# STABILITY OF PROBIOTIC BACTERIA IN THE PRODUCTION OF PROBIOTIC AND SYNBIOTIC SALAMI

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Key Words: Fermented sausage, starter culture, probiotic bacteria, prebiotic, shelf life

## Introduction

Human nutrition and wellbeing is highly influenced by the composition and activity of the microflora present in the intestine which is the largest immune generating organ with 80% of all antibody-producing cells (Biancone et al, 2002). Therefore dietary modulation by intake of an adequate amount of probiotic bacteria can induce beneficial effect to health e.g. by stimulating the immune system. Dairy products are the best known group of probiotic food products but also salami produced with applied probiotic bacteria could be an additional source to obtain the daily intake of more than 10<sup>8</sup> CFU viable probiotic bacteria (Guidelines FAO/WHO, 2002) perceived needed to obtain positive health effects. Among tested probiotic strains it has previously been proven that the best suited culture to survive during the processing of salami is *Lactobacillus paracasei* (Andersen, 1998. Andersen, 1999). Selected non-digestible oligosaccharides, prebiotics, can stimulate the development of probiotic bacteria in the colon (Coppa, 2005), and consequently, the synbiotic concept, to combine probiotic bacteria with prebiotic ingredients, could prove to be of added value if the prebiotic does not influence the processing and final product negatively.

# Objectives

The aim of this study was to explore the influence of synbiotic versus probiotic concept in the production of small calibre salami. Furthermore, the shelf life of the applied probiotic bacteria in both concepts was investigated over 10 weeks to study the stability of *Lb. paracacei* during storage.

## **Material and Methods**

As the experiment was conducted at a commercial manufacturer, KMOTR-Masna Kromeriz, all details in composition of the ingredients used were not revealed. Neither were the details in the production procedure. The sausages were produced with the normal recipe and processing procedure. The survival of *Lb. paracasei* was investigated in product with prebiotic (Zeus) and without prebiotic (Hades) applied.

# Recipe and processing

- Meat: pork and beef meat with approximately 23% back fat added.
- Ingredients approximately: 2.8% curing salt and salt, 0.4% dextrose, ascorbate, and spices.
- Starter cultures: 0.025% Lyocarni THM-17 (*Pediococcus pentosaceus* and *Staphylococcus xylosus*) and 5 doses Lyofast BGP 2 (*Lb. paracasei*) providing 5x10<sup>6</sup> CFU probiotic bacteria/g meat.
- Prebiotic: 2.5% DERACEL FC 20 (inulin-type fructans).
- Casing: approximately 45 mm cellulose casing.
- Processing: the mince was produced in a bowl chopper and stuffed with about 300 g/sausage. The sausages were fermented at 25°C and total processing time was 16 days with a weight loss of 41%.
- Codes: Zeus (with synbiotic) and Hades (with probiotic bacteria and without prebiotic).

# Bacteriological investigation

LAB were detected by surface plating on MRS (Merck), anaerobically incubated for three days at 30°C. *Lb. paracasei* was detected by surface plating on MRS + moxalactam (Andersen, 1998) and HHD (Homo-Heterofermentative Differentiation, internal recipe) in parallel, both anaerobically incubated for three days at 30°C. Staphylococci were detected by surface plating on S-110 (Oxoid), aerobically incubated for two days at 30°C. For verification purposes, a proportion of each colony type deviating in colony morphology was microscopically examined. All analyses were performed on a sample size of approximately 35 g finely chopped sausage.

#### Analytical programme

After the industrial production was completed, the sausages were vacuum-packed, and 15 randomly picked sausages of each code were collected at the manufacturer. They were sent to Italy for bacteriological survey. They were analysed on day of receipt (3 weeks after initiated production), and after 2, 4 and 10 weeks of refrigerated (5°C) storage (illustrated as 5, 7 and 13 weeks, respectively). LAB, *Lb. paracasei* (probiotic), staphylococci, and pH (Knick instrument) were monitored.

### **Results and Discussion**

The growth dynamics of LAB, *Lb. paracasei* (probiotic), and staphylococci during shelf life are illustrated in Figure 1 for Zeus and Figure 2 for Hades, respectively (left Y-axe). Furthermore, the pH kinetics are included (right Y-axe).

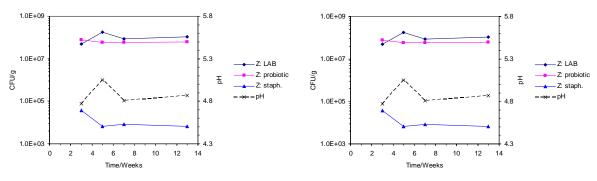




Figure 2. Bacteriological investigations of Hades

During processing LAB and *Lb. paracasei* grew to approximately 10<sup>8</sup> CFU/g. This is normal growth kinetics of starter culture LAB and demonstrated that *Lb. paracasei* grew and survived well in both types of sausages. Starter culture pediococci dominated LAB development all through shelf life. The level of *Lb. paracasei* was slightly higher in Zeus than in Hades indicating that the application of prebiotic might enhance the development in the salami. Nevertheless, in both products *Lb. paracasei* was very stable through out 10 weeks of refrigerated storage. The level of staphylococci was as anticipated at this stage of the processing and slightly further decline was observed. Probably due to additional fermentable sugars when prebiotic was applied the pH was lower in Zeus (average pH 4.88, stdev. 0.12) than in Hades (average pH 5.05, stdev. 0.03). Depending on the required final pH the amount of fermentable sugars should be adjusted when prebiotic is added. The application of DERACEL FC 20 did not influence the taste of Zeus negatively. The most notable sensory difference was that Zeus had slightly softer texture without influence on the slice-ability of the product.

#### Conclusions

It has been demonstrated that it is possible to produce salami with *Lb. paracasei* applied as probiotic culture jointly with a starter culture controlling the acidification. *Lb. paracasei* developed during production and was stable throughout 10 weeks of storage in Zeus and Hades. These findings indicate that probiotic and synbiotic salami could be a supplementary source of daily intake of probiotic bacteria as 5 g of salami, with a survival rate of *Lb. paracasei* as in Zeus and Hades, could give a substantial intake of beneficial bacteria. The addition of prebiotic influenced the final pH and should be taken into account when developing synbiotic products.

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