USE OF TWO PEDIOCOCCUS STRAINS ISOLATED FROM SOUR VEGETABLES AS STARTERS IN RAW HAM

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INTRODUCTION

The pH value of fermented whole-meat products do not decrease to levels as low as in fermented sausage, and texture of hams does not develop as firm as that of fermented sausage when using common starters (Petäjä and Kuusela 1978). By using acid effectively producing lactic acid bacteria isolated from sour vegetables the pH-values of fermented whole-meat products would obviously decrease to lower levels than those achieved by common starters currently in use. In a preliminary study, the activities of 2 Pediococcus strains isolated from sour vegetables as starters in dry sausage were examinated (Petäjä 1997). The strains proved to grow well, to form acid and thus decrease pH effectively, and affect ripening and quality favourably. The effects of these strains on the ripening of fermented ham (raw ham) have preliminarily been investigated here. This paper is a researsch note of the study.

MATERIAL AND METHODS

Lactic acid bacteria: The first preliminary studies dealing with the isolation and examination of lactic acid bacteria from sour vegetables were described in Petäjä (1997). On the basis of acid production (pH decrease), two strains were selected for testing as starters in dry sausage; these 2 strains, MLHK (from sour cabbage with marine algae) and POHK (from sour carrot stripes), were also used here.

Preparation of raw hams: Four experimental series of raw hams were prepared. Each series consisted of control ham inoculated with RM 2000 starter (Pediococcus pentosaceus + Staphylococcus carnosus) (Rudolf Müller & Co., Giessen, Germany), ham inoculated with Pediococcus strain MLHK and Baktoferment 61 starter (Staphylococcus carnosus) (Rudolf Müller & Co.), and ham inoculated with Pediococcus strain POHK and Baktoferment 61 starter.

The experimental hams (Musculus adductor) were cured by injecting brine in amounts corresponding to 5% of the weight of the meat. The injected brine contained 26% NaCl, 10% glucose, 0.5% ascorbic acid, 0.35% KNO_3, and 0.012% $NaNO_2$. Curing was completed by dry-salting hams with 2% coarse salt for 1 day at 6°C.

The RM 2000 starter, the strains MLHK and POHK and Baktoferment 61 starter were inoculated into the hams in addition to the brine. The strains MLHK and POHK were used as APT broth cultures, RM 2000 and Baktoferment 61 as commercial preparates. The aim was to inoculate lactic acid bacteria at doses of 10^7 colony forming units (cfu)/g, RM 2000 staphylococci at 10^7cfu/g and Baktoferment 61 staphylococci at 10^6cfu/g into ham.

Dry-salted hams were placed in a net and hung in a ripening chamber. The ripening program of the hams was as follows:

Ripening time	Temperature	Humidity	Smoking
1 day	22°C	96%	
2 - 7 days	21 - 20°C	96 - 90%	3 h/day
8 - 16 days	15°C	80%	

Determinations on the experimental hams:

Sensory evaluation: The texture, aroma and flavour were evaluated after 4, 8 and 16 days of ripening using a scoring system and descriptive method as follows:

- Texture (scores 6 - 0; 6 firm, 4 quite firm, 2 slightly firm, 1 - 0 soft)
- Aroma (scores 6 - 0: 6 excellent, 4 good, 2 odourless, 1 - 0 unpalatable)
- Flavour (scores 6 - 0; 6 excellent, 4 good, 2 moderate, 1 - 0 unpalatable)

The evaluation was performed in the laboratory by 2 technicians (the hams were too small for panel evaluation), who determined the scores by consulting with each other.

pH-value and titrated acid: The pH-value was measured directly from the hams at 3 different sampling sites (the mean was used as a result) and from a 1:9 dilution (0.9% NaCl solution) of sample hams.

Acid titration was conducted of the filtrate obtained from 1:9 dilution, and the results were calculated as percentages of ham.

Weight loss: Weight loss was measured as percentages of the original weight.

Microbiological determinations: Each experimental series was studied microbiologically after manufacture (day 0) and after 2, 4, 8 and 16 days of ripening. The following determinations were performed: total plate count of aerobically growing bacteria (APT agar, BBL 10918, 4 days at 30°C), inoculated lactic acid bacteria (APT agar, BBL 10918, 4 days at 30°C), staphylococci (Baird-Parker agar, Labm 85 and X085, 2 days at 37°C), pseudomonads (GSP agar, Kielwein 1969, 4 days at 25°C) and yeasts and molds (Rose-Bengal agar, Labm lab36 and X009, 2-4 days at 30°C).

RESULTS AND DISCUSSION

Sensory evaluation:

Texture: The mean texture values of all 3 experimental hams were the same after 4 (3.8/6) and 8 days (4.8/6) of ripening (Table 1). After 16 days of ripening the texture values of MLHK and POHK hams were higher than the values of RM 2000 hams; the differences, however, were not significant.

Aroma: The aroma values of the experimental sausage groups were not significantly different after 4, 8 and 16 days of ripening (Table 1). The strongest aroma appeared in POHK sausages.

Flavour: The flavour values of experimental hams were not significantly different after 4, 8 and 16 days of ripening (Table 1). The strongest flavour appeared in POHK hams.

pH-value: At the beginning of ripening the pH values ranged 5.55 - 5.92 (means 5.76-5.80; Table 2). During the first 2 days of ripening (1 day at 6°C followed by the 1 day at 22°C) only the pH value of control hams (RM 2000) decreased by a mean of 0.24 pH. During the following 2 days the pH values of all ham groups decreased distinctly. After 8 days of ripening the pH of MLHK and POHK hams decreased to <5 (means 4.75 and 4.92) and the pH of control hams to >5.

It can be concluded that Pediococcus strains from sour vegetables decrease the pH of dry hams to lower levels (pH <5) than those attained with pediococci of the RM 2000-preparate. By sour vegetable lactic acid bacterial strains the decrease of pH value can be confirmed in dry hams.

Titrated acid content: The mean content of titrated acid of raw ham material was 0.89% in all experimental ham groups (Table 2). During the first 2 days of ripening the acid content did not increase. During the following 2 weeks of ripening the content of titrated acid increased to >1.5% the highest contents being in hams inoculated with sour vegetable pediococci.

Weight loss: The range of weight loss percentages was 25.0-32.3 after 16 days of ripening which level was aspired. The hams inoculated with sour vegetable pediococci dried slightly more rapidly (means: MLHK ham 28.4%, POHK ham 29.5%) than control hams (mean 26.5%).

Microbiological determinations: The mean count of inoculated RM 2000 pediococci was 7.3 log cfu/g, the inoculated counts of sour vegetable pediococci being 1 log unit less (Tables 3). The inoculated lactic acid bacteria formed the predominant part of the microbial flora of experimental hams during the ripening period. During the first 2 days of ripening (1 day at 6°C followed by 1 day at 22°C) the mean count of RM 2000 pediococci increased by 1.3 log cfu/g, increasing thereafter only slightly. The count of vegetable pediococci increased by log unit during the first 2 days of ripening, increasing further during the second 2 days by more than 1 log unit/g to the final level of >8.5log cfu/g.

The mean count of staphylococci in RM 2000 hams was 7.4 log cfu/g after inoculation, increasing only slightly during ripening. In hams inoculated with sour vegetable pediococci the mean Staphylococcus count was 5.5 log cfu/g after inoculation, remaining roughly at this level throughout the ripening period.

During the first 2 two days of ripening the experimental hams contained pseudomonads at counts ranging 2.0 - 5.7 log cfu/g. During the second 2 days of ripening the pseudomonad counts decreased to <2.0 log cfu/g and remained at that level.

The experimental hams contained yeasts at counts ranging <2.0 - 5.4 log cfu/g. The yeasts appeared throughout the 16-day ripening period showing variable counts.

CONCLUSIONS

Two lactic acid bacterial strains (Pediococcus) isolated from sour vegetables can be applicable for use as starters in fermented ham on the following bases:

1. The 2 Pediococcus strains isolated from sour vegetables grew to levels of >8.5 log cfu/g in raw ham.
2. The sour vegetable strains formed more acid in raw ham than pediococci of commercial starter preparate RM 2000, thus also decreasing the pH. The pH of the hams prepared with sour vegetable pediococci decreased to <5.0 during ripening.
3. The texture of the sausages prepared by sour vegetable pediococci was firmer than the texture of RM 2000 hams. The mean aroma and flavour scores also were higher than those of RM 2000 sausages.

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TABLE 1. Texture, flavour and aroma of experimental hams after 4, 8, and 16 days of ripening.

	4 days		8 days		16 days	
Ham group	X	s	X	s	X	s
Texture						
1. RM 2000	3.8	0.5	4.8	0.5	5.0	0.0
2. MLHK	3.8	0.5	4.8	0.5	6.0	0.0
3. POHK	3.8	0.5	4.8	0.5	5.5	1.0
Aroma						
1. RM 2000	4.0	0.0	4.5	0.6	5.0	0.9
2. MLHK	4.3	0.5	4.8	1.5	5.3	1.0
3. POHK	4.5	0.6	5.3	1.0	5.5	1.0
Flavour						
1. RM 2000	4.0	0.0	4.8	0.5	5.3	0.5
2. MLHK	4.3	1.0	5.0	1.4	5.8	0.5
3. POHK	4.8	0.5	5.3	1.0	5.8	0.5

X = mean

s = standard deviation of mean

1) Means within the vertical line not followed by the same letter are significantly different 8($p < 0.05$). If no letters are listed after the means, no differences are present among them.

TABLE 2. The pH values and titrated acid contents(%) of experimental hams after 0, 2, 4, 8, and 16 days of ripening.

	0 days		2 days		4 days		8 days		16 days	
Ham group	X	s	X	s	X	s	X	s	X	s
pH-value										
1. RM 2000	5.80	0.14	5.56	0.13	5.18	0.18	5.08	0.18	5.08[a]	0.12
2. MLHK	5.76	0.16	5.76	0.13	5.16	0.25	4.75	0.19	4.72[b]	0.16
3. POHK	5.76	0.17	5.73	0.10	5.31	0.16	4.92	0.20	4.99[ab]	0.21
Titrated acid (%)										
1. RM 2000	0.89	0.14	0.90	0.03	1.16	0.20	1.35	0.10	1.53	0.20
2. MLHK	0.89	0.14	0.89	0.02	1.19	0.21	1.44	0.19	1.62	0.13
3. POHK	0.89	0.14	0.96	0.10	1.20	0.24	1.34	0.08	1.66	0.15

TABLE 3. Total count of lactic acid bacteria (log colony forming units/g = log cfu/g) corresponding to counts of inoculated lactic acid bacteria in experimental hams after 0, 2, 4, 8, and 16 days of ripening.

	0 days		2 days		4 days		8 days		16 days	
Ham group	X	s	X	s	X	s	X	s	X	s
1. RM 2000	7.3	0.3	8.6	0.3	8.7	0.3	8.9	0.2	8.9	0.1
2. MLHK	6.4	0.2	7.3	0.5	8.5	0.1	8.7	0.5	8.6	0.2
3. POHK	6.3	0.2	7.4	0.7	8.7	0.4	8.6	0.5	8.5	0.4