



Article

Hybrid Meat Products: Incorporation of White Bean Flour in Lean Pork Burgers

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Abstract: The effect of partial lean pork-meat replacement by white *Phaseolus vulgaris* L. flour in hybrid burgers was studied. A multivariate regression model was used to test different bean flour levels (BF: 8–15 g/100 g) and water/bean flour ratios (W/BF: 1.2, 1.6, and 1.8 g/g). Process yield, texture profile analysis, color parameters, thermal transitions, and microstructure of the systems were analyzed. Respond Surface Methodology was used to model the response behaviors and optimization. Burgers with BF showed yields higher than 88%. Hardness and cohesiveness decreased as the BF level increased, with a more noticeable effect when the W/BF ratio became larger. Regarding color, the higher the BF and the W/BF ratio in burgers, the higher the L* obtained. The desirability optimization predicted an optimum formulation consisting of 15 g BF/100 g and 1.36 g/g W/BF with similar attributes to a commercial pork burger. The thermal analysis showed an increase in the enthalpy associated with the myosin denaturation and the interactions between meat proteins and BF led to higher temperatures for the starch gelatinization and protein denaturation. The microstructure of BF burgers presented a more stable coarse gel matrix derived from coagulated meat proteins combined with the flour components. The mathematical procedure adequately predicted the hybrid burger quality attributes.

Keywords: hybrid burgers; pulse flour; functional ingredients; alternative proteins



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1. Introduction

Meat is recognized as the highest-quality protein source and is highly appreciated for its taste. In addition, pork is the most widely eaten meat in the world, and several investigations have found potential health benefits associated with its consumption [1,2]. Nevertheless, meat products may also contain added fat, saturated fatty acids, cholesterol,

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high levels of salt, etc., causing consumers to perceive them as unhealthy foods [3]. To change this perception, different strategies for healthier product development must be promoted. Some of them could study formulation or processing modifications to avoid or restrict the presence of certain potential unhealthy compounds; others could evaluate the incorporation of ingredients that have possible health-promoting effects [4,5].

Moreover, the worldwide consumption of meat is increasing, with adverse consequences for the environment, global food security, and human health, which have triggered calls for reduced meat consumption [6]. Therefore, there is an increasing consumer awareness that linked the shift toward plant-based dietary patterns with health and environmental benefits [7,8].

In this sense, partial meat substitution through plant-based ingredients in popular meat-based products is an emerging strategy to reduce the proportion of consumed meat [9]. The concept of a hybrid meat product has appeared in this context. Traditionally, several functional ingredients (high carbohydrate and/or protein sources) have been added to processed meats as extenders, fillers, or binders. However, hybrid meat products have a favorable implication based on mixing plant and meat proteins, rather than the economic or technological reasons that has driven their inclusion in the past. The primary purpose of hybrid meat foods is to focus on a lower environmental impact, the product's health improvement, and the general idea of decreasing meat consumption [10].

Among different nutritious plants and alternative protein sources, legumes are presented as the most conventional, nutritious, and accessible protein sources [11]. Within legumes, beans are a low-priced source of a high level of protein, amino acids, carbohydrates, dietary fiber, vitamins, phenolic acids, and flavonoids [12]. In addition, consistent with Lin & Fernández-Fraguas [13], raw bean flours demonstrate better emulsifying properties than reported in lentil and chickpea flours [14] and close to bean protein isolates behavior [15]. Therefore, bean flours could be functional ingredients in the development of reformulated healthy meat products with fat replacement using pre-emulsified marine or vegetable oils, where bean components could act as emulsifiers.

This work aimed to evaluate the effect of the partial replacement of pork meat with white bean flour in hybrid burgers with pre-emulsified sunflower oil on the main quality attributes and to find the level that optimizes its characteristics. Additionally, the thermal and microstructural properties of hybrid products were evaluated.

2. Materials and Methods

2.1. Materials

Pork meat (top round cut including *Adductor femoris* and *Semimembranosus* muscles) was obtained from a local supplier. White beans (*Phaseolus vulgaris* L.) were obtained from Estación Experimental "Cerrillos" (INTA, Salta, Argentina), cleaned, and classified to remove dirt and defective beans. A cyclonic mill (Udy Corporation, Fort Collins, CO, USA) was used to ground whole bean seeds and a 1 mm stainless steel mesh to obtain the final flour. Refined high oleic acid (HO) sunflower oil (82.6 g C18:1n-9/100 g; Granix S.A., Buenos Aires, Argentina) was used as the lipid source. Cold distilled water (4 °C) and analytical-grade sodium tripolyphosphate (TPP) and NaCl (Anedra, Argentina) were used.

2.2. Experimental Design

The experimental design was developed to analyze the effect of water (W) and bean flour (BF) contents on the quality of hybrid burgers. Two BF levels (8 and 15 g/100 g) and three water/bean flour (W/BF) ratios (1.25, 1.6, and 2 g/100 g) were tested. A general constraint that all BF-burger (BF-B) formulations kept an equal amount of pork meat + BF + W was included (88.5 g/100 g; Table 1). The other ingredients (HO-sunflower oil, 10 g/100 g; NaCl, 1 g/100 g; TPP 0.5 g/100 g) were kept constant. Additionally, a control-burger formulation (C-B) was included to evaluate the effect of incorporating bean flour into the system: a laboratory-prepared burger with 78.5 g/100 g pork meat, 10 g/100 g HO-sunflower oil, 10 g/100 g water, and salts (NaCl 1 g/100 g; TPP 0.5 g/100 g).

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BF-B Formulation	Pork Meat (g/100 g)	BF (g/100 g)	W/BF Ratio
1	70.50	8	1.25
2	67.70	8	1.6
3	64.50	8	2
4	54.75	15	1.25
5	49.50	15	1.6
6	43.50	15	2

Table 1. Pork meat, bean flour (BF), and water/bean flour ratio (W/BF) to prepare 100 g of raw bean flour pork burgers (BF-B).

An available Argentinean commercial pork burger (COTO C.I.C.S.A, Buenos Aires, Argentina) was used as a target for texture and color variables in the optimization stage.

2.3. Burger Manufacture

The excess fat and connective tissue were removed from pork muscles. Pork meat passed through a grinder (80 mm diameter and 8 mm-thick plate with 21 holes of 95 mm diameter) (Meifa 32, Cimbra; Buenos Aires, Argentina). Meat portions of 500 g were vacuum-packed in bags (PO₂: 19.6 cm³/m².day.bar at 23 °C; Maraflex, Bemis., Buenos Aires, Argentina), frozen, and stored at -20 °C until used (up to 3 weeks). Before use, the meat was thawed (18 h, 4 °C).

Sunflower oil was incorporated into the burgers as a pre-emulsion using an aqueous phase with a fixed ratio of flour/water (1 g/2 mL). Emulsification was done using a handheld processor (Braun, Argentina) for 1.5 min. Lastly, the meat, emulsified oil, sodium salts, and the additional flour or water according to the formulation were mixed in a food processor (Universo, Rowenta, Germany) for 4 min. Batters were stored for 1 h at 4 $^{\circ}$ C.

Approximately 40 g (± 1 g) of the formulation was used to mold each burger (height: 1.2 cm; diameter: 5 cm). Samples were wrapped separately in polyethylene film, frozen, and stored at -20 °C until analysis (up to 21 days).

The manufacturing process of all burger formulations (nine samples/formulation) was replicated (duplicate) on 2 different days, using different batches of meat.

The cooking process was carried out using a double-sided electric household grill (Oster, Xiamen, China) at 210 $^{\circ}$ C for 3 min to assure an internal burger's temperature of 71 $^{\circ}$ C [16]. The samples were then cooled immediately at room temperature over absorbent paper.

2.4. Process Yield

Process yield was determined according to Andrés et al. [17] as the percentage of retained weight after the cooking treatment.

2.5. Texture Measurements

Cooked specimens of each formulation were analyzed following the protocol previously adopted by Argel et al. [18]. Briefly, at least 10 samples with fixed dimensions were taken from the center of the burgers (height—1.5 cm; diameter—1.7 cm) and compressed twice to 30% of their original height to perform a Texture Profile Analysis (TAXT2i Texture Analyzer, Stable Micro Systems, London, UK). Hardness, springiness, cohesiveness, chewiness, and resilience were determined using a probe with a 75 mm diameter and a speed test of 0.5 mm/s.

2.6. Color

The color was measured at room temperature on the internal surface of transversally slices of cooked burger recently cut using a Chroma Meter CR-400 colorimeter (Minolta Co., Osaka, Japan) and CIELAB parameters (L*, a*, and b*) were determined. A total of 10 measures were taken for each formulation.

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2.7. Thermal Analysis

To evaluate thermal transitions in the systems, raw and cooked burger formulations of the extreme BF or W levels (1, 3, 4, and 6; Table 1) were analyzed using a Differential Scanning Calorimeter (DSC, TA Instruments, New Castle, DE, USA). Additionally, to verify the major burger ingredients on an individual basis, bean flour and a mix of pork meat plus salt were studied.

Bean flour samples were evaluated according to Dolores-Alvarez et al. [19] and pork meat plus salt and burgers were prepared according to Marchetti et al. [20]. All samples were weighed into aluminum DSC pans and hermetically sealed, before being subjected to a heating program over a temperature range from 25 to 120 °C and a heating rate of 10 °C/min. The equipment was calibrated with indium (m.p. = 156.61 °C and $\Delta H = 28.54 \text{ J/g}$) and an empty pan was used as a reference. At least two replicates were conducted for all samples. Thermograms of different samples were obtained and data were analyzed with Universal Analysis 2000 (TA Instruments, USA).

2.8. Microstructure Observation

A scanning electron microscope (SEM, FEI Quanta 200, Hillsboro, OR, USA) was used to observe the microstructure of raw and cooked, control, and optimized burgers. Samples were taken from the center and near the surface of raw or cooked products, fixed with Carnoy fluid (60% ethyl alcohol, 30% chloroform, glacial 10% acetic acid, v/v) at 4 °C for 24 h. Then, serial dehydrations were carried out using increasing concentrations of ethanol (70%, 12 h, 95%, 2 h, 100%, 2 h), and the critical point drying technique was applied before any observation [21]. The samples were mounted on aluminum stubs using double-sided sticky tape and vacuum coated with gold film. Two replicates of each formulation were observed and at least five representative fields were obtained from each replicate.

2.9. Statistical Analysis

A second-order complete polynomial equation was used to fit the behavior of each measured variable as a function of bean flour content (C_B) and water/flour ratio (C_W), expressed as coded variables:

$$\hat{Y} = \alpha_0 + \alpha_1 C_B + \alpha_2 C_W + \alpha_{11} (C_B)^2 + \alpha_{22} (C_W)^2 + \alpha_{12} C_B C_W$$
 (1)

where \hat{Y} corresponded to each response variable, α_0 is the constant coefficient, α_1 and α_2 , corresponded to the linear terms, α_{11} and α_{22} are the quadratic coefficients, and α_{12} corresponded to the interaction term. A backward stepwise methodology was adopted to determine the significant variables (p < 0.05) of each response. After the surface responses were obtained, the "lack of fit" test and the "adequate precision" coefficient were chosen to evaluate the acceptability of the model proposed [22].

After the textural and quality attributes were regressed as a function of BF and W/BF ratio, the calculation of the optimal levels of ingredients was performed using the desirability function [23]. This optimization method incorporates desires and priorities for each of the variables, combined into an overall desirability function (D) defined as the geometric mean of each individual desirability (d_i) .

Optimization's main objective was to determine the levels of the independent variables (formulation components) that would give the best product characteristics, considering the quality parameters of the commercial product as a target.

To validate the performance of the predictive equations, an additional batch of samples with the target composition of the optimum BF-B was prepared. Process yield, color, and texture were measured and statistically compared to the predicted values (Marchetti et al., 2015).

Regression analysis, response surfaces, and the corresponding optimization were done using Design-Expert (Stat-Ease Inc., Minneapolis, MN, USA). ANOVA and pairwise comparisons for thermal analysis using Tukey's test were computed using the Infostat software. Differences in means were considered significant when p < 0.05.

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3. Results and Discussion

3.1. Physicochemical Properties

Individual predictive equations were calculated (Equation (1)) and the model results are shown in Table 2. It is important to notice that most of the proposed models showed highly significant probability values (p < 0.0001). Additionally, the adequacy of the models was corroborated with the non-significant lack of fit (p > 0.05) and the adequate precision coefficient larger than 4 (Table 2). This statistical analysis indicates that the variations observed can be well explained with the proposed equations and the model can be used to navigate the design space, considering the significant model discrimination showed by the adequate precision [22]. However, there was not a significant correlation (p > 0.05) between independent variables and the burger's redness (a^*).

Table 2. Regression coefficients of the proposed model for the variables: process yield, texture variables (hardness, cohesiveness, springiness, chewiness, resilience), and color parameters (L* and b*) expressed in terms of the coded level of the bean flour (BF) and water/bean flour ratio (W/BF) in the BF-burgers. Statistical significance of the models (P), lack of fit, and "adequate precision" coefficient, are also included.

Regression Coefficients	Process Yield (%)	Hardness (N)	Cohesiveness	Springiness	Chewiness (N)	Resilience	L*	b*
Constant	89.58	15.07	0.53	0.84	6.62	0.36	77.65	12.06
BF	-0.12	-2.18	-0.013	-0.024	-1.17	-0.023	0.31	0.39
W/BF	-1.16	-2.32	-7.90×10^{-3}	-0.025	-1.23	-0.013	0.81	-0.21
$\text{BF} \times \text{W/BF}$		-0.94	-4.41×10^{-3}				0.271	
Significance of the model (P)	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Lack of fit (P)	0.137	0.385	0.911	0.922	0.300	0.165	0.995	0.446
Adequate precision	11.57	14.46	16.49	15.30	15.05	19.70	8.04	8.08

Figure 1a–g show the representation of the predicted values of the response surface models as 3D surfaces.

All BF-B process yields were higher than 88% (Figure 1a) regardless of the flour content in the matrix. Response surface showed that process yield was mainly controlled by the W/BF ratio showing a linear coefficient one order larger than the one for BF content (-1.16 vs. -0.12). Both factors negatively affected the burger process yield, i.e., increasing moisture content led to an increase in cooking losses. Conversely, an increase in the flour level, at the same W/BF ratio, was accompanied by a decrease in moisture, and this had a direct influence on the reduction in cooking loss (higher yields). It is interesting to point out the low value of the coefficient of BF level since it shows that the partial replacement of the pork meat with the bean flour did not produce a marked decrease in the burger cooking yield.

On the other hand, C-B that comprised meat + salt + oil presented the lowest process yield (mean value 76.4 ± 0.6 %). A burger is generally composed of more or less intact meat fibers and fiber bundles, randomly distributed [24]. Meat comminuting in combination with salt addition resulted in some extraction into the water phase of myofibrillar proteins that, with the subsequent heating, ended in a complex meat gel system together with other burger components. As the C-B was prepared in an equivalent procedure to the BF-B, the higher yields (lower cooking losses) of the latter would be related not only to the ability of the extracted meat proteins to form a gel and the low shrinkage of whole or pieces of fibers but principally with the BF components, chiefly fiber and starch, either alone or in interaction with the meat protein that could form a network and prevent the liquid losses [25]. These findings confirmed our previous studies in burgers with different pulse flours [18], where pulse protein and starch could imbibe water and interact with the other matrix components, while the pulse fiber could reinforce it by hydration and increasing its viscosity.

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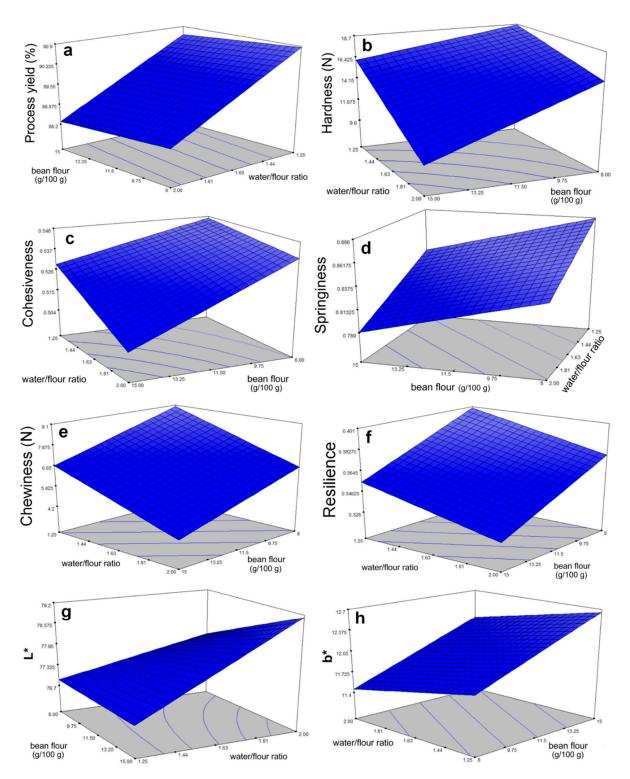


Figure 1. Contour plots of the variation of (a) process yield (g/100 g); (b) hardness (N); (c) cohesiveness; (d) springiness; (e) chewiness (N); (f) resilience; (g) lightness (L^*) ; (h) yellowness (b^*) , as a function of bean flour (BF) level (g/100 g) and water/bean flour ratio (W/BF) of BF-burgers. Darker colors indicate lower values of the modeled responses.

As can be seen in the coefficients in Table 2, the behavior of hardness and cohesiveness was controlled both by the main effects of the W/BF ratio and BF content and also by the interaction between them, with coefficients higher in magnitude for hardness. Therefore, hardness and cohesiveness response surfaces showed similar trends concerning the ingre-

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dient levels (Figure 1b,c): the 3D surfaces plot showed that both parameters decreased as BF level increased, with a more noticeable effect when the W/BF ratio became larger. This effect was previously observed when different pulse flours were used in pork burgers [18]. These results were consistent with studies that had shown that some meat substitutes resulted in minced meat products with better texture characteristics because they were dissolved in the meat protein matrix, absorbed water, and resulted in softener products [26,27]. In this sense, Baugreet et al. [28] attributed the softening of beef burgers to the rapid hydration of the lentil flour particles with low density.

When analyzing the different BF-burgers, a very good positive correlation was observed between pork meat content and hardness ($R^2 = 0.85$). However, when evaluating the hardness of the C-B, a drastic reduction in its value was observed that did not fit to previous regression for the other burgers (Figure 2a). The spatial arrangement of the fibers is of utmost importance for the textural behavior of the meat. Minced meat is constituted by disintegrated muscle bundles and fibers of different sizes. These components are arbitrarily scattered in the matrix together with cracks between them, and are held together by the extracted myofibrillar mass [24]. Thus, the differences between BF-B and C-B hardness could be attributed to a structural change in the matrix. It could be inferred that the BF components would be interacting with the meat proteins, reinforcing the meat gel.

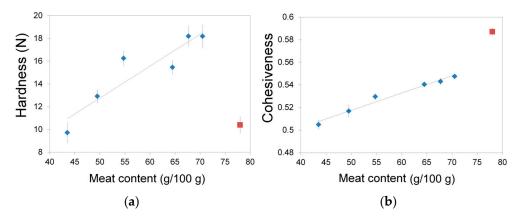


Figure 2. Burger 's hardness (**a**) and cohesiveness (**b**) variation with the pork meat content. (♦) BF-B; (■) C-B; (−) linear regression.

A similar result was obtained by Tahmasebi et al. [29], who found significantly higher values of hardness when pigeon pea flour was added to sausages, and Choi et al. [30], who reported that the addition of fiber increased the hardness in comparison to controls with only meat.

On the other hand, the cohesiveness of the C-B showed a higher value than those observed in BF-B (Figure 2b). This could imply that although the interaction between the meat proteins and the flour components could increase the hardness, it would reduce the tighter connections that occurred among meat particles, leading to a less dense and uniform structure, decreasing the burger's cohesiveness [31]. Similar results were found with rice bran fiber that significantly decreased the cohesiveness of a meat system [32] and with quinoa seed flour in goat meat nuggets [33].

Concerning chewiness, springiness, and resilience, linear models were fitted (Table 2). The three parameters were negatively correlated with BF level and W/BF ratio, but among them, chewiness showed a larger dependence on both factors.

Regarding color parameters, lightness (L^*) was dominated by the BF level, the W/BF ratio, and also the interaction between them, while for the yellowness (b^*), the interaction was not significant. As can be seen in Figure 1f, the higher the BF level and the W/BF ratio in the burger formulation, the higher the L^* obtained. This increase could be explained in terms of the lightness of white bean flour and the higher water amount in the burgers. Similar results were reported in pork sausages containing starch when the water content was increased in the formulation [34,35]. In addition, compared with the control (C-B,

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 $L^* = 66.0$), white bean flour inclusion resulted in burgers with higher lightness (L^* ranged between 77.1 and 78.3). This could also be attributed to the milky appearance imparted by the oil emulsification process which was also reported by other authors [36,37].

Concerning b*, a linear relationship for yellowness (b*) was observed for BF (positively) and W/B ratio (negatively). As it was mentioned before, it was not possible to establish a mathematical model for a*, finding an average redness value of $a^* = 3.23 \pm 0.08$.

3.2. Optimization and Validation of Pork Burger Formulation

Product optimization allows setting conditions that simultaneously satisfy the requirements placed on each of the responses and conditions. To obtain simultaneous optimization, individual parameters can be maximized, minimized, or set within limits. Particularly, for this hybrid burger system, the optimization criteria used are shown in Table 3. Bean flour level and process yield were maximized, while significant dependent factors such as texture and color parameters were fixed in the range of the values determined for commercial pork burgers. Within the studied response variables of the hybrid burgers, those that resulted significantly (p < 0.05) and with an adequate fit were selected for simultaneous optimization. Therefore, the a* parameter was not included. In addition, L* was not considered due to the corresponding value of the commercial product (70.08) being out of the observed range for the hybrid burgers (74.75–81.45). Concerning chewiness and resilience parameters, because they presented similar tendencies to hardness and springiness they were not included.

Table 3. Optimization criteria and predicted and experimental values of process yield, hardness, cohesiveness, springiness, and yellowness (b*) obtained for the optimized BF-burger.

Dagmana	Ontinination	Predicted	Experimental		
Response Variable	Optimization Criteria	Values	Mean	Confidence Interval ($\alpha = 0.05$)	
BF	Maximum	15	_	_	
Process yield (%)	Maximum	90.29	90.40 (0.5)	88.40-92.17	
Hardness (N)	12	15.23	12.5 (0.8)	10.49-19.97	
Cohesiveness	0.545	0.536	0.513 (0.004)	0.51-0.55	
Springiness	0.81	0.832	0.76 (0.01)	0.78 - 0.89	
b*	13	12.60	12.8 (0.1)	11.30-13.89	

According to the results, the optimal formulation of a BF-burger consisted of BF at $15 \, \text{g}/100 \, \text{g}$ and a W/F ratio of $1.36 \, \text{g/g}$. Individual desirability functions led to an overall D value of 0.756, which was considered satisfactory since it is close to the maximum (D = 1). It is important to point out that BF-burgers with a similar level of bean flour had been sensorial analyzed with adequate acceptability [18].

Table 3 also shows the predicted values using the mathematical model for textural and quality properties for the optimal formulation and the experimental results of the same sample prepared as an external validation of the mathematical model. Both predicted and measured results were not statistically different (p > 0.05) for all the analyzed variables. Therefore, it may be concluded that the chosen mathematical procedure adequately predicted the quality attributes of the bean flour-added pork burger.

3.3. Thermal Properties

To evaluate the effect of composition on the thermal properties of BF-burgers, thermograms corresponding to formulations in extreme BF or W/BF levels (1, 3, 4, and 6) were analyzed. All formulations exhibited five endothermic transitions without significant differences among their peak temperatures (Tp): 54.56 ± 0.08 °C; 67.3 ± 0.2 °C; 73.7 ± 0.1 °C; 80.6 ± 0.2 °C; and 96.4 ± 0.2 °C. As an example, Figure 3 shows one of the thermograms. To assign thermal transitions with the different components in the burger, differential scanning calorimetry was also performed on pork meat/salts mix and bean flour (Figure 3).

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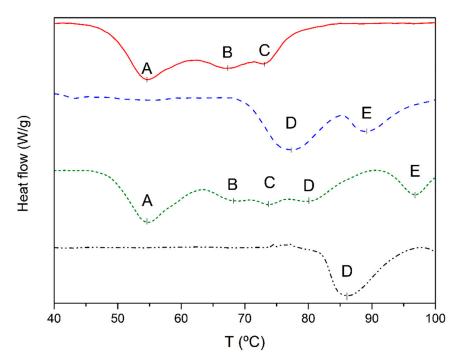


Figure 3. DSC normalized traces of pork meat/salts mix (—), bean flour (—-), and raw (******) and cooked (********) BF-burgers. A, B, C: endothermic peaks associated with thermal transitions in pork meat. D, E: endothermic peaks associated with thermal transitions in bean flour.

The three first transitions (A, B, and C in Figure 3) were also observed in the pork meat/salts mix thermogram at similar Tp (p > 0.05) and could be associated with the denaturation of different pork meat protein fractions in presence of salt [38], corresponding as follows: A to myosin (Tp = 54.14 °C), B to sarcoplasmic proteins and collagen (Tp = 66.34 °C), and C to actin (Tp = 73.42 °C). To analyze the effect of bean flour in the enthalpies associated with these thermal transitions, after peak integration, ΔH were corrected to express all per gram of pork meat. Results showed that the addition of bean flour produced a significant increase (p < 0.05) in the energy required to make myosin denaturation, which was increased when comparing the BF-burgers formulations with lower (1 and 3) and higher (4 and 6) flour levels. Enthalpies were 0.52, 0.64, and 0.95 W/g pork meat for 0, 8, and 15 g BF/100 g, respectively. The second transition enthalpy did not show significant differences and for the third peak enthalpy was difficult to measure due to a noticeable overlapping with the fourth transition.

The fourth and fifth transitions in BF-B (D and E in Figure 3) were associated with bean flour components as they were similar to those in the bean flour thermogram (located at 77.5 \pm 0.2 °C and 90.1 \pm 0.3 °C). The fourth transition (D) was attributed to starch gelatinization and the latter (E) related to protein denaturation [39,40]. The shift of these peak temperatures would be attributed to the disruption of starch crystallites that may form complexes with other molecules in the meat system [41] or to the presence of salt and the influence of a protein network system [42].

Thermal transitions in the cooked BF-B were also analyzed, both in the center and on the surface. No peaks were observed in the thermograms of the surface of the cooked BF-B (figure not shown). Conversely, thermograms of the center of cooked BF-burgers showed one transition at 85 °C (Figure 3). As the burger center reached 71 °C during the cooking process, meat proteins were almost denaturized, but the bean starch would not gelatinize and persist natively in the cooked product. Probably, as a consequence of the lower water availability or/and stronger interactions with the system, the starch gelatinization temperature was observed at higher values.

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3.4. Microstructure Observation

SEM images with different magnifications of both raw and cooked C-B and optimized BF-burger are shown in Figures 4 and 5, respectively. As can be seen at $1000 \times$ magnification the raw and cooked matrix appearances of both products were noticeably different (Figures 4a,c and 5a,c,e). In C-B, a coarse, smooth, and scaly protein matrix was observed, which was more notorious with cooking and probably related to the low yields. On the contrary, BF-B showed a stable coarse gel matrix derived from coagulated meat proteins combined with the BF components that when cooked could give a firm, viscoelastic, and smooth texture to this product, associated with higher yields and lower hardness.

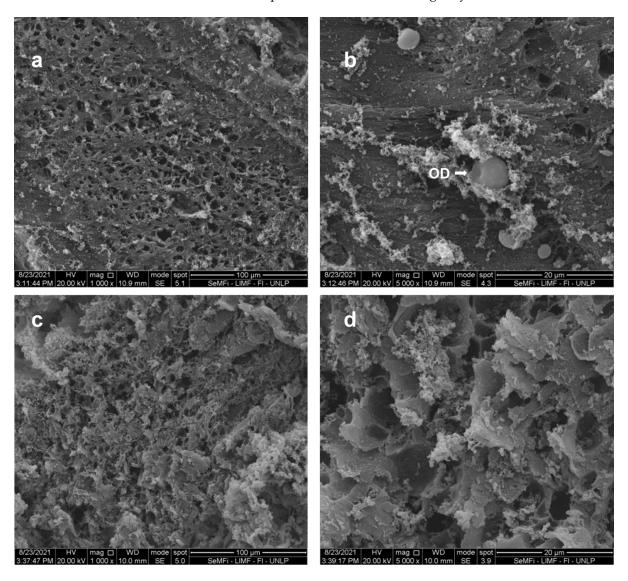


Figure 4. Scanning electron micrographs of the control burger (C-B): raw (**a**,**b**) and cooked (**c**,**d**). OD: oil droplet.

Embedded in the BF-B protein matrix, for both raw and cooked samples, some oil droplets could be found, the result of the HO–sunflower oil emulsification, and their retention by the matrix. Although in the C-B the same oil level was added, very few oil droplets were found in raw burgers and were very difficult to observe when cooked. In addition, in BF-B, a high number of starch granules could be observed that were integrated into the protein-aggregated matrix. Native starch granules with ovoid shapes could be seen in raw BF-B (Figure 5a,b). Some of those granules persisted in the cooked burger ungelatinized, retaining their appearance in the center (Figure 5c,d) but with some shape

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distortion in the burger surface (Figure 5e,f). Corresponding DSC data (Figure 3) were consistent with these facts.

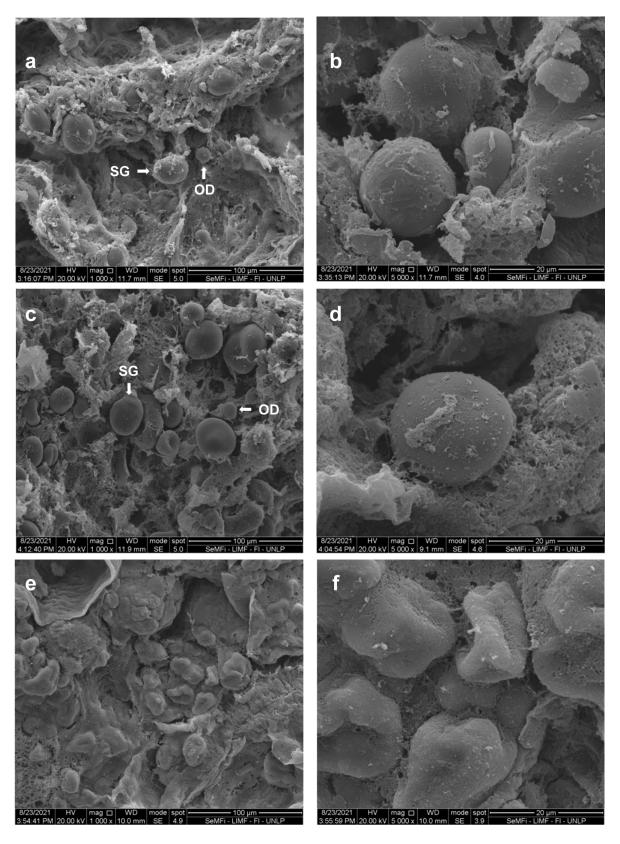


Figure 5. Scanning electron micrographs of the optimized BF-burger: raw (a,b); cooked—center (c,d) and surface (e,f). OD—oil droplet; SG—starch granule.

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4. Conclusions

This study demonstrated that through an adequately chosen mathematical procedure, the quality attributes of a low-fat pork burger with bean flour as a partial replacement of meat could be predicted and an optimal formulation could be proposed. Although the bean flour and water contents affected the product properties, the thermal analysis demonstrated that their relative levels were not the significant factor, as the bean flour added to the meat system and the possible interactions among its components with the protein meat matrix.

Both from a textural point of view and through thermal and microstructural tests it was possible to infer the existence of interactions between the bean flour and the meat proteins. The incorporation of BF led to products with higher cooking yield and greater hardness; both factors allow the incorporation of higher water content in the formulation to achieve a hybrid meat product with characteristics similar to those available on the market made with 100% pork. Further studies are necessary to characterize the nutritional and compositional characteristics of reformulated hybrid burgers, as well as carry out the sensory evaluation to corroborate the acceptance of this product by the consumer.

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