BIOPROTECTION: AN ADDITIONAL SAFETY HURDLE

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Background

In previous publications the application of commercially available bioprotective starter cultures has been described. In these papers the benefit of bioprotection in various meat products is documented. The application areas can roughly be divided into two main groups. One being fermented, dry sausages in which Bactoferm™ F-LC is a starter culture with an anti-listerial effect (Andersen, 1999; Christensen and Jørgensen, 1994). The added value is obtained due to the two lactic acid bacteria (LAB) that are used in this culture. They both produce bacteriocins in addition to appropriate acidification, aroma and colour formation. The other product group includes cooked, vacuum-packed or modified atmosphere packed (MAP) sliced meats to which Bactoferm ™ B-2 (formerly FloraCarn L-2) being a *Lactobacillus sakei* can be applied (Andersen, 1995a; Andersen 1995b). Furthermore, the culture Bactoferm™ B-FM, consisting of the same strain as B-2 together with *Staphylococcus xylosus*, has successfully been applied to fresh sausages such as British sausages and Chorizo (Andersen, 1997).

Meat spoilage may occur at any stage from "soil to table". The spoilage may be caused by a wide range of reactions, including physical, chemical, enzymatic and microbiological interactions (Huis in't Veld, 1995). Microbial spoilage can be delayed by preservation techniques. If the bacteriological environment is disturbed sufficiently by preservative factors, the bacteria are not able to multiply or even survive. Nevertheless, normally some types of bacteria will adapt to the new environmental conditions over time although with delayed initiate growth. It may be advantageous to combine milder preservation methods in order to avoid stressing the bacteria to an extent where they build up defence mechanisms (Gould, 1996). This is normally done by a combination of several preserving factors or hurdles (Leistner, 1994). Furthermore, there is a consumer trend towards products that are preserved to a lesser extent, of better quality, are more natural, free from additives, and nutritionally healthier (Stiles, 1996). To ensure food safety in for example organic foods is a challenge.

Application of selected strains of LAB as a competitive flora to inhibit undesirable bacteria, such as spoilage organisms and pathogens, should be considered as an additional biological hurdle. Hereby a higher degree of product safety can be obtained. Deliberate use of hurdle technology will be an additional factor to ensure more stable foods through synergistic effects from a controlled microflora and anti-microbial metabolites produced. To enhance efficiency bioprotection should be combined with other inhibiting factors such as packaging, avoidance of temperature abuse and GMP. An obstacle of adding a bioprotective culture to meats is the initial high cell count in the inoculated meat products, which is often perceived as an indication of poor hygiene during processing.

In a figurative sense bioprotection can be described as follows: A MAP or vacuum-packed meat product can be considered as a well-defined room such as a cinema. Generally cooked meat products contain only a low initial level of bacteria. Nevertheless, depending on storage conditions, the initial approx. 10^2 CFU/g will develop to approx. 10^8 CFU/g over time and will fill out the room. The composition of the initial flora may be pathogenic, spoilage, and/or harmless bacteria. It is impossible to predict which of these bacteria will dominate during the shelf-life and that is part of the problem (Jay, 1996). By adding a bioprotective culture the empty space (from 10^2 to 10^7 CFU/g) will be filled out with a rather inactive LAB strain and consequently inhibit the indigenous bacteria from developing so fast. This *Lac. sakei* is isolated from vacuum-packed beef and does not produce bacteriocin (Juven, 1998). The mode of action is considered as being competitive exclusion which inhibits the indigenous bacteria by using easy fermentable nutrients and rest oxygen.

A number of deaths caused by outbreaks of foodborne infections have once again drawn the public's attention to *Listeria monocytogenes* (Food Safety & Security, 2000). Listeria is widely spread and problematic because it can adapt to a wide range of environmental factors and therefore is difficult to control. Infections can be fatal for people with for example compromised immune systems, unborn foetuses, small children, and elderly people.

Objective

The aim of this work was to investigate the growth of *L. monocytogenes* and the anti-listerial effect of Bactoferm™ B-2 in three commercially available cooked meat products. Hereby it was possible to explore whether bioprotection as an additional hurdle could improve the safety of these products. The three products were: vacuum-packed cooked diced ham, MAP (20%N_2/80% CO_2) small emulsion sausages, and cooked MAP (60% CO_2/40% N_2) sliced meat product called "rullepølse" (rolled cooked pork belly). The latter is produced as an organic product with a low level of hurdles.

Methods

The following bacterial strains were added to relevant codes: Bactoferm™ B-2 at the level of approx. 10^7 CFU/g and *L. monocytogenes* V80 at the level of 10^2-10^3 CFU/g.

The newly produced meat products were inoculated with and without B-2 and *L. monocytogenes* at 5°C. As mentioned above the cooked ham was vacuum-packed, and emulsion sausages plus "rullepølse" were MAP in different combinations. The packed products were stored at 5 and 10°C, respectively. The products were analysed the day of inoculation and once a week for 4 weeks.

LAB were detected by pour plating (enumeration) as well as spread plating (visual recognition and microscopic examination) on MRS (Oxoid), anaerobically incubated for three days at 30°C. Listeria was enumerated on Palcam (Merck) with 2.5% egg yolk emulsion (Oxoid) added, incubated microaerophilic for two days at 37°C. Low levels of Listeria were examined semi quantitatively after enrichment (modified method after McClain & Lee, 1988 and Campanini et al, 1993).

Results and discussion

Bactoferm™ B-2 dominated as expected and developed to a level of approx. 10^9 CFU/g homogeneously in all samples. As shown in figure 2 the development in the emulsion sausages was slower than in the two other products. In the uninoculated samples the indigenous LAB developed to the same level as the inoculated except in the emulsion sausages. The growth was faster in cooked ham than in "rullepølse". The data are displayed below.

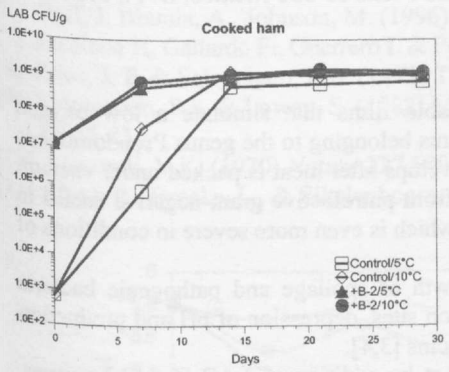

Figure 1. Development of LAB in cooked ham

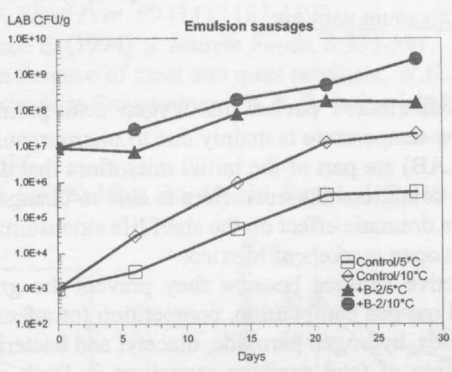

Figure 2. Development of LAB in emulsion sausages

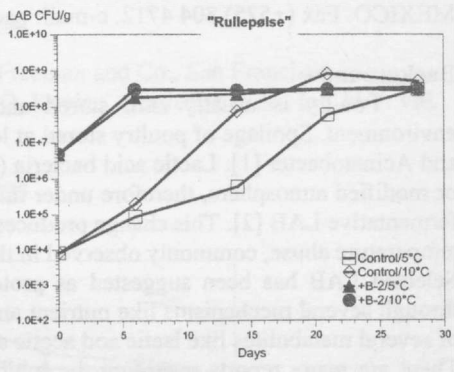

Figure 3. Development of LAB in "rullepølse"

It is difficult to make comparison between the products because there are different hurdles involved by which the producer wants to ensure product quality. A reason for the development of LAB being slower in the emulsion sausages might be that they are smoked. In previous tests it has been proved that MAP normally does not influence the LAB development (unpublished data). The indigenous LAB grow faster in all three products at 10°C than 5°C. LAB in the organic "rullepølse" do not grow as fast as in the cooked ham. This could indicate that the MAP used efficiently delays the development.

The development of *L. monocytogenes* is depicted in figure 4-6.

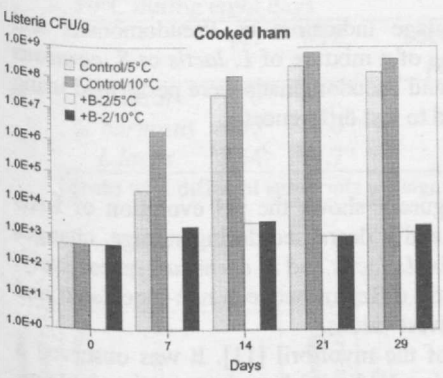

Figure 4. Development of Listeria in cooked ham

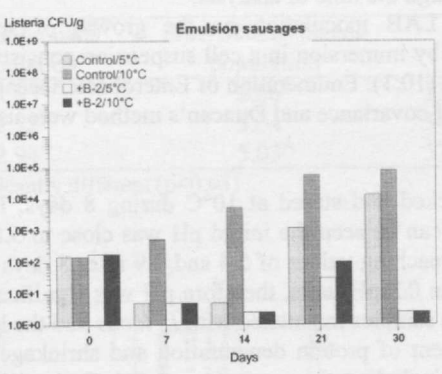

Figure 5. Development of Listeria in emulsion sausages

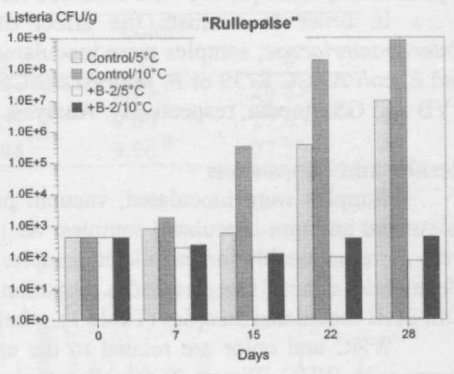

Figure 6. Development of Listeria in "rullepølse"

It is demonstrated that *L. monocytogenes* grows in all three meat products although to a different degree. As expected the same pattern as with the growth of LAB is seen. In respect to Listeria contamination the most vulnerable product is cooked diced ham. Both at 5 and 10°C Listeria increases fast. Although the inoculation level of *L. monocytogenes* is higher than an expected natural contamination level cooked ham manufactured in this way can cause problems. Again the development in emulsion sausages is the slowest. Still, it is proved that by temperature abuse Listeria will grow. Also in "rullepølse" the hurdles involved delay the development of Listeria as well. On the other hand, after two weeks of storage a rapid growth is seen. In all product/temperature combinations Bactoferm™ B-2 inhibits the development of *L. monocytogenes* efficiently. The trials have demonstrated that B-2 is able to act as an additional hurdle when the culture is applied to commercially produced cooked meat products. This implies that using bioprotection will enhance the product safety during the self-life.

Conclusion

Listeria is a potential problem in cooked meat products. Addition of Bactoferm™ B-2 is able to inhibit the development of *L. monocytogenes* in the three tested cooked meat products. The bioprotective properties of the added culture thus provide a way to enhance the control of Listeria and consequently to improve the safety of the products.

Literature list is available upon request.