

# MICROBIOLOGICAL QUALITY OF FRESH MEAT PRODUCTS STORED UNDER MODIFIED ATMOSPHERES

P. Tremonte<sup>1</sup>, E. Sorrentino<sup>1</sup>, M. Succi<sup>1</sup>, T. Di Renzo<sup>1</sup>, A. Reale<sup>1</sup>, G. Maiorano<sup>2</sup> and R. Coppola<sup>1</sup>

<sup>1</sup>Dipartimento di Scienze e Tecnologie Agro-Alimentari, Ambientali e Microbiologiche, Università degli Studi del Molise, Via De Sanctis, 86100 Campobasso, Italy

<sup>2</sup>Dipartimento di Scienze Animali, Vegetali e dell'Ambiente, Università degli Studi del Molise, Via De Sanctis, 86100 Campobasso, Italy

### Background

The high quality and the safety of fresh meat products are important features to be conserved during the production, the distribution and the storage.

During the last years, several studies were carried out in order to reduce the spoilage bacteria and, consequently, to extend the shelf-life of the products. Djenane *et al.* (2002) showed small but significant inhibition of the growth of psychrotrophic microorganisms with the application of rosemary extract on the surface of beef steaks, although Sanchez-Escalant *et al.* (2001) demonstrated a lack of effect of the antioxidants on microbial growth. Zhagh *et al.* (1996) reported that the storage of beef in presence of lactic acid reduces the initial number of spoilage bacteria. Moreover, the modified atmosphere packaging (MAP) is well known as a method for extending the shelf-life of meat products (Luno *et al.*, 2000; Djenane *et al.*, 2002; Jajasingh *et al.*, 2001).

In a recent study, Djenane *et al.* (2003) showed that either lactic acid bacteria and *B. termosphacta* are significantly inhibited in beef steaks packaged in  $CO_2 40\%$  and the shelf-life of these products, previously treated with lactic acid and a mixture of natural antioxidants, could be extended. Even though, the use of modified atmosphere with high  $CO_2$  and low  $O_2$  levels could cause meat bleaching (Ledward, 1970; 1984).

Jayasingh *et al.* (2001) reported that the colour stability of ground beef, packaged with CO 0.5%, was strongly improved. However, the consumers require safe and natural products (Hugas *et al.*, 2002) without additives such as preservatives and antioxidants and without the use of toxic gasses.

In this perspective, the present study aimed to compare three different gas mixtures. The screened MAP were obtained with low  $CO_2$  and high  $O_2$  concentration (MAP1), high  $CO_2$  and  $O_2$  concentration (MAP2), or high  $CO_2$  and low  $O_2$  concentration (MAP3).

## Objectives

The aim of this study was to investigate the effect of different modified atmosphere packaging on the shelflife of fresh meat products in order to harmonise the safety with the sensorial quality of the products.

#### Materials and methods

Use of modified atmosphere packaging. A mix of triturated pork lean, fat (10%), NaCl (2.5%) and black pepper (0.15%) was used to fill casing. After filling, the products were divided into four batches. The first (MAP1) was packaged in modified atmosphere containing CO<sub>2</sub> 20%, O<sub>2</sub> 70% and N<sub>2</sub> 10%; the second (MAP2) was packaged in modified atmosphere containing CO<sub>2</sub> 40% and O<sub>2</sub> 60%; the 3<sup>rd</sup> (MAP3) was packaged in modified atmosphere containing CO<sub>2</sub> 40% and N<sub>2</sub> 30%; the last batch was used as a Control (C) and packaged without gas mixtures. Each batch was stored at +4 °C for 12 day.

pH measurement. Potentiometric measurement of pH was made at 0, 3, 6 and 12 days of storage by inserting a pin electrode of a pH-meter (Crison 2001) directly into each sample. The results were expressed as the mean of four determinations performed on different parts of the samples.

Colour determination. Colour was measured on sausages at 0, 3, 6, and 12 days of storage, using the Hunter L\*, a\*, b\* system (CIE, 1978) with a reflectance spectrophotometer (Minolta CR300b).

Microbial analyses. Total and faecal coliforms were counted on VRBLA after incubation for 48 h at 37 °C and 44 °C respectively.

Total mesophylic bacteria were counted on Plate Count Agar (Oxoid) after incubation at 28°C for 48 h.



*Brochothrix thermosphacta* was enumerated on STA Agar base (Oxoid) with STA selective supplement (Oxoid) after incubation at 37°C for 48 h.

*Pseudomonas* spp. were counted on Pseudomonas Agar (Oxoid) with SR102E supplement (Oxoid) after incubation at 22°C for 24 h.

Water holding capacity, cooking loss and shear force analyses. Water holding capacity (WHC) was measured with the procedure described by Monetti (1997), and values were expressed as meat expulsed water. For cooking loss determination, sausages were weighed and cooked at 177 °C in a convection oven to an internal temperature of 74 °C. The temperature was measured with thermocouple probe inserted into the center of the sample. After cooling at room temperature, sausages were weighed, and cooking loss was determined as percentage reduction of weight.

For Warner-Bratzler shear force determination cooked sausages were cooled, wrapped in freezer paper and held overnight at 4 °C before WB shear values were obtained on four 1.27 cm-diameter cores. Each core was sheared three times at a crosshead speed of 230 mm/min. Average shear values for each sample were calculated on the basis of 12 measurements.

## **Results and discussion**

The packaging without modified atmospheres caused an increase in undesirable microorganisms producing alterations. In fact, the samples from batch C showed an appreciable increase in psycrotrophic microorganisms, such as *Pseudomonas* spp., able to degrade the proteins producing off-flavours and free amines.

*B. thermosphacta* is described by several Authors (Grau, 1980; Nissen *et al.*, 1996) as an important and undesirable microorganism in the fresh meat products. This species occurs constantly in the samples packaged without modified atmospheres and in the present work showed an increase in counts during the storage time in the Control samples. In this batch an increase of all the other microbial groups was also observed.

In this research we observed that  $CO_2$  produces an appreciable action on microbial growth in batches stored in modified atmosphere, as previously demonstrated by other Authors (Djenane *et al.*, 2003), but the effect and the entity of the antimicrobial activity are both due to the  $CO_2$  concentration and to the presence of other gasses. In fact, the batches treated with a modified atmosphere of  $CO_2$  20% (MAP1) showed microbial counts very similar to those of the Control batch (without modified atmosphere).

 $CO_2$  40% determined the best results on the assayed fresh sausages but this  $CO_2$  percentage showed different effects on microbial growth when combined with different  $O_2$  concentrations: the mixture  $CO_2$  40% and  $O_2$ 60% (MAP2) caused a sensitive inhibitory effect on microbial growth but no effect was revealed in the samples packaged with  $CO_2$  40% and  $O_2$  30% (MAP3). In fact this combination evidenced a lack of effect on microbial counts and the samples showed an increase of the altering microorganism, even though their levels were slightly lower when compared to those of MAP1 and of the Control batch. Concentration of  $CO_2$  20% (MAP1) didn't seem to cause any inhibitory effect on the spoilage bacteria.

These results confirmed that microorganisms are  $CO_2$ -sensible in presence of a high  $O_2$  concentration while they are not  $CO_2$ -sensible in a low  $O_2$  concentration (Mano *et al.*, 2000).

The gas composition of the MAP2 determined also a positive effect on the definition and maintenance of the red colour, the most important parameter for the consumer's choice. In the samples from MAP2 the high  $O_2$  concentration permitted the presence of myoglobine in oxidative state, which is responsible for the red colour, and at the same time, the high  $CO_2$  concentration assured the safety of the product. The above phenomena have not been recorded in the samples from the other batches.

In conclusion, the gas mixture of MAP2 determined a good inhibitory effect on the microorganisms, assuring the prevention of alterative factors, and positively affecting some rheological parameters.

In fact, greater WHC showed by those sausages determined less cooking loss, while the treatment didn't modify the juiciness. In addition, sausages from MAP2 showed a more reduced shear value.

## Conclusions

By the results obtained from microbiological, physical and rheological analyses it was possible to understand the playing role of the different atmosphere compositions on the microbial evolution and on the quality of sausages during the storage. In particular the use of techniques that modify in a very significant way the microbial growth in fresh sausages allowed us to assure the preservation of the most important quality parameters and the extension of the shelf-life.



## References

CIE (1978). Recommendations on uniform color spaces-color difference equations psycometric color terms. Commission international de l'Eclairage, Paris. Supplement n°2 to CIE Publication N°15 (E-1.3.1) 1971/(TC-1.3.).

Djenane, D., Sánchez-Escalante, A., Beltrán, J. A., Roncalés, P. (2002). Ability of  $\alpha$ -tocopherol, taurine and rosemary, in combination with vitamin C, to increase the oxidative stability of beef steaks packaged in modified atmosphere. *Food Chemistry*, *76*, 407-415;

Djenane, D., Sánchez-Escalante, A., Beltrán, J. A., Roncalés, P. (2003). The shelf-life of beef steaks treated with DL-lactic acid and antioxidants and stored under modified atmospheres. *Food Microbiology*, *20*, 1-7.

Grau, F. H. (1980). Inhibition of the anaerobic growth of brochotrix thermosphacta by lactic acid. *Appl. Envirn. Microbiol.* 40, 433-436.

Hugas, M., Garriga, M., Monfort, J.M. (2002). New mild technologies in meat processing: high pressure as a model technology. *Meat Science*, *62*, 359-371.

Jayasingh P., Cornforth, D.P., Carrpenter, C.E. (2001). Evaluation of carbon monoxide treatment in modified atmosphere packaging or vacuum packaging to increase color stability of fresh beef. *Meat Science*, *59*, 317-324.

Ledward, D. A. (1970). Metamyoglobin formation in beef stored in carbon dioxide enriched and oxygen depleted atmospheres. *Journal of Food Science*, *35*, 33-37.

Ledward, D. A. (1984). Haemoproteins in meat and meat products. In B.J.F. Hudson, Delevopments in food proteins – III (pp. 33-64). London: Applied Science Publishers;

Luno, M. Roncalés P., Djenane, D., Beltrán, J.A. (2000). Beef shelf life in low O<sub>2</sub> and high CO<sub>2</sub> atmospheres containing different low CO concentrations. *Meat Science*, *55*, 413-419.

Mano, S.B., Ordonez, J. A., Garcia de Fernando, G.D. (2000). Growth survival of natural flora and Aeromonas hydrophyla on refrigerated uncooked pork and turkey packaged in modified atmospheres. *Food Microbiol.* 17, 183-191.

Monetti P.G. – Appunti di suinicoltura (1997) pp. 107-108.

Nissen, H. Sorheim, O., Dainty, R. (1996). Effects of vacuum, modified atmospheres and storage temperature on the microbial flora of packaged beef. *Food Microbiol.* 13, 183-191.

Sánchez-Escalante A., Djenane D., Torrescano G., Beltrán J. A., Roncales P. (2001). The effects of ascorbic acid, taurine, carnosine and rosemary powder on colour and lipid stability of beef patties packaged in modified atmosphere. *Meat Science*, *58*, 421-429;

Zhang, Y.M., Jia, J.G., Zhou, Z.Q., Yan, R.Q., Wang H. (1996). Delevopment of preservatives for fresh pork. Meat Hyg. 8, 1-4, 9