein efficiency ratio (PER).

RESULTS AND DISCUSSION

The percent composition of WY and of PPC is shown in **Table 1**. The protein contents of WY and of PPC of yeast from an ethanol distillery were lower than the one determined in the brewing industry yeast (*13*).

This finding can be accounted for the fact that although distillery yeast is recycled several times (three to four times), the brewing yeast is used normally only once. In addition, brewery fermenting mixtures are normally purer than the ones from distilleries. Therefore, distillery yeast is more exhausted of its cell components, including proteins, and cell walls are thicker and more resistant to proteolytic enzymes. Total lipids are normally low in yeast cells and tend to form complexes and precipitate with proteins and RNA. Ashes in the PPC were over two >2 times the concentration in WY. This could be explained by the addition of STMP in the phosphorylation reaction. Protein and fiber are the two main components of yeast biomass, accounting for about two-thirds of the cell components. This has been found also by other workers (8, 11, 13). It is important to point out the high predominance of soluble fiber in the WY.

Table 2. Mineral Elements in the Whole Yeast Cells (WY) and Phosphorylated Yeast Protein Concentrate (PPC)^a

	results (n	results (mg/100 g)		
element	WY	PPC	RDA ^b (mg/day)	
Na	6.3 ± 0.3	308.0 ± 40	2000	
Ca	147.7 ± 1.4	13.3 ± 0.3	1200	
Mg	143.5 ± 0.3	8.2 ± 0.4	350	
Ρ	1516.0 ± 20.0	5248.0 ± 101	1200	
K	2035.0 ± 5.0	68.0 ± 3	2500	
Fe	38.0 ± 0.7	12.3 ± 0.3	10–15	
Mn	1.4 ± 0.01	0.2 ± 0.0	2–5	
Zn	12.7 ± 0.1	2.0 ± 0.1	10–15	
Cu	4.9 ± 0.03	3.6 ± 0.1	2–3	

^a Mean of three analytical determinations ± standard deviation. ^b Recommended daily allowance, according to NAS (*32*), for young adults.

Table 3. Essential Amino Acid Profile and Score (EAE), Protein True Digestibility (TD), and Corrected Amino Acid Scoring (PDCAAS) of Whole Yeast Cells (WY) and Phosphorylated Yeast Protein Concentrate (PPC)

amino acid (g/100 g of protein)	WY	PPC	FAO/WHO (ref) ^a
threonine	4.7	5.0	3.4
methionine + half-cystine	2.4	2.3 ^b	2.5
valine	4.8	6.0	3.5
isoleucine	4.2	5.1	2.8
leucine	6.0 ^b	8.5	6.6
tyrosine + phenylalanine	6.5	9.2	6.3
lysine	8.0	9.2	5.8
histidine	4.2	2.4	1.9
tryptophan	1.2	1.8	1.1
EAE (% ref)	91.7	93.2	
TD ^c (%)	68.0 ± 5.0 b	90.0 ± 1.1 a	
PDCAÁS (%)	62.0	84.0	

^a FAO/WHO (27), reference for children 2–5 years old. ^b Most limiting amino acid. ^c Casein control: TD = 93.5 ± 0.8 a; PDCAAS, 94%.

As shown in **Table 1** the content of RNA (10.4%) is quite high in the PPC. Phosphorylation of yeast protein has been used, among other reasons, with the purpose of decreasing RNA content in the concentrate (11, 30, 31). However, in the present work it was observed that RNA content depended on the pH of the protein precipitation. It decreased in the PPC as the pH increased above the pI, with a concurrent decrease in PPC yield. On the other hand, at lower pH, in this work 3.2, there was a substantial gain in yield and a concurrent increase in RNA in the precipitate. At pH 3.2 the yield of PPC, on a dry basis, was 20 kg of PPC/100 kg of WY.

Regarding mineral elements (**Table 2**) the PPC showed an excess of sodium and phosphorus, compared with WY, a consequence of the phosphorylation with STMP. All of the other elements were in accordance with published data (4). It is important to note the high content of iron, manganese, zinc, and copper in the WY, which is also an excellent source of selenium (4).

The essential amino acid profiles for the WY and PPC are shown in **Table 3**. The most limiting amino acid in WY was leucine, and those in PPC were the sulfur amino acids. Limitation was small because the EAE was 91.7% for WY and 93.7% for PPC. Except for histidine, all essential amino acids were slightly higher in PPC compared with WY. This might suggest higher degradation of amino acids in WY, during acid hydrolysis or, more likely, a selective precipitation of proteins in the PPC. It is worth noting the high content of lysine and

Table 4. Fatty Acid Composition of Whole Yeast Cells (WY) and Phosphorylated Yeast Protein Concentrate $({\rm PPC})^a$

fatty acid (% of total)	WY	PPC
caprylic (C8.0)	2.01	nd
capric (C10:0)	0.73	0.60
hundecanoic (C11:0)	0.33	0.20
lauric (C12:0)	2.03	2.00
myristic (C14:0)	0.97	0.60
pentadecanoic (C15:0)	0.33	0.20
palmitic (C16:0)	24.60	30.80
not identified	nd	0.60
palmitoleic (C16:1 ω7)	5.77	14.40
margaric (C17:0)	nd	0.30
cis-10-heptadecenoic	nd	0.20
stearic (C18:0)	9.03	14.80
elaidic (C18:1 ω9T)	1.57	0.20
oleic (C18:1 ω9)	22.47	14.60
trans-linoleic (C18:2 ω6T)	nd	0.20
linoleic (C18:2 \omega6)	29.90	12.20
α -linolenic (C18:3 ω 3 α)	0.53	0.60
arachidic (C20:0)	5.03	0.80
not identified	nd	0.20
behenic (C22:0)	nd	0.70
arachidonic (C20:4 ω 6)	nd	0.40
not identified	nd	0.30
eicosapentaenoic (C20:5 ω 3)	nd	0.70
docosahexaenoic (C20:5 ω 3)	nd	4.90
saturated	42.71	52.00
monounsaturated	28.31	29.20
polyunsaturated	28.90	19.20
1 - 7		

^{*a*}T, trans; ω , omega; nd, not detected.

tryptophan in the yeast proteins, which makes them good candidates to enrich cereal proteins.

Table 3 also shows a significant difference in TD between WY and PPC. The lower TD for WY, compared with PPC, can be explained by two major factors, the rigid cell wall and the high content of fiber in WY (4, 11, 13). The PPC true protein digestibility (TD) did not differ from that of casein (see footnote). Both higher TD and higher EAE resulted in much higher PDCAAS for PPC, compared with WY (**Table 3**).

Table 4 illustrates the fatty acid composition of WY and PPC. With some variations, the predominant fatty acids are the same in both WY and PPC, namely, palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1 ω 9), and linoleic acid (C18:2 ω 6). Oleic, linoleic, and arachidic acids appeared in higher concentration in WY, whereas palmitic, palmitoleic, stearic, and docosahexaenoic acid (DHA) showed higher concentrations in the PPC. Nine fatty acids were not detected (nd) in the WY but were found in low concentrations in PPC. This might be explained by the solubility of these fatty acids in the protein extraction and/or phosphorylating media or by complexation with the precipitated protein. It is worth noting the reasonable concentration (5%) of DHA in the PPC, which was not detected in WY. Three small peaks (not identified) appeared in the PPC that were not detected in WY. Overall, there was a predominance of saturated fatty acids in both WY and PPC. In WY the distribution was more adequate in terms of saturated, monounsaturated, and polyunsaturated fatty acids, whereas in the PPC the saturated fatty acids were found to be in the highest and the polyunsaturated in the lowest concentration.

According to Halász and Lásztity (4) unsaturated fatty acids predominate in yeast during the exponential phase of growth, but there is a shift to saturated fatty acids with aging of the cells or when some kind of stress is imposed to the culture. In the case of alcoholic fermentation considerable stress is imposed to the cells, both by the high concentration of alcohol and by

Table 5. Viscosity (Centipoise \times 10⁻²) of Phosphorylated Protein Concentrate (PPC), Texturized Soy Protein (TSP), and Soy Protein Isolate (SPI) at Two Protein Concentrations and Various Temperatures^a

concen-			he	ating pha	ise		cooling phase ^b
tration (% w/v)	product	25°C	33°C	60°C	70°C	80°C	30°C
6	PPC	44.0	24.0	19.0	17.5	17.0	18.0
	TSP	51.0	31.0	27.0	24.5	25.0	29.0
	SPI	134.5	106.0	76.0	51.0	24.0	13.5
15	PPC	194.0	139.5	72.0	60.0	52.5	70.0
	TSP	202.0	179.5	201.5	316.5	483.0	1007.0

^a Results represent the mean of two analytical determinations. ^b Cooling phase, viscosity at 30 °C after heating to 80 °C.

Table 6. Solubility (Percent w/v) of Phosphorylated Yeast Protein Concentrate (PPC), Soy Protein Isolate (SPI), and Texturized Soy Protein (TSP), at Various pH Values^a

	product	
PPC	SPI	TSP
3.1 ± 1.0 bC 5.9 ± 0.5 aB	14.9 ± 2.2 aB 3.2 ± 0.4 aC	4.1 ± 1.8 bB 4.1 ± 0.7 aB
8.1 ± 0.5 aB	2.7 ± 0.4 bC	5.3 ± 1.4 aB 8.2 ± 1.9 cA
	3.1 ± 1.0 bC 5.9 ± 0.5 aB 8.1 ± 0.5 aB	PPC SPI 3.1 ± 1.0 bC 14.9 ± 2.2 aB 5.9 ± 0.5 aB 3.2 ± 0.4 aC

^a Results represent the mean of three analytical determinations \pm standard deviations. Different lower case letters (rows) indicate statistical difference (p < 0.005). Different capital letters (columns) indicate statistical difference (p < 0.05).

the successive use of the same cells in repeated cycles of fermentation.

In **Table 5** the viscosities of PPC, TSP, and SPI at 6 and 15% (w/v) concentrations were compared at different temperatures.

At lower concentration (6%) the viscosity of all three products studied decreased with increasing temperature. SPI showed higher viscosity at all temperatures tested up to 70 °C, but presented the lowest viscosity at 30 °C, in the cooling phase, probably due to protein denaturation and coagulation at 80 °C.

At 15% protein concentration only PPC and TSP were tested, and both showed considerably higher viscosity at all temperatures with the difference that while PPC viscosity decreased as the temperature increased, TSP showed a decrease in viscosity at 33 °C in the heating phase. At 60 °C the viscosity of TSP was identical to that of 25 °C, but at 70 and 80 °C a substantial increase in viscosity was observed. The viscosity of TSP at 30 °C, the cooling phase, was 5.6 times higher than at 33 °C in the heating phase and was 5 times higher for PPC, comparing 30 and 33 °C in the cooling and heating phases, respectively. Although at 6% protein concentration, viscosities at 30 °C, cooling phase, were similar to those at 80 °C, at 15% protein concentration, viscosities at 30 °C, cooling phase, were 1.3-2.0 times higher than those at 80 °C in the heating phase. This phenomenon might suggest protein denaturation and/or hydration followed by gelation at higher temperatures (70 and 80 °C).

The solubility in aqueous media of PPC, SPI, and TSP, after adjustment to various pH values and agitation for 1 h at 25 °C, is shown in **Table 6**. At pH 3.0 solubility was higher for SPI (14.9%). At pH 4.0 solubility was very low, ranging from 3.2% for SPI to 5.9% for PPC. At pH 5.0 highest solubility was 8.1% for PPC and lowest (2.7%) for SPI. Solubility increased **Table 7.** Water-Holding Capacity (Grams of Water per Gram of Protein)^{*a*} at Different pH Values of 1% Dispersion of Phosphorylated Yeast Protein Concentrate (PPC), Texturized Soy Protein (TSP), and Isolated Soy Protein (SPI) at Three pH Values^{*a*}

pН	PPC	TSP	SPI
5.0 6.0 7.0	$\begin{array}{l} 4.56 \pm 0.58 \text{ bB} \\ 8.42 \pm 1.11 \text{ aA} \\ 9.16 \pm 0.67 \text{ bA} \end{array}$	$\begin{array}{c} 7.66 \pm 0.68 \text{ aBC} \\ 9.15 \pm 1.17 \text{ aAC} \\ 10.22 \pm 1.62 \text{ bA} \end{array}$	$\begin{array}{c} 5.60 \pm 0.39 \text{ bC} \\ 9.75 \pm 0.99 \text{ aB} \\ 16.75 \pm 2.87 \text{ aA} \end{array}$

^a Mean of six analytical determinations \pm standard deviation. Different lower case letters (rows) indicate statistically different results ($p \le 0.05$). Different capital letters (columns) indicate statistically different results ($p \le 0.05$).

 Table 8. Emulsifying Capacity (Milliliters of Oil per Gram of Protein) of

 Chuck Roll with 20 and 40% Meat Protein Replacement by

 Phosphorylated Yeast Protein Concentrate (PPC), Texturized Soy

 Protein (TSP), or Soy Protein Isolate^a

	replacement	replacement (% of protein)		
product	20	40		
chuck roll + PPC chuck roll + TSP chuck roll + SPI	$408.93 \pm 28.44 \text{ bB} \\ 440.95 \pm 20.61 \text{ aA} \\ 431.20 \pm 15.78 \text{ bA} \\$	463.09 ± 21.68 aA 397.98 \pm 10.30 bA 447.75 \pm 10.22 aA		

^a Results are the mean \pm standard deviation of three analytical determinations. Different lower case letters (columns) indicate statistically different results (p < 0.05) among treatments. Different capital letters (rows) indicate statistically different results (p < 0.05) among % replacement.

significantly at pH 7.0 for all three proteins, ranging from 8.2% for TSP to 20.7% for SPI. Both the pH and the nature of the protein preparation influenced solubility significantly.

Table 7 presents the WHC for PPC, TSP, and SPI at pH 5.0, 6.0, and 7.0. For all three protein sources WHC increased with increase in pH. Higher WHC (grams of water per gram of protein) was at pH 7.0, being 9.2, 10.2, and 16.7 for PPC, TSP, and SPI, respectively, with statistical difference for SPI. At pH 5.0 the WHC was lowest due to proximity of the pI of these proteins, when the material becomes more compact, holding lower net charges, both factors contributing to diminish water retention. At pH above pI the protein becomes more flexible, with an increase in net negative charges, favoring hydration and water retention.

The values found for WHC for PPC in the present work at pH 5.0 and 7.0 were smaller than the ones found by Pacheco and Sgarbieri (*12*) of 5.1 and 18.6 g of water/g of protein, respectively, for PPC from brewing industry yeast; however, they were higher than the values obtained for nonphosphorylated brewing yeast protein concentrate extracted with sodium perchlorate of 4.3 and 6.1 at pH 5.0 and 6.0, respectively (*12*).

The emulsifying capacity (EC) and emulsion stability of PPC produced in this research were compared with a commercial SPI. Emulsifying capacity (milliliters of oil per gram of protein) was 424.6 for PPC and 447.6 for SPI with no statistical difference between them (p > 0.05). The emulsion stability (percent) was 83.5 for PPC and 73.2 for SPI, also with no statistical difference between the two values.

Pacheco (33) found for PPC from brewing yeast a value of 492 mL of oil/g of protein, a value that did not differ from SPI and was slightly higher than the one reported in this paper for alcohol yeast.

Results of replacement of 20 and 40% meat (chuck roll) protein by one of the meat extenders (PPC, TSP, SPI) are shown in **Table 8**. With 20% meat protein replacement the TSP showed the best EC, differing from PPC and SPI, with no difference

Table 9. Diet Consumption, Protein Consumption, Body Weight Change, Protein Efficiency Ratio (PER), and Net Protein Ratio (NPR) for Three Sources of Dietary Protein: Whole Yeast Cells (WY), Phosphorylated Yeast Protein Concentrate (PPC), Commercial Casein (CAS), and a Protein-free Diet (PFD)^a

	sources of protein			
measured parameter	WY	PPC	CAS	PFD ^b
diet consumed (g)	183.4 ± 33.2 b	340 ± 39.6 a	332.4 ± 1.4 a	101.0 ± 22.5
protein consumed (g)	$18.3 \pm 3.3 \text{ b}$	36.0 ± 4.2 a	33.3 ± 3.1 a	\sim 0
body weight change (g)	25.4 ± 7.5 b	135.7 ± 11.9 a	123.4 ± 10.2 a	(-14.9 ± 1.3)
PER	1.4 ± 0.6 b	$3.9 \pm 0.6 \text{ a}$	4.0 ± 0.6 a	ŇD
NPR	2.1 ± 0.6 b	4.3 ± 0.6 a	4.4 ± 0.6 a	ND

^a Results represent mean ± SD of eight rats per treatment. Different letters (rows) indicate statistical differences (*p* < 0.05). ^b Protein-free diet used for the calculation of NPR and the true protein digestibility (TD), **Table 3**.

between the two latter ones. However, with 40% meat protein replacement EC was higher and identical for PPC and SPI, being superior to and statistically different from TSP. EC was higher at 40% meat protein replacement for PPC and SPI than at 20% replacement, but for TSP EC was higher with 20% replacement than with 40%.

All values of EC obtained by replacement of either 20 or 40% of chuck roll protein (**Table 8**) were statistically identical with 100% chuck roll protein with an emulsifying capacity of 430.7 \pm 9.2 mL of oil/g of protein.

Results of the biological (rat) assay are presented in **Table 9**. It compares some nutritional parameters from WY, PPC, and commercial casein (CAS) as the control. For all parameters studied WY showed statistically lower values than PPC and CAS, which did not differ between themselves. PER and NPR can be considered high for both PPC and CAS.

The nutritional superiority of PPC, compared to WY, is graphically illustrated in **Figure 1**, showing the continuous growth lines for WY, CAS, and PPC during the entire experimental period. At the end of 21 days of feeding, body weight gain for the rats on CAS and PPC was 4.8 and 5.3 times higher, respectively, than for WY. No statistical difference was found between CAS and PPC, and both were superior to WY protein. The very low growth-promoting property of WY, compared with CAS and PPC, can be explained by the low true digestibility (TD) and protein digestibility corrected amino acid scoring of WY (**Table 3**).

It is important to mention the higher growth-promoting capacity (89.8 g) and NPR (3.65) found for whole yeast cells from the brewing industry (33) compared to 25.4 g and 2.1, respectively, from the yeast alcohol industry (**Table 9**). Digestibility was 83% in brewing yeast (33) and 60% for yeast from an ethanol distillery (**Table 3**).

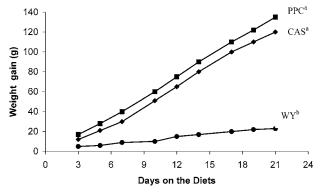


Figure 1. Continuous grouth curve for rats (eight per treatment) fed diets with the following protein sources: WY, whole yeast cells; CAS, commercial casein; PPC, phosphorylated yeast protein concentrate.

This suggests that yeast from an alcohol distillery should have a much thicker and tougher cell wall to be digested than the yeast cells from breweries and also contributes to explain the much lower growth-promoting capacity of the yeast from the alcohol industry. Probably the thickening of the cell wall from the alcohol distillery yeast is related to recycling of the same cells in various fermentation batches and the high ethanol concentration to which the cells are submitted.

One can conclude that industrial yeast from alcohol distilleries is a good source of essential nutrients. Digestibility of the whole cells is low due to thickening of the cell wall, which makes the action of digestive enzymes more difficult. The nutritive value of the protein concentrate is quite high, equivalent to that of casein; however, the yield of protein as concentrate is low, \sim 20% on weight basis, making the industrial manufacturing of yeast protein concentrate not competitive with other protein sources such as soy, for example.

At the moment the best uses of yeast as a byproduct from breweries and distilleries are as derivatives in the form of whole autolysate, for nutritional and functional purposes, and as yeast extracts, for nutritional and flavoring applications (34, 35). Utilization of the yeast cell wall fraction, rich in glycan, mannan, and oligosaccharides (MOS), has been of great interest recently (36).

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