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MICROBIOLOGICAL CHARACTERISTICS OF A TYPICAL ITALIAN SALAME PRODUCED WITH POTATOES

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Abstract

Italy has an ancient sausage tradition; in recent years there has been an increasing interest in the production of "salami", typical Italian products generally made of pork and spices, manufactured by artisanal methods.

The present research concerns "Salame di patate", a typical product coming from a small area in Piedmont (Northwest Italy), made of boiled potatoes and pork.

The aim of the research was the microbiological characterization of this product as regards lactic acid bacteria and *Micrococcaceae*.

Lactic acid bacteria represented the predominant bacterial group and the strains isolated were biochemically characterized; it was also carried out the genetic identification of few isolates.

Keywords: Lactic acid bacteria, microbiological characterization, *Micrococcaceae*, salami, salame di patate.

Résumé

En Italie il y a une ancienne tradition de saucissons; dans les derniers ans il y a eu une augmentation d'intérêt vers la production artisanale des "saucissons", typiques produits italiens constitués de viande de cochon et épices.

La présente recherche concerne les "Salame di patate", un saucisson fait de viande de cochon et des pommes de terre, produit dans une petite zone du Piémont (nord- Ouest Italie).

Le but de cette recherche a été la caractérisation microbiologique de ce produit avec considération particulière pour les bactéries lactiques et les *Micrococcaceae*.

Les bactéries lactiques ont représenté le groupe bactérien prédominant et les souches isolées ont été caractérisées biochimiquement; on a même entrepris l'identification génétique de quelques flots.

Mots clés: bactéries lactiques, caractérisation microbiologique, *Micrococcaceae*, saucissons, salame di patate.

Auszug

Italien hat eine sehr alte Salami-tradition, in den letzten Jahren ist die Interesse für die handwerkliche Produktion von typischen italienischen Produkte mit Schweinefleisch und Gewürze grösser geworden.

Objekt dieser Forschung ist der "Salame di patate", eine Wurst aus Schweinefleisch und Kartoffeln, der in einer kleinen Zone von Piedmont (Norden-Westen Italien) hergestellt worden ist.

Zweck dieser Forschung ist die mikrobiologische Charakterisierung dieses Produktes mit besonderem Interesse für Milchbakterien und alle *Micrococcaceae*.

Milchbakterien sind die wichtigere bakterielle Gruppe und die isolierten Stämme wurden biochemischerweise charakterisiert; es wurde auch die genetische Identifizierung von einigen isolierten gemacht.

Schlüsselwörter: Milchbakterien, mikrobiologische Charakterisierung, *Micrococcaceae*, salami, salame di patate.

1. Introduction

In recent years, according to the growing interest in local gastronomic traditions, many studies have been carried out on microbiological characterization of different Italian sausages (Cantoni and Comi, 1986; Campanini *et al.*, 1987; Pirone and Manganelli, 1990; Torriani *et al.*, 1994; Coppola *et al.*, 1995; Coppola *et al.*, 1997). Piedmont (Northwest Italy) has an ancient sausage tradition; more than 70 traditional products composed of meat have been registered in this area; in particular there is an increasing interest in "salami", typical Italian products generally made of pork and spices, manufactured by artisan methods.

The present research concerns "Salame di patate", a typical product coming from Canavese, a restrict area in Piedmont. The historical information about this product are almost absent and exclusively based on oral tradition.

The mixture of "Salame di patate" is composed of boiled potatoes, lean pork, lard or bacon coming from other productions; the percentage in composition varies according to producer although generally pork and potatoes are 1:1 present.

It must be underlined the use of additives added following a secret recipe varying according to every producer; salt, white and black pepper, garlic, nutmeg and red wine are the most important.

The "Salame di patate" is sold as raw sausage to be eaten within 2-3 days after manufacturing or subsequently a short ripening.

By tradition, the "Salame di patate" is typically produced in autumn and winter when the farm-workers slaughter pork and it originated from the need of poor people to transform not valuable pieces of pork in a pleasant and nourishing food.

At present the production of this sausage is quite widespread in Piedmont where it is considered a traditional product to be protected

and to be attributed a protected designation origin (PDO).

The aim of the present research was the microbiological characterization of fresh "Salame di patate" as regards lactic acid bacteria which are important in the technology of sausages.

2. Materials and methods

Samples of "Salame di patate"

20 samples of "Salame di patate" have been analysed. They have been produced in the area of Canavese (Northwest Italy) by 20 different producers totally representing the whole output.

Microbiological analysis

"Salame di patate" samples have been analysed within 3 days from their production.

To count microorganisms, 25g of each sample were added to 250mL of sterile saline solution and homogenized in Stomacher. Serial dilutions were prepared and plated in the following media: plate count agar (PCA), incubated at 37°C for 48h for total microbial load, MRS agar at pH 5.8, incubated anaerobically (Gas-Pak system) at 37°C for 48h and mannitol salt agar (MSA), incubated at 30°C for 48h.

Finally in order to evaluate the hygienic-sanitary safety of "Salame di patate", coliform growth, particularly *E. Coli*, has been appreciated by Petrifilm.

Characterization and genetic identification of lactic acid bacteria

After incubation, randomly selected colonies were purified by two subsequent subcultures and then submitted to microscopic examination, Gram staining, catalase test, production of gas from D-glucose, growth at 15°C and 45°C.

Biochemical profile of lactic acid bacteria isolates has been determined by API 50 CH galleries (Biomérieux).

Finally genetic identification of few isolates was carried out by molecular techniques based on PCR.

The protocol described by Chagnaud *et al.* (2001) was used in order to genetically identify isolates belonging to *Lactobacillus fermentum* species, while the method suggested by Mora *et al.* (1997) was employed to identify *Pediococcus pentosaceus* isolates.

3. Results and discussions

The microbial loads found in 20 samples of "Salame di Patate" analysed are shown in figure 1; the minimal value of total microbial load was 3.1×10^3 cfu/mL, the maximum value 3.2×10^7 cfu/mL.

The bacteria group showing the highest load values was lactic acid bacteria with values between 1.0×10^2 cfu/mL and 2.7×10^7 cfu/mL; *Micrococcaceae* had lower loads ranging from a minimal value of 1.0×10^2 cfu/mL to a maximal value of 8.2×10^5 cfu/mL.

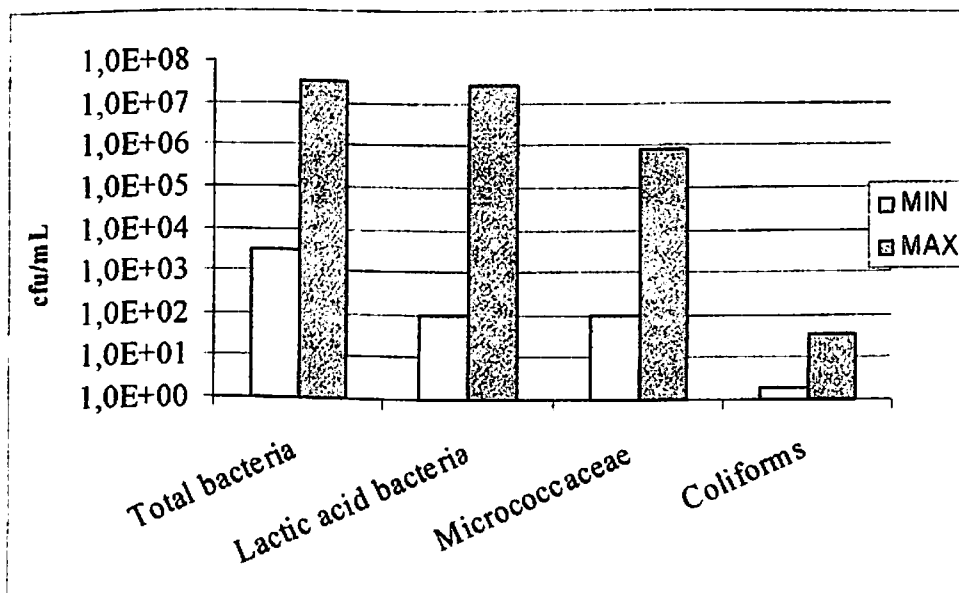


Fig.1. Minimal and maximal values of cfu/mL of total bacteria, lactic acid bacteria, *Micrococcaceae* and coliforms present in the 20 "Salame di patate" samples.

Coliforms reached the maximum load value of 3.4×10^1 cfu/mL and no colonies of *E. coli* were isolated.

At the moment 16 lactic strains have been biochemically characterized; 8 species belonging to the genera *Pediococcus*, *Leuconostoc*, *Lactobacillus* and *Lactococcus* have been identified.

Among the strains characterized, *Leuconostoc mesenteroides mesenteroides* showed the highest frequency value of 31.3% (figure 2).

The genetical identification confirmed the results obtained by the biochemical characterization of the species *Pediococcus pentosaceus* and *Lactobacillus fermentum*. In figure 3 the amplification band of 889bp specific for *Lactobacillus fermentum* species is showed.

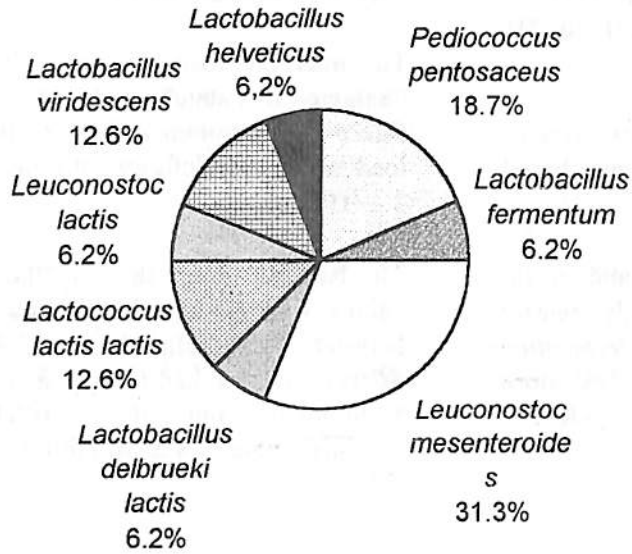


Fig.2. Percentage of frequency of 8 lactic acid bacteria species among the 16 characterized isolates.

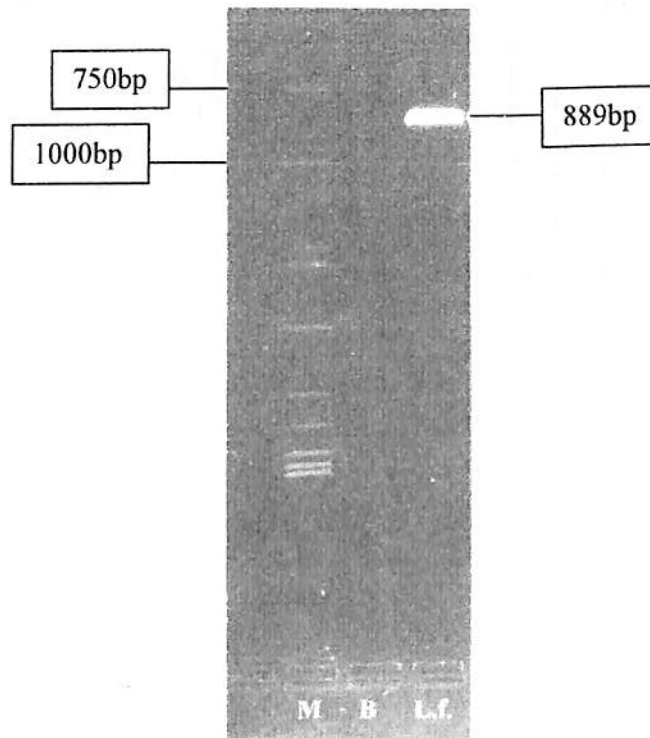


Fig.3. PCR profile of *Lactobacillus fermentum* strain by species-specific primers Ferml-Lowlac. M: marker; B: blank; L.f.: *Lactobacillus fermentum*.

4. Conclusions

The microbiological characterization of fresh "Salame di patate" samples showed the predominant presence of lactic acid bacteria;

Micrococcaceae with good load values were present.

It must be noticed a good variability among lactic bacteria isolates analysed; 8 species were represented among 16 strains. *Leuconostoc*

mesenteroides mesenteroides showed the highest values of frequency; *Pediococcus pentosaceus*, *Lactococcus lactis lactis* and *Lactobacillus viridescens* were quite present while only one isolate was found for each of the following species: *Lactobacillus delbrueckii lactis*, *Lactobacillus fermentum*, *Lactobacillus helveticus* and *Leuconostoc lactis*.

Many authors refer the presence of these species of lactic acid bacteria in sausages (Sakhare *et al.*, 2003; Senne *et al.*, 2003; Tyopponen *et al.*, 2003; El-Fadaly *et al.*, 2002; Liu and He, 2002; Borch *et al.*, 1988).

Lactic acid bacteria, present in "Salame di patate", seem to be important for ripening of the product; they probably contribute to the sensory characteristics of the ripened sausage. According to these observations it will be interesting to state lactic bacteria load of the product at different stages of ripening process.

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