

## DETERMINATION OF THE DECOMPOSITION MICROBIAL FLORA IN PORTUGUESE SMOKED DRY SAUSAGE AFTER 4 MONTHS SHELF LIFE IN M.A.P. (MODIFIED ATMOSPHERE PACKAGE)

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### Background

The consumer wishes to purchase food that is both convenient and yet appears fresh without any taste of artificial additives. However reducing the processing and use of preservatives in food may allow the growth of pathogens, especially if the food is stored incorrectly<sup>1</sup>. Modified atmosphere packaging is one of the modern techniques which provide sufficient shelf life to foods to allow their distribution, while still meeting the demands of the consumers for convenience and fresh-like quality<sup>2</sup>. With the purpose of extending shelf life there are various methods of modified atmosphere packaging that are used to alter the gaseous environment on and around foods. Three basic requirements are requested for successful application of MAP to meat products. First, is that the gas or gases surrounding the product contain at least 20% (v/v) CO<sub>2</sub> compared to the normal 0,03% found in air. The second is that the product and modified atmosphere (MA) be contained in a package, which prevents or inhibits the exchange of the gases with the exterior environment. Lastly, the storage temperature must be controlled to insure the effectiveness of the gas mixture at controlling microbial growth. Several different gas mixtures have been investigated for MAP of meats but nearly all successful tests use some combination of CO<sub>2</sub>, N<sub>2</sub>, and in a few cases O<sub>2</sub>. Of these gases, CO<sub>2</sub> is the most important because it is the most inhibitory to the growth of spoilage microorganisms<sup>3</sup>.

### Objective

To determine changes in some microbial genera of decomposition in two types of Portuguese smoked dry sausages during shelf life period in modified atmosphere package.

### Methods

Two types of Portuguese smoked dry sausage were studied after their commercial manufacture and after shelf life period. These "chouriço" contained pork (86.03%), red pepper paste (6.21%), water (3.1%), garlic paste (1.1%), salt (1.03%), olive oil (0.52%), sugar (0.21%), spices (0.17%), liquid smoke (0.1%), additives [sodium chloride containing 0.6% of sodium nitrite, E250 (1.03%) and commercial sausage sodium polyphosphate, E452(i) (0.5%)] for type "Alentejano", and pork (88.01%), red pepper paste (5.74%), water (1.27%), garlic paste (1.13%), salt (1.0%), paprika (0.5%), white wine (0.39%), sugar (0.21%), liquid smoke (0.14%), spices (0.11%), additives [sodium chloride containing 0.6% of sodium nitrite, E250 (1.0%) and commercial sausage sodium polyphosphate, E452(i) (0.5%)] for type "Ribatejano". The technological process of "chouriço" consisted in a mixture of pork meat and fat minced ( $\pm 20$  mm) with the formulation ingredients. This paste is matured for one day at 5°C. The stuffing is filled into pork gut in *Ribatejano* sausage type and into dehydrated bovine gut for *Alentejano* sausage type, subject to the drying effect of smoke and temperature. Thermal processing was divided in two phases. First phase is in industrial cooked/smoker chambers with temperature between 50 and 60°C and relative humidity and smoke production automatically controlled for 4 hours. Second phase is in a traditional smokery for 3 days. Final product is kept one day in a stabilisation room at 17-19°C with RH of 75% until packaging. The product was packed separately in modified atmosphere with 45% of CO<sub>2</sub> and 55% of N<sub>2</sub>. The material package is composed by plastic polymers laminate of several very thin polymers. It was used a Combitherm film, 70-300 mm, PA/EVOH/PE/SY - coextruded, laminate [EVOH (ethylene vinyl alcohol), PE (polyethylene) and PA (polyamide polymer)]. Shelf life period recommended was 4 months at room temperature (20 to 25°C). The experiments were conducted at a commercial meat plant and divided in four groups. The first group was accomplished in 840 kg of "chouriço" type A (AC2). The second group was performed in 560 kg of "chouriço" Type R (RC2). The third group of experiments was carried out in 840 kg of "chouriço" type A (AC1). The last experiment was made in 560 kg of "chouriço" type R (RC1). In all experiments were taken 6 samples of sausages. From these 6 samples, 3 were immediately prepared for microbial analyses and the other 3 were kept at room temperature and analysed after four months, shelf life period. The preparation involved the mix of 8.33 g of each sample. Sausage samples were not peeled to accomplish the microbial analyses. Counting of microorganisms, aiming at the technological aspects included: Total count of Mould and Yeast (CRBA, Oxoid CM549 with chlorophenicol, Oxoid SR78E - 5 days at 25°C); Total count of Lactic Acid Bacteria (MRSA, Merck 1.10660 with 2 ml/ 1000 ml of 5% solution tallium acetate and 1 ml/ 1000ml of 1% solution trifenil-tetrazolium - incubation in anaerobic jars at 30°C for 72 hours. Total count of Mesophylic Sporeformer Bacteria (After 80°C for 10 minutes the enumeration was performed in TGEA, Oxoid CM127 sealed with 0,08% of Agar, Himedia RM026 - 72 to 120 hours at 30°C. Viable counts per gram were transformed to logarithms (base 10).

### Results and Discussion

The results of microbiological analyses of Lactic Acid Bacteria (LAB), Total Mesophylic Anaerobic Count (TMAC), Total Viable Count (TVC) and viable counts of mould are presented in Figures 1-4. All the values obtained in LAB, TMAC and TVC after 4 months in modified atmosphere were inferior to those reported in the final product, in the beginning of shelf life period. Modified atmosphere (45% CO<sub>2</sub> / 55% of N<sub>2</sub>) has influence on microbial flora decrease. Modified atmosphere with CO<sub>2</sub> extend shelf life even when compared to vacuum packaging in high barrier films<sup>4</sup>. The type of packaging system greatly influenced the microorganisms present in the product. The effectiveness of CO<sub>2</sub> as a bacteriostatic is also dependent on its solubility in the aqueous portion of the food. As the storage temperature increases, solubility and thus ability of the gas to inhibit growth decreases. For this reason, MA containing elevated concentrations of CO<sub>2</sub> are more effective at low temperatures<sup>5</sup>. Several researchers have demonstrated that physiological state of the microorganisms also determines the effect of CO<sub>2</sub>. Bacteria in the lag phase of growth are especially susceptible to CO<sub>2</sub>, while those in the exponential phase are less so. The earlier the application of CO<sub>2</sub> to a food, the longer its potential shelf life<sup>6</sup>. Other factor that influenced the effectiveness of MAP is the ability of the package to contain the MA. Gases used in MAP permeate all common food packaging film by diffusion and so the shelf life is directly affected by the rate of diffusion. This fact has great influence on the survival and growth of mould and, on the basis of results shown in Fig. 4, mould counts increased during shelf life in two samples. However, the cost of the films increases with increasing barrier<sup>7</sup>. Thus, a trade off in shelf life versus cost must be

made. Higher barrier films contain the CO<sub>2</sub> longer than low barrier films, but at an increase in cost. The total amount, rather than concentration of CO<sub>2</sub> relative to the mass of food in a package, the length of time the gas is in contact with the food, the condition of the product when the CO<sub>2</sub> is applied, and the storage temperature all contribute to the effectiveness of the MA. The amount of CO<sub>2</sub> required for maximum shelf life is, in part, a function of the ratio of headspace volume to mass of product. If this ratio is low (i.e., <1) then a higher concentration of CO<sub>2</sub> may be required. If the ratio of headspace to product volume is large (i.e., >1), the concentration of CO<sub>2</sub> can be as low as 10-20% and still effective. N<sub>2</sub> is used primarily as a filler gas, to keep the package from collapsing as CO<sub>2</sub> is dissolved into the product<sup>8</sup>.

**Conclusions**

Furthermore investigation concerned with the effect of different combinations of CO<sub>2</sub>/N<sub>2</sub> in dry meat products has to be developed. Only with this data is it possible to determine the optimum concentration of CO<sub>2</sub>/N<sub>2</sub> to determine shelf life period with safety on these type of traditional meat products.

**References**

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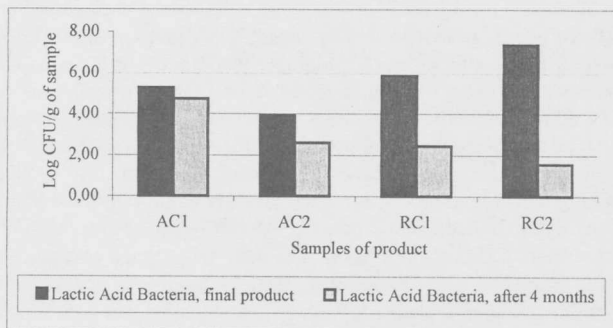


Figure 1. Total counts of Lactic Acid Bacteria.

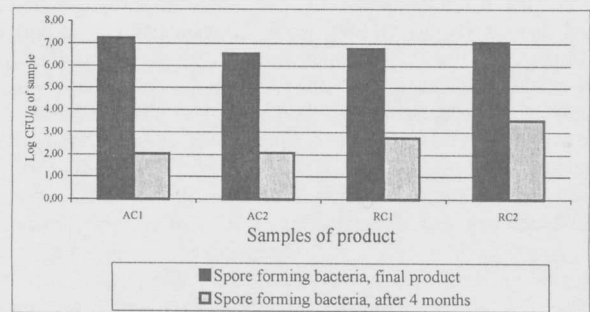


Figure 2. Total counts of spore forming bacteria.

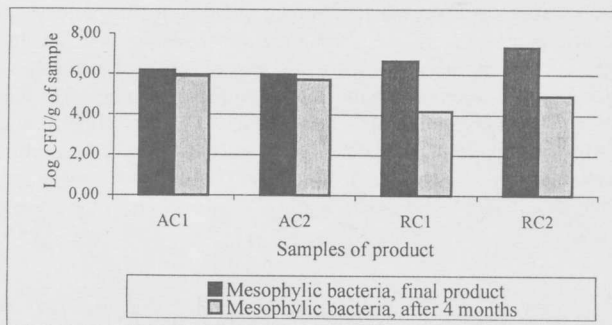


Figure 3. Total counts of mesophylic bacteria.

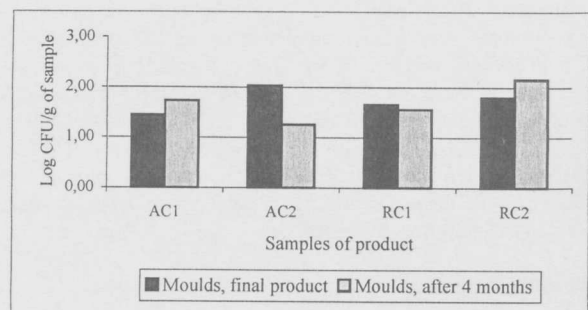


Figure 4. Mould counts.