PE8.36 Effect of Packaging Conditions on Shelf-Life of Bologna Sausages Made with Citrus Fibre Washing Water and Rosemary Essential Oil. 400.00

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Abstract— The aim of this work was to study the effect of (i) the addition of citrus fibre washing water (CFWW) obtained as ecoefficient process and rosemary essential oil (REO) and (ii) storage conditions on the chemical, microbiological and sensorial properties of bologna sausages. Traditional formula was used as control sample and experimental bolognas contained 5% CFWW and 0.02% REO. Samples were packed either in vacuum or air pouches and stored for 28 days Samples from each treatment and storage conditions were taken at 0, 6, 12, 18 and 24 days and analyzed on the same day. Lipid oxidation was assessed by TBA method, for sensory evaluation a quantitative descriptive analysis was carried out. Microbiological counts were also determined. Samples with CFWW+REO stored on vacuum packaging showed the lowest TBA values. Enterobacteriaceae and psychrotrophic bacteria were not detected in any sample. Samples with CFWW+REO stored on vacuum packaging showed the lowest counts of aerobic bacteria and lactic acid bacteria. Sensory evaluation results showed similar quality scores for samples added with CFWW+REO and stored either on air or vacuum packaging. The lowest scores were for control bolognas stored on air packaging.

I. INTRODUCTION

Everyday, the scientific literature contains new references to the beneficial effects of different ingredients and/or bioactive compounds with functional properties, and it is interesting that many of these functional ingredients are obtained from the coproducts of the agro-food industry itself. The preparation of extracts rich in dietary fibre and natural antioxidants from the co-products of the citrus processing industry could be used as a functional ingredient in, among others, the meat [1] and dairy product [2] industries. The process of obtaining the fibre from citric fruits also generates other co-products, among them the washing water used since this process uses large quantities of water, which, besides having economic importance, is important from an environmental point of view. One way of avoiding this problem would be to re-cycle the water to take advantage of the large quantity of potentially beneficial substances it contains and to re-use the water, once depurated, which would represent a saving both in economic and environmental terms. Research is currently being undertaken to improve techniques for extracting the bioactive compounds found in such coproducts [3] for use in the production of functional foods. The aim of this work was to study the effect of (i) the addition of citrus fibre washing water (CFWW) and rosemary essential oil (REO) and (ii) storage conditions on the chemical, microbiological and sensorial properties of bologna sausages. Traditional formula was used as control sample and experimental bolognas contained 5% CFWW and 0.02% REO.

II. MATERIALS AND METHODS

Sausage manufacture Bolognas were manufactured according to a traditional formula (only the meat percentages add up to 100% while the percentage of the other ingredients are related to meat): 50% lean pork meat and 50% pork backfat; 15% water (ice, w/w), 3% potato starch (w/w), 2.5% sodium chloride (w/w), 300 mg/kg sodium tripolyphosphate, 500 mg/kg sodium ascorbate, 150 mg/kg sodium nitrite, spices (0.01% black pepper, 0.005% nutmeg and 0.2% garlic powder). This original mixture was used as control sample while to assess the influence of the concentration of citrus fibre washing water and rosemary essential oil, water content were replaced in the control formula by citrus fibre washing water (5%) and rosemary essential oil (0.02%) was added. The citrus fibre washing water was obtained by the method described by Lario et al., [4] and rosemary essential oil (REO) was supplied by Ravetllat Aromatics (Barcelona, Spain).

The products were prepared in a pilot plant and followed industrial processing techniques. Frozen raw

material of animal origin, except pork backfat, was transferred to the cutter (Tecator 1094 Homogeneizer, Tekator, Höganäs, Sweden) with the sodium chloride to extract salt-soluble proteins; after comminution, the other ingredients and additives were added. Then, pork backfat, previously divided into cubes 10 x 10 x 10 cm, was added. After homogenization, the mixture was stuffed into artificial casing Fibran-Pack (Fibran, Girona, Spain) 100 x150 mm long, clipped at both ends (Polyclip system/Niedecker, Germany) and cooked in a water bath. The sausages were kept in the bath until the coldest point reached 72 °C (geometric centre of bologna, which corresponds to the thickest part of the product). A thermocouple probe (Omega Engineering, Inc., Stamford, Conn., U.S.A.) positioned in the geometric centre of the bologna was used to monitor product temperature. When the end-point temperature was achieved, the sausages were immediately chilled in ice. After reaching room temperature, the product was transferred to the lab in insulated boxes containing ice.

Storage conditions Immediately after manufacture, some slices (1.5 cm thick) were aseptically removed from each bologna sausage. The trays containing the slices were packed either in vacuum, or air pouches made of polyethylene and polyamide laminate of water vapour permeability 1.1 g/m2/24 h at 23 °C, nitrogen permeability 10 cm3/m2/24 h at 23 °C, carbon dioxide permeability 140 cm3/m2/24 h at 23 °C, and oxygen permeability 30 cm3/m2/24 h at 23 °C (Fibran, Girona, Spain). The pouches were heat-sealed and stored at $4 \pm$ 1 °C in a cabinet simulating supermarket retail conditions. The cabinet was illuminated by a standard supermarket fluorescent lamp (OSRAM, Germany). All the samples were exposed to continuous lighting at 1000 lux at the surface, measured using a luxometer Lutron LX-102 (Taiwan). The positions of the samples in the cabinet were rotated every 24 h to minimize light intensity differences and possible temperature variations on the meat surface. The packs were stored for 24 d. Samples from each treatment and storage conditions were taken at 0, 6, 12, 18, and 24 d (storage time) and analyzed on the same day.

Lipid oxidation Lipid oxidation was assessed in triplicate by the 2-thiobarbituric acid (TBA) assays following the recommendations of Buege and Aust [5]. TBARS values were calculated from a standard curve of malonaldehyde (MA) and expressed as mg MA/kg sample.

Sensory evaluation Non-trained panellists (30) were recruited from the staff and students of the Miguel Hernández University, Alicante, Spain. Panellists were chosen on the basis of previous experience in consuming traditional bolognas. Furthermore, a preparatory session was held prior to testing, so that each panel could thoroughly discuss and clarify each attribute to be evaluated in bolognas. Testing was after the panellists initiated agreed on the specifications. A Quantitative Descriptive Analysis was carried out [6]. All sensory work was carried out in the sensory laboratory at the University, which fulfils requirements according to the international standards [7, 8]. During evaluation, the panellists set in private booths under incandescent/fluorescent light, with an intensity of approximately 350 lux. Rectangular pieces approximately $1.5 \text{ cm} \times 2 \text{ cm}$ were cut from the centre of bologna slices and served at room temperature [9]. Each panellist evaluated three replicates of all the treatment groups; the sample presentation order was randomized for each panellist. Tap water was provided between samples to cleanse the palate. The sensory attributes were measured in unstructured scales with descriptors at both ends, no standards were provided. The attributes measured and their descriptors were as follows: for "external evaluation": global appearance (from conventional bologna appearance to unexpected appearance), colour (from extremely light to extremely dark), shine (from dull to bright), hue (from pale pink to brown) and homogeneity perception (from particulate to no particles observed); for odour (from imperceptible to extremely intense); for "taste": acid taste, saltiness, and fatness (from imperceptible to extremely intense); and for "texture": hardness (from extremely soft to extremely tough), juiciness (from extremely dry to extremely moist). At the end of the test, panellists were asked to give a score for product quality from 0 to 10. Microbiological analysis Samples (25 g) were excised from the interior of the sausages with a sterile scalpel and forceps and then homogeneized with sterile 1.5% peptone water in a Stomacher 400 (Colworth, London, U.K.) for 1.5 min. Total viable counts were determined on Plate Count Agar, Enterobacteriaceae using Violet Red Bile Glucose Agar (VRBGA) and Lactic acid bacteria (LAB) were counted on double layer MRS Agar at pH 5.6. In all cases, plates were incubated at 35 °C for 48 h. Psychrotrophic microbiota was determined on Plate Count Agar, and the plates were incubated at 7 °C for

10 d. Culture media were from Oxoid (Oxoid Unipath Ltd. Basingtoke, Hampshire, U.K.).

III. RESULTS AND DISCUSSION

Lipid Oxidation Figure 1 shows how lipid oxidation (TBARS) compared during the experiment in both treatments and in the different types of packaging. At day 0, the CFWW+REO samples showed lower oxidation values (P<0.05) than the control in all two types of packaging. At day 6, the lowest oxidation values recorded were in the CFWW+REO samples packed in vacuum (5.30 mgMA/kg), while no statistically significant differences were observed between the air packed CFWW+REO sample and the control sample packed under vacuum. At days 12 and 18, vacuum-packed CFWW+REO samples continued to show the lowest oxidation values, while the airpacked CFWW+REO sample showed higher (P<0.05) values than the Vacuum-packed control (6.41 and 6.18 mgMA/kg sample, respectively). At the end of the experiment (24 days), the vacuum-packed CFWW+REO treated sample showed the lowest oxidation degree (P<0.05) of all the samples, regardless of packaging type, while both the air-packed control and air-packed CFWW+REO samples showed the highest values for this parameter.



Figure 1. Evolution of TBA in bolognas (control and formulated with CFWW+REO) exposed to light and stored for 24 d.

Chopping and heating can catalyze the lipid oxidation because of disrupts cellular protective compounds contained in cell membranes such as vitamin E, electron, and hydrogen donors [10]. The lower lipid oxidation values mentioned above obtained with the CFWW+REO treatments would be due to the protective effect of the same. The agents responsible for the antioxidant activity in both orange fibre and oregano essential oil are the bioactive compounds they contain and, mainly, polyphenols. This antioxidant activity is related with the capacity of polyphenols to act as metal-chelators, free radical scavengers, hydrogen donators and inhibitors of the enzymatic systems responsible for initiating oxidation reaction. Furthermore, they can act as substrate for free radicals like superoxide or hydroxyl or intervene in propagation reactions [11].

Microbiological analysis The microbiological stability of cooked meat products depends on intrinsic factors, such as their composition, and extrinsic factors, especially the packaging and storage temperature. In our experiment, no enterobacteria nor psycotrophic bacteria were found in either of the treatments (control and CFWW+REO), regardless of packaging method or time of storage, probably due to the sodium chloride and phosphates contained in the products [12]. The growth of aerobic and lactic bacteria was greater (P<0.05) in the control samples than in those with added CFWW+REO (Figures2 and 3) at all times and all packaging conditions, except in the in CFWW+REO sample packed in air at six days, which showed higher values than the controls packed in vacuum, a difference that was maintained until the end of the experiment (24 days).



Figure 2. Evolution of counts of aerobic bacteria in bolognas (control and formulated with CFWW+REO) exposed to light and stored for 24 d.



Figure 3. Evolution of counts of lactic acid bacteria in bolognas (control and formulated with CFWW+REO) exposed to light and stored for 24 d.

In all the samples, the total aerobic bacteria and lactic acid counts at the end of the experiment were below those considered as representing a degraded product Sensorial analysis



Figure 4 shows the results of a sensorial evaluation of the different samples assayed.

For global appearance CFWW+REO vacuum packed showed the highest values while control air packed showed the lowest scores in this respect. For shine and hue CFWW+REO vacuum packed showed the highest values follow by control sample vacuum packed. In the case of odor, similar values were obtained in all samples, despite the marked odour of rosemary, this was not judged negatively by the panellists who scored the samples containing REO in a similar way or even higher than the rest of the samples. Colour homogeneity, acid taste, saltiness and fatness obtained similar values The sample containing 5% CFWW and 0.02% REO was best regarded by the panellists. Figure 4. Results of quantitative descriptive analysis carried out at day cero of bolognas (control and formulated with CFWW+REO) exposed to light.

IV. CONCLUSION

The addition of citrus fibre washing water and rosemary essential oil is a technologically viable alternative in fine paste meat products, since they improve their acceptance and have desirable effects as regards oxidative stability and reduced microbial growth, which contribute to prolonging their shelf-life. Also maintain polyphenolic compounds, such as flavonoids, which may have a beneficial effect on human health since they have been linked with the prevention of various illnesses.

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REFERENCES

[1] Fernández-López, J., Viuda-Martos, M., Sendra, E., Sayas-Barberá, E., Navarro, C., & Pérez-Alvarez, J. A. (2007). Orange fibre as potential functional ingredient for dry-cured sausages. European Food Research and Technology, 226 (1-2), 1-6.

[2] Sendra, E., Fayos, P., Lario, Y., Fernández-López, J., Sayas-Barbera, E., & Pérez-Alvarez, J. A. (2008). Incorporation of citrus fibres in fermented milk containing probiotic bacteria. Food Microbiology, 25(1), 13-21.

[3] Viuda-Martos, M., Fernández-López, J., Sayas Barberá, M. E., Pérez-Álvarez, J. A., & Sendra-Nadal, E. (2007). Aprovechamiento de co-productos de las industrias procesadoras de zumos cítricos. Alimentación, Equipos y Tecnología, 221, 64-67.

[4] Lario, Y., Sendra, E., Garciìa-Peirez, J., Fuentes, C., Sayas-Barberaì, E., Fernaindez-Loipez, J., & Peirez-Alvarez, J. A. (2004). Preparation of high dietary fiber powder from lemon juice by-products. Innovative Food Science and Emerging Technologies, 5(1), 113-117.

[5] Buege, J. A. & Aust, S. D. (1978). Microsomal lipid peroxidation. Methods in Enzymology, 52, 302-304

[6] IFT 1981. Institute of Food Technologists. Sensory evaluation guide for testing food and beverage products. Journal of Food Science, 11, 50-59

[7] ASTM. (1986). Physical requirements. Guidelines for sensory evaluation laboratories, STP 913. Philadelphia, Pa.: American Society for Testing and Materials.

 [8] ISO. 1988. International Standard 8589. Sensory analysis general guidance for the design of test rooms. Ref. nr ISO 8589:1988 (E). Genevè: International Organization for Standardization. [9] ASTM (1988). Standard and Sensory Evaluation of Materials and Products. RC Storer, editor. ASTM Manual Series. Philadelphia, Pa.: American Society for Testing and Materials.

[10] Keokamnerd, T., Acton, J. C. Han, I. Y., & Dawson, P. L. (2008). Effect of commercial rosemary oleoresin preparations on ground chicken thigh meat quality packaged in a high-oxygen atmosphere. Poultry Science, 87(1), 170-179.

 [11] Al-Mamary, M., Al-Meeri, A., & Al-Habori, M.
(2002). Antioxidant activities and total phenolics of different types of honey. Nutrition Research, 22, 1041–1047.

[12] Borch, E., Kant-Muermans, M. L., & Blixt, Y. (1996). Bacterial spoilage of meat and cured meat products. International Journal of Food Microbiology, 33, 103-120.