MEAT STARTER CULTURES IN SALAMI MANUFACTURE

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SUMMARY

P.pentosaceus, *L.plantarum*, *L.sake*, *S.carnosus* and an atypical *M.luteus* were characterised with respect to growth and acid production and their viability during salami manufacture. Growth and acid production were determined at constant pH (4.7, 5.5 and 6.3) and *P.pentosaceus*, *L.plantarum* and *L.sake* showed similar characteristics over the pH range studied while *S.carnosus* and M.varians were sensitive to the lower pH. Also, *P.pentosaceus* and *L.sake* showed greater psychrotrophic growth than *L.plantarum*.

Salami made with *P.pentosaceus* maintained higher viable numbers in the product over six weeks than did *L.plantarum*. In all cases, *S.carnosus* did not proliferate in the presence of the more pH-tolerant organisms. Further, the proliferation of *P.pentosaceus* and *L.plantarum* did not prevent the development of non-starter flora, an important factor in determining product quality.

INTRODUCTION

The requirements of modern dry-sausage manufacture and the increasing size of production runs demands consistency of product quality. The traditional process is a less defined and slower process than that required by today's manufacturers who need to 'push' the product and reduce holding times which constitute an important cost factor. Since the inception of commercial starters, the search for better preparations incorporating faster acid- producing strains has continued (Bacus 1984). The aim of this study was to investigate strain characteristics of five bacteria to identify the most suitable strains for use in salami manufacture.

EXPERIMENTAL METHODS

1. Cultures And Starter Preparations

Five cultures (strains JC1, LP2, LS1, SS1 and JC3) were obtained from the Bacterial Products Division of Burns Philp Pty Ltd. Strains JC1, LP2 and LS1 were routinely propagated at 30°C in Mann Rogosa and Sharpe (MRS) broth and strains SS1 and JC3 in a peptone-yeast-extract (PYE) broth, consisting of 1% peptone, 0.5% Yeast-extract, 0.5% sodium chloride and 1% glucose. Bacterial preparations for use in salami manufacture were freeze-dried and dry mixed with ground food grade salt as the carrying medium.

2. Identification Of Bacteria

Identification of organisms was confirmed by gram and catalase reactions and testing with API kits, No. 5OCHL (Lactobacilli and pediococci) or kit No. 2050 (Staphylococci or Micrococci). Additional biochemical tests were performed for identification as required.

3. Growth And Acid Production

(i) pH Profiles.

Cultures were grown in broth (MRS or PYE) containing 1% glucose at a constant pH of 4.7, 5.5 or 6.3. Growth temperatures were maintained at 25°C or 30°C. Bacterial growth was determined by absorbance at 650nm with reference to a standard curve of absorbance vs dry weight. Specific growth rates were estimated by determining the slope of Ln dry weight vs time and expressed as h^{-1}. Acid production was determined by measuring the volume of 2.5M NaOH, added to maintain pH during growth.

(ii) Temperature Profiles

Growth of cultures (at inoculum levels of 10^6 cfu/mL) in 10 mL of MRS or PYE broth containing 1% glucose were determined between 13 and 32°C. The pH of the culture and growth (6lOnm) were determined after incubation for 16h.

4. Salami Manufacture

Salamis were manufactured using either a Milano or Mettwurst spice mixture (1.0%) (Mauri Flavours Group)

TABLE 1 - Salami manufacture utilising P.pentosaceus /S.carnosus and L.plantarum/S.carnosus culture preparations

BATCH	DAY	pH	Bacterial Counts (log cfu g^{-1})		
			MRS (anaerobic)		MSA (aerobic)
			Starter	Non-starter	SS1
JC1/SS1	0	5.81	7.22	<5.00	6.93
METIWURST	2	4.78	8.04	7.86	6.54
	8	4.64	7.88	7.10	6.57
	43	4.96	7.30	<5.00	5.41
LP2/SS1	0	5.80	7.02	<5.0	6.90
METIWURST	2	4.80	7.51	8.15	6.60
	8	4.58	6.49	7.91	6.41
	43	4.79	6.56	<5.0	5.45
LP2/SS1	0	5.78	7.26	<5.00	7.15
MILANO	2	5.14	7.62	7.63	6.93
	8	4.72	6.58	7.05	6.18
	43	4.83	6.46	<5.00	6.23

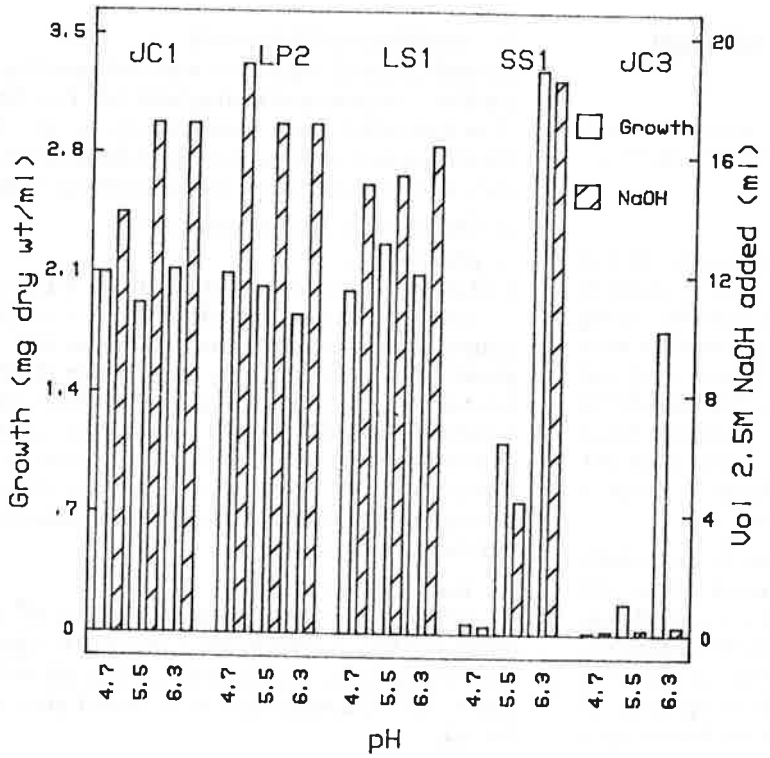

FIG.1. The effect of pH on growth and acid production.

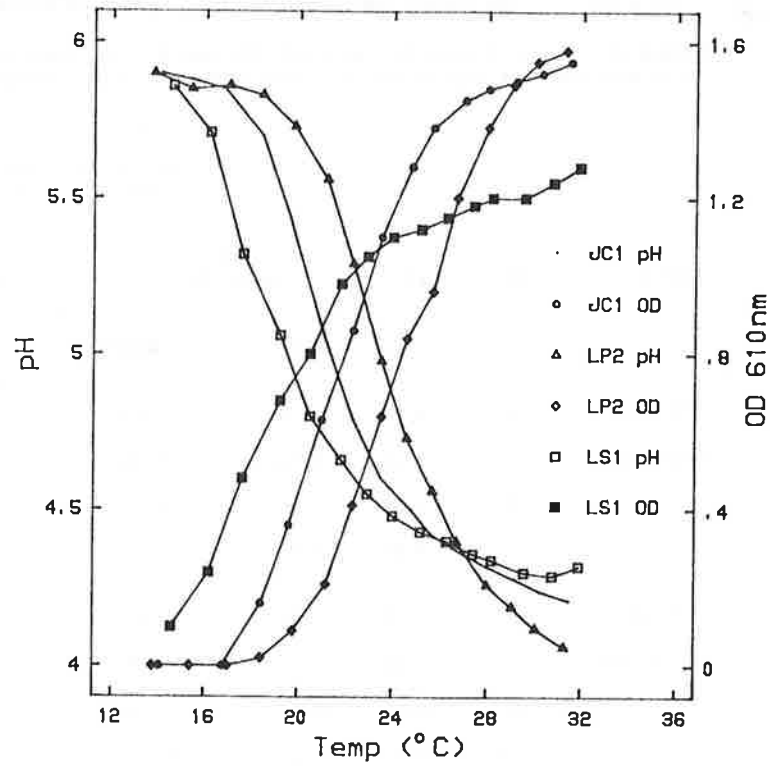

FIG.2. The effect of temperature on growth and acid production of P.pentosaceus JC1, L.plantarum LP2 and L.sake LS1.

with freeze-dried culture at $10^7 g^{-1}$. The mixture consisted of 67% beef and 29% pork back fat, 2.0% sodium chloride, 0.5% glucose and 0.018% (w/w) sodium nitrite. Synthetic casings (65 mm diameter) were filled and the salamis cured at 24°C for 24h and relative humidity (RH) 85 to 90% followed by 24h at 22°C and 80 to 85% RH. After 48h the salamis were cold-smoked at 24°C for 4h, then held at 20°C and 75 to 80% RH for 3 days followed by 18°C and 70 to 75% RH for 1 week.

5. *Analytical Methods*

Ammonia was determined on culture supernatants after centrifugation using Boehringer Kit No. 542946. The pH of meat mixtures was determined on filtrates obtained by blending 10 g of sample with 30 mL of distilled water and filtering.

6. *Enumeration*

Colony forming units were enumerated on homogenised samples using MRS agar (lactic acid bacteria) and Mannitol Salt Agar (MSA) (staphylococci and micrococci). MRS agar was incubated anaerobically at 30°C for 2 days for lactobacilli and pediococci and MSA media were incubated aerobically at 30°C for 2 to 7 days for staphylococci or micrococci.

RESULTS

1. *Strain Identification*

The organisms were identified as *P.pentosaceus* (JC1), *L.plantarum* (LP2), *L.sake* (LS1), *P.pentosaceus* (JC1), *S.carnosus* (SS1) and *M.luteus* (JC3). *M.luteus* JC3 produced a yellow pigment and showed an inability to grow on nutrient agar containing 7.5% NaCl which is atypical of this species (Sneath et al. 1986).

2. *Growth And Acid Production*

The effect of pH on growth and acid production indicated that *S.carnosus* SS1 and *M.luteus* JC3 were more pH sensitive than the lactobacilli and pediococcus (Fig.1). *S.carnosus* SS1 showed a higher cell mass and consequently acid production at pH 6.3 than the lactobacilli or pediococcus (Fig.1). At all pH'S, growth and acid production by *M.luteus* JC3 was the lowest (Fig.1). When cultures of *S.carnosus* SS1 and *M.luteus* JC3 were grown without pH control, PYE media was preferred over MRS media and generally supported greater culture growth, particularly for *M.luteus* JC3.

Ideally, fermentation temperatures don't exceed 25°C and this temperature was sub-optimal for all strains investigated. At temperatures below 23°C, *L.sake* LS1 showed more psychrotrophic growth than either *P.pentosaceus* JC1 or *L.plantarum* LP2 (Fig.2). At this temperature, both *P.pentosaceus* JC1 and *L.sake* LS1 had about twice the cell mass of *L.plantarum* LP2, although there was little difference in the final pH. At 30°C, *L.sake* LS1, had less cell mass than *L.plantarum* LP2 or *P.pentosaceus* JC1 and at 25°C *P.pentosaceus* JC1

and *L.plantarum* LP2 demonstrated the largest difference in cell growth (Fig.2).

The specific growth rates at 25°C and pH 5.5 (a pH mid-phase in salami fermentation) of *P.pentosaceus* JC1 ($0.75h^{-1}$) and *L.sake* LS1 ($0.80h^{-1}$) were about 1.5 times that of *L.plantarum* ($0.48h^{-1}$). These differences are consistent with the more psychrotrophic growth properties of *P.pentosaceus* JC1 and *L.sake* LS1 shown in Figure 2. The growth and acid production of *S.carnosus* SS1 and *M.luteus* JC3 between 17 and 32°C was also investigated (Fig.3). *M.luteus* JC3 grew better than *S.carnosus* SS1 above 25°C but there was no difference below 25°C. This organism produced ammonia which accounted for the increase in pH during growth (Fig.3).

3. Salami Manufacture

Salami products were manufactured with freeze-dried culture preparations of *S.carnosus* SS1 in combination with the more psychrotrophic *P.pentosaceus* JC1 and the less psychrotrophic *L.plantarum* LP2. Salami made with Mettwurst spice mixture and an *L.plantarum/S.carnosus* culture reached 5.14 at 48h but the Milano mixture slowed acid development by 0.34 pH units over the same period (Table 1). Growth of *L.plantarum* at 48h was unaffected by the Milano mix which suggested that acid production rather than growth had been inhibited (Table 1). Salami made with *P.pentosaceus/S.carnosus* culture maintained higher viable numbers over 6 weeks than did the *L.plantarum/S.carnosus* culture (Table 1). In all cases, *S.carnosus* did not proliferate in the presence of the pH tolerant organisms. Growth of *P.pentosaceus* JC1 and *L.plantarum* LP2 did not prevent growth of non-starter flora in the Mettwurst product, and this effect was greater with *L.plantarum* LP2 (Table 1).

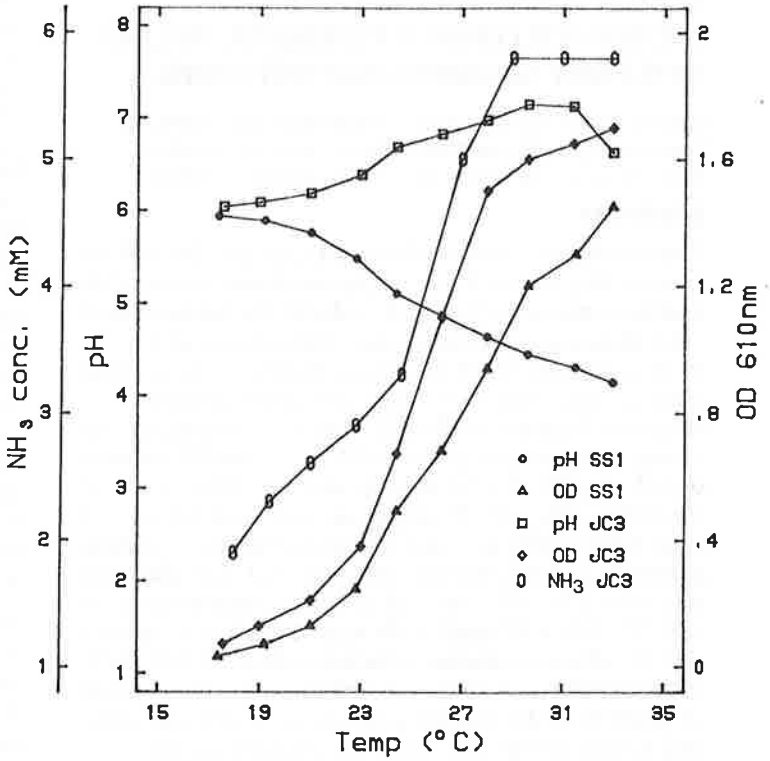

FIG.3. The effect of temperature on growth, acid production and ammonia production by S.carnosus SS1 and M.luteus JC3.

DISCUSSION

In recent times, emphasis has been placed on the selection of fast acid producing starters with the ability to maintain viability at low pH. For the lactic acid bacteria studied, no differences in pH-sensitivity could be detected. However, it was demonstrated that *S.carnosus* SS1 was more pH-sensitive than the lactic acid bacteria tested and that *M.luteus* JC3 was more pH-sensitive than *S.carnosus* SS1. The pH-sensitivity of *S.carnosus* SS1 can explain the failure of this organism to proliferate in salami when added in the presence ot *P.pentosaceus* and *L.plantarum*. The inability of staphylococci and micrococci to grow in the presence of lactic acid bacteria suggests that they must be metabolically active early in the fermentation (when added to the product mix). Also in this study, the advantages of psychrotrophic strains of lactic acid bacteria in salami fermentations has been considered. Although no differences occurred with the final pH of the product at 48h, the higher cell growth of *P.pentosaceus* was attributed to its psychrotrophic growth properties. Lactic acid bacteria strains showed a strong viability in the product over a period of six weeks.

CONCLUSIONS

The use of psychrotrophic lactic acid bacteria where fermentations less than 25°C are employed, should ensure a greater predominance of starter flora over non-starter flora. The ability to utilise any factors to control this ratio in favour of the starter will be important in determining product quality.

ACKNOWLEDGEMENTS

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