10 Conditions for the use of lactic acid as a decontaminant in the meat industry

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Introduction

Good manufacturing practices (G.M.P.) during slaughter include all measures necessary to produce meat with the lowest possible microbial contamination. This is only attainable if the whole process is strictly controlled (W.H.O. VPH/83.42). Microbiological examination of carcass surfaces should be used to identify critical hazard points in slaughter lines (Gerats et al., 1981). Data thus obtained may be used in steering the process in order to attain a satisfactory product.

Usually pig skin is heavily contaminated with bacteria. During scalding and dehairing and particularly during singeing a substantial reduction of the bacterial load is achieved (Snijders, 1976). In the blackscraper and polishing machinery, however, pig carcasses are often severely recontaminated (Snijders, 1976; Gerats et al., 1981). If cleaning and disinfection are inadequate, considerable quantities of dirt (haires, parts of the epidermis etc.) remain in the machinery. Consequently, the blackscraping and polishing machinery acts as a continuous source of bacterial contamination. In cattle and veal slaughter lines the skinning process entails severe hygienic problems. Mechanical skinning is therefore preferable.
Evisceration is another critical point, Carcasses should be opened carefully to prevent severing of the intestines. The rectum should be cut out and tied off.
Furthermore, an integral part of G.M.P. is the training, instruction, and motivation of personnel working at slaughter lines (Gerats et al., 1982).

Decontamination is another possibility for reducing the contamination of carcasses, cuts, and by-products. However, it has often been asked whether decontamination procedures are really required when advanced hygiene measures and adequate refrigeration are applied along slaughtering and processing lines. Whenever and wherever, in spite of hygienic practices, meat surfaces or cuts do become (cross) contaminated, such a terminal decontamination may be useful (Smulders and Woolthuis, 1983).
Lactic acid (LA) is an acceptable decontaminant, because i) it is a natural product; ii) it is physiological and not toxic; iii) it is often used in the meat industry, and also produced fortuitously in meat products as a result of fermentation (Barendsen et al., 1984).
This paper is an attempt to interrelate the data obtained by our Department in several experiments on lactic acid decontamination.

Materials and methods

Experiments were conducted on pig, veal, and cattle slaughter lines. Carcasses were sprayed with LA solutions at approximately 45 minutes post mortem (p.m.), and subsequently chilled in conventional chill rooms (3 ± 1°C). In one experiment hot deboned pig bellies were sprayed with 5% v/v lactic acid, 3 hours p.m., and subsequently stored under refrigeration at 7°C for 8 days. Porcine livers were dipped for 5 minutes in a 0.20% v/v lactic acid solution, allowed to drain for 30 s,and subsequently vacuum packaged and stored at 3 ± 1 °C. Veal brains were sprayed with 1.25 LA after manual extirpation. In a special cleaning and disinfection unit (a perspex tube with a diameter of 6 cm and two flat spray nozzles) tests with and without LA were carried out on stainless steel plates. LA solutions were prepared by diluting a 90% L-lactic acid stock solution (Chemie Combinatie Amsterdam).
The surface pH of carcasses was assessed with a pH meter and combined electrodes (Russell pH Ltd, Auchtermuchty, Fife, Scotland). The colour score of carcasses after LA treatment was assessed visually by two trained graders. Veal longissimus chops were evaluated sensorically by a 15 member consumer panel.

Sampling for bacteriological examination was carried out using an excision technique (Snijders, 1976; 1984). Tissue discs were sampled by means of sterile cork borers, scalpels, and tweezers, after which they were collected in plastic bags. After peptone-saline solution had been added, samples were macerated in a Stomacher for 2 minutes. Colony counts were expressed in colony forming units (c.f.u.) per cm² (except those from veal brains which were expressed per gram) and then converted to logarithms base$_{10}$. The following criteria for meat samples were determined: Mesophilic aerobic colony counts (ACC) in plate count agar or Tryptone Glucose Beef Extract Agar (poured plate method, incubation at 30°C for 3 days); Enterobacteriaceae colony count (ECC) in violet red bile glucose agar (poured plate method with overlayer, incubation at 37°C for 1 day)

As a substitute for knifeblades, stainless steel plates (9 x 4,5 cm) were used. They were overlayered with buffered Tryptone Soya Broth Agar (Direct Surface Agar Plate method). For this examination colony counts were expressed in c.f.u. per knife. Differences between c.f.u. counts were assessed using a Student t-test and a Wilcoxon test.

Results and Discussion

The importance of cleaning and disinfection as part of GMP is clearly shown in table 1. Aerobic colony counts of 4.2 log N per cm² were found on pork skins after they had passed polishing machinery which had not been cleaned regularly (Snijders 1976).

When cleaning was carried out more systematically but without daily disinfection, the contamination of the carcasses was reduced to 3.3 log N per cm². Mechanizing the cleaning procedure by using rotating spraying devices combined with a daily disinfection programme resulted in a further reduction of contamination. When the carcasses were sprayed with a 1 % v/v lactic acid solution (2 L of solution for 25 sec per carcass) after having passed the polishing machinery, an additional reduction of 0.5 log N per cm² was obtained. The effect of LA decontamination depends among other things on concentration, application time, temperature, the attachment of micro-organisms to surfaces, and the application method (spraying or immersing) (van Netten and Mossel, 1980, Notermans and Kampelmacher, 1983).
A 5 % LA spray results in a high reduction (table 2), but causes an unacceptable discolouration of the treated cuts.

Nevertheless, the data in table 2 show very clearly the immediate and delayed bacteriological effects of LA decontamination on hot deboned pig bellies under experimentally inadequate storage conditions (7°C). Especially Enterobacteriaceae were strongly inhibited in their development. In further studies (van Netten et al., 1984) it was found that LA decontamination effects a shift on the microflora towards the Gram positive population and provides a higher protection against the possible enteropathogenic Gram negative association. Spraying of hot carcasses at 45 minutes p.m. resulted in a greater effect than spraying of chilled carcasses. This is probably due to the fact that on hot carcasses the bacteria are still in the waterfilm and have not yet become attached to the surface (Notermans and Kampelmacher, 1983).

Table 1: The effect on the aerobic colony counts (ACC) of pig carcasses of cleaning and disinfection of the polishing machinery with or without an additional 1 % v/v lactic acid spray on the carcasses (applied 45 min p.m.).

Treatment	ACC in log N/cm²	
1. Irregular cleaning	4.2 ± 0.5 *	a
2. systematic foam cleaning	3.3 ± 0.3	b
3. Mechanisation of foam cleaning and disinfection	2.8 ± 0.3	c
4. as 3, and subsequently LA spray	2.3 ± 0.4	d

* Each mean value comprises 20 observations. From each carcass samples were pooled from the rind on the cheek, the breast and the back. The values with different superscripts differ significantly ($p < .01$).

The breast of a cattle carcass is contaminated significantly more than the shoulder (Table 3). Therefore lactic acid decontamination is most effective at this spot (Snijders et al., 1979). After 3 days of chilling some delayed effect of lactic acid decontamination is still noticeable, although not as clearly as shown in Tabel 2.

Table 2: The effect of 5 % v/v lactic acid (applied 3 h p.m.) on aerobic (ACC) and Enterobacteriaceae colony counts (ECC) (log N/cm²) of hot deboned pig bellies stored at 7°C.

Days	0	2	3	6	8
ACC					
controls	4.7 ± 0.4	7.4 ± 0.2	8.0 ± 0.1	8.7 ± 0.3	9.2 ± 0.1
treated	3.8 ± 0.4	4.0 ± 0.7	5.2 ± 1.3	6.4 ± 0.7	7.6 ± 0.5
Δ	0.9	3.4	2.8	2.3	1.6
ECC					
controls	2.7 ± 0.5	3.1 ± 1.5	4.6 ± 0.6	6.0 ± 0.5	5.5 ± 0.4
treated	< 1.3	< 1.3	1.7 ± 0.7	2.7 ± 1.5	3.3 ± 1.3
Δ	≥ 1.4	≥ 1.8	2.9	3.3	2.2

Δ significant reduction ($p < .01$) n=8

This delayed effect, which is probably the result of a prolonged lag phase of acid-injured micro-organisms surviving LA decontamination, can also be seen in veal carcasses during prolonged storage time (Table 4, Smulders and Woolthuis, 1984).

Experiments have shown the ultimate value for LA spraying which does not cause permanent discolouration of the meat to be 1 % v/v for beef and 1.25 % v/v for veal. However, immediately after LA spraying a slight discolouration occurs, which disappears after 1 day of chilling. The LA concentration may be slightly higher for veal than for beef because veal is paler. Nevertheless, LA concentrations higher than 1.25 % v/v will produce an undesirable discolouration of the subcutaneous fat cover on the carcass. However, after this fat has been trimmed, primal cuts may be treated with concentrations as high as 2 % v/v without causing discolouration (Smulders and Woolthuis, 1984). High concentrations of LA caused blood to coagulate leaving rusty brown spots, which are commercially unacceptable. To prevent this rusty brown discolouration the carcasses must be rinsed before treatment in order to remove these blood spots. For pork the ultimate LA concentration is also 1 % v/v. But if little liquid is used, for instance as a result of applying electrostatic spraying, concentrations of up to 1.5 % v/v can be used (Labots et al., 1983). The difference in LA concentrations resulting in minimal discolouration may be attributed to dilution of LA in the waterfilm covering pig carcasses. A 2.4 % v/v LA spray on pork carcasses gives clear discolourations, especially in the thorax, cavities, and locations in which lactic acid solutions are collected.

Table 3: The effect on the bacteriological condition of cattle carcasses of a 1 % v/v lactic acid spray (applied 45 min. p.m.)

	Aerobic colony count		Enterobacteriaceae colony count	
Days	0	3	0	3
Breast				
controls	4.7 ± 0.4	5.3 ± 0.9	2.0 ± 0.6	75%*2.1 ± 0.8
treated	2.9 ± 0.8	2.9 ± 0.7	< 1.3	< 1.3
Δ	1.8	2.4	≥ 0.7	≥ 0.7
shoulder				
controls	3.1 ± 0.7	3.9 ± 1.4	13 % 1.9	13 % 2.2
treated	2.5 ± 0.7	2.6 ± 0.8	< 1.3	< 1.3
Δ	0.6	1.3		

Δ significant reduction $p < .05$ (n=8)

* percentage of plates appropriate for enumeration from which means have been calculated (% : log N cm² ≥1.3)

Table 4: The effect on the bacteriological condition (log N/cm²) of veal carcasses of a 1.25 % v/v lactic acid spray (applied 45 min p.m.)

	Aerobic colony count		Enterobacteriaceae colony count	
Days	0	14	0	14
shoulder				
controls	3.2 ± 0.5	4.6 ± 1.2	24 %*1.9 ± 0.4	29 % 2.7 ± 0.2
treated	2.5 ± 0.5	3.3 ± 0.8	6 % 1.3	6 % 2.2
Δ	0.7	1.3	≥ 0.6	≥ 0.5

Δ significant reduction ($p < .01$) n=17

* percentage of plates appropriate for enumeration from which means have been calculated (%: log N cm² ≥ 1.3)

The pH of the surface of veal carcasses treated with 1.25 % LA decreases

significantly ($p < .01$) from 7.0 to 3.7. At 24 h post mortem the pH was 5.6, and after 72 h no significant difference in surface pH could be observed between treated and untreated carcasses.
Experiments with consumer taste panels have shown veal longissimus chops treated with 2 % v/v LA spray during 30 s not to be significantly different from the controls, whereas a treatment with 4 % v/v LA could be identified (Woolthuis and Smulders, 1984).

Besides spraying, cuts or organs can also be immersed in LA solutions (Table 5). However, if the immersion time is prolonged, the LA concentration has to be reduced to inhibit discolouration (Woolthuis et al., 1984). Spraying veal brains with 1.25 % v/v LA gave a small immediate effect (reduction of 0.3 log N per g), but should be discouraged because after one week of storage brains showed an inacceptable discolouration, whereas the bacterial load was not significantly different from that of controls (Smulders et al., 1984).

Table 5: Combined effects of decontamination by immersion in 0.20 % v/v lactic acid for 5 min followed by vacuum packing on the bacteriological condition at the surface of porcine liver (log N/cm²)

Days	Aerobic colony count		Enterobacteriaceae colony count	
	1	5	1	5
Controls	4.4 ± 0.4	5.2 ± 0.5	2.1 ± 0.6	2.2 ± 0.6
treated	2.2 ± 0.3	2.4 ± 0.6	17 %*1.7 ± 0.1	17 %1.5 ± 0.1
Δ	2.2	2.8	≥ 0.4	≥ 0.7

Δ significant reduction ($p < .001$) n=12

* percentage of plates appropriate for enumeration from which means have been calculated (%: log N/cm² ≥ 1.3)

According to EEC regulations the temperature of water used for cleaning and disinfection of tools during slaughtering has to be at least 82°C (180°F). Immersing a knife for 15 sec in water of 82°C effects a reduction of 2.2 log N, which is significantly lower than the reduction obtained in the disinfection unit with LA (Table 6). Disinfection of knives at 82°C is far from optimal if it is not preceded by mechanical cleaning. The latter is possible if one uses a disinfection unit in which water is sprayed onto the knifesurface through 2 nozzles (Snijders et al., 1984). By adding LA to the water its temperature may be reduced, and disinfection by spraying resulted in a better effect than was obtained in standing water.

Table 6: The effect of cleaning and disinfection of knives in a disinfection unit on the aerobic colony count with or without 2.7% v/v lactic acid and a pressure of 15 ato at different temperatures and spraying times

time S	temperature °C	reduction Δ log N	reduction with LA Δ log N
5	20	0.2* a	2.6 b
15	20	0.4 a	3.2 d
5	45	0.7 b	3.1 d

* Each mean value comprises 20 observations
The values with different superscripts differ significantly ($p < .01$). The mean level of contamination on the knives which were used as controls was 3.3 ± 0.2 log N.

Conclusion

The use of LA as a terminal decontaminant linked to perfect slaughter line hygiene could bring important advantages. LA produces both an immediate (bactericidal) and a delayed (bacteriostatic) effect, which results in an extended shelf life of meat. The level of contamination with enteropathogenic micro organisms may be reduced through an increased suppression of the Gram negative bacteria. However, the use of lactic acid as a decontaminant in the meat industry must never result in neglect of hygiene in slaughtering and processing lines.

References

Barendsen, P., Smulders, F.J.M., Woolthuis, C.H.J. en Korteknie, F. Enkele chemische en fysiologische aspecten van produktie en gebruik van melkzuur. Een literatuuroverzicht. VVDO-rapport 8401, 1984.

Gerats, G.E., Snijders, J.M.A., Van Logtestijn, J.G. Slaughter techniques and bacterial contamination of pig carcasses. Proc. 27th Eur. Meet. Meat. Res. Work, Vienna 1: 198-200, 1981.

Gerats, G.E., Steensma, H.O., Tazelaar, F. Arbeidsomstandigheden en arbeidsvoldoening in Nederlandse varkensslachterijen. Rijks Universiteit Utrecht, 1982.

Labots, H., Logtenberg, H., Stekelenburg, F.K., Snijders, J.M.A. Onderzoek naar de mogelijkheid om onder bedrijfsomstandigheden het aantal Salmonella bacteriën op het oppervlak van varkenskarkassen te verminderen. CIVO-rapport T83.183, 1983.

Netten, P. van, Mossel, D.A.A. The ecological consequences of decontaminating raw meat surfaces with lactic acid. Arch. Lebensm. Hyg. 31, 190-191, 1980.

Netten, P. van, Zee, H. van der and Mossel, D.A.A. A note on catalase exchanced recovery of acid injured cells of gram negative bacteria and its consequences for the assessment of the lethality of L-lactic acid decontamination of raw meat surfaces. J. Appl. Bacteriol. in press, 1984.

Notermans, S., Kampelmacher, E.H. Haften von Bakterien bei der Fleischverarbeitung, 63, 83-88, 1983.

Smulders, F.J.M., Woolthuis, C.H.J. Influence of two levels of hygiene on the microbiological condition of veal as a product of two slaughering/Processing sequences. J. Food Prot., 46, 1032-1035, 1983.

Smulders, F.J.M., Woolthuis, C.H.J. The immediate and delayed microbiological effects of lactic acid decontamination on calf carcasses. J. Food Prot. in press, 1984.

Smulders, F.J.M., Korteknie, F., Woolthuis, C.H.J., Snijders, J.M.A. The effect of lactic acid decontamination and frozen storage on the keeping qualities of calf brain. Proc. 30th Eur. Meat. Meat Res. Work Bristol,... 1984.

Snijders, J.M.A. Pig slaughtering hygiene. Vet. Med. Diss. 1976. Utrecht.

Snijders, J.M.A., Schoenmakers, M.J.G., Gerats, G.E., Pijper, F.W. de. Dekontamination schlachtwarmer Rinderkörper mit organischen Säuren. Fleischwirtschaft, 59, 656-663, 1979.

Snijders, J.M.A., Janssen, M.H.W., Gerats, G.E., Corstiaensen, G.P. The need for a good bacteriological sampling technique in slaughter lines. International J. of Food Microbiol., in press, 1984.

Snijders, J.M.A., Janssen, M.H.W., Corstiaensen, G.P., Gerats, G.E. Reiniging en desinfektie van messen in de vleesindustrie. Vleesdistrib. en Vleestechnol., 4, 10-12, 1984.

WHO 1983. Guidelines on prevention and control of salmonellosis. WHO Document VPH/8342.

Woolthuis, C.H.J., Mossel, D.A.A., Logtestijn, J.G. van, Kruijf, J.M. de Smulders, F.J.M. Microbial decontamination of porcine liver with lactic acid and hot water. J. Food Prot., 47, 220-226, 1984.

Woolthuis, C.H.J. and Smulders, F.J.M. Microbial decontamination of calf carcasses by lactic acid sprays. J. Food Prot., in press, 1984.