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Research Article

Strategies to Reduce Purge Losses in Meat Products Stuffed in Plastic Casings

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Two different meat emulsions were prepared with different physical stability: R_1 with 6.28 ± 1.13% total expressible fluid and R_2 with 17.7 ± 1.48%. The emulsions were placed in plastic casings at three different surface tensions (ST), expressed as contact angle, and three distinct overstuffing percentages (OS). The stuffed samples were cooked in an industrial oven. After cooling, purge losses (PL) and texture profile analysis (TPA) were measured. The reduced surface tension of the plastic casings significantly decreased the PL of both recipes. In the case of R_2, a combination of high OS and low ST was necessary to reduce PL in a 60%. In the case of TPA, OS had a statistical influence on parameters like chewiness, cohesiveness, and hardness. Plastic casings with different surface tension (to increase adherence of meat emulsion to the casing) stuffed at different levels of overstuffing percentages (to reduce free space between meat emulsion and casing) represent a potential tool to reduce PL of products based on low stability meat emulsions.

1. Introduction

Nowadays, consumers are paying more attention to the health consequences of food intake [1]. As a result, producers are opting for "clean label" alternatives that commonly attribute to food that is based on the presence or absence of particular ingredients, such as certain preservatives [2–4].

In the case of the meat industry, products like sausages are generally considered unhealthy. The excessive consumption of particular ingredients used in the production of these products produces negative impacts on health [5]. Excessive consumption of sodium is the primary cause of increased blood pressure [6] and cardiovascular disease (CVD) [7]. Moreover, a high intake of phosphates has also been shown to promote the growth and progression of lung cancer [8]. Healthier meat products which can be potentially sold in the "clean label" market could be produced by reducing the levels of sodium and phosphates [2]. However, reducing these components represents a significant technological challenge, as some of the product's functional properties can be negatively affected [9]. Reduction of sodium chloride and phosphates has a direct impact on the microstructure, stability, instrumental texture, and water-holding capacity (WHC) of meat emulsions. As a consequence of these changes a decrease in consumer acceptance and technological efficiency of the cooking process, due to an increase in cooking or purge losses, can occur [10–12].

To counteract the adverse effects of the reduction of sodium and phosphate on the functional and organoleptic properties of meat emulsions, researchers have currently been looking for alternative components which include edible seaweeds [13], microbial transglutaminase [14, 15], soy protein isolate [16], and bamboo salt [6], along with rice and potato starches [17]. Other researchers have been focused on emerging technologies including high hydrostatic pressure (HHP) processing [9] or ultrasound treatments [18] aimed at improving functional properties and minimizing cooking water losses. Furthermore, the combination of HHP with microbial transglutaminase [19] or ultrasound treatments with potassium chloride [20] have provided effective results. Nevertheless, no published research has been found that concerns the use of plastic casings to maintain quality and reduce purge losses in stuffed products filled with meat emulsion with low stability, which is the focus of this publication.

To understand the potential of these materials, it is important to know the popularity that plastic casing has nowadays. Unlike natural and other artificial coverings made from collagen, cellulose, fibrous, or textile fabric, the plastic casing can potentially reduce water losses during cooking due to low permeability. Plastic casings also enable the production of sausages with greater volume, constant diameter, and are cost-effective [21]. In addition, the different synthetic polymers used in their production (PVDC, polyester, polyethylene, polyamide, and polypropylene) make monolayer and multilayer structures possible [22] As they have low water vapor and oxygen permeabilities, stability and storage of the final product are facilitated [23]. Nevertheless, a combination of low stability emulsion (like low salt and phosphate emulsions) in a plastic casing can generate purge losses because of the incompatibility of the emulsion's WHC. The water expelled by the emulsion during cooking can get trapped between the surface and the high barrier envelope [24]. In order to study this problem properly, some researchers found it necessary to develop an emulsion model system to keep this variable controlled [25]. In the case of purge losses, a reduction of the pH of the meat is necessary to reduce its WHC [26] and enhance stuffed products' water losses during cooking which makes the final analysis of this parameter possible.

Some authors have suggested solutions for other meat products packaged in plastic materials. Goodmann et al. [27] suggested that adhesion between plastic packaging in cook-in ham is required to prevent purge. Other authors found that the surface tension of the plastic material between the casing and the meat was the principal factor to enhance this effect [28, 29]. It is possible to modify the surface tension of tubular casings to make them more compatible with the surface characteristic of the food product. This modification can be carried out by subjecting the surface of the plastic material to radiation in the presence of oxygen (like corona discharge, flame, plasma, ultraviolet, and beam radiation). As a result, oxidation of the surface of the plastic material takes place and an increase in surface tension is made possible; expressed as decreased contact angle [30, 31]. On the other hand, the reduction in "free space" between the plastic material and the meat product is studied to solve purge losses. A firmer wrap of vacuum skin pack (VSP) reduces the space between the meat and the plastic; thus, a lower percentage of the purge is collected in comparison to traditional vacuum packaging [32, 33]. In plastic casings with a good shrinkage capacity, the same effect can be reproduced during cooking when the pressure exerted by the casing can potentially reduce the risk of water loss from the meat emulsion [21].

In this context, this study aims to develop strategies to reduce purge losses of low-stability meat emulsions filled in plastic casings. The effects of the surface tension of the plastic material (to increase adherence of meat emulsion to the casing) and different levels of overstuffing percentages (to reduce free space between meat emulsion and casing) on purge losses were evaluated. Changes in the texture profile of final meat products were also analyzed.

2. Materials and Methods

2.1. Materials. Pork shoulder (fat/lean: 6.25 ± 0.35 g/100 g), in square pieces, was purchased from a local butcher (Cárnicas Kiko, Navarra, Spain) and was stored at −18°C until required for product manufacture. Additives (sodium chloride, phosphate, nitrite, and vinegar) were purchased from Pimursa (Murcia, Spain).

Multilayer plastic casings based on polyamide (BTC, θ 50 mm) were provided by Viscofan, Mexico S, RL, CV (San Luis de Potosí, México). The supplier produced three separate types of plastic material by treating each with one level of corona treatment (high-frequency electrical discharge) to modify the inner surface tension (ST) of the casing. To check the effect of corona treatment on ST, the supplier measured the contact angle (CA) of a drop of water on the plastic surface of each treated casing through image analysis. Results are shown in Table 1.

2.2. Methods

2.2.1. Preparation of Unstable Raw Meat Emulsions. Preliminary experiments were performed to develop the procedure described below to prepare unstable meat emulsions to secure high purge losses (PL) after a cooking cycle of stuffed products.

The semifrozen pork shoulder was cut up in an industrial bowl chopper with a 15 mm plate (Castellvall Industrial machinery, Girona, Spain) until meat pieces reached a homogeneous size. At the same time, brine was prepared by mixing water at 10°C (96,88%) with the rest of the additives (1.80% sodium chloride, 0.30% phosphate, 0.02% nitrite, and 1% vinegar) using an Ultra-Turrax® for 10 minutes at 70% of the mixer's top speed (10,500 rpm). Vinegar was used to achieve a final brine with a pH of 4.70, and acetic acid content of 0.04%, to minimize the level of WHC and stabilize the meat emulsion as preliminary experiments showed. After these preparations, portions of the chopped pork shoulder and brine were mixed together in distinct ratios (Table 2). One batch of each recipe was blended separately in a vacuum blend machine (Castellvall Industrial machinery, Girona, Spain) for 2 hours at 32 rpm and 0.9 bar of pressure to remove the air from the mixtures. The vacuum blend machine is provided with a water jacket system that secures that after blending meat emulsion temperature was around 3 ± 1°C.

Forty-five kilograms of each recipe were prepared and taken directly to the stuffing machine. In addition, five hundred grams of each recipe were stored after preparation to be used in different raw meat emulsion laboratory analyses.

Table 1: Surface tension of BTC subjected to distinct corona treatments.

Identification	Corona treatment (watts: W)	Contact angle* (θ_c)
CA_1	0	51
CA_2	200	26
CA_3	400	24

*Contact angle was measured between plastic casing using water like testing liquid.

Table 2: Meat emulsion recipes compositions.

Identification	Pork shoulder (%)	Brine (%)
R_1	77	23
R_2	53	47

2.2.2. Stuffing and Cooking Procedures. Stuffed samples were prepared following the schematic diagram of a split-plot design represented in Figure 1. Thus, casings were stuffed with emulsions R_1 or R_2 at three different levels of overstuffing percentage (OS) defined for the diameter of each stuffed sample (Table 3). Each combination of recipe (R_1 or R_2), overstuffing percentages (OS_1, OS_2, and OS_3), and casing type (CA_1, CA_2, or CA_3) were stuffed twice (Figure 1) independently with a Poly-clip system ICA stuffing machine (Hattersheim am Main-Germany).

After stuffing, samples were cooked for 1.5 hours in an industrial oven UKM.2001E-Mauting (Valtice-Czech Republic) with a dry bulb temperature of 80°C (heating rate of 14°C/min) and relative humidity around 90%. Finally, samples were cooled with water at approximately 10°C in the same oven for 1 hour. After this process, the final core temperature of the samples was around 30°C.

Stuffed samples ($n = 5$) of each sub-plot (18 subplots per recipe) were stored at 4°C for 24 hours until final analysis (purge losses and texture profile analysis).

2.3. Analyses

2.3.1. pH and Stability of Raw Meat Emulsions. Within the most studied parameters in raw meat emulsions, pH and stability, expressed as total expressible fluid (TEF), were analyzed in this study.

After the preparation of raw meat emulsions, pH was measured when the meat emulsion temperature reached 20–22°C. Firstly, the pH-meter (Hanna H199161) with a penetration electrode and temperature sensor (FC2023-Hanna) was calibrated using buffers pH = 4 and pH = 7. Then, the penetration electrode was introduced directly into the meat emulsion at three different points. The pH was calculated as the mean value of the three measurements.

Emulsion stability was determined according to Glorieux et al. [34] with some modifications. Raw meat emulsion (20 ± 1 g) was placed in a preweighed falcon tube (θ 3.5×10 cm) and the initial mass of the fresh sample was measured (**sf**). The sample was heated in a water bath (WNB 14-Memmert) at 80°C for 20 minutes (final core temperature around 75°C). Then, after cooling it down in a water bath (16 ± 1°C), the sample was centrifugated ($10000 \times g$, 30 min, 20–22°C, Centric MF 48-DOMEL) and the supernatant was separated from the cooked sample. The weight of the cooked sample was registered (**sc**). Total expressible fluid (TEF) was calculated by the following equation:

$$\mathbf{TEF}\,[\%] = \left(\frac{\mathrm{sf} - \mathrm{sc}}{\mathrm{sf}}\right) \mathrm{x}\,100. \quad (1)$$

Four samples were prepared for each recipe (R_1, R_2). TEF was expressed as the mean value of the four measurements.

2.3.2. Purge Losses (PL) and Texture of Cooked Samples. PL and texture of the stuffed meat samples were evaluated.

PL was calculated by first weighing the stuffed meat samples (**wi**) using a COBOS balance (model C-1500 CBJ. Max $1{,}500 \pm 0.10$ g). The purge of samples was removed by making a vent on the surface of the plastic casing using a box cutter. After removing the purge, the stuffed samples were weighed again (**wf**). PL was calculated by the following equation:

$$\mathbf{PL}\,[\%] = \left(\frac{\mathrm{wi} - \mathrm{wf}}{\mathrm{wi}}\right) \mathrm{x}\,100. \quad (2)$$

PL per each subplot was calculated as the mean of 4 samples.

The texture was analyzed by performing a Texture Profile Analysis (TPA). TPA conditions described by Pires et al. [11] were used with some modifications. One sample of each subplot was cut longitudinally into five 2 cm thick pieces and compressed axially (TA.XT.plus Texture Analyzer: stable microsystem) in two consecutive cycles of 50% of compression (speed 2 mm/s). A 75 mm diameter probe to completely cover the surface of each sample was used. Hardness (g), cohesiveness, chewiness (g), and springiness were calculated by the Software Exponent Connect (stable microsystem).

2.3.3. Statistical Analysis. Statistical analyses were made using Minitab® Statistical Software (Minitab Inc. v20).

Design of Experiments (DoE) was used with a Split-plot structure considering OS (α) as the main plot factor with three levels ($i = 6$, 14 and 22%), CA (β) as the subplot with three levels ($j = 24$, 26 and 51 θc) (Figure 1) and PL (Y) as response. To allow a correct analysis of the Split-plot design we carried out the analysis by using ANOVA (General Lineal Model). In this case, a new variable, Whole Plot (η), was defined as random variable with 6 levels (k) that correspond with 3 overstuffing treatments replicated twice. The model used could be described using the following equation:

$$Yijk = \mu + \alpha i + \eta ki + \beta j + \alpha\beta ij + \varepsilon ki. \quad (3)$$

The normality of the residuals was tested using the Anderson–Darling test. The significant differences were calculated by Tukey's test ($P < 0.05$).

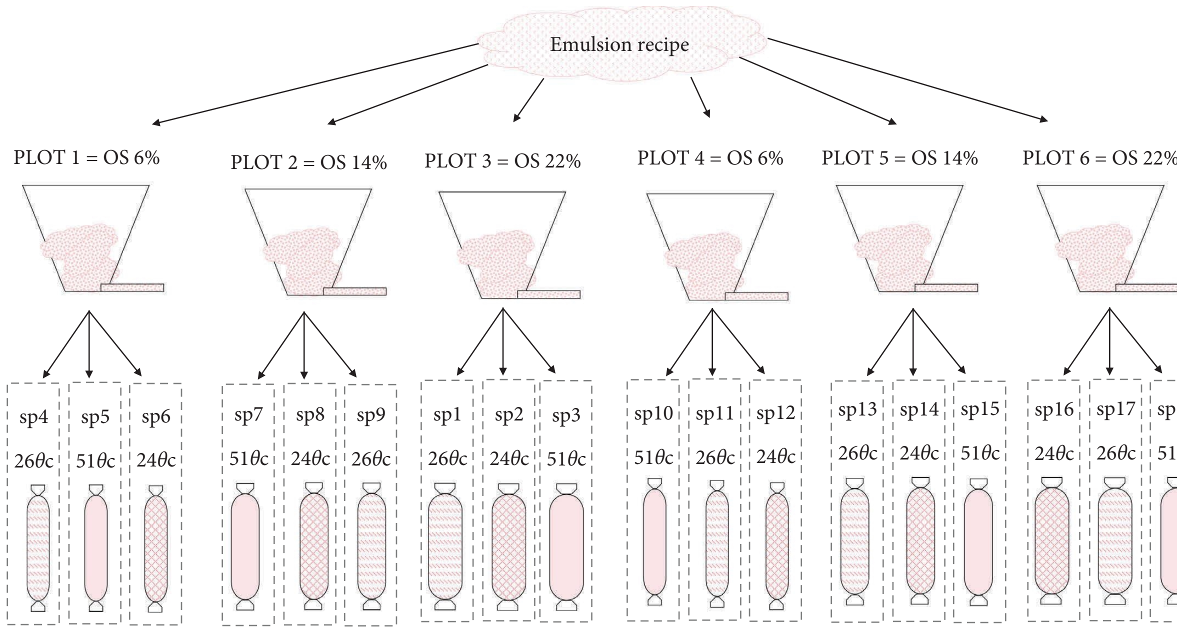

Figure 1: Schematic diagram of the experimental plan for the split-plot structure used to verify the effects of over-stuffing (OS) and contact angle (θc) of plastic casing on purge losses. (sp = subplot; $n = 5$ samples per each sp). Same diagram was used to prepare stuffed samples of meat emulsion recipes R_1 and R_2.

Table 3: Relation between over-stuffing percentage and diameter of stuffed samples.

Identification	Over-stuffing (%)	Diameter (mm)
OS_1	6	54
OS_2	14	58
OS_3	22	62

The same method was used for TPA analyses (hardness, chewiness, springiness, and cohesiveness).

3. Results and Discussions

3.1. pH and Stability in Meat Emulsions. The pH and TEF of meat emulsions R_1 and R_2 are shown in Figure 2. The pH of R_2 was lower than that of R_1 which was probably due to the addition of a higher percentage of brine (Figure 2(a)). In this case, the pH of the final meat emulsion was affected by the interaction of the brine (with $pH = 4.70 \pm 0.01$) and the buffering capacity of the raw meat used in the meat emulsion formulation. Previous studies have demonstrated that as higher amounts of acid solution are added to minced pork meat, lower pH levels are reported in the final product [35]. This lower pH tendency is due to the rapid combination of the protons in the acidic solution with the exposed meat matrix [36].

In addition, the shrinkage in myofibrillar structure caused by lowing the pH combined with the denaturation of the meat protein during cooking in a water bath has an influence on WHC and on meat emulsion stability [12, 37]. This tendency is illustrated here with the TEF values obtained from R_2 with lower pH (5.68 ± 0.02) which presented higher TEF ($17.70 \pm 1.48\%$), indicating higher emulsion instability in comparison with R_1 (pH: 6.13 ± 0.02//TEF: $6.28 \pm 1.13\%$) (Figure 2(b)). On the other hand, the percentage of meat in each recipe had an influence in the TEF values obtained. According to Carballo et al. [38], an increase in protein content causes an increase in the number of locations in the polypeptide chains of meat which are capable of interacting during heating. This makes the formation of a much more stable protein gel matrix possible while reducing the release of water and therefore lowering cooking loss values. In this case, recipe R_1 with a higher percentage of protein content (77%) presented less TEF values.

The range of TEF values reported by Glorieux et al. [34] and Pires et al. [11] in meat emulsions with reduced phosphate and salt are consistent with values obtained in this study. For this reason, both emulsions are considered appropriate analogs of low-salt and low-phosphate meat emulsions to study the effect of modified surface tension and overstuffing percentage (OS) of plastic casing on purge losses in this kind of product.

3.2. Purge Losses (PL) and TPA in Cooked Stuffed Samples. PL values for the stuffed samples containing emulsion R_1 and R_2 are shown in Figures 3(a) and 4(a), respectively. R_1 emulsion stuffed products presented lower PL values than those prepared with the R_2 recipe, regardless of casing treatment, as seen by comparing the figures. These results can be attributed to the higher TEF values and therefore higher instability in emulsions prepared with the R_2 recipe. Thus, as expected, the higher the TEF of the meat emulsion, the higher the PL values in the final stuffed products.

The effects of increasing OS and decreasing CA of the casings on the PL values are shown in Figures 3 and 4. Variation in OS did not significantly affect the PL values of

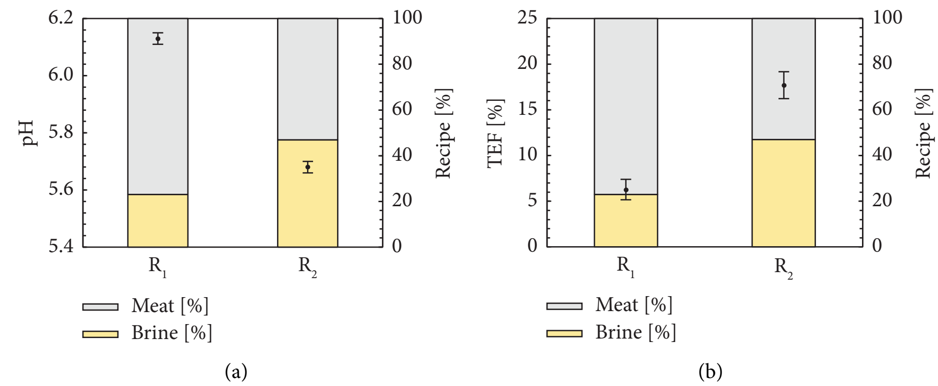

Figure 2: Mean ± SE of pH (a) and total expressible fluid (TEF) (b) of raw meat emulsions R_1 and R_2. Content of meat (grey) and brine (yellow) in each recipe are represented in column charts in secondary axis.

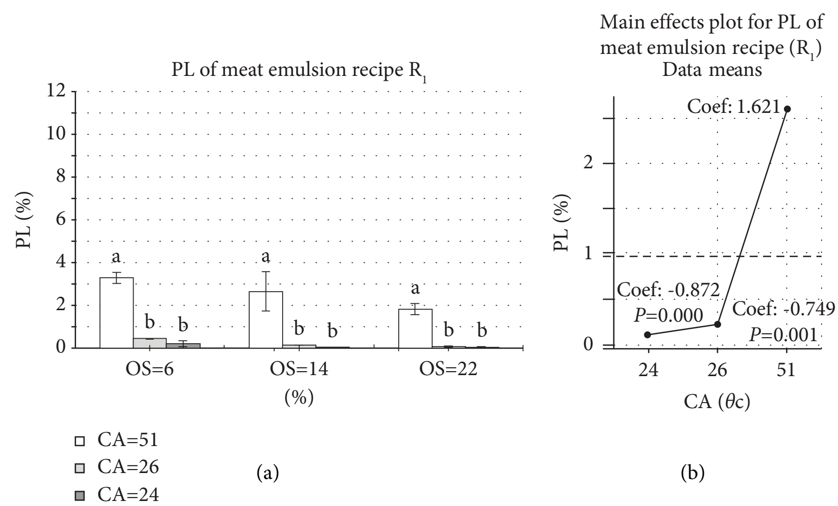

Figure 3: (a) Purge losses (PL) of stuffed samples with meat emulsion recipe R1 (TEF: 6.28 ± 1.13%). Different lowercase letters indicate a significant difference ($P < 0.05$) among casings with different CA (contact angle). (b) Main effects plot for PL of meat emulsion recipe R_1. Coefficients and P-valor included in the figure were taken of the general lineal model analyze (ANOVA). By default, minitab make comparisons with respect of extreme level of each factor (CA: 51 θc).

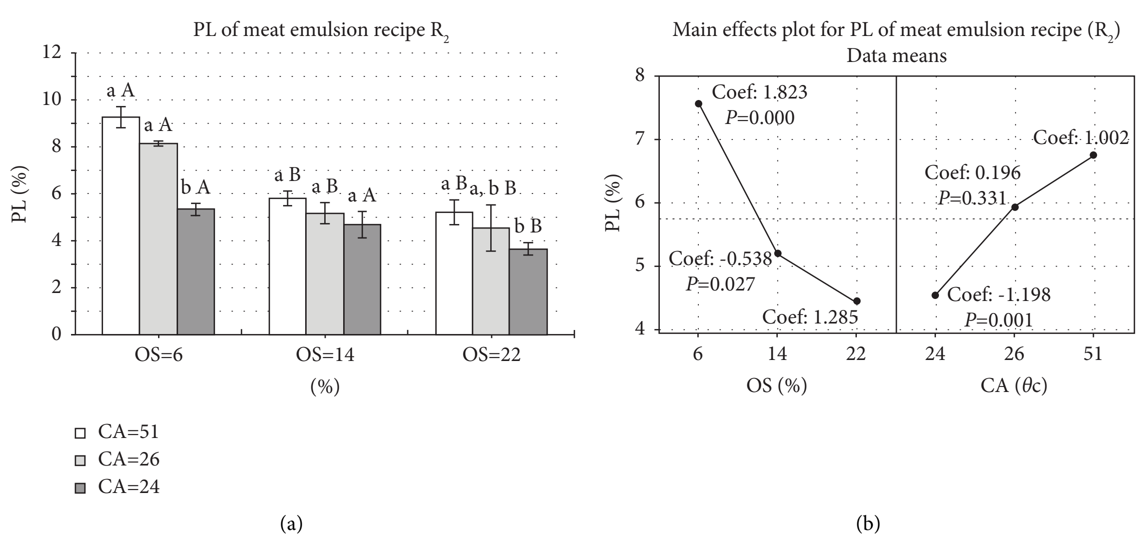

Figure 4: (a) Purge losses (PL) of stuffed samples with meat emulsion recipe R2 (TEF: 17.7 ± 1.48%). Different lowercase letters indicate a significant difference ($P < 0.05$) among casings with different CA (contact angle) and different capital letters indicate a significant difference among casings with different OS (overstuffing). (b) Main effects plot for PL in meat emulsion recipe R_2. Coefficients and P-valor included in the figure were taken of the general lineal model analyze (ANOVA). By default, minitab make comparisons with respect of extreme level of each factor (OS = 22%; CA: 51 θc).

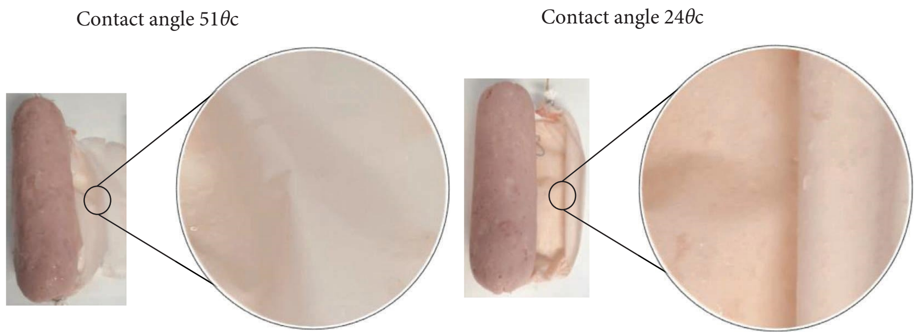

Figure 5: Comparison of meat emulsion adhesion to plastic casing with distinct surface tension. Both samples were stuffed with meat emulsion recipe R_1 at 6% of overstuffing.

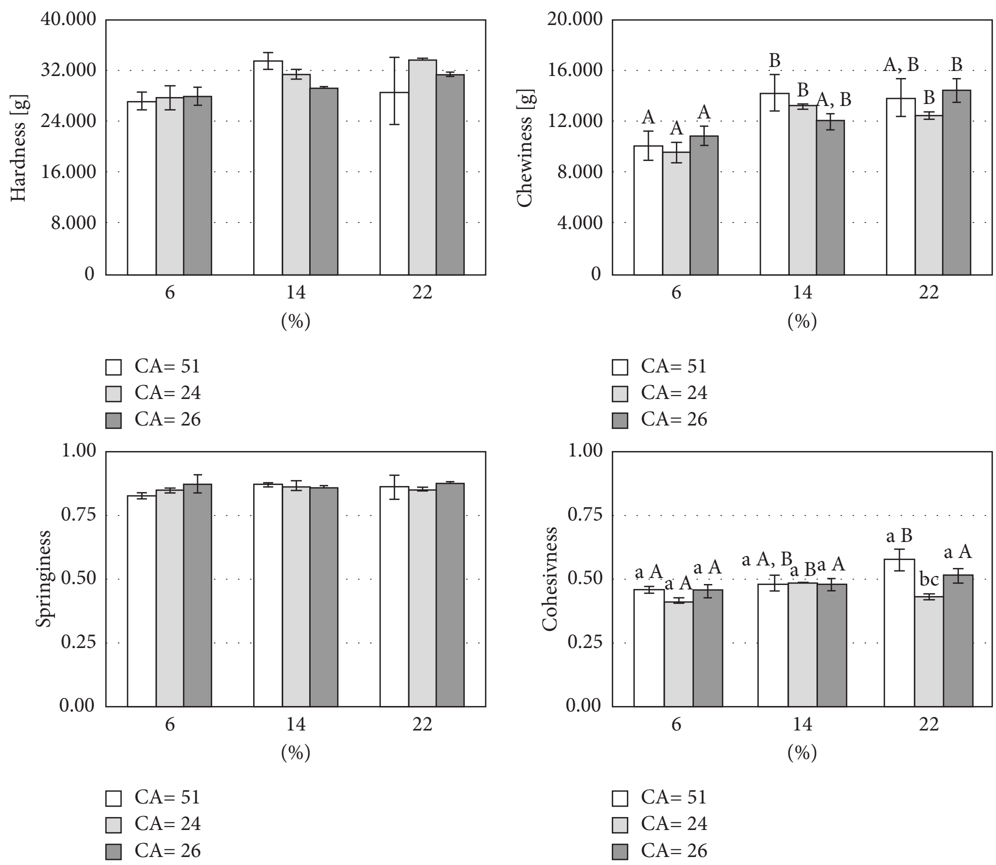

Figure 6: Texture profile analysis parameters for recipe R_1 (X-axes: overstuffing percentage). Means between each same split-plot parameters combination ± standard error. Different lowercase letters indicate that the average values differ significantly ($P < 0.05$) by CA (contact angle). Different uppercase letters indicate that the average values differ significantly ($P < 0.05$) by OS.

the stuffed samples prepared with the more stable emulsion (Supplementary material, Table 1). However, when using the less stable emulsion (R_2), a greater OS resulted in significantly lower PL values. In this case, as shown in the main effects plot for R_2 (Figure 4(b)), increasing OS from 6 to 14% was more critical than increasing it from 14 to 22% (no significant differences). Thus, for samples based on the R_2 emulsion in a plastic casing with CA = 51 θc, an OS increase from 6% to 14% resulted in 37% less PL. However, an OS increase from 14 to 22% presented only a 10% PL reduction.

In addition, a significant decrease in PL values for both studied emulsion recipes (Figures 3 and 4) were also observed as the result of decreasing the CA of the casing (that is, increasing their ST). In R_1 products, as shown in Figures 3(a) and 3(b), the reduction of CA from 51 θc to 26 θc was critical, regardless of the OS used. In this case, no significant differences were found between casings with CA equal to 26 or 24 θc. Thus, when OS was 6%, a decrease of CA from 51 θc to 26 θc resulted in a reduction of PL values from 3.30 ± 0.27 to 0.46 ± 0.02% (an 86% reduction). When using

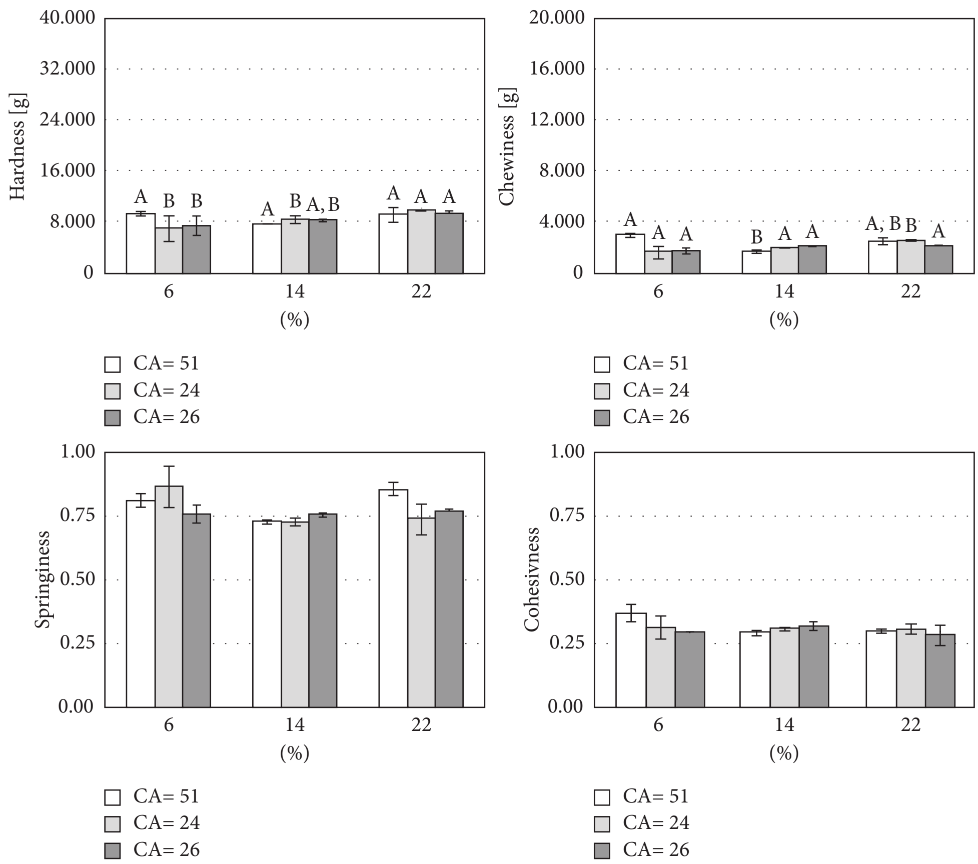

Figure 7: Texture profile analysis parameters for recipe R_2 (X-axes: overstuffing percentage). Means between each same split-plot parameters combination ± standard error. Different uppercase letters indicate that the average values differ significantly ($P < 0.05$) by OS.

casings with CA = 24 θc, an extra, but not significant, reduction of PL was observed; regardless of the OS applied (Figure 3(a)). In the case of the less stable emulsion, R_2, the CA reduction from 51 to 26 θc was as important as from 26 to 24 θc and seemed to be dependent on OS. However, no statistically significant interaction between the effects of CA and OS on PL values for R_2 emulsion products was found (Supplementary material, Table 2).

The results illustrate the importance of these two factors (OS and CA) on PL of unstable emulsions stuffed into plastic casings. When using the more stable emulsion R_1 (TEF = 6.28), reducing the CA of the casing was enough to almost eliminate completely purge loss (98% reduction). When using the less stable emulsion (TEF = 17.70), a combination of increasing OS and reducing CA brought about a dramatic reduction of PL (60% reduction). In the latter, improved results could have probably been achieved by applying even lower CA and higher OS.

A reduced CA constituted the increase in ST caused by the corona treatment on the inner surface of the casing. Increasing surface tension increased adherence between the meat emulsion and the plastic casing which presented, as a consequence, more protection against fluid loss in both studied emulsions. This observation is consistent with the research performed by Goodmann et al. [27] in which the strategy was used to reduce PL in cook-in ham. However, adherence between meat emulsion and the plastic casing must be controlled because, as can be seen in Figure 5, an excessive adherence could cause the meat to stick to the casing during product removal generating meat cling problems [37, 39].

Increasing OS resulted in a reduction of the “free space” between meat and plastic material which has been shown to reduce purge losses significantly, yet only when the most unstable emulsion was used. This effect was observed by Lagerstedt et al. [32] and Strydom and Hope-Jones [33] in VSP. Moreover, during the stuffing process, it was possible to observe that an increase in OS generated a higher stiffness on the surface of the sample likely due to the increased pressure applied by the plastic casing to the product. This fact could improve the retention of the water by the meat emulsion matrix during the cooking cycle.

The TPA values of R_1 and R_2, at distinct levels of OS and corona treatment, are shown in Figures 6 and 7, respectively. Different ranges were observed in hardness and chewiness between emulsion recipes. The highest values for hardness and chewiness were observed for recipe R_1. Thus, the higher percentage of meat in the emulsion recipe, the higher the values for these two texture parameters. Rust–Olson [40] and Pietrasik [41] suggested that increased protein content leads to a denser gel/emulsion matrix, and because of this, the hardness is higher in the higher protein-content sausages. P-values (with statistical effect) taken for ANOVA analyze for each TPA parameter (Supplementary material, Table 3) allows to determine than for R_1, OS had a significant effect on chewiness and cohesiveness, whereas CA only on cohesiveness. Significant interactions (OS $*$ CA) were found only for cohesiveness. In the case of R_2, we did not observe any significant influence of CA on TPA parameters. However, OS and the interaction of OS $*$ CA were found to have a significant effect on hardness and chewiness.

OS was the factor that had a significant effect on more TPA parameters. Parameters like chewiness, cohesiveness, and hardness presented higher values at 22% of OS. In the case of recipe R_2, higher values of chewiness could be related to the greater retention of water by the meat emulsions matrix because of the less purge values founded.

4. Conclusions

This study revealed the potential of plastic casings to reduce purge losses during the cooking of stuffed products. However, certain limits strongly influenced by the stability of the raw meat emulsion used were observed. For meat emulsion with TEF less than 7%, the increased surface tension of the plastic material was enough to reduce purge with a statistical effect ($P < 0.05$). In the case of meat emulsions with approximately 17% TEF, the combination of increased surface tension of the plastic material and a reduction of the "free space" between the raw meat emulsion and plastic casing (by an increase in an overstuffing percentage) was necessary to reduce purge losses.

In the case of TPA, the percentage of meat in the recipe played a determining role for the resulting values of hardness and chewiness. As for the two plastic casing factors evaluated, overstuffing has proven to have a greater incidence on most parameters (such as hardness, chewiness, and cohesiveness) in comparison with changes in the surface tension of the plastic material.

In the same way as emerging technologies and alternative ingredients, plastic casings with different surface tension (to increase adherence of meat emulsion to the casing) stuffed at different levels of overstuffing percentages (to reduce free space between meat emulsion and casing) represent a potential tool to reduce purge losses and improve production yields of products based on low stability meat emulsions, comparable with low-salt or low-phosphate products.

Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

Disclosure

The manuscript "Strategies to reduce purge losses in meat products stuffed in plastic casing" had been developed as part of Ph.D. thesis in Industrial modality where student, Tatiana Flores, had coworking with the University Public of Navarra and Viscofan S.A. In any part of the manuscript "Strategies to reduce purge losses in meat products stuffed in plastic casings" had been suggested that results obtained were exclusively related to specific products manufactured by Viscofan. The result shows a possible application for "plastic casings" materials in general.

Conflicts of Interest

The authors declare that there are no conflicts of interest.

Acknowledgments

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Supplementary Materials

Results of analysis of the Split-plot design by using ANOVA (General Lineal Model). OS (overstuffing) as the main plot factor with three levels (6, 14 and 22%), CA (contact angle) as the sub-plot with three levels (24, 26 and 51 θc) and purge losses as response (Tables 1 and 2). A new variable, Whole Plot, was defined as random variable with 6 levels that correspond with 3 overstuffing treatments replicated twice. Same method was used for TPA parameters analysis (Table 3). Table 1: Variance Analyze to meat emulsion recipe R_1. OS: Over-stuffing; CA: Contact angle; WP: Whole plot. Significance level was defined as $P < 0.05$. Table 2: Variance Analyze to meat emulsion recipe R_2. OS: Over-stuffing; CA: Contact angle; WP: Whole plot. Significance level was defined as $P < 0.05$. Table 3: P-values (ANOVA analyze) with statistics effects in TPA parameters. OS: over-stuffing; CA: contact angle; WP: whole plot. Each TPA parameter was analyzed independently of the rest. (*Supplementary Materials*)

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