CHARACTERIZATION OF LACTIC ACID BACTERIA USED IN COMMERCIAL MEAT STARTER CULTURES: STRAIN IDENTIFICATION AND EXOCELLULAR POLYSACCHARIDE PRODUCTION

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INTRODUCTION

Meat fermentation process is the result of microbial activity and involve 3 distinct steps: acidification, maturation and drying. Addition of starter cultures is nowadays a common practice in meat fermentation industry and tended to give products a greater uniformity from batch to batch (Schillinger and Lücke, 1991). Starter cultures used in fermented meat technology may include bacteria (Lactic acid bacteria and *Micrococcaceae*), yeasts and moulds. Lactic acid bacteria (LAB) are responsible for the meat acidification which is important for product preservation, and control of spoilage and pathogenic bacteria. They could also produce waste products (lactic acid, exocellular polysaccharide, biogenic amines, etc…) which can act more or less positively on the final product.

Exocellular polysaccharides (EPS) are polysaccharides synthesized by bacteria and are excreted outside the cell wall. EPS can either remain associated with the cell surface as a capsule or be secreted into the environment (Cerning, 1990). Slime production and/or the presence of a ropy texture caused by LAB in meat products (Korkeala et al., 1988) is an interesting spoilage phenomenon and of considering economic importance. Ropy slime production is often associated to *Lactobacillus sake* strains which are also used in some commercial starter culture preparations (Hammes et al., 1990). This species is considered as a major spoilage bacteria in vacuum-packaged cooked meat products (Mäkelä et al., 1992; Holley et al., 1996). Cooked meat products and fermented sausages are commonly handled in the same rooms in meat processing plants. Fermented sausages could then contribute to contamination by ropy slime-producing LAB as mentioned by Mäkelä (1992).

Numerous studies have been done on slime producing bacteria contaminating meat products but only few on the impact of meat starter culture strains as spoiling agents. Therefore, the objective of this study was to isolate and identify LAB strains used in commercial meat starter preparations and to test their ability to produce ropy slime under various processing conditions.

MATERIALS AND METHODS

BACTERIAL STRAINS. Commercial meat starter cultures were obtained from 14 different manufacturers. Sixty seven LAB strains were isolated as pure culture and were subsequently identified using API rapid 50 CH tracks (Bio-Merieux, France). Stock cultures were maintained at 4°C on slants of Man, de Rogosa & Sharp (MRS) medium.

EXOCELLULAR POLYSACCHARIDE PRODUCTION. To reproduce final fermentation parameters observed in North America and in Europe, NaCl (3%) was added to MRS broth and pH was adjusted to 5.2 (North America) and 5.5 (Europe). Bacterial strains were first subcultured from colonies grown on MRS plates. Subcultures were done in duplicate and were incubated at 20°C for 24 hrs. Before use, strains were resubcultured at 0.1% in fresh medium and suspensions were reincubated in the same conditions. EPS production was evaluated by ethanol precipitation (Otsuji et al., 1994). Three volumes of cold ethanol (4°C) 95% were added to each sample. Suspensions were mixed by inversion and kept at room temperature for 30 min. Precipitates were easily detected when compared to uninoculated samples used as negative control.

RESULTS AND DISCUSSION

Ninety three commercial meat starter cultures are currently available on the market from 14 different manufacturers. Identification results (figure 1) performed at the Food Research and Development Centre (FRDC) indicate that the 67 different LAB strains isolated from commercial cultures belonged to one of the 5 following species: *Lactobacillus plantarum* (10 strains), *Lactobacillus sake* (5 strains), *Lactobacillus sp.* (7 strains), *Pediococcus acidilactici* (14 strains) and *Pediococcus pentosaceus* (30 strains). One strain of *Lactococcus lactis* subsp. *lactis* was also found. Data showed that strain identification provided by commercial meat starter manufacturers is often inadequate or obsolete.

The percentages of LAB strains producing EPS under different processing conditions are shown in table 1. Experimental data suggests that among commercial strains tested, *Pediococcus pentosaceus* was the only one producing EPS. Precipitate formation was highly reproducible within repetitions under the same growth conditions. Furthermore, the same *Pediococcus pentosaceus* strains were found to produce EPS under both processing conditions tested. Under the defined maturation conditions used, representing the fermentation processes in North America or in Europe, EPS production was only observed with *Pediococcus pentosaceus* where 26.7% of the strains tested were positive for both cases. Surprisingly, no Lactobacilli strains were producing EPS under those conditions. Expression of the EPS phenotype appears to be highly linked to environmental conditions such as temperature, pH, salt concentration, etc… (Mäkelä and Korkeala, 1992). It is therefore possible that under those defined maturation conditions, which varied only at the pH level, the Lactobacilli strains tested were unable to express their EPS production traits. Modifying some of

those environmental parameters, one by one, should allow a complete identification of the EPS-producing strains among the commercial meat starters.

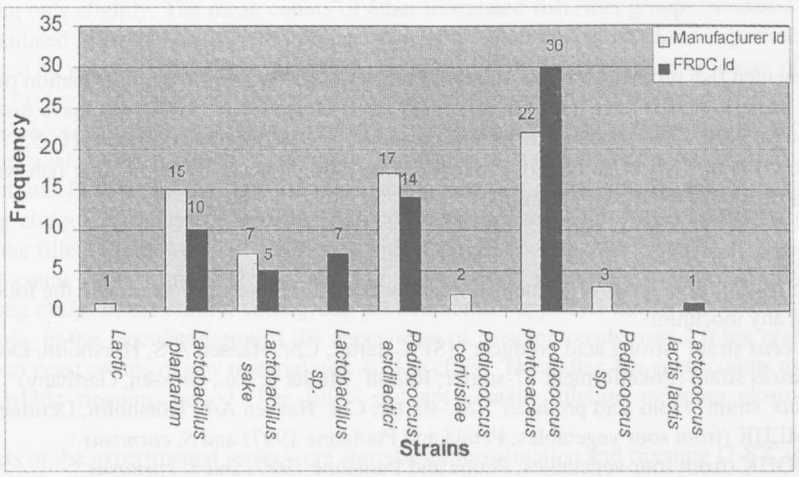

Figure 1. Comparison of the identification provided by manufacturers with the Food Research and Development Centre (FRDC) identification of LAB strains present in commercial meat starter cultures.

Table 1. Percentage of LAB strains producing EPS used in meat starter cultures under defined maturation conditions representing processes used in North America or in Europe

Species	North America[a] (%)	Europe[b] (%)
P. pentosaceus	26.7	26.7
P. acidilactici	0	0
Lb. plantarum	0	0
Lb. sake	0	0
Lb. sp.	0	0

[a]pH 5.2, T 20°C, NaCl 3%
[b]pH 5.5, T 20°C, NaCl 3%

CONCLUSION

From a technical point of view, ability of commercial meat starter cultures to produce EPS was demonstrated but seems unlikely to occur in fermented sausages. Therefore, slime production and/or ropy texture problem caused by LAB in cooked meat products could not only be attributed to LAB present in raw materials. Commercial meat starter cultures should not be excluded as a potential source of contamination by LAB producing ropy slime in cooked meat products.

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