# MICROBIOLOGICAL CHANGES IN *MORCILLA DE BURGOS* PASTEURIZED BY HOT WATER IMMERSION

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## Introduction

Blood sausages are very traditional meat products manufactured all around the world. Recently, the increasing demand for ethnic specialties has risen up the interest for these products and the need to ensure their safety and longer shelf-life in order to expand their market place. *Morcilla de Burgos* is a typical blood sausage from Spain. It is made by a mixture of onion, rice, animal fat (mainly lard), blood and spices. All these ingredients are mixed and stuffed in natural casings and then cooked in boiling water during 45-60 min. After this cooking step they are air cooled at room temperature and usually sold without packaging in local markets and vacuum packaged in retail shops. Cooked meat products, as *morcilla de Burgos*, are liable to contamination during post-cooking handling, particularly during the chilling step, just before vacuum packaging. Shelf-life of vacuum packaged *morcilla* is around 14 to 21 days depending on the initial contamination and the storage conditions (Santos et al., 2005a). Producers are interested in increasing the shelf-life of this product in order to reach new distant markets and to reduce losses attributed to spoilage. Thermal processing has long been used to control microorganisms associated with food products. Moist heat is much more effective than dry heat in deactivating microorganisms because proteins, which may be destroyed during thermal processing, are more stable in a dry state (Hansen and Riemann, 1963). The aim of this work was to study the effect of the different pasteurization treatments on the bacterial spoilage populations of *morcilla de Burgos* during storage at 4ºC.

## Materials and Methods

Treatments: Two different and independent studies were carried out: pasteurization at 75ºC, during 10, 20 and 30 min (75P10, 75P20 and 75P30), and pasteurization at 95ºC, during 5, 10 and 15 min (95P5, 95P10 and 95P15), in both studies were included air control samples (air) and vacuum control samples (vacuum).

Samples and processing of the sausages: *morcillas* stuffed in natural beef casings were selected for the different preservation experiments. Sausage emulsion of all ingredients was stuffed into 35-45 mm natural beef casings ready to use. The blood sausages were then transferred to a cooking vessel and boiled in water at 95ºC for around 1 h (day 0). After cooking, *morcillas* were air cooled overnight at room temperature (8–10 ºC). On day 1, samples were individually packed in PA/PE bags (Cryovac Grace S.A, Sealed Air Corporation, Barcelona, Spain) for 75ºC experiments and CN300 bags (Cryovac) for 95ºC experiments and transported under refrigeration to the laboratory, where samples were divided into five batches, being one paper wrapped (air storage control), while the rest of the samples were vacuum packed. *Morcilla* were pasteurized by packages immersion at the in a water bath at treatment temperatures of 75 or 95ºC for the corresponding time. At the end of the treatment samples were immediately cooled by dipping packages into an ice water bath. During storage two packages from each treatment were randomly selected for the pH and microbiological analyses every five days from the 2nd to the 47th day and every week from 47th to 105th day, (air and vacuum *morcillas* sampling treatments were stopped when product showed evidences of spoilage).

pH measurement: pH was measured by blending 25 g of product with 225 ml of distilled water for 2 min. A digital pH-meter Micro pH 2000 (Crison, Barcelona, Spain) was used for the measurement.

Microbial analyses: 25 g slices of *morcilla* (including the skin) were taken aseptically and homogenised with 225 ml of sterile Ringer's solution (Oxoid, Basingstoke, UK). Serial decimal dilutions in sterile Ringer's solution (Oxoid) were prepared and 1 ml or 0.1 ml samples of appropriate dilutions were poured or spread onto total count and selective agar plates in duplicate. Total viable count (TVC) was determined on Plate Count Agar (Oxoid); lactic acid bacteria (LAB) on MRS Agar (Oxoid); pseudomonads on Pseudomonads Agar Base (Oxoid) with CFC supplement (Oxoid); enterobacteria on Violet Red Bile Glucose agar (Oxoid); yeasts and molds on Sabouraud Chloramphenicol Agar (Biokar Diagnostics, Beauvais, France); enterococci on Slanetz and Bartley Medium (Oxoid); Baird Parker microbiota on Baird-Parker Agar Base (Oxoid) with Egg Yolk Tellurite Emulsion (Oxoid); sulfite-reducing clostridia in tubes containing 20 ml of melted TSN Agar (Tryptone Sulfite Neomycin, Biokar). For experimental purposes, the lowest detection limit of the above techniques was $10^2$ cfu/g, except for total viable count and enterobacteria whose limit was 10 cfu/g.

Statistical analyses: Microbial and pH data were statistically analysed using ANOVA procedures. Data analyses were conducted using the statistical package Statgraphics Plus for Windows ver. 4.0.

## Results and Discussion

The initial pH average of the *morcillas* before packaging was around 6.30, the pH of vacuum and air samples decreased significantly (P<0.05) from 17th and 27th day, respectively. The pH of pasteurized samples did not show significant differences (P>0.05) during storage, except 75P10 samples which decreased from 67th day to the end of storage.

Sulfite-reducing clostridia, pathogenic staphylococci and enterococci were not detected in any sample during storage. At day 0 only TVC counts were detected mainly composed by *Bacillus* species. Before pasteurization (day 1) pseudomonads, enterobacteria, molds and yeasts counts were observed (1-3 Log cfu/g) as well as LAB, the most abundant group (3-5 Log cfu/g). After pasteurization microbial groups previously mentioned were not detected except LAB, which were observed in 75P10 samples from 22nd day to the end of study, and occasionally in 75P20, 75P30 and 95P5 and TVC (Fig. 1). LAB were also the main spoilage bacteria in vacuum packaged samples as it has been reported in several studies (Santos et al., 2005a, Santos et al., 2005b) while pseudomonads (main spoilage genera in aerobic samples), enterobacteria, moulds and yeast counts were restricted and tend to diminish during the storage of vacuum items.

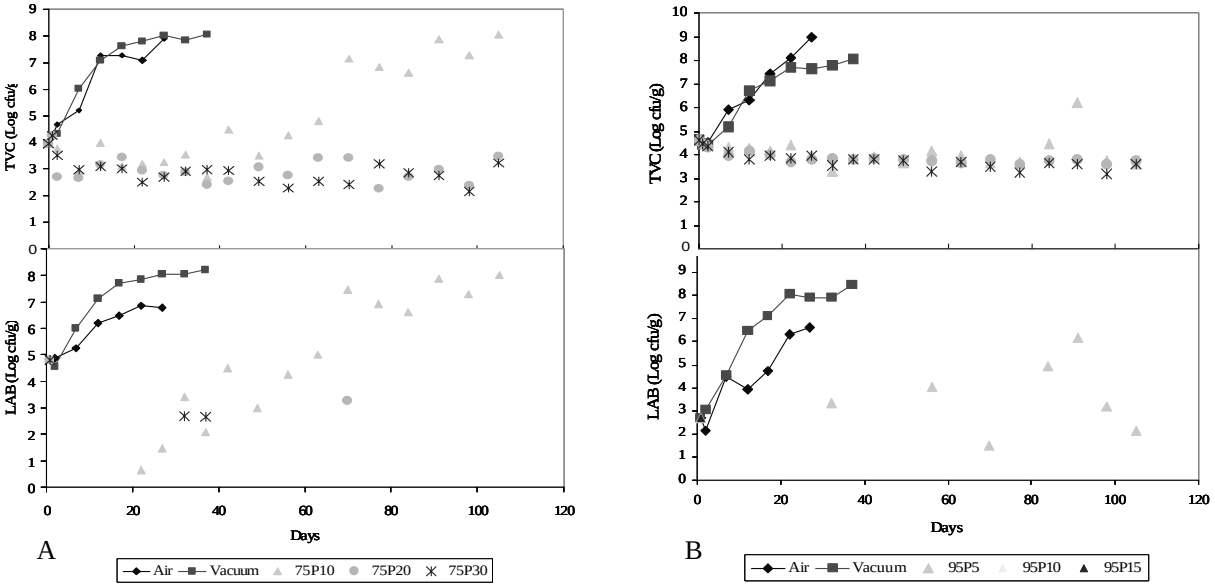


Figure 1. TVC and LAB counts evolution during storage of morcillas after pasteurization treatment at 75ºC (A) and 95ºC (B)

**Conclusions**

Pasteurization at 95ºC was the most effective treatment to reduce the spoilage bacterial in *morcilla de Burgos*, but both pasteurization treatments (75 and 95ºC) were not effective reducing TVC, presumably composed by *Bacillus* species which are thermotolerants. LAB strains can resist a mild pasteurization treatment probably to the protective effect of the rich nutrient composition of this product, which contains carbohydrates, proteins and fat.

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