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# Short communication

# Effects of sodium lactate and other additives in a cooked ham product on sensory quality and development of a strain of *Lactobacillus curvatus* and *Listeria monocytogenes*

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## Abstract

Cooked cured ham products were produced according to a standard recipe for cooked ham with various levels of sodium lactate, sodium diacetate or buffered sodium citrate. They were compared with a reference ham product with respect to sensory quality and growth of *Lactobacillus curvatus* and *Listeria monocytogenes*. For this, a part of the products was sensory analysed directly after preparation. Another part of the cooked ham products was minced and homogeneously inoculated with *L. curvatus*  $(10^4/g)$  and *L. monocytogenes*  $(10^2/g)$  and filled in 60-g plastic pouches. After vacuum packaging, the pouches were stored at 4°C for up to 40 days.

Between the different ham compositions, only minor differences were found for appearance, internal colour, structure and firmness. The addition of 0.2% Na-diacetate had a negative effect on the odour and taste of the ham product. The addition of 2.5% to 3.3% Na-lactate inhibited the growth of *L. curvatus* compared to the reference, while 0.1% and 0.2% Na-diacetate did not. *L. monocytogenes* was best inhibited by the addition of Na-lactate but also by the addition of 0.2% Na-diacetate. On the other hand, the growth of *L. monocytogenes* was stimulated by the addition of 1% buffered Na-citrate. © 2001 Elsevier Science B.V. All rights reserved.

Keywords: Sodium lactate; Lactobacillus curvatus; Listeria monocytogenes

# 1. Introduction

Cooked cured ham is a popular meat product of which an important part is sliced and prepackaged in vacuum or gas atmosphere to be sold with a "sellby-date" of 3 to 4 weeks at 7°C. Despite stringent hygienic conditions during finishing, handling and slicing of the cooked ham, it is almost impossible to avoid a minor contamination. From this general contamination, it will be mainly lactic acid bacteria that preferably develop under the anaerobic circumstances in the sliced prepackaged cooked meat product at temperatures below 7°C (Muermans et al., 1993, 1994). Above numbers of 10 million/g, these bacteria can be responsible for sensory deviations, like souring and gas formation. Beside lactic acid bacteria, pathogenic *L. monocytogenes* bacteria are also capable to develop under these conditions. Although *L. monocytogenes* are mostly found in very

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low numbers in only 1% to 2% of cooked meat products directly after packaging, they can form a significant health hazard when they grow out during storage of the product. Recent cases of food poisoning, linked to meat products, demonstrate the importance to incorporate extra safety hurdles in these type of products (Anon, 1999; Ryser and Marth, 1999).

The growth rate of lactic acid bacteria and L. monocytogenes is dependant mainly on the water activity  $(a_{w})$  of the product and the storage temperature and to a smaller extent on the amount of nitrite in the product (Muermans et al., 1994). The  $a_{\rm w}$  is determined largely by the amount of sodium chloride added to the meat product. In literature, sodium lactate is also mentioned as water activity decreasing additive (Brewer et al., 1991; Chen and Shelef, 1992: Miller and Acuff, 1994: Papadopoulos et al., 1990). An additional inhibitory effect of lactates has been reported as well (Blom et al., 1997: Debevere, 1989; Houtsma et al., 1993; Shelef and Yang, 1991; Shelef, 1994; Stillmunkes et al., 1993; de Wit and Rombouts, 1990). Furthermore, antimicrobial activity of sodium salts of other short-chain organic acids, like citric acid, acetic acid and combinations of these salts have been reported (Buchanan et al., 1993; Schlyter et al., 1993; Shelef and Addala, 1994; Shelef et al., 1997). All these substances are generally recognised as safe (GRAS) by the U.S. Code of Federal Regulations, 21 CFR. However, published studies on the antimicrobial activity of these substances on L. monocytogenes in cooked cured meat products are limited (Blom et al., 1997; Ovist et al., 1994; Shelef and Addala, 1994; Wederquist et al., 1994: Weaver and Shelef, 1993).

To obtain more information about the effect and the practical application, a comparative study was carried out with cooked ham products with Nalactate, Na-diacetate and buffered Na-citrate as additives.

# 2. Materials and methods

## 2.1. Product preparation

A general recipe of a Dutch industrial cooked-inthe-bag ham was used as a reference. The chemical composition of the raw ham used was 70% moisture, 7.5% fat and 20.5% protein. The basic composition of the brine was 64% moisture, 12.5% starch, 10% colorozo, 10% malto dextrine, 2.5% phosphate, 0.5% sodium ascorbate and 0.5% sodium glutamate. The proposed ratio ham:brine was 80:20. Based on these estimations, the calculated moisture/protein ratio of the end product will be 4.3.

Apart from the reference, five batches of cooked ham products were prepared with the following additives: 2.5% Na-lactate (60% syrup, Purasal, PURAC Biochem), 3.3% Na-lactate, 0.1% Na-diacetate, 0.2% Na-diacetate or 1% buffered Na-citrate (15 parts Na-citrate, 1 part citric acid w/w), respectively.

For each composition, five cooked-in-the-bag hams of approximately 5 kg were prepared according to a standard production process. One lot of 150-kg raw hams was deboned, defatted and demembraned, then cut into small pieces of circa 300 g. These pieces were randomly divided over six batches of 25 kg and stored at 0°C. Six brines were made according to the recipe mentioned above with the different test substances being added, respectively. In order to maintain a total of 100%, the amount of water in the brine was reduced with the weight of the test substance added in every separate occasion. After preparation, the brines were stored at 0°C. At production, each brine was injected in a batch of raw mechanically tenderized ham in the ratio mentioned. Each batch was tumbled separately overnight and filled in five vacuum shrink foil bags. The bags were put into moulds and pasteurised in a steam cabinet during 6 h and 10 min at 72°C, resulting in a temperature of 70°C in the coldest spot for 5 min. After cooling with cold water, the hams were stored at 0°C until further examination.

#### 2.2. Chemical analyses

The pH of the cooked ham products was measured using a Consort pH-meter type D214, with a combined glass electrode (Scott Geräte, type N 5800A). Water activity ( $a_w$ ) values were determined at 25°C with a Novasina electric hygrometer (type ER84/3H/63T with sensors enBSK-4). The formulation of the different ham compositions was checked by analysing the amounts of moisture, fat, protein, salt, nitrite, carbohydrates, phosphate and Na-lactate or acetic acid as required. The brine percentage in the final ham product was calculated as:

Brine percentage =  $\frac{\text{Salt\%}}{\text{Salt\%} + \text{moisture\%}} 100.$ 

#### 2.3. Sensory analyses

After preparation, the ham products with different additives were sensory analysed by an experienced panel of five members. The panelists were asked to judge the products for characteristics: appearance, colour, structure, firmness, odour and taste.

## 2.4. Challenge testing

Another part of the cooked ham products was inoculated with both L. curvatus (code LAB 962) and L. monocytogenes, type A (ATCC 19114). One ham of each composition was placed in the bowl of a disinfected laboratory cutter (Stephanal, type FA 30). cut into small pieces and inoculated with a suspension of L. curvatus and L. monocytogenes bacteria to a final level of about  $10^4$  and  $10^2/g$  product, respectively. After inoculation of the ham, the product was minced and homogenized for 2 min. Subsequently, the minced product was divided into 30 portions of 60 g and vacuum packaged in plastic pouches with an oxygen permeability of less than  $1.5 \times 10^{-11}$  m<sup>3</sup> m<sup>-2</sup> Pa<sup>-1</sup> day<sup>-1</sup> at 20°C. The packages obtained were stored at 4°C for up to 40 days. During the experiment, the temperatures were registered using a Temptimem<sup>®</sup> data logger.

#### 2.5. Microbiological analyses

At appropriate time intervals, 20 g of minced ham product was taken aseptically from a single package, diluted 10-fold in physiological peptone saline (PPS) and homogenised in a stomacher for 1 min. Additional serial dilutions were made in PPS. Numbers of *L. curvatus* were estimated by pour plating using de Man-Rogosa-Sharpe Agar (MRSA, Oxoid CM 359). Plates were incubated anaerobically (BBL, Gaspak plus) at 30°C and counted after 3 days. Numbers of *L. monocytogenes* were determined by pour plating using Palcam agar (Oxoid CM 877 and SR150E). Plates were incubated at 37°C for 2 days. Occasionally, presumptive colonies developed on Palcam agar were confirmed by catalase reaction, dark field microscopy and production of  $\beta$ -haemolysine. Final identification was performed with the *Listeria* identification system of Medvet, Microbact 12L strip.

# 3. Results and discussion

## 3.1. Chemical composition

In Table 1, the results are given of the chemical analyses of the cooked ham products with different additives. The overall results show that the intended composition of the different cooked ham products was reasonably approximated, considering the small-scale production process. However, the fat content of all products was lower than expected, resulting in somewhat higher moisture and protein contents. The brine percentage of the reference product was a little low compared to the other compositions. This was also expressed in the relative high  $a_w$ -value of this product.

The reference ham product contained 0.18% acetic acid and 0.76% Na-lactate. These amounts are naturally present in meat. The determined amounts of acetic acid in the product agreed fairly well with the amounts aimed at, considering the recovery rates of 94% and 106%, respectively. The recovery rates of the added amounts of Na-lactate in the product were 78% and 101%. The addition of Na-diacetate as well as buffered Na-citrate slightly lowered the pH values of the final cooked products. These lower pH values probably also account for the lower nitrite levels found in these products.

# 3.2. Sensory quality

The results of the sensory analysis of the different cooked ham products are summarised in Table 2. Between the different ham compositions, only minor

Table 1			
Chemical analyses of prepared	ham products	with different add	litives

Ham product	Reference	With 2.5% Na-lactate	With 3.3% Na-lactate	With 0.1% Na-diacetate	With 0.2% Na-diacetate	With 1% buffered Na-citrate
Moisture (%)	73.9	72.4	72	74.2	73.9	72.7
Fat (%)	2.5	3.1	2.8	2.8	2.5	1.9
Protein	18.1	16.9	16.7	16.9	17.4	17.8
(% total N $\times$ 6.25)						
Carbohydrates	4.1	4.4	4.6	4.2	4.5	4.8
(% as glucose)						
Total phosphate	0.64	0.66	0.71	0.71	0.79	0.70
(% as $P_2O_5$ )						
Salt (% as NaCl)	2.0	2.2	2.3	2.2	2.2	2.2
Brine (%, calculated)	2.6	2.9	3.1	2.9	2.9	2.9
Nitrite (mg/kg)	11	11	11	6	4	3
Acetic acid (%)	0.18	n.d.	n.d.	0.26	0.35	n.d.
Sodium lactate (%)	0.76	1.93	2.76	n.d.	n.d.	n.d.
Moisture/protein ratio	4.1	4.3	4.3	4.4	4.2	4.1
$a_{\rm w}$ -value at 25°C	0.977	0.969	0.961	0.973	0.972	0.974
pH	6.2	6.2	6.1	6.0	5.9	6.0

n.d. = not determined.

differences were found for appearance, internal colour, structure and firmness. The structure of the product with Na-citrate was rather irregular and the firmness of the products with 3.3% Na-lactate and 0.1% Na-diacetate was slightly tough. All of these deviations were considered to be within the normal variation of this type of product.

No significant differences (P < 0.05) of odour and taste were observed between the reference ham product and products with 2.5 Na-lactate, 3.3% Nalactate, 0.1% Na-diacetate and buffered Na-citrate, respectively. However, significant deviations were found for the product with 0.2% Na-diacetate.

## 3.3. Effects on bacterial growth

The results of the growth of *L. curvatus* are graphically displayed in Fig. 1. The inoculated *L. curvatus* grew well in all different ham products. The cut off level of  $10^7$  cfu/g was reached within 2 to 2.5 weeks in the reference product and the products containing 0.1% and 0.2% Na-diacetate or 1%

Table 2					
Sensory assessment	of ham	products	with	different	additives

Ham product	Ratings (scale $10 =$ excellent to $1 =$ unacceptable)							
	Reference	With 2.5% Na-lactate	With 3.3% Na-lactate	With 0.1% Na-diacetate	With 0.2% Na-diacetate	With 1% buffered Na-citrate		
Appearance	9	8.5	8.5	9	8.5	9		
Internal colour	8	8	8.5	8.5	8	8		
Structure	8.5	8	8.5	8.5	8.5	7.5		
Firmness	8.5	8.5	7	7	7.5	8.5		
Odour	8	8	8.5	7.5	6.5 *	8.5		
Taste	7.5	7.5	7	7	6.5 *	8.5		

\* Significantly lower.

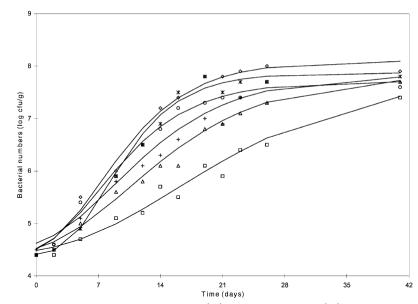


Fig. 1. Growth of *L. curvatus* in ham product with 2.5% Na-lactate ( $\triangle$ ), with 3.3% Na-lactate ( $\square$ ), with 0.1% Na-diacetate ( $\diamondsuit$ ), with 0.2% Na-diacetate (+) and with 1% buffered Na-citrate (**X**) versus the reference product ( $\bigcirc$ ) at 4°C.

buffered Na-citrate, and in 3 and 5 weeks for products containing 2.5% and 3.3% Na-lactate, respectively. In the reference product, *L. monocytogenes* showed a one-log increase in numbers during the total storage period (Fig. 2). This indicates that a cooked ham product containing salt and nitrite is not an optimal growth medium for the *L. monocytogenes* strain tested. In the products containing Na-lactate or 0.2% Na-diacetate, *L. monocytogenes* did not grow. However, in the ham product containing buffered Na-citrate, *L. monocytogenes* growth was observed, reaching maximum numbers of  $10^6$  cfu/g within 3 weeks storage at 4°C.

The maximum specific growth rate ( $\mu_{max}$ , in h<sup>-1</sup>), the lag time ( $\lambda$ , in h) and the maximum cell number ( $N_{max}$ , in cfu/g) of the growth curves, modelled using the modified Gompertz equation (Muermans et al., 1993), are presented in Table 3. These results show that the presence of Na-lactate in the ham product decreased the growth rate and increased

Table 3

Estimates of the maximum specific growth rate ( $\mu_{max}$ , in h<sup>-1</sup>), the lag time ( $\lambda$ , in h) and the maximum cell number ( $N_{max}$ , in cfu/g) at 4°C using the modified Gompertz equation

Ham product	$a_{\rm w}$ -value	Inoculum	Growth curve parameter		
			$\mu_{\rm max}$ (h <sup>-1</sup> )	$\lambda$ (h)	N <sub>max</sub> (cfu/g)
Reference	0.977	L. curvatus	0.020	28	$5.2 \times 10^{7}$
With 2.5% Na-lactate	0.969	L. curvatus	0.014	41	$6.7 \times 10^{7}$
With 3.3% Na-lactate	0.961	L. curvatus	0.010	89	$6.6 \times 10^{7}$
With 0.1% Na-diacetate	0.973	L. curvatus	0.023	36	$1.3 \times 10^{8}$
With 0.2% Na-diacetate	0.972	L. curvatus	0.016	34	$6.8 \times 10^{7}$
With 1% buffered Na-citrate	0.974	L. curvatus	0.027	81	$7.4 \times 10^{7}$
	0.974	L. monocytogenes	0.041	153	$2.3 \times 10^{6}$

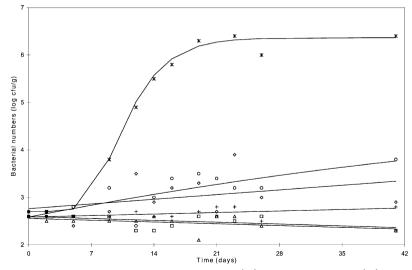


Fig. 2. Growth of *L. monocytogenes* in ham product with 2.5% Na-lactate ( $\triangle$ ), with 3.3% Na lactate ( $\Box$ ), with 0.1% Na-diacetate ( $\diamond$ ), with 0.2% Na-diacetate (+) and with 1% buffered Na-citrate ( $\mathbf{X}$ ) versus the reference product ( $\bigcirc$ ) at 4°C.

the lag time of *L. curvatus*. The shelf life of cooked ham product at 4°C can be more than doubled by the addition of 3.3% Na-lactate. The addition of buffered Na-citrate seemed not to influence the growth of *L. curvatus*. In ham products containing 0.1% or 0.2% Na-diacetate, growth of *L. curvatus* was almost comparable to the reference product.

Growth of *L. monocytogenes* was observed only in the product with buffered Na-citrate, with growth rates being even faster than the growth rate of *L. curvatus*. On the contrary, the lag time of *L. monocytogenes* was approximately twice as long compared to *L. curvatus*. The reason for the observed growth stimulating effect of Na-citrate on *L. monocytogenes* is not clear yet, but will be subjected to further study.

In summary, the addition of sodium lactate to a cooked ham product, in amounts between 2.5% and 3.3%, showed a shelf life extending effect, without clear sensory deviations. In addition, the development of *L. monocytogenes* was inhibited in the presence of sodium lactate. The addition of sodium diacetate in amounts above 0.1% influenced the sensory quality of the ham product adversely, while shelf life was not markedly increased compared to the reference.

Although the addition of 1% buffered sodium citrate did not show any adverse effect on sensory

properties, the growth of the *L. monocytogenes* strain tested was stimulated by this additive.

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