

## MICROBIOLOGICAL PROFILE AND HAZARDS OF TRÁS-OS-MONTES

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**Introduction**

Northern Portugal is known for its wide variety of highly appreciated traditional meat products, and the Trás-os-Montes region is particularly rich in its diversity and specificity. These products were traditionally made with characteristics of aw and/or pH that ensured its stability without refrigeration. However, changes in manufacturing scale, associated to the high variety of raw materials, other than pork meat and fat, used in the preparation of some of them (viscera, blood, bones, bread, pumpkin, flour, chicken), might lead to products changes, and eventually, to hazards for the consumer. Amongst microbiological hazards usually associated to this kind of products, *Staphylococcus aureus*, *Salmonella* and *Listeria monocytogenes* are notorious. Different types of fermented sausages have been implicated in staphylococcal poisoning outbreaks (Lücke, 1985). This organism, which is salt and nitrite tolerant, is also able to grow under a wide range of environmental conditions. Given the proper conditions, therefore, its growth occurs with enterotoxin production (González - Fandos *et al.*, 1999). In 1998, data presented at the World Congress of Foodborne Diseases, refer *Salmonella spp.* as the main responsible for the outbreaks registered in Europe. In recent years, meat products have attracted increased attention as an important source of foodborne listeriosis.

**Objectives**

The main goal of this work was the evaluation of some aspects of the microbiological profile of traditional Trás-os Montes sausages, particularly those concerning hygienic quality and safety for the consumer, as a preliminary approach to the problem of microbiological hazards in this type of products.

**Methods**

161 samples of 9 different types of sausage products were analysed (10 Moura de Calda Quente; 10 Chouriço Doce; 11 Bocha; 10 Chouriço de Abóbora; 10 Chouriço Azedo; 10 Botelo; 32 Alheira; 38 Linguiça and 30 Salpicão). These products should be studied in two separated groups: those that must be heat processed before consumption, and dry meat products, such as Linguiça and Salpicão, that can be eaten without any domestic treatment. It was performed total count of mesophilic microorganisms and search for coliform bacteria, *Escherichia coli*, *S. aureus*, according to Portuguese standard methods. *Salmonella spp.*, *Listeria spp.*, *L. monocytogenes* and *Staphylococcus enterotoxin* were detected using an Enzyme Linked Fluorescent Assay in an automatic system (miniVIDAS-Biomerieux 99088), using the test sets and the preparation of the samples recommended by the manufacturer. The pH was measured directly on the paste with a crison pH meter model microph 2002. The aw was determined in a Rotronic Higrroskop DT at 25°C.

**Results and discussion**

All samples presented a high counting of mesophilic microorganisms, near 8 log c.f.u. /g. In traditional sausage products from Trás-os-Montes, no starter microorganisms are used, nor fermentable sources are added to the dough, except for some products in which bread, blood, sugar or honey are obligatory ingredients. In some earlier studies concerning traditional sausage products from Trás-os-Montes, it was observed high levels of lactic acid bacteria, and a less counting of micrococcus and staphylococcus, as well as relatively low pH values. The values of these parameters are close to those found in products previously inoculated, and could explain the results we found in mesophilic microflora counting in some of these products.

Table 1 shows the contamination levels of the samples, considering coliform bacteria, *E. coli* and microorganisms responsible for foodborne diseases. Figure 1 shows the average values found for aw and pH, for each kind of analysed product.

29% of the analysed samples showed coliform contents, indicating a deficient hygienic quality (Coliform present in 0,001g), presenting 23% evidence of a faecal contamination (*E. coli* in 0,01g of analysed product).

Considering the integral results of the analysed products, it doesn't seem to exist a constant relationship between the hygienic quality of the products and the presence (or absence) of the searched pathogenic microorganisms, except for the presence of *Salmonella*. In fact, 86% of the products contaminated by *Salmonella* showed a microbiological profile indicating a deficient hygienic quality and evidence of a faecal contamination (Coliform present in 0,001g and *E. coli* in 0,01g).

No connection was found between the presence of *L. monocytogenes* or *S. aureus* and the hygienic quality of the products. Now, one of the main food contamination sources by *S. aureus* is man, and the presence of this microorganism is usually connected to a deficient handling (Eley, 1996), and not to a deficient hygienic quality. The origin of meat products contamination by *Listeria* is sometimes difficult to identify, due to its ubiquitous character. The contamination with this microorganism is associated to water, handlers, several raw materials (particularly meat) and other ingredients. Once inside the industry, *Listeria* multiplies throughout the facilities surfaces, equipments and tools, making it possible to contaminate products at different production stages (Moreno & Garcia, 1993).

Amongst microorganisms that can cause foodborne diseases, *S. aureus* stood out for its frequent occurrence, being present in 1g in 40% of the analysed products. 126 of the samples monitored on the search for *S. aureus*, were also subject to search for stafilococcic toxin. Results show, as expected, that the presence of the microorganism alone doesn't necessarily imply the existence of an effective risk to the consumer's health. Despite 42% of these samples showed *S. aureus*, only 5% showed positive on the search for stafilococcic toxin. Such results can be due to the fact that aw in these products is low enough to inhibit the production of stafilococcic enterotoxin. (Bergdoll, 1989). The opposite was also observed, i.e., the absence of *S. aureus* doesn't necessarily imply there is no danger for the consumers health. 5 positive samples at the search for stafilococcic toxin were free of *S. aureus*. These samples of dry products (ready to eat), have a smoking/drying stage during the processing that might contribute to inactivate the microorganism, without the inactivation of its toxin.

The potential hazard represented by the presence of staphylococcal toxin, ready to eat products are apparently the most hazardous; 82% (9 in 11) of the samples with detectable toxin are from this group. 13% of the ready to eat products presented *Listeria*, 5% of them were *L. monocytogenes*. *Salmonella* was only present in 3% of the ready to eat products. According to criteria defined by Leistner and Rodel (1976), in these products the aw alone is low enough ( $aw \leq 0,91$ ) (Fig. 1), to ensure its stability. In the products that need cooking before consumption, *Listeria* was found in 10%, *L. monocytogenes* in only 3%, and *Salmonella* in 6%. The hazard that these products might represent is conditioned by thermal processing that occurs during the domestic preparation.

### Conclusions

Considering the integral results of the analysed products, it doesn't seem to exist a constant relation between the products hygienic quality and the presence - or absence - of the searched pathogenic microorganisms. Apparently, ready to eat products deserve special attention concerning the danger of causing foodborne diseases. Between the 11 samples positive at the staphylococcal toxin, 9 belonged to ready to eat products. From the 5 samples presenting *Listeria monocytogenes*, 3 belonged to this group of products. This shows the importance of eliminating and/or reducing the level of these pathogenic agents in food products, specially in ready to eat products. The increase of foodborne diseases recently verified results namely from the lack of hygienic-sanitary conditions at the places where food products are prepared, and from the deficient formation of the handling personnel. Foodborne diseases have serious consequences, both in public health and in general economy. So, control rules should be applied. Proper education of food industry handlers, adequate production techniques, and the implementation of HACCP systems, are examples of measures that might eliminate and/or reduce to an acceptable level the occurrence of these pathogenic agents in sausage products.

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### Acknowledgements.

This work was supported by PAMAF - 3056.

Table 1 - Results of the search for microorganisms related to hygienic quality and responsible for foodborne diseases (number and % of samples where the microorganism was found, maxim dilution or dilution interval in which it was found).

	N° of analysed samples	Present in 25g	Absent in 1g	Present 1-0,1g	Present 0.01g	Present $\geq 0.001$ g
Coliforms	161		31 (19%)	57 (35%)	26 (16%)	47 (29%)
<i>E. coli</i>	161		90 (56%)	34 (21%)	15 ( 9%)	22 (14%)
<i>S. aureus</i>	160		96 (60%)	25 (16%)	39 (24%)	
<i>Salmonella</i>	147	7 (5%)				
<i>Listeria spp.</i>	147	17 (12%)				
<i>L. monocytogenes</i>	147	5 (3%)				

Figure 1 - Average values of aw and pH of different types of analysed products

