




# Incorporation of natural and mechanically ruptured brewing yeast cells in beef burger to replace textured soy protein

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**Abstract** The use of brewer's yeast to replace textured soy protein (TSP) in burgers was investigated. Three formulations were made, corresponding to a control formulation with 4% TSP, a formulation containing 4% yeast cells in their natural state, and a formulation made with 4% mechanically ruptured yeast cells, which were characterized for the chemical, technological, and sensory properties. Significant differences were observed for pH and instrumental color between the formulations, with no changes in the visual color evaluation by the untrained assessors. The addition of yeast cells resulted in a higher cooking yield and lower reduction in diameter, contributing to maintaining the shape and juiciness of burgers, which is a positive aspect from the technological point of view. The TSP-based formulation presented higher overall appearance and flavor scores when compared with the other formulations, with no significant differences for the other

sensory attributes. The results showed that debittering of yeast-cell biomass is required to remove hop resins and tannins before using in burgers, aimed to improve the product's acceptance and the purchase intent. The debittered yeast cell biomass can be used in burger formulations with great potential to replace TSP, as an alternative to obtain a free allergen meat product.

**Keywords** Brewery biomass · By-product · Burger · Technological characteristics · Sensory evaluation

## Introduction

Non-meat proteins are used in the manufacture of meat products to improve the texture (bonding strength and firmness) and the emulsion stability, as well as to decrease cooking losses, increase yield, and reduce costs (Pancrazio et al. 2016). Textured soy protein (TSP) is one of the most used non-meat proteins in mortadella, sausages, and burgers (Yamada et al. 2010). However, soy protein is one of the major allergenic proteins, especially for infants (Savage et al. 2010; Meinschmidt et al. 2016).

Alternatively, the use of brewing yeast cells as a protein source in meat products has been evaluated with positive results due to its emulsifying properties, flavor (due to the great amount of glutamic acid) and color enhancing, and water-holding capacity (Yamada et al. 2010; Pancrazio et al. 2016). It can be used in the food industry to produce yeast protein concentrates and isolates, with the maintenance of the functional properties and nutritional values. Moreover, brewer's yeast is considered safe, has a considerable protein profile (between 40 and 58%), with the presence of essential amino acids (lysine, leucine), carbohydrates, mineral salts, particularly selenium and B

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vitamins (B1, B2, B6, pantothenic acid, niacin, folic acid, and biotin) (Ferreira et al. 2010).

Meat products are complex food systems, and the water absorption, gelation, and emulsion formation can influence the stability and texture of the cooked product. However, meat proteins provide low functionality to these products, generally requiring supplementation with other emulsifying compounds to form a stable emulsion (Dutra et al. 2012). Several studies have shown the advantages of the use of non-meat additives with this purpose, such as food hydrocolloids (Wu and Lin 2011), brewing yeast extract (Pancrazio et al. 2016), and vegetable oils (Kang et al. 2017), among others.

Burgers consist of simple mixtures of ground meat and animal fats (beef, pork, poultry meat, fish, or their blends). Other animal tissues such as the connective tissue/tendons can also be part of the mixture. Burgers are characterized by the addition of salt and spices, mainly black and white pepper, and also herbs, garlic, or onion during mincing and blending steps, as well as meat extenders, which can also be used in the formulations. In industrial meat processing, the most used extender is soy concentrate in medium to coarse granular shape as textured vegetable protein. Textured soy protein is commonly used as a non-meat ingredient in some formulations in quantities up to 25%. Other non-meat ingredients suitable for this purpose include rusk, breadcrumbs, and dried flakes from roots and tubers (Heinz and Hautzinger 2007).

Previous studies of our research group have evaluated the functional and digestibility properties of yeast samples subjected to different cell disruption methods. The result showed better emulsifying properties and greater oil retention capacity for the yeast cells subjected to mechanical rupture by ultrasound (MRY) (Bertolo et al. 2019). In this context, this study aimed to use the biomass of *Saccharomyces* sp. yeast cells used for beer production, in both their natural state and mechanically ruptured form, in burger formulations, and to evaluate the chemical, technological, and sensory characteristics when compared to the conventional formulation made with textured soy protein.

## Material and methods

### Obtaining brewer's yeast

The yeast cells suspension used in the tests, resulting mainly from the production of Pilsen beer, was donated by a brewery in the city of Chapecó, Santa Catarina, Brazil. The brewer's yeast was reused in the brewery up to five times.

The natural yeast cells (NY) were obtained as described by Bertolo et al. (2019). First, the cell suspension was centrifuged (Centribio, Cienlab, Brazil) for ten minutes at  $349 \times g$  (2500 rpm) to remove excess water and other suspended solids. After discarding the supernatant, distilled water was added in a 2:1 ratio (v/v) to eliminate the remaining impurities. The washing and centrifugation steps were performed twice, thus obtaining the clean natural yeast cells (NY).

NY was divided into two portions. A portion was stored in an ultra-freezer at  $-86\text{ }^\circ\text{C}/24\text{ h}$  and freeze-dried (benchtop freeze-dryer TFD5503, Ilshin Lab. Co. (Ltd., Korea) at  $-61\text{ }^\circ\text{C}$  and 67 mbar. The freeze-dried mass was manually crushed and sieved (mesh 32 mm  $\mu\text{m}^{-1}$ ). The other portion was subjected to cell rupture through a low-frequency ultrasound to obtain the mechanically ruptured yeast cells (MRY). For that, NY was mixed with distilled water at a 1:1 ratio (w/v) and subjected to an ultrasound bath (Q335D, Quimis, Brazil) for 1 h, at 40 kHz and maximum temperature of  $30\text{ }^\circ\text{C}$ . Then, the MRY was placed in Petri dishes, frozen in an ultra-freezer, freeze-dried, crushed, and sieved, using the same parameters for obtaining the NY.

### Application of brewer's yeast in burgers

NY and MRY were incorporated into the beef burger formulations, according to the proportions shown in Table 1. A control formulation made with textured soy protein (TSP) was used for comparison purposes since it is the most used vegetable protein in meat products. The TSP was obtained from a local market, as well as the other

**Table 1** Beef burger formulations: standard, with addition of natural yeast and with addition of mechanically ruptured yeast

Ingredients (% , m/m)	Formulations		
	S-F	NY-F	MRY-F
Lean beef	75	75	75
Ice water	12.30	12.30	12.30
Beef fat	7	7	7
TSP	4	0	0
Brewery yeast	0	4	4
Salt	1.5	1.5	1.5
Black pepper powder	0.1	0.1	0.1
Onion powder	0.5	0.5	0.5
Garlic powder	0.5	0.5	0.5

TSP textured soy protein, S-F standard formulation with addition of 4% of textured soy protein (TSP), NY-F formulation with addition of 4% of natural yeast (NY), MRY-F formulation with addition of 4% of mechanically ruptured yeast (MRY)

ingredients (lean beef, beef fat, salt, black pepper, onion, and garlic powder). The Brazilian legislation (Brasil 2000) allows the addition of up to 4% of non-meat-based protein to burgers as aggregated protein.

The burger processing was performed according to the following stages: acquisition of raw-material, manual removal of connective and fat tissues, cutting, and milling of raw material in industrial grinder with a 3 mm disk (MSI-10, Becker, Brazil). The amounts of raw materials and ingredients used in the formulations are shown in Table 1, which were placed in a recipient in the following order: lean beef, beef fat, water, salt, black pepper, onion and garlic powder, and textured soy protein (S-F) or brewer's yeast (NY-F and MRY-F), and manually homogenized. Then, the burgers (50 g) were molded in stainless-steel molds 8 cm in diameter, taken out from the mold, packed, and stored in plastic containers at  $-18\text{ }^{\circ}\text{C}$  until the analysis.

The three formulations were manufactured in two batches ( $n = 2$ ). The chemical, technological, and microbiological characterization was performed in triplicate.

### Chemical characterization

The chemical characterization of the different burger formulations was performed according to the methodologies of the AOAC International (2016), as follows: ash content by incineration in a muffle (Q-318M24, Quimis, Brazil), method 923.02; lipids content by the Soxhlet extract (LUCA-145/6, Lucadema, Brazil), method 920.39; crude protein content by micro Kjeldahl (LUCA-341/02, Lucadema, Brazil), method 960.52, using the factor 6.25 to convert nitrogen to crude protein; moisture content by the gravimetric method 925.45, with sample drying in an oven (Centribio, Cienlab, Brazil); the carbohydrate content was calculated by difference.

### Physical and technological properties

The burger formulations were evaluated for cooking yield, diameter reduction, water holding capacity, shear force, color, water activity, and pH.

The cooking yield (CY) and the diameter reduction (DR) was determined as described by Berry (1992). The burgers were packed in aluminum foil and kept in an industrial electric griddle (C-80, Venâncio, Brazil) for approximately 12 min at  $150\text{ }^{\circ}\text{C}$ , turning each 2 min until the internal temperature of the burgers reached  $72 \pm 2\text{ }^{\circ}\text{C}$ , monitored by a digital thermometer. The percent cooking yield was calculated by the ratio between the mass of the cooked sample and the mass of the raw sample. The percent diameter reduction was expressed as a function of the

raw sample diameter and the cooked sample diameter, measured using a caliper.

Water holding capacity (WHC) was determined according to the methodology described by da Silva Sobrinho et al. (2005) with adjustments. The burger samples ( $12 \pm 0.5\text{ g}$ ,  $3 \times 3 \times 0.8\text{ cm}$ ) were placed between two pieces of Whatman grade 1 filter paper with 115 mm in diameter, previously dried, between two acrylic-plastic plates, and a force of 5 kg was applied for 10 min. Then, the sample was weighed, and the amount of water lost was calculated by difference.

The texture analysis was performed using raw burger samples of 1.0 cm wide, 4.0 cm long and 0.8 cm high. The shear force (N) required for cutting the samples was measured in a CT3 texture analyzer (Brookfield, Brazil), with a Warner Bratzler blade, speed of 5 mm/s, Trigger load of 1.0 N, and distance of 50% for the depth of cut, as reported by Abularach et al. (1998) with adaptations.

The instrumental color of the raw and cooked burgers was determined by colorimetry (EZ 0374 4500L, Hunter Lab MiniScan, Brazil), operating in the CIE system ( $L^*$ ,  $a^*$ ,  $b^*$ , in which  $L^*$  represents lightness, and  $a^*$  and  $b^*$  are the chromaticity coordinates). The color variation ( $\Delta E$ ) was calculated according to lightness ( $L^*$ ), chromaticity  $a^*$  (green to red), and chromaticity  $b^*$  (blue and yellow) of the standard formulation (S-F) when compared with the formulations with the replacement of TSP for brewer's yeast (NY-F and MRY-F), according to Lopes et al. (2005), as shown in Eq. 1

$$\Delta E^* = \left[ (\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2 \right]^{1/2}. \quad (1)$$

Water activity ( $a_w$ ) at  $25\text{ }^{\circ}\text{C}$  was measured in a water activity apparatus (AquaLab Pre, Decagon, Brazil).

The pH was measured in a benchtop pH meter (mPA210, MS Technopon, Brazil) according to the method 943.02 of AOAC (2016)

### Microbiological characterization

The microbiological characterization of the burgers was performed according to the Normative Instruction 62 from August 26, 2003, Ministry of Agriculture, Livestock, and Supply before the sensory evaluation. The Brazilian Legislation (Brasil 2001) has established the microbiological sanitary standards for meat products, cooked or not, sausages or not, as  $10^2\text{ MPN g}^{-1}$  for coliforms at  $45\text{ }^{\circ}\text{C}$ ,  $10^2\text{ CFU g}^{-1}$  for coagulase-positive staphylococci,  $10^2\text{ - CFU g}^{-1}$  for sulfite-reducing *Clostridium* at  $46\text{ }^{\circ}\text{C}$ , and the absence of *Salmonella* sp. in 25 g.

## Sensory evaluation

The sensory tests were performed with 50 untrained assessors, at a single session. Smokers, underage, and those who do not consume the product and/or reported intolerance or allergy to the product, or its components were excluded from the study. The analyses were conducted at the Sensory Analysis Laboratory of the State University of Santa Catarina (UDESC), Pinhalzinho campus, in compliance with the appropriate laws and institutional guidelines, after the ethics committee approval (register number: 75617517.5.0000.0118). A statement that informed consent was obtained from each individual that participated of the tests.

Each assessor received a portion of the sample (~ 10 g) and a response sheet in individual booths. The samples were cooked on a conventional electric oven (Grill, Fischer, Brazil) at 230 °C, until reaching an internal temperature of 72 °C at the geometric center. Water and cracker biscuits were provided to cleanse the palate between samples. The samples were coded with three-digit codes and individually served in a randomized order.

The burgers were analyzed for the acceptability using a structured hedonic scale of nine points, varying from 1 (dislike it extremely) to 9 (like it extremely), regarding the attributes overall acceptance, appearance, color, flavor, aroma, and texture. The purchase intent test was also performed using a 9-point structured scale, varying from 1 (would certainly not buy it), 5 (maybe would buy it) and 9 (would certainly buy it) (Teixeira et al. 1987; Dutcosky 2007).

The acceptability index (AI) of each formulation was calculated according to Eq. 2 (Teixeira et al. 1987)

$$AI (\%) = \frac{\text{average score obtained for the product}}{\text{maximum score given to the product}} \times 100. \quad (2)$$

## Statistical analysis

A mixed model analysis of variance (mixed model ANOVA) was used to analyze all variables of the study. The parameters were set as dependent variables, the formulation was considered as fixed effects, and the different batches and replicates were considered as random effects. The comparisons of treatments were performed by the Tukey's test ( $p \leq 0.05$ ), using the STATISTICA 13.2 Trial software (Statsoft). The values were expressed as mean values and standard deviation.

Data from the acceptance test and the purchase intention were analyzed by the same software considering a mixed linear model, including the different formulations (S-F,

NY-F, MRY-F) as fixed effects and the assessors as a random effect. Means were compared by the Tukey's test ( $p \leq 0.05$ ).

## Results and discussion

The results of the chemical characterization of the different burger formulations with replacement of TSP (textured soy protein) for NY (natural yeast) and MRY (mechanically ruptured yeast) are presented in Table 2. No significant differences were observed for moisture, lipids, protein, ash, and carbohydrates levels ( $p > 0.05$ ) among the samples, which shows that replacing vegetable protein for microbial protein did not affect the chemical characteristics of the product, indicating, therefore, the possibility of using brewer's yeast in beef burger formulations.

Regarding the protein contents, the formulations were in accordance with the technical regulation for identity and quality of burgers (Brasil 2000), which establishes the minimum content of 15% of protein in the final product. De Oliveira et al. (2014) also obtained similar protein contents, which ranged from 18.81 to 20.45% for beef burgers with reduced sodium and the addition of 4% TSP.

Previous studies of our research group have shown protein levels of  $42.81 \pm 1.03$ ;  $42.83 \pm 0.11$ ; and  $43.80 \pm 0.62$  in g 100 g<sup>-1</sup> of product, for TSP, NY, and MRY, respectively (Bertolo et al. 2019). Similar non-meat proteins were used in the burger formulations of the present study, and no changes were observed in the protein contents of the burgers (Table 2). It is worth mentioning that the replacement of TSP for yeast cells provided an increase in essential amino acids such as lysine, once TSP has a smaller amount of this amino acids when compared with the yeast cells, with values of 6.1 g 100 g<sup>-1</sup> (Weingartner 1987) and 7.8 g 100 g<sup>-1</sup> (Yamada et al. 2003), respectively. Additionally, the replacement of TSP by the yeast minimized the allergenic effect caused by soy protein, while maintaining the same protein content in the final product.

Concerning the lipid levels, the samples of the present study were in accordance with the Brazilian legislation, which has recommended a maximum fat content of 23% in beef burgers (Brasil 2000). A similar amount of fat was used in all samples, which was confirmed by the results of the lipid determination of the different formulations (7%).

Regarding the total carbohydrates, the formulations were in accordance with the Brazilian legislation, which has established a maximum carbohydrate content of 3% (Brasil 2000). Although the carbohydrate contents of the natural yeast cells and the TSP were 53.9% and 48.6%, respectively, as reported by Bertolo et al. (2019), these

**Table 2** Chemical characterization of the standard burger and the ones added of natural yeast and mechanically ruptured yeast replacing the textured soy protein

Component (g/100 g of d.b.)	S-F	NY-F	MRY-F
Moisture	69.56 ± 0.82 <sup>a</sup>	69.77 ± 0.68 <sup>a</sup>	71.09 ± 0.44 <sup>a</sup>
Lipid	8.48 ± 0.18 <sup>a</sup>	8.07 ± 0.08 <sup>a</sup>	7.23 ± 0.62 <sup>a</sup>
Protein (N × 6.25)	18.00 ± 0.75 <sup>a</sup>	17.25 ± 0.80 <sup>a</sup>	17.38 ± 0.33 <sup>a</sup>
Ash	2.46 ± 0.06 <sup>a</sup>	2.30 ± 0.02 <sup>a</sup>	2.29 ± 0.13 <sup>a</sup>
Total carbohydrates	2.08 ± 0.13 <sup>a</sup>	2.62 ± 0.50 <sup>a</sup>	2.00 ± 0.81 <sup>a</sup>

Values expressed as mean ± standard deviation. Different letters in the same row indicate significant difference ( $p \leq 0.05$ ) by the Tukey test

S-F standard formulation with addition of 4% of textured soy protein (TSP), NY-F formulation with addition of 4% of natural yeast (NY), MRY-F formulation with addition of 4% of mechanically ruptured yeast (MRY), d.b. dry basis

differences in the non-meat protein sources did not affect the carbohydrates of the burgers.

The results of the instrumental color of the burger formulations are presented in Table 3. Significant differences ( $p \leq 0.05$ ) were observed for the lightness value ( $L^*_{\text{raw}}$ ) of the sample MRY-F, indicating a lighter shade when compared with the other raw samples, probably due to the interaction between the mechanically ruptured yeast (MRY) and the burger constituents. On the other hand, no significant differences were observed for  $L^*_{\text{(cooked)}}$  between the samples ( $p > 0.05$ ).

Regarding the red coordinate ( $a^*$ ), significant differences were observed for the sample S-F ( $p \leq 0.05$ ) when compared with the samples with yeast cells, for both the raw and cooked form. The raw burger S-F presented a higher  $a^*$  value, indicating a more reddish shade when compared with NY-F and MRY-F, probably due to the

differences in the composition (ash, carbohydrates, and lipids) of NY and MRY when compared with the control (TSP) (Bertolo et al 2019), which may have caused the dilution of the myoglobin pigment (Do Prado et al. 2019). Moreover, the difference among the raw samples remained after cooking ( $p < 0.05$ ), once lower  $a^*_{\text{(cooked)}}$  values were observed for S-F when compared with NY-F and MRY-F. When comparing the raw and cooked samples, all formulations exhibited lower  $a^*$  values after cooking, probably due to the effect of the Maillard and caramelization reactions that occurred during the heat treatment (do Prado et al. 2019).

Concerning the yellowness ( $b^*$ ), there was no significant difference ( $p > 0.05$ ) between the samples, for both  $b^*_{\text{(raw)}}$  and  $b^*_{\text{(cooked)}}$ . In previous studies, no differences were observed between the NY and MRY samples for all parameters evaluated ( $L^*$ ,  $a^*$  and  $b^*$ ) (Bertolo et al 2019), which was also observed in the results in Table 3, except for  $L^*_{\text{(raw)}}$ .

For the  $\Delta E$ , according to Francis and Clydesdale (1975), values close to zero indicate that the samples resulted in a product with characteristics similar to the control, while  $\Delta E \geq 2$  between two samples can be considered as noticeably different to the human eye. The raw formulations made with brewer’s yeast (NY-F and MRY-F) had  $\Delta E$  values  $> 2$ , indicating a noticeable change in color when compared with the control formulation (S-F). However, after cooking, there was no longer this perception since the results were close to zero, indicating a difference not perceptible to the human eye.

Concerning the cooking yield (CY), the sample NY-F statistically differed ( $p \leq 0.05$ ) from S-F and MRY-F (Table 4), with CY value of approximately 82%, which was higher when compared with the other samples (S-F and MRY-F), probably due to the higher oil holding capacity of the natural yeast when compared to textured soy protein. Bertolo et al. (2019) reported higher OHC of natural yeast cells when compared with textured soy protein, with values of 8.44 mL g<sup>-1</sup> and 4.28 mL g<sup>-1</sup>,

**Table 3** Instrumental color and color variation of the standard burger and the ones added of natural yeast and mechanically ruptured yeast replacing the textured soy protein

Parameters	S-F	NY-F	MRY-F
$L^*_{\text{(raw)}}$	43.64 ± 0.95 <sup>b</sup>	43.27 ± 0.98 <sup>b</sup>	46.35 ± 0.08 <sup>a</sup>
$L^*_{\text{(cooked)}}$	35.52 ± 0.93 <sup>a</sup>	35.58 ± 0.67 <sup>a</sup>	35.18 ± 0.64 <sup>a</sup>
$a^*_{\text{(raw)}}$	13.89 ± 0.75 <sup>a</sup>	9.52 ± 0.08 <sup>b</sup>	9.74 ± 0.18 <sup>b</sup>
$a^*_{\text{(cooked)}}$	6.97 ± 0.09 <sup>b</sup>	7.52 ± 0.13 <sup>a</sup>	7.35 ± 0.12 <sup>a</sup>
$b^*_{\text{(raw)}}$	16.93 ± 0.31 <sup>a</sup>	16.59 ± 0.70 <sup>a</sup>	17.08 ± 0.11 <sup>a</sup>
$b^*_{\text{(cooked)}}$	13.74 ± 0.45 <sup>a</sup>	13.77 ± 0.29 <sup>a</sup>	13.38 ± 0.20 <sup>a</sup>
$\Delta E_{\text{(raw)}}$	–	4.39	4.95
$\Delta E_{\text{(cooked)}}$	–	0.67	0.53

Values expressed as mean ± standard deviation. Different letters in the same row indicate significant difference ( $p \leq 0.05$ ) by the Tukey test

S-F standard formulation with addition of 4% of textured soy protein (TSP), NY-F formulation with addition of 4% of natural yeast (NY), MRY-F formulation with addition of 4% of mechanically ruptured yeast (MRY). Lightness ( $L^*$ ), chromaticity  $a^*$  (green to red), chromaticity  $b^*$  (blue and yellow), and color variation ( $\Delta E$ )

**Table 4** Technological and chemical properties of the standard burger and the ones added of natural yeast and mechanically ruptured yeast replacing the textured soy protein

Parameters	S-F	NY-F	MRY-F
Cooking yield (%)	67.90 ± 1.18 <sup>b</sup>	82.03 ± 2.89 <sup>a</sup>	72.74 ± 2.10 <sup>b</sup>
Diameter reduction (%)	20.42 ± 1.15 <sup>a</sup>	13.30 ± 1.89 <sup>b</sup>	16.28 ± 2.59 <sup>b</sup>
Water holding capacity (%)	94.21 ± 1.54 <sup>a</sup>	95.03 ± 0.58 <sup>a</sup>	94.78 ± 0.94 <sup>a</sup>
Shear force (N)	2.13 ± 0.18 <sup>a</sup>	1.97 ± 0.26 <sup>a</sup>	1.88 ± 0.15 <sup>a</sup>
Water activity	0.979 ± 0.006 <sup>a</sup>	0.987 ± 0.002 <sup>a</sup>	0.979 ± 0.005 <sup>a</sup>
pH	5.74 ± 0.02 <sup>a</sup>	5.59 ± 0.01 <sup>b</sup>	5.51 ± 0.01 <sup>c</sup>

Values expressed as mean ± standard deviation. Different letters in the same row indicate significant difference ( $p \leq 0.05$ ) by the Tukey test

*S-F* standard formulation with addition of 4% of textured soy protein (TSP), *NY-F* formulation with addition of 4% of natural yeast (NY), *MRY-F* formulation with addition of 4% of mechanically ruptured yeast (MRY)

respectively, and lower water holding capacity (WHC) with values of  $7.69 \text{ g g}^{-1}$  and  $8.69 \text{ g g}^{-1}$ , respectively. Although the modified rupture yeast cells (MRY) had similar OHC and WHC when compared with NY, it was submitted to the ultrasound process, which can modify the protein conformation by affecting the hydrogen bonds and the hydrophobic interactions.

Some authors reported CY values of 69.83% in burgers made with TSP (Filho et al. 2012), which is close to the results of S-F obtained in this study. De Oliveira et al. (2014) found 72.75% of CY in beef burger made with 10% of animal fat and 4% of TSP. The CY values of the burgers made with TSP reported by those authors indicated that the burgers with the addition of NY exhibited excellent CY when compared with the burgers made with TSP. Shahiri Tabarestani and Mazaheri Tehrani (2014) reported that the simultaneous addition of soy flour and starch provided a positive effect on cooking yield of hamburger, with values reaching 91.68%. This result is due to the ability of soy flour and starch to bind water and fat, and to retain these components in the meat, which was also observed for NY of the present study.

Significant differences ( $p \leq 0.05$ ) were observed for the diameter reduction (DR) among the samples, with higher values for S-F when compared with NY-F and MR-Y-F (Table 4). The sample NY-F presented a lower DR, probably due to the higher OHC of the natural yeast, as also reported for CY. The diameter reduction and, consequently, the lower cooking shrinkage, which is considered one of the most important physical quality changes that occur in beef burgers during the cooking process, is due to the denaturation of meat proteins and water and fat release from beef burger patties (Shahiri Tabarestani and Mazaheri Tehrani 2014). The use of textured soy protein in blended ground beef patties was associated with substantial reductions in cooking loss (Smith et al 1976). In the present study, the brewer's yeast also led to a reduction of cooking loss, which was more pronounced when compared with

TSP probably due to its higher oil holding capacity. Do Prado et al. (2019) reported DR of 20.54% in cooked beef burgers made with soy protein isolate, while Filho et al. (2012) reported 13.27% in beef burgers made with TSP. All those results are similar to the findings of this study.

No significant differences were observed for the water holding capacity (WHC) ( $p > 0.05$ ) among the burger formulations (Table 4), with values above 94%, which suggests a high protein functionality that leads to immobilization of free water, thus leading to an increase in the product stability, with positive impacts on cooking yield (Kenney et al. 1992). The water holding capacity influences the sensory quality of meat since the water loss during cooking may impair its succulence and tenderness. The physical properties of the meat (color and texture of raw meat) and the consumers' acceptance depend on its ability to retain water (Forrest and Pérez 1979). The addition of yeast to the burger formulations did not affect this property, which shows that the replacement of TSP by brewer's yeast may be an effective alternative while maintaining WHC. Filho et al. (2012) reported 67.66% of WHC in beef burgers, which is lower than the values obtained in this study.

The pH values differed significantly ( $p > 0.05$ ) among the samples (Table 4), and the sample S-F presented the higher value, followed by NY-F and MR-Y-F. The change in pH depends upon the pH of the non-meat source added, as reported for fiber source by Mehta et al (2015). Keeton and Melton (1978) evaluated ground beef containing 0, 10, 20, and 30% of textured soy protein, and reported pH values of 5.75, and 6.0–6.5 for the treatments containing 0, and 10–30% TSP, respectively. Angiolillo et al. (2015) found a pH value of 5.68 in beef burgers made with minced meat, salt, and oregano, and pH values ranging from 5.55 to 5.60 in beef burgers enriched with fructooligosaccharides, inulin, and an oat bran-loaded protein foam. Therefore, we can state that the burger S-F presented higher pH due to the pH of TSP, while NY-F and MR-Y-F reduced the

pH burger probably due to the pH of the brewer's yeast and its interactions with the meat matrix.

Regarding the water activity and shear force values (Table 4), no significant differences ( $p < 0.05$ ) were observed among the samples indicating that the replacement of the TSP by brewer's yeast did not affect these characteristics.

In the meat industry, soy protein is the most widely used vegetable protein, due to its biological value, the emulsifying and stabilizing properties, and its ability to increase the water holding capacity and improve the texture of the final product (Macedo-Silva et al. 2001). The primary function of the protein is to improve the dimensional stability of the patties, preserve the structural integrity of the ground meat pieces during heat treatment, and help to retain meat juices (decreased cooking losses) (Singh et al. 2008). In this study, the addition of brewer's yeast to the burger formulations provided an improvement in the different properties. The formulations containing natural or mechanically ruptured yeast cells presented the best results for cooking yield and diameter reduction of beef burgers, showing the possibility of replacing a vegetable protein with allergenic potential (TSP) for another of microbial origin (yeast cells), maintaining or improving important technological characteristics in the final product. In addition, the possibility of using brewer's yeast in its natural form was demonstrated, which becomes more viable from the technological and economic point of view.

The microbiological characterization showed that all burgers were within the limits established by the Brazilian legislation (Brasil 2001), indicating the absence of *Salmonella* sp. in 25 g of sample,  $< 3.0$  MPN  $g^{-1}$  for coliforms, maximum of  $2.3 \times 10^2$  CFU  $g^{-1}$  for coagulase-positive staphylococci, and  $< 1.0 \times 10^2$  CFU  $g^{-1}$  for sulfite-reducing *Clostridium* at 46 °C. According to these results, all burgers were safe for human consumption from a microbiological standpoint.

The sensory scores for all the attributes evaluated and the results of the purchase intention tests of the burger formulations are presented in Table 5. No significant differences were observed for the attributes appearance and color among the samples ( $p > 0.05$ ). The control formulation statistically differed ( $p < 0.05$ ) from the other samples, with higher scores for the overall acceptance, aroma, flavor, and purchase intent. No differences were observed between the formulations NY-F and MRY-F for the overall acceptance, aroma, texture, and purchase intent, while significant differences were observed for the attribute flavor, with lower values for the sample NY-F when compared with MRY-F.

The most prominent differences were observed for the attribute flavor, once the consumers preferred the burger made with textured soy protein when assessing this

**Table 5** Means of the sensory attributes in the Hedonic Scale Test and the purchase intent of standard burgers and the ones added of natural and mechanically ruptured yeast replacing textured soy protein

Attributes	Formulations		
	S-F	NY-F	MRY-F
Overall acceptance	7.65 ± 0.80 <sup>a</sup>	6.43 ± 1.06 <sup>b</sup>	6.55 ± 1.18 <sup>b</sup>
Appearance	7.65 ± 0.74 <sup>a</sup>	7.63 ± 0.74 <sup>a</sup>	7.58 ± 1.01 <sup>a</sup>
Color	7.48 ± 0.85 <sup>a</sup>	7.28 ± 0.88 <sup>a</sup>	7.40 ± 0.93 <sup>a</sup>
Aroma	7.65 ± 0.86 <sup>a</sup>	6.93 ± 1.07 <sup>b</sup>	7.03 ± 1.23 <sup>b</sup>
Flavor	8.03 ± 0.73 <sup>a</sup>	5.30 ± 1.54 <sup>c</sup>	5.98 ± 1.23 <sup>b</sup>
Texture	7.88 ± 0.82 <sup>a</sup>	7.38 ± 0.93 <sup>b</sup>	7.55 ± 0.96 <sup>ab</sup>
Purchase intent	8.50 ± 1.34 <sup>a</sup>	3.60 ± 1.93 <sup>b</sup>	4.30 ± 1.79 <sup>b</sup>

Values expressed as mean ± standard deviation. Different letters in the same row indicate significant difference ( $p \leq 0.05$ ) by the Tukey test

S-F standard formulation with addition of 4% of textured soy protein (TSP), NY-F formulation with addition of 4% of natural yeast (NY), MRY-F formulation with addition of 4% of mechanically ruptured yeast (MRY)

attribute. Whereas the range of acceptance threshold is 6 to 9, the rejection from 1 to 4 and the score 5 is considered as an indecision zone, only the flavor scores of the formulations containing brewer's yeast were close to the indecision zone.

For the purchase intent, the formulations containing yeast cells showed scores in the range of the rejection threshold. This result may be due to the extremely bitter flavor of the yeast to the presence of bitter compounds, such as resins and tannins from the hops used in brewing (Alvim et al. 1999), indicating the need for a previous treatment to debitter the biomass before using in meat products such as burgers.

According to Teixeira et al. (1987) and Dutcosky (2007), to be considered as sensory accepted, the product should present an acceptability index (AI) of at least 70%. Higher acceptability indexes were observed for the samples S-F and MRY-F for all attributes evaluated through the hedonic scale and the purchase intent test, among the different burger formulations (Table 6), while the NY-F was below the range of acceptance threshold for the attribute flavor.

## Conclusion

This study investigated the suitability of brewer's yeast to completely substitute textured soy protein in burgers. The results demonstrated that the yeast cells both in the natural state and subjected to mechanical rupture improved the

**Table 6** Acceptability indexes (%) of the standard burger and the ones added of natural yeast and mechanically ruptured yeast replacing the textured soy protein

Attributes	Formulations		
	S-F	NY-F	MRY-F
Overall acceptance	85.00	80.31	81.88
Appearance	85.00	84.72	84.17
Color	83.06	90.94	82.22
Aroma	85.00	86.56	78.06
Flavor	89.17	66.25	74.69
Texture	87.50	81.94	83.89

S-F standard formulation with addition of 4% of textured soy protein (TSP), NY-F formulation with addition of 4% of natural yeast (NY), MRY-F formulation with addition of 4% of mechanically ruptured yeast (MRY)

cooking characteristics (higher cooking yield and lower diameter reduction), with a positive effect on the chemical composition and the physical properties of the burgers. On the other hand, it negatively affected the sensory characteristics of the product, reducing its acceptability with replace the texture soy protein by brewery yeast.

To guarantee the sensory acceptability of the product, previous debittering treatment of the biomass is recommended to improve the consumers' acceptance and purchase intent. Further studies are required in the preparation of brewer's yeast, due to its potential to replace textured soy protein, in addition to be an alternative to the use of residues from the brewing industry.

The present findings suggest that the replacement of 100% textured soy protein for brewer's yeast in burger formulations may be a promising alternative, though more studies are required to improve the attribute flavor and the product's acceptance. In addition to providing an allergen-free burger as an alternative to the traditional burger, the brewer's yeast can be used as a safe, natural, and valuable non-meat protein in the meat industry.

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**Authors' contributions** APB conceptualized the work, adapted the methodologies, investigated, and wrote and revised the MS. APK co-supervised the work. ER co-administrated the project, conceptualized the work. GARS wrote and revised the MS. DC administrated the project, supervised, and conceptualized the work, wrote, and revised the MS.

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**Availability of data and material** The data and material will be available to the journal on request.

## Declarations

**Conflict of interest** The authors declare that they have no conflict of interest.

**Ethical approval** This manuscript contains studies with human. The sensory tests were performed with 50 untrained assessors, at the Sensory Analysis Laboratory of the State University of Santa Catarina (UDESC), Pinhalzinho campus, after the ethics committee approval (register number: 75617517.5.0000.0118).

**Consent to participate** The consent form is made in two copies, one of which will remain with the researcher and the other with the research participant.

**Consent for publication** The consent form is made in two copies, one of which will remain with the researcher and the other with the research participant.

## References

- Abularach MLS, Rocha CE, de Felício PE (1998) Características de qualidade do contrafilé (m. *L. dorsi*) de touros jovens da raça nelore. *Food Sci Technol* 18(2):205–210. <https://doi.org/10.1590/S0101-20611998000200012>
- Alvim ID, Vilela ESD, Baldini VLS, Bragagnolo N, Sgarbieri VC (1999) Produção piloto de derivados de levedura (*Saccharomyces sp*) para uso como ingrediente na formulação de alimentos. *Braz J Food Technol* 2(1,2):119–125
- Angiolillo L, Conte A, Del Nobile MA (2015) Technological strategies to produce functional meat burgers. *LWT* 62:697–703. <https://doi.org/10.1016/j.lwt.2014.08.021>
- AOAC (2016) Official methods of analysis of the association of official agricultural chemists, 20th edn. AOAC International, Rockville
- Berry BW (1992) Low fat level effects on sensory, shear, cooking, and chemical properties of ground beef patties. *J Food Sci* 57(3):537–540. <https://doi.org/10.1111/j.1365-2621.1992.tb08037.x>
- Bertolo AP, Biz AP, Kempka AP, Rigo E, Cavalheiro D (2019) Yeast (*Saccharomyces cerevisiae*): evaluation of cellular disruption processes, chemical composition, functional properties and digestibility. *J Food Sci Technol* 56(8):3697–3706. <https://doi.org/10.1007/s13197-019-03833-3>
- Brasil (2001) Agência Nacional de Vigilância Sanitária. Resolução n. 12, de 2 de janeiro de 2001. Regulamento técnico sobre os padrões microbiológicos para alimentos. Diário Oficial da União. Brasília
- Brasil (2000) Ministério da Agricultura e do Abastecimento. Instrução Normativa n. 4, de 31 de março de 2000. Aprova os Regulamentos Técnicos de Identidade e Qualidade de carne mecanicamente separada, de mortadela, de linguiça e de salsicha. Diário Oficial da República Federativa do Brasil. Brasília, DF, 05 abr. 2000. Seção I, 6–10
- da Silva Sobrinho AG, Purchas RW, Kadim IT, Yamamoto SM (2005) Características de qualidade da carne de ovinos de diferentes genótipos e idades ao abate. *R Bras Zootec* 34(3):1070–1078
- de Oliveira DF, Mileski JPF, de Carli CG, Marchi JF, Silva DC, Coelho AR, Tonial IB (2014) Golden flaxseed flour as a substitute for animal fat in reduced-sodium beef hamburgers.



- Braz J Food Technol 17(4):273–282. <https://doi.org/10.1590/1981-6723.0714>
- do Prado MEA, Queiroz VAV, Correia VTDV et al (2019) Physicochemical and sensorial characteristics of beef burgers with added tannin and tannin-free whole sorghum flours as isolated soy protein replacer. *Meat Sci* 150:93–100. <https://doi.org/10.1016/j.meatsci.2018.12.006>
- Dutcosky SD (2007) Análise sensorial de alimentos. Champagnat, Curitiba
- Dutra MP, Cardoso GP, Ramos EM, Ramos ADS, Pinheiro ACM, Fontes PR (2012) Technological and sensory quality of restructured low-fat cooked ham containing liquid whey. *Cienc e Agrotecnologia* 36(1):86–92. <https://doi.org/10.1590/S1413-70542012000100011>
- Ferreira IMPLVO, Pinho O, Vieira E, Tavarela JG (2010) Brewer's *Saccharomyces* yeast biomass: characteristics and potential applications. *Trends Food Sci Tech* 21(2):77–84. <https://doi.org/10.1016/j.tifs.2009.10.008>
- Filho RB, de Oliveira CP, Gomes QO (2012) Preparation of veal burger with added prebiotic inulin as functional ingredient and fat substitute. *Revista Verde* 7(4):33–37
- Forrest JC, Pérez BS (1979) Fundamentos de la ciencia de la carne. *Ciencia y Tecnología de la Carne*, Acribia, Zaragoza
- Francis FJ, Clydesdale FM (1975) Food colorimetry: theory and applications. AVI, Westport, CT
- Heinz G, Hautzinger P (2007) Meat processing technology. For small-to medium-scale producers. Food and Agriculture Organization (FAO) of the United Nations, Regional Office for Asia and the Pacific. Bangkok. <http://www.fao.org/3/a-ai407e.pdf>. Accessed 18 June 2020
- Kang ZL, Zhu D, Li B, Ma H, Song Z (2017) Effect of pre-emulsified sesame oil on physical-chemical and rheological properties of pork batters. *Food Sci Technol* 37(4):620–626. <https://doi.org/10.1590/1678-457x.28116>
- Keeton JT, Melton CC (1978) Factors associated with microbial growth in ground beef extended with varying levels of textured soy protein. *J Food Sci* 43:1–125. <https://doi.org/10.1111/j.1365-2621.1978.tb15250.x>
- Kenney PB, Kastner CL, Kropf DH (1992) Raw and pre-heated epimysium and gelatin affect properties of low-salt, low-fat, restructured beef. *J Food Sci* 57(3):551–554. <https://doi.org/10.1111/j.1365-2621.1992.tb08040.x>
- Lopes AS, Mattietto RA, de Menezes HC (2005) Estabilidade da polpa de pitanga sob congelamento. *Ciênc Tecnol Aliment* 25(3):553–559
- Macedo-Silva A, Shimokomaki M, Vaz AJ, Yamamoto YY, Tenuta-Filho A (2001) Textured soy protein quantification in commercial Hamburger. *J Food Compos Anal* 14:469–478. <https://doi.org/10.1006/jfca.2001.1014>
- Mehta N, Ahlawat SS, Sharma DP (2015) Novel trends in development of dietary fiber rich meat products—a critical review. *J Food Sci Technol* 52:633–647. <https://doi.org/10.1007/s13197-013-1010-2>
- Meinlschmidt P, Ueberham E, Lehmann J, Reineke K, Schlüter O, Schweiggert-Weisz U, Eisner P (2016) The effects of pulsed ultraviolet light, cold atmospheric pressure plasma, and gamma-irradiation on the immunoreactivity of soy protein isolate. *Innov Food Sci Emerg* 38:374–383. <https://doi.org/10.1016/j.ifset.2016.06.007>
- Pancrazio G, Cunha SC, de Pinho PG, Loureiro M, Meireles S, Ferreira IM, Pinho O (2016) Spent brewer's yeast extract as an ingredient in cooked hams. *Meat Sci* 121:382–389. <https://doi.org/10.1016/j.meatsci.2016.07.009>
- Savage JH, Kaeding AJ, Matsui EC, Wood RA (2010) The natural history of soy allergy. *J Allergy Clin Immunol* 125(3):683–686. <https://doi.org/10.1016/j.jaci.2009.12.994>
- Shahiri Tabarestani H, Mazaheri Tehrani M (2014) Optimization of physicochemical properties of low-fat hamburger formulation using blend of soy flour, split-pea flour and wheat starch as part of fat replacer system. *J Food Process Pres* 38:278–288. <https://doi.org/10.1111/j.1745-4549.2012.00774.x>
- Singh P, Kumar R, Sabapathy SN, Bawa AS (2008) Functional and edible uses of soy protein products. *Comp Rev Food Sci Food Saf* 7:14–28. <https://doi.org/10.1111/j.1541-4337.2007.00025.x>
- Smith GC, Marshall WH, Carpenter ZL, Branson RE, Meinke WW (1976) Textured soy proteins for use in blended ground beef patties. *J Food Sci* 41:1148–1152. <https://doi.org/10.1111/j.1365-2621.1976.tb14405.x>
- Teixeira E, Meinert E, Barbata PA (1987) Análise sensorial dos alimentos. UFSC, Florianópolis
- Weingartner KE (1987) Processing, nutrition and utilization of soybeans. In: Sing SR, Rachie K, Daschiel KE (eds) *Soybeans for the tropics: research, production and utilization*. Wiley-Interscience Publications, Chichester, UK, pp 149–178
- Wu YB, Lin KW (2011) Influences of xylooligosaccharides on the quality of Chinese-style meatball (kung-wan). *Meat Sci* 88(3):575–579. <https://doi.org/10.1016/j.meatsci.2011.02.018>
- Yamada EA, Alvim ID, Santucci MCC, Sgarbieri VC (2003) Composição centesimal e valor protéico de levedura residual da fermentação etanólica e de seus derivados. *Rev Nutr* 16(4):423–432. <https://doi.org/10.1590/S1415-52732003000400006>
- Yamada EA, Cipolli KMVAB, Harada MM, Sgarbieri VC (2010) Utilização de extrato de levedura (*Saccharomyces* spp.) de destilaria de álcool em salsicha. *Braz. J. Food Technol.* 13(3):197–204. <https://doi.org/10.4260/BJFT2010130300026>

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