EFFECT OF A COMMERCIAL STARTER CULTURE ON SURVIVAL OF SALMONELLAE IN METTWURST

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SUMMARY

Only the highest (10^8 cells/gr) tested inoculation level of L. plantarum reduced Salmonella contamination in all three inoculation levels (10, 100 and 1000 cells/gr) tested to about 1 cell per gram of Mettwurst. However, since this only happend when fermentation was followed by a seven-day-refrigiration period, it is therefore concluded that these three conditions (inoculation of 10^8 Lactobacilli per gram, fermentation and refrigiration) must be fulfilled in order to inhibit Salmonella growth in Mettwurst contaminated with up to 1000 cells per gram.

INTRODUCTION

The idea of using lactic acid bacteria for the control of pathogenic bacteria in raw hams and sausages is not at all new: The American National Academy of Sciences (NAS, 1975) had recommended using them against S. aureus, which was later carried out by Bartholomew and Blumer (1977, 1980), while Marcy et al. (1985) made similiar tests in reduced-sodium sausages. A general review of this idea is given by Mossel et al. (1985). Salmonella is one of the most important meat poisoning bacteria, due to its high pathogenesis, especially in the very young, old or debilitated humans (ICMSF,1978). Due to their high sensitivity to heat, acidity and low water activity (Sinell,1986) these organisms are rarely found in either cooked (Andres, 1985) or long fermented meat products (Johnston and Elliot, 1976; Johnson,1980; Luecke, 1986). However, they may be a problem in fresh uncooked products, such as Mettwurst (Sinell, 1986). The aim of this work was to find a correlation between the number of inoculated Lactobacilli and the number of surviving Salmonellae in Salmonella-inoculated Mettwurst.

MATERIALS AND METHODS

26 Mettwurst sausages weighing ca. 150 grams each were inoculated with a commercial starter culture (Combi Start 1505, Christian Hansen's Laboratories) and a group D Salmonella sp. according to the following 2x(4x3+1) design: Starter culture was standardized to give 4 inoculation levels of L. plantarum (10^5,10^6,10^7 and 10^8 cells per gram of sausage) and the Salmonella sp. (which had previously been isolated from raw turkey meat and confirmed as group D by the Veterinary Institute of Beit Dagan, Israel) was standardized to give 3 inoculation levels (10, 100 and 1000 cells per gram sausage). A thirteenth combination was inoculated with 10^7 Lactobacilli per gram and no Salmonella and served as a control. Each of these combinations consisted of 2 sausages,of which one was tested right after processing while the other was kept at 4 deg. C. for seven days after processing and then tested. Each test was carried out in duplicate. Sausages were formulated to contain ca. 32.5% moisture, 9% protein, 54% fat, 3% salt, 0.5% lactose, 0.8% spices, 500 ppm sodium erythorbate and 200 ppm sodium nirite.

Sausage batters were stuffed into 43-mm-diameter,water-and-smoke-permiable cellulose casings and put in a smokehouse for 4 days at 22-18 deg. C. and 92-85% relative humidity. They were smoked each day for 4 hours on days 2,3 and 4, and then put in a refriserator and/or tested as above.

Microbiological analyses consisted of total aerobic count (APT agar, Difco, 48 hr. at 30 deg.C.), Lactobacillus count (Rogosa bios (R) agar, Biolife, two layers, 72 hr. at 37 deg. C.) and Salmonella count (Brilliant Green(BG) agar, Difco, 18-24 hr at 37 deg.C.). Decimal dilutions were made with a phosphate buffer solution (pH 7.2). Only typical red colonies were counted as Salmonellae while at least two of them (if present) were transfered to Kligler Iron agar (Difco) and Lysine Iron agar (Difco) slant tubes and incubated 18-24 hr. at 37 deg.C.. Only the colonies that appeared to be Salmonella-positive on both agars (Difco Manual, 1977; Biolife Manual, 1985; Merck, 1982; Poelma and Silliker, 1976; ICMSF, 1978) were counted as such and the BG agar readings were corrected accordingly. In case of doubt, serological tests with Salmonella polyvalent antiserum were made. In any case where the Salmonella count was less then 10 per gram (negative on the 10^{-1} dilution plate) an enrichment was made by incubating a 10 gram sample in 90 ml Selenite Cystine broth (Difco) for 18-24 hr. at 42 deg. C. followed by streaking a loopful of the incubated broth on BG agar and identification of suspected colonies as above.

pH measurments were carried out by inserting a needle combination glass electrode attached to a Knick PORTAMESS ES 651 digital pH-meter - directly into the sausages.

RESULTS AND DISCUSSION

Results are shown in figures 1 through 7. The symbols APT, R and BG refer to the culture media and incubation periods described above. The numbers 1 and 2 refer to testing the product immediately after processing(series 1) or after 7 days at 4 deg.C. (series 2) respectively. All figures are expressed in logarithmic scales and represent the logarithmic means of the respective duplicates.

Fig. 1 (Salmonella inoculated at 10/gr) shows that the total count (APT) and Lactobacillus count (R) curves were almost parallel, which means that Lactobacilli were indeed the dominant flora. The pH curves, however, show a negative correlation to both of them, as could be expected. In the 10^6/gr inoculation there was an unexplained factor that inhibited all flora. Salmonella growth did not exceed 10/gr, yet it is not correlated to the Lactobacillus inoculation.

Fig. 2 (Salmonella inoculated at 100/gr) shows similar characteristics with a negative correlation between the R and the BG1 curves, but not with the BG2 curve.

Fig. 3 (Salmonella inoculated 1000/gr) also shows these relationship between the APT+R curves and the pH curves. However, the correlations between these and the BG curves are more distinct. There is,in fact,a consistent decrease in the BG2 counts as Lactobacillus inoculations increase.

Fig. 4 (Salmonella counts vs. inocula, series 1) shows distinctly increasing functions at all Lactobacillus inoculations. At 1000/gr the effect of Lac. inoculation (except for the 10^6/gr) on Salmonella growth is also very clear.

Fig. 5 (Sal. counts vs. inoc. series 2) shows the same effects even more distinctly, with the addition of the fact that the inoculation of 10^8 Lactobacilli/gr completely inhibited Salmonella growth at all Salmonella inoculations tested.

Fig. 6 (Lac. and Sal. growth at inoc. of 10^7 Lac./gr) shows, along with the control group, that only Sal. growth is (positively) correlated to Sal. inoculation, i.e. neither of them had any effect on Lactobacillus growth. This was expected because the number of Salmonella cells was negligible relative to that of the Lactobacilli.

Fig. 1: Microbial Growth and pH at 4 Inoculation Levels of L. plantarum and 10 Salmonellae per Gram Mettwurst, Series 1 and 2.

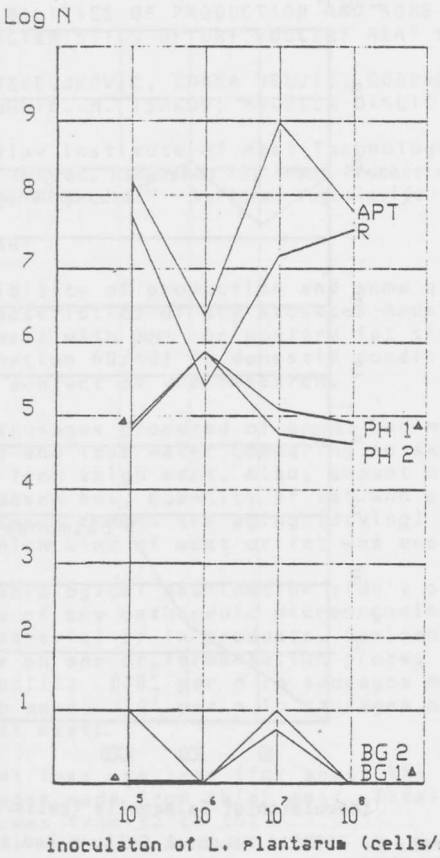


Fig. 2: Microbial Growth and pH at 4 Inoculation Levels of L. plantarum and 100 Salmonellae per Gram Mettwurst, Series 1 and 2.

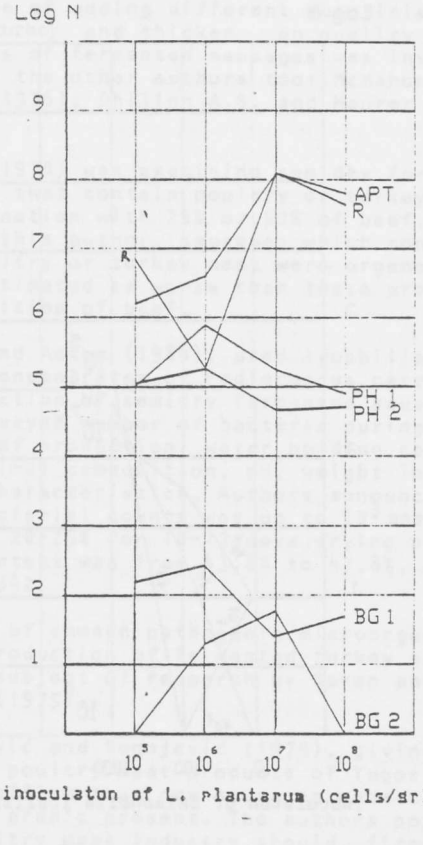


Fig. 3: Microbial Growth and pH at 4 Inoculation Levels of L. plantarum and 1000 Salmonellae per Gram Mettwurst, Series 1 and 2.

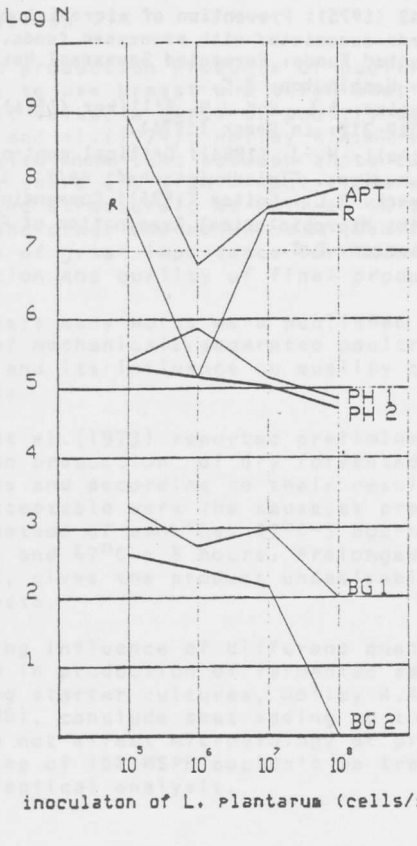


Fig. 4: Salmonellae - Growth vs. Inoculation at 4 Inoculation Levels of L. plantarum, Series 1.

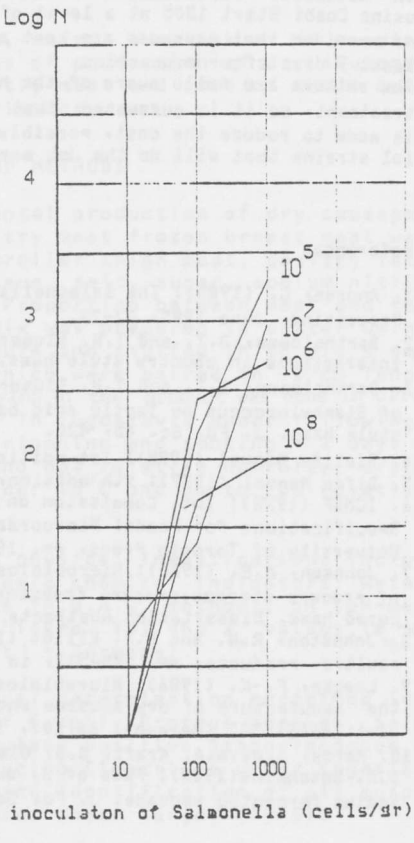


Fig. 5: Salmonellae - Growth vs. Inoculation at 4 Inoculation Levels of L. plantarum, Series 2.

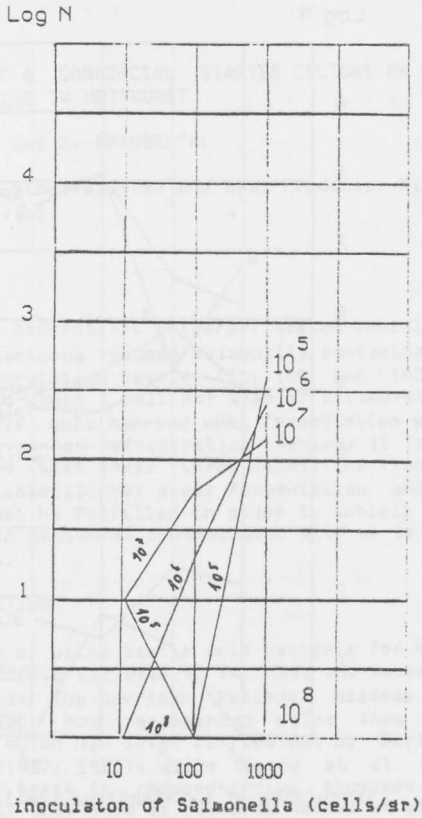
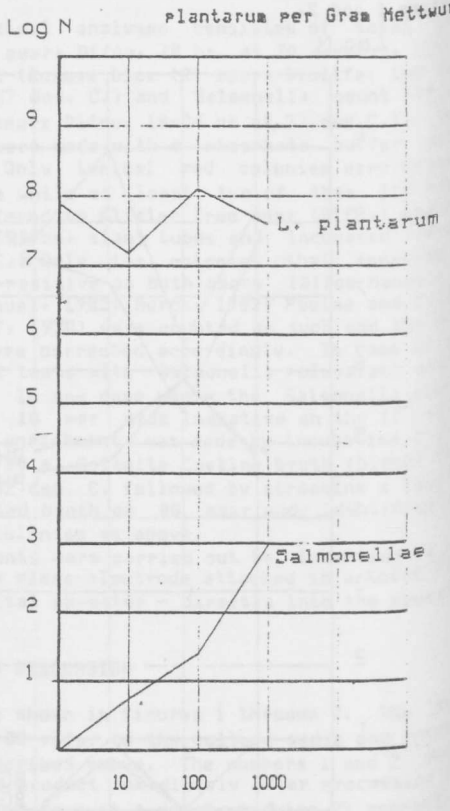


Fig. 6: Growth of L. plantarum and Salmonella at 4 Inoculation Levels of Salmonella and 10^7 L. plantarum per Gram Mettwurst.



CONCLUSIONS

It has been found possible to inhibit Salmonella growth in Mettwurst contaminated by up to 1000 cells/gr by using Combi Start 1505 at a level of 10^7 Lactobacilli/gr provided that sausages are kept at 0-4 deg.C. for at least 7 days after processing.
The authors are fully aware of the high cost of such a treatment, so it is suggested that further reasearch is made to reduce the cost, possibly by finding bacterial strains that will do the job more effectively.

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