Products for weaners
Benzoic acid or the combination of lactic acid and formic acid

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Hanne Maribo, Lars Egelund Olsen, Bent Borg Jensen[1] and Nuria Miquel[1]
The National Committee for Pig Production,
DANISH BACON AND MEAT COUNCIL

Abstract

The following has been tested for weaners in the age 4-10 weeks: the combination of 0.7% formic acid and 0.7% lactic acid in diet 1 (4-6 weeks) and diet 2 (6-10 weeks), and benzoic acid with 2% added to diet 1 and 1% added to diet 2. The acid products were compared to a control mix. The test was carried out in one herd with 160 pigs per group divided into 20 blocks (replicates) per treatment.

Mortality in the test averaged 1.5%, and 2.5% was on average removed due to disease. Treatment frequency for diarrhoea was significantly lower for the pigs given benzoic acid compared to the other two groups. Prevalence of diarrhoea did not differ between the control group and the group given the combination of lactic and formic acid.

Production value per pig (4-10 weeks) for the individual mixes was calculated on the basis of the achieved production results using the same price per FUp for all feed mixes.

Overall the test showed that the pigs given feed containing benzoic acid had a significantly higher production value than the control pigs and the pigs given feed containing the combination of lactic acid and formic acid. Furthermore, the group with the combination of formic and lactic acid had a significantly higher production value than the control pigs. The benzoic acid used in this test is a food quality and therefore fairly expensive, and the achieved increase in productivity could not pay for the product. However, the calculation does not include costs for medication in connection with treatments for diarrhoea. The group with the combination of lactic and formic acid had a production value level with the control group when the prices of the acids are included in the calculations.

Microbiological studies revealed that adding benzoic acid to the feed significantly reduces the studied populations of micro organisms and the microbial activity in the entire gastro-intestinal tract. This causes a reduction in the production of organic acids in the intestine. This applies to both the control group and the group with the combination of lactic and formic acid. The bacteriological results did not differ between the control group and the pigs given feed added lactic and formic acid. However, this group had a lower content of lactobacillus in the stomach.

Presently, benzoic acid is not approved as an additive/preservative for pig feed, however the acid and some of its salts may be used for preserving food. Several of the salts from benzoic acid are also approved as preservatives for pet food. Lactic acid and formic acid are both approved as preservatives for pig feed.

[1] The Danish Institute of Agricultural Sciences.

Background

In the efforts to find alternatives to antibiotic growth promoters, the National Committee for Pig Production has tested various pure acids for weaner feed. A test with acid dosage has previously been carried out where formic acid in doses of 0.7% and 1.4% and lactic acid in doses of 0.7, 1.4 and 2.1% (Report no. 469) were tested. The test did not result in an effect on production results, but the microbiological studies revealed that adding lactic acid and formic acid to the feed had a positive effect on the gastro-intestinal system's micro flora and acid content. On the basis of the results from these microbiological studies, a combination of 0.7% formic acid and 0.7% lactic acid in weaner feed was tested. The reason for choosing this combination was to achieve a high concentration of lactic acid in the gastro-intestinal tract. The background for this choice was that the results from the test of acid dosages showed that adding formic acid to the feed caused a reduction in the number of yeast in the gastro-intestinal tract, and yeast uses lactic acid when growing. Addition of lactic acid caused an increase of the number of lactic acid bacteria and thereby the production of lactic acid in the gastro-intestinal tract. It has been observed that an increased content of lactic acid has a reducing effect on the number of coli bacteria in the gastro-intestinal tract. Benzoic acid was chosen as it has shown positive results in an in vitro test conducted by the Danish Institute of Agricultural Sciences.

The aim of this test was to examine whether addition of a combination of formic acid and lactic acid or addition of benzoic acid to diet 1 (4-6 weeks) and diet 2 (6-10 weeks) affected production value. The effect of the products was primarily measured on production value calculated on the basis of the production results: daily gain and feed conversion. Secondarily the effect was measured on the number of disease treatments.

Materials and methods

The test was carried out at Experimental station Grønhøj. All pens were equipped with concrete slatted floor in approx. two thirds of the pen and solid floor in the last third by the trough, and there was one nipple drinker per pen. 160 pigs were penned in all groups divided into 20 blocks (replicates). Post-weaning at the age of four weeks the pigs were initially divided according to size and hereafter randomly divided in the three groups so the groups within each block (replicate) were identical re. number of pigs per pen and weight. Seven, eight or nine pigs were penned in each pen. Average weight on penning and finish was 7.5 kg and 27 kg, respectively.

Table 1. Test design

Group	1	2	3
Diet 1	Control feed	0.7% lactic acid and 0.7% formic acid	2.0% benzoic acid
Diet 2			1.0% benzoic acid

The diets in all groups were optimised so the content of the amino acids: lysine, methionine, cystine, threonine and tryptophan was 5% above the present standard to ensure against movements in the content of crude protein of the ingredients. The feed's content of other nutrients met the present standards (cf. FOKUS PÅ Normer for Næringsstoffer, The National Committee for Pig Production, 6th edition, 1998). The diets were produced at Aarhusegnens Andel in Malling, and were heat-treated and pelleted at a minimum temperature of 81 degrees Celsius. The composition of the feed and a more detailed description of the added products can be seen from appendices 1 and 2.

At each delivery of feed, all feed mixes were analysed for FUp, energy content (enzyme-digestible organic matter), crude protein and for the amino acids: lysine, methionine, cystine, threonine and for calcium, phosphorus and zinc. Diet 1 in all groups was screened for antibiotics. Appendix 3 shows the analysis results.

In all groups feed was given once a day. All pigs had access to feed and water 24 hours a day. The first two weeks post-weaning the pigs were given diet 1. From 6 to 10 weeks they were given diet 2. In both periods feed intake, gain, disease treatments and mortality were registered.

Microbiological studies

The microbiological studies comprised eight pigs per test treatment, a total of 24 pigs. They were removed 14 days post-weaning. Pigs from eight litters were used (three pigs per litter), ie. one pig from each litter in each treatment. The pigs were anaesthetized and collected at Grønhøj at approx. 9 o'clock and put down with pentobarbital at Research Centre Foulum between 10 and 11 o'clock. After being put down, the gastro-intestinal tracts were immediately dissected free and divided into eight segments: stomach ("stomach"), three identical sections of the small intestine ("small 1-3"), caecum ("caecum"), and three identical sections of the large intestine (large 1-3). Total content in each of the eight sections was removed and weighed, and pH was immediately measured. Weight and length of the intestinal tract were registered as were pathological lesions in the pars proventricularis. Samples from gastro-intestinal content from all eight sections were analysed for content of dry matter, concentration of lactic acid, formic acid and other volatile fatty acids (VFA): butyric acid, propionic acid and ethanoic acid.

The composition of the micro flora was examined in four sections of the gastro-intestinal tract: stomach ("stomach"), the last third of the small intestine ("small 3"), caecum ("caecum") and the middle of the large intestine ("large 2"). Analyses were made of the content of total anaerobic bacteria, coliform bacteria, lactose-negative enterobacteria, lactic acid bacteria, lactobacillus (constituting part of the lactic acid bacteria), enterococci, and yeast cells.

Calculations

Production value was calculated as: (kg gain x DKK per kg gain) – (number of analysed FUp x DKK per FUp). Value of gain (DKK6.00 per kg gain) was calculated partly on the basis of the average weight on penning and transfer in the entire test and partly on the basis of the average price in the last five years (September 1, 1995 to September 1, 2000) for 7 kg's pigs (DKK228 per pig, +/- DKK7.16 per kg) and 30 kg's pigs (DKK364 per pig, +/- DKK4.98 per kg). The feed prices are averages of the last five years (September 1, 1995 to September 1, 2000), diet 1: DKK2.11 per FUp, and diet 2: DKK1.52 per FUp.

A calculation (including the price of the product) was made on the basis of the last five weeks' prices (weeks 40-45, 2000). Value of gain (DKK6.18 per kg gain) was here calculated on the basis of the following prices: 7 kg's pigs (DKK240 per pig, +/- DKK7.75 per kg) and 30 kg's pigs (DKK381 per pig, +/- DKK5.40 per kg), diet 1: DKK2.42 per FUp, and diet 2: DKK1.46 per FUp. In the calculation of the actual production value, the feed price is added the price of the product.

Production value was analysed as primary parameter with weight on penning as co-variable, and the model comprised the following variables: block, housing unit, and group. Registrations of disease were analysed as secondary parameter.

Data were subjected to an analysis of variance in SAS under the GLM procedure. Significant differences are stated at 5% level adjusted for three comparisons in pairs (all groups with each other) at a Bonferroni t-test. The results are shown as averages for each group adjusted to the same weight on penning.

Data for examination of the gastro-intestinal tract were subjected to an analysis of variance in SAS under the GLM procedure. Adjustments were made for sex, block and litter. Treatments were compared mutually and to the control block. Significant differences are stated at 5% level.

Results and discussion

Feed analyses

Analyses of diet 1 (4-6 weeks) revealed lower contents of energy, crude protein and thereby amino acids than calculated. Except for the content of energy in the control feed, the contents of energy, protein and amino acids in diet 1 were within the latitude of Danish feedstuff legislation. Contents of energy, protein and amino acids were at the same level in the three groups, and therefore these deviations are not thought to have influenced the test result.

The analysis in the control group revealed a zinc content of 317 mg/kg, ie. higher than the allowed 250 mg/kg. A deviation of this size is not estimated to influence the prevalence of diarrhoea.

Analyses of diet 2 (6-10 weeks) corresponded well with the calculated content. The screening analysis for antibiotic growth promoters in the feed did not show antibiotics in any of the diets.

Disease

Mortality in the test averaged 1.5%, and 2.5% was on average removed due to disease. There was no difference between the groups in the number of dead or removed pigs.

Treatment frequency for diarrhoea was significantly lower for the pigs given benzoic acid compared to the control group and the group given feed containing the combination of lactic and formic acid (table 2). There tended to be fewer diarrhoea treatments in the first two weeks post-weaning for the pigs given benzoic acid in the feed compared to the pigs given lactic acid and formic acid in the feed. Treatment frequency expresses how large a part of the pigs that has received treatment. Collective treatment of a pen via the feed for 2-3 days was made when more than 50% of the pigs in the pen had clinical symptoms of diarrhoea. Furthermore, individual treatments were made via injection.

Table 2: Frequency of diarrhoea treatments (4-10 weeks)

Group Product	1 Control	2 0.7% formic acid 0.7% lactic acid	3 2.0% benzoic acid in diet 1 1.0% benzoic acid in diet 2
Treatment frequency	117[a]	113[a]	60[b]

1) When comparing treatment frequency for diarrhoea between the groups there has to be a minimum difference of 41% units in order for there to be a significant difference.

Production results

The production results are stated for the periods 4-6 weeks, 6-10 weeks and for the entire test period (see table 3).

Table 3. Production results			
Group Product	1 Control	2 0.7% formic acid 0.7% lactic acid	3 2.0% benzoic acid in diet 1 1.0% benzoic acid in diet 2
No. blocks No. pigs penned	20 160	20 160	20 160
4-6 weeks Daily feed intake, FUp Daily gain, g FUp per kg gain	 0.22 117 2.02	 0.25 148 1.67	 0.25 138 1.96
6-10 weeks Daily feed intake, FUp Daily gain, g FUp per kg gain	 0.87 445 1.92	 0.90 476 1.89	 0.95 514 1.85
4-10 weeks Daily feed intake, FUp Daily gain, g FUp per kg gain	 0.66 343 1.92	 0.70 377 1.86	 0.73 397 1.85

Production value has been calculated on the basis of the achieved production results. Production value using five years' prices can be seen from table 4.

Table 4. Production value using 5 years' prices and 5 weeks' prices, respectively			
Group Product	1 Control	2 0.7% formic & 0.7% lactic acid	3 Benzoic acid 2.0% in diet 1 and 1.0% in diet 2
Production value using 5 years' prices[1]: DKK/pig Index	 54.1[a] 100	 60.0[b] 111	 65.2[c] 121
Actual production value using 5 weeks' prices incl. the price of the product[2]: DKK/pig Index	 58.2 100	 58.5 101	 55.7 96
[1] When comparing production value between one of the groups and the control group there has to be a minimum difference in production value of DKK5.5 or 10 index points in order for it to be significant. [2] **No** statistical calculations have been made of differences in gross margin using 5 weeks' prices.			

The pigs given the combination of lactic acid and formic acid in the feed had a significantly better production value than the control block. This was due to a higher daily gain before intermediate weighing and for the entire period (4-10 weeks) and a lower feed consumption for the entire period compared to the control group.

The pigs given benzoic acid had a significantly better production value compared to both the control group and to the pigs given the combination of lactic acid and formic acid. Compared to the control pigs, the pigs given benzoic acid had a higher feed intake, a higher daily gain, and lower feed consumption for the entire test period. Compared to the pigs given the combination of lactic and formic acid, the pigs given benzoic acid had a higher daily gain before and after intermediate weighing while the pigs given the combination of lactic and formic acid had a higher daily gain before intermediate weighing compared to the pigs given benzoic acid.

The calculation of actual prices is based on the last five weeks' average prices and feed prices added the cost of adding the product to the feed. The benzoic acid used in this test was food quality and therefore fairly expensive. The price of a feed quality will be lower. The achieved increase in production that was obtained by adding benzoic acid could not pay for the product. However,

the calculation does not include costs for medication in connection with treatments for diarrhoea. A diarrhoea treatment for five days for Coli diarrhoea costs approx. DKK1-4 per pig. The group where the feed contained the combination of lactic and formic acid has a production value level with the control group when the prices of the acids have been included in the feed price. The product prices (farmer's price) used can be seen from appendix 3 and have been supplied by the supplier.

Microbiological examinations

The results of these examinations include only the period until two weeks post-weaning as the pigs were slaughtered 14 days post-weaning before switching to diet 2.

Stomach lesions

The result shows no difference between the three groups regarding prevalence of stomach lesions; on average index was 2 for all groups. Ulcer examinations comprised only eight pigs per group and that is very few for this type of examination. However, the result may be used as an indicator of whether there are any problems in using the acid products in question.

pH

pH in intestinal content varied between the various intestinal sections. pH was increasing from the start of the small intestine and to the caecum. From caecum to the start of the large intestine pH drops and again increases towards the end of the large intestine (figure 1). There was no significant difference in pH between the groups in the various gastro-intestinal sections with the exception of a significantly lower pH in the caecum in the pigs fed the combination of lactic and formic acid compared to the control block (figure 1).

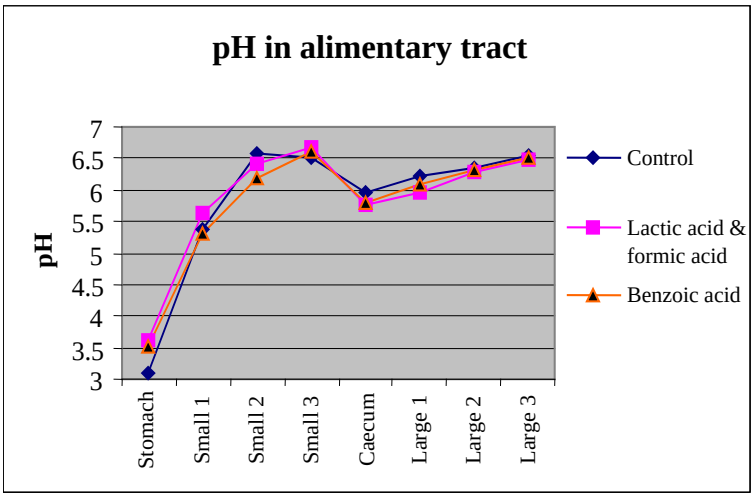

Figure 1. pH measured in the content of various sections in the alimentary tract.

Dry matter

The content of dry matter did generally not differ between the three groups for any of the examined intestinal sections. The control pigs had a significantly lower content of dry matter in the last part of the small intestine compared to the pigs fed the combination of lactic and formic acid.

Microbiology

The microbiological examinations comprise the number of lactic acid bacteria, lactobacillus, enterococci, coliform bacteria, lactose-negative entero bacteria, yeast, mould and total number of anaerobic bacteria in the stomach, last part of the small intestine, caecum and the middle of the large intestine.

Addition of 2% benzoic acid resulted in a significantly lower number of lactic acid bacteria in the stomach, the last part of the small intestine, caecum and the last part of the large intestine compared to the control feed and the group where the feed had been added 0.7% lactic acid and 0.7%

formic acid. The number of lactic acid bacteria did not differ between the control group and the group where the feed had been added 0.7% lactic acid and 0.7% formic acid (figure 2 and table 3 in appendix 4).

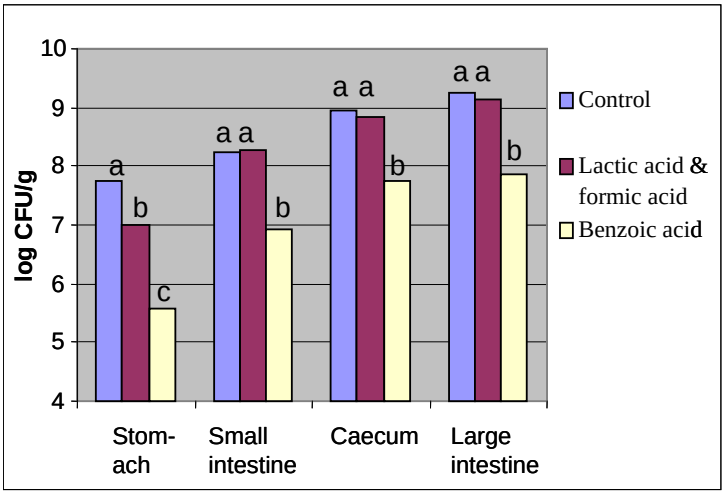

Figure 2. Content of lactic acid bacteria in the gastro-intestinal tract.

In the stomach the number of lactobacillus was highest in the control block and lowest in the group given benzoic acid. The pigs given the combination of lactic and formic acid were intermediate. The number of lactobacillus in the small intestine, caecum and large intestine was lowest in the group where the feed was added benzoic acid compared to the other two other groups (figure 3, table 3 in appendix 4).

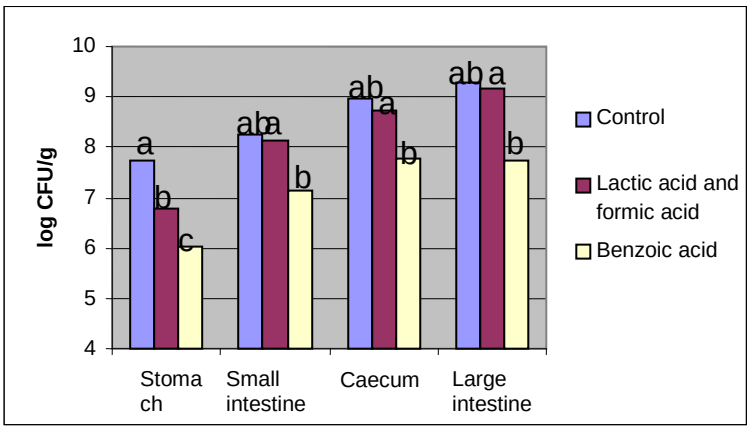

Figure 3. Content of lactobacillus in the gastro-intestinal tract.

The number of coliform bacteria did not differ between the groups in the various sections of the alimentary tract (figure 4, table 3 in appendix 4). Even though the amount of coli bacteria in the group where the feed was added benzoic acid was lower than the control group, the difference was not significant. The explanation may be that there was great variation between the pigs in the test and that the number of coliform bacteria in the gastro-intestinal tract generally was very low in this test. The number of coliform bacteria in the gastro-intestinal tract in this test was reduced by up to a factor 100 compared to the results in Report no. 469.

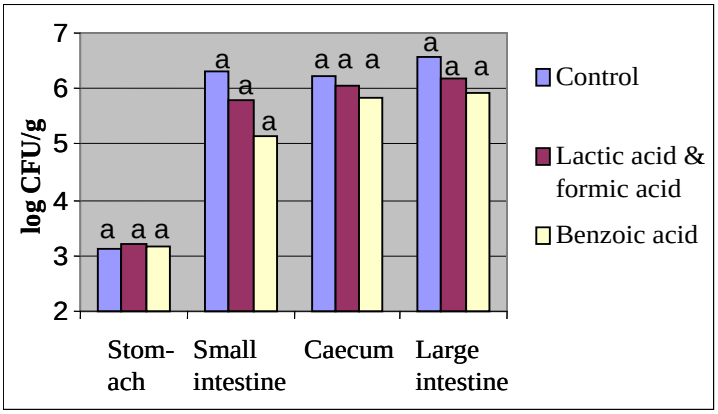

Figure 4. Content of coliform bacteria in the gastro-intestinal tract.

The number of yeast was significantly lower in the gastro-intestinal tract in the pigs fed benzoic acid compared to the other two groups. There was no difference in the number of yeast in the group fed the combination of lactic acid and formic acid compared to the control group (figure 5, table 2 in appendix 4).

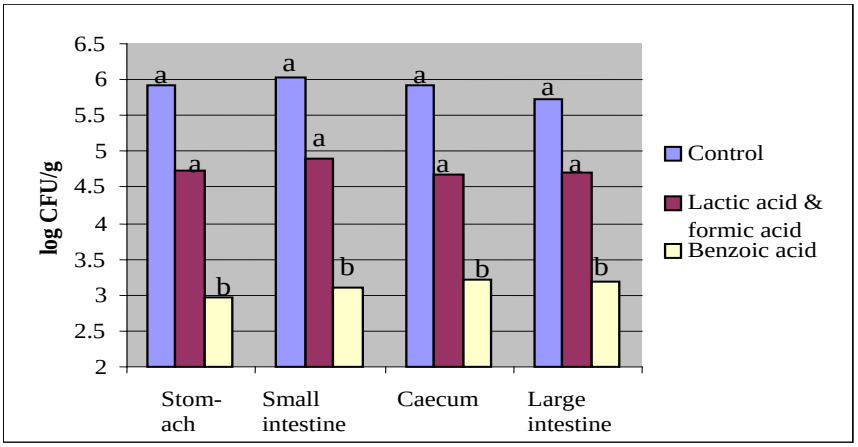

Figure 5. Content of yeast in the gastro-intestinal tract.

The total number of anaerobic bacteria in the stomach was significantly highest in the control group compared to the groups added acid. In the last section of the small intestine there was a significantly lower content of anaerobic bacteria in the pigs fed benzoic acid compared to the control group and the group fed the combination of lactic and formic acid. In caecum and large intestine there was no difference in the number of anaerobic bacteria between the groups (figure 6, table 3 in appendix 4).

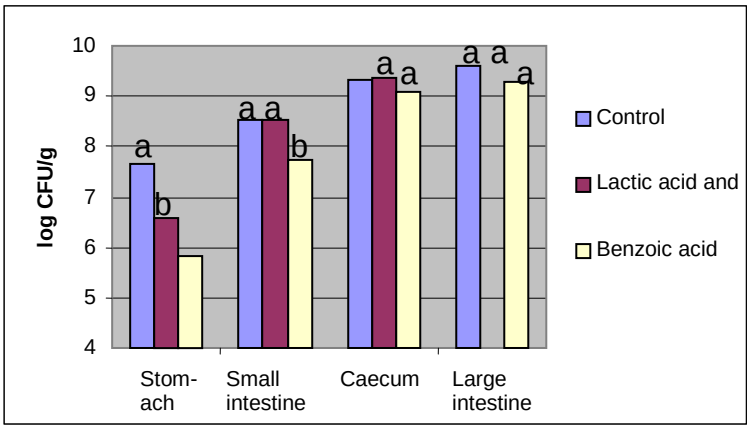

Figure 6. Total content of anaerobic bacteria in the gastro-intestinal tract.

Content of acid in the gastro-intestinal tract

Lactic acid, formic acid and benzoic acid

In the group given benzoic acid in the feed, the content of lactic acid is constant throughout the stomach and small intestine, whereas considerable amounts of benzoic acid were found in the stomach. Minor amounts of benzoic acid were also found in the entire small intestine of the pigs fed benzoic acid in the feed (figure 7, table 4 in appendix 4). The existence of benzoic acid in the small intestine may have influenced the microbiology in the small intestine. The content of lactic acid was the same in the control group and the group given lactic acid and formic acid in the feed. The content of lactic acid was lower in the last part of the small intestine in the pigs given benzoic acid in the feed compared to the other two groups. This corresponds to a lower content of bacteria in the small intestine in the pigs given feed added benzoic acid.

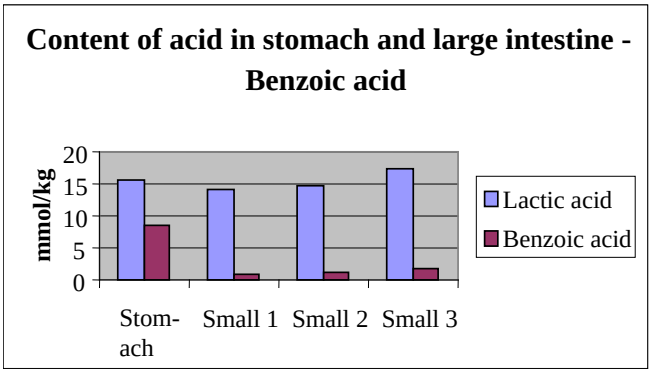

Figure 7. Content of lactic acid and benzoic acid in the gastro-intestinal tract in pigs fed feed added benzoic acid.

Volatile fatty acids (VFA)

The content of volatile fatty acids varied throughout the gastro-intestinal tract. The total content of VFA drops from the stomach and to the first section of the small intestine. The content then increases from the beginning to the end of the small intestine. Between the last part of the small intestine and the caecum, the content of VFA increases by 12% where the content is at a maximum, and then it drops throughout the large intestine (table 4 in appendix 4).

In the stomach, the content of VFA was significantly lower in the group fed benzoic acid compared to the other two groups. There was no difference between the control group and the group fed the combination of lactic and formic acid. There was no difference between the groups in the small intestine, caecum and the first two sections of the large intestine. In the last part of the large intestine, the content of VFA was higher in the control group compared to the group fed the combination of lactic and formic acid, while there was no difference between the two test groups.

Conclusion

Overall, the test showed that the pigs fed feed containing benzoic acid had a significantly higher production value than the control pigs and the pigs fed the combination of lactic and formic acid. Furthermore this group (lactic and formic acid) had a significantly higher production value than the control pigs. The pigs fed benzoic acid had a significantly lower treatment frequency for diarrhoea compared to the other two groups. The price of the benzoic acid used in this test was relatively high as it was food quality. When adding the price of benzoic acid to the feed price, the achieved increase in production cannot pay for adding the acid. The calculation does not include costs for medication in connection with diarrhoea treatments. Addition of the combination of lactic and formic acid achieved an actual production value level with the control group when the prices of the acids were included in the actual feed price.

Microbiological studies revealed that adding benzoic acid to the feed significantly reduces the studied populations of micro organisms and the microbial activity in the entire gastro-intestinal tract. This causes a reduction in the production of organic acids in the intestines. This applies to both the control group and the group with the combination of lactic and formic acid. The bacteriological results did not differ between the control group and the pigs given feed added lactic and formic acid. However, this group had a lower content of lactobacillus in the stomach.

Presently, benzoic acid is not approved as an additive/preservative for pig feed, however the acid and some of its salts may be used for preserving food. Several of the salts from benzoic acid are also approved as preservatives for pet food. Lactic acid and formic acid are both approved as preservatives for pig feed.

Participant

Agricultural technician Tommy Nielsen, The National Committee for Pig Production.

Test no. 586/File no. 4523.

Appendix 1

Diet 1 (4-6 weeks) and diet 2 (6-10 weeks), composition of ingredients, %

	Diet1			Diet 2		
Group **Product**	1 Control	2 0.7% lactic acid + 0.7% for-mic acid	3 2.0% benzoic acid	1 Control	2 0.7% lactic acid + 0.7% for-mic acid	3 1.0% benzoic acid
Wheat	67.83	65.17	64.03	53.85	51.66	51.97
Barley	-	-	-	10.00	10.00	10.00
Soybean meal, toasted	9.51	10.12	10.38	19.61	20.06	20.04
LT Fishmeal	8.00	8.00	8.00	-	-	-
Fishmeal, regular	-	-	-	7.00	7.04	7.00
Animal fat	4.05	4.71	5.00	4.04	4.65	4.51
Molasses, beet	2.00	2.00	2.00	2.00	1.70	2.00
Potato protein conc.	5.00	5.00	5.00	-	-	-
L-lysine 99%	0.35	0.35	0.34	0.39	0.38	0.38
Methionine 40%	0.15	0.15	0.16	0.22	0.22	0.22
Threonine 50%	0.06	0.05	0.05	0.15	0.14	0.14
Tryptophan 40%	0.02	0.02	0.02	-	-	-
Chalk	1.14	1.14	1.13	1.06	1.06	1.06
Monocalcium phosphate	1.20	1.20	1.20	1.11	1.11	1.11
Sodium chloride	0.34	0.34	0.34	0.34	0.35	0.34
Vitamins + minerals	0.35	0.35	0.35	0.23	0.23	0.23
Lactic acid	-	0.7	-	-	0.7	-
Formic acid	-	0.7	-	-	0.7	-
Benzoic acid	-	-	2.0			1.0

Appendix 2

Product description stated on the basis of information supplied by the companies

Group 2	
Product name	**Lactic acid: Lafeed 80** **Formic acid 85%**
Supplier	HCI Nordic Agro
	Frydenlundvej 30, DK-2950 Vedbæk, Denmark
	Tel.: +45 4329 2888
Contents	**Lafeed 80** is a 80% lactic acid (CH3CHOHCOOH). The product is liquid with a density of 1.20-1.30g/ml. The content of lactic acid in the natural (L+) form is minimum 95%. pKa value for lactic acid is 3.86. **Formic acid 85%** is a 85% acid, colourless and with an acrid smell. The product is liquid. pKa value is 3.75 and density is 1.19g/ml.
Price	Lactic acid and formic acid: DKK7.50 per kg.

Group 3	
Product name	**Benzoic acid**
Supplier	HCI Nordic Agro
	Frydenlundvej 30, DK-2950 Vedbæk, Denmark
	Tel.: +45 4329 2888
Content	Benzoic acid is a colourless crystal or white crystalline powder with a weak odour and sour, pricking flavour. The product is difficult to dissolve in water (1:400). Benzoic acid comes in two qualities: feed quality and a pharmaceutical quality.
Price	The pharmaceutical quality: approx. DKK25 per kg. A feed quality is expected to be somewhat cheaper.

Appendix 3

Diet 1: calculated and analysed content of nutrients – one production during the test

Group **Product**		**1** **Control**	**2** **0.7% lactic acid** **0.7% formic acid**	**3** **2.0% benzoic acid**
	Calculated	**Analysed**[1]		
DM content, %[2]	87	86	86	86
FUp per 100 kg[2]	119	114	117	117
Crude protein, %[2]	21.0	20.1	20.5	20.6
Lysine, g/kg	14.7	13.2	14.7	14.3
Methionine, g/kg	4.8	4.3	4.3	4.3
Meth.+cyst., g/kg	8.0	7.4	7.3	7.4
Threonine, g/kg	8.7	7.7	7.8	8.1
Calcium, g/kg	8.9	9.1	9.2	10.0
Total-phosphorus, g/kg	7.0	6.4	7.3	7.4
Zinc mg/kg	237	317	230	230
Screening for antibiotics	-	Negative	Negative	Negative

[1] Analysed content is stated on the basis of two analyses unless otherwise stated.
[2] Average of four analyses.

Diet 2: calculated and analysed content of nutrients – two productions during the test

Group **Product**		**1** **Control**	**2** **0.7% lactic acid** **0.7% formic acid**	**3** **2.0% benzoic acid**
	Calculated	**Analysed**[1]		
DM content, %[2]	87	87	86	86
FUp per 100 kg[2]	116	116	115	114
Crude protein, pct.[2]	20.3	20.7	20.5	20.5
Lysine, g/kg	13.8	13.3	13.9	13.8
Methionine, g/kg	4.5	4.0	4.1	4.1
Methionine + cystine g/kg	7.6	7.1	7.4	7.3
Threonine, g/kg	7.9	7.7	7.3	7.8
Calcium, g/kg	8.7	9.2	9.2	8.5
Total-phosphorus, g/kg	7.2	7.6	7.7	7.1
Zinc, mg/kg	184	165	154	161

[1] Analysed content is stated on the basis of two analyses unless otherwise stated.
[2] Average of four analyses.

Appendix 4

Table 1: Stomach lesions and weight of the various intestinal sections (8 pigs per group)

Group	1	2	3
	Control	0.7% lactic acid 0.7% formic acid	2.0/1.0% benzoic acid
Ulcer index			
Ulcer index	2.8	0.6	2.6
Weight of intestinal sections			
Stomach	75.9	81.4	74.0
Small intestine	488.6	491.0	429.6
Caecum	30.9	28.8	26.8
Large intestine	121.9	117.5	107.5

Table 2: Dry matter content and pH in the individual intestinal sections (8 pigs per group).
Values marked with different letters are significantly different ($p<0.05$).

Group	1	2	3
	Control	0.7% lactic acid 0.7% formic acid	2.0/1.0% benzoic acid
Dry matter content (%)			
Stomach	19.5	18.9	16.6
Small intestine 1	7.6	6.1	8.3
Small intestine 2	10.8	8.1	10.5
Small intestine 3	8.9	10.3	11.0
Caecum	9.8	10.4	11.0
Large intestine 1	20.0	16.3	19.3
Large intestine 2	21.6	22.0	23.8
Large intestine 3	24.5	25.5	25.1
pH in alimentary tract			
Stomach	3.10	3.61	3.51
Small intestine 1	5.38	5.62	5.31
Small intestine 2	6.57	6.40	6.18
Small intestine 3	6.50	6.68	6.61
Caecum	5.95	5.76	5.81
Large intestine 1	6.21^{a}	5.95^{b}	6.08^{a}
Large intestine 2	6.36	6.28	6.31
Large intestine 3	6.56	6.48	6.52

Table 3: Content of bacteria in the individual intestinal sections (log CFU/g intestinal content) (8 pigs per group). Values marked with different letters are significantly different ($p<0.05$).

Group	1	2	3
	Control	0.7% lactic acid 0.7% formic acid	2.0/1.0% benzoic acid
Lactic acid bacteria			
Stomach	7.75[a]	7.00[b]	5.58[c]
Small intestine 3	8.24[a]	8.27[a]	6.91[b]
Caecum	8.95[a]	8.85[a]	7.74[b]
Large intestine 2	9.24[a]	9.12[a]	7.88[b]
Lactobacillus			
Stomach	7.72[a]	6.77[b]	6.02[c]
Small intestine 3	8.24[ab]	8.15[a]	7.12[b]
Caecum	8.98[ab]	8.74[a]	7.76[b]
Large intestine 2	9.29[ab]	9.15[a]	7.73[b]
Coliform bacteria			
Stomach	3.12	3.20	3.15
Small intestine 3	6.29	5.81	5.13
Caecum	6.23	6.05	5.83
Large intestine 2	6.56	6.16	5.93
Yeast			
Stomach	5.93[a]	4.74[a]	2.98[b]
Small intestine 3	6.04[a]	4.91[a]	3.10[b]
Caecum	5.93[a]	4.68[a]	3.21[b]
Large intestine 2	5.73[a]	4.71[a]	3.18[b]
Total number of anaerobic bacteria			
Stomach	7.67[a]	6.59[b]	5.82[c]
Small intestine 3	8.53[a]	8.53[a]	7.27[b]
Caecum	9.31[a]	9.38[a]	9.08[a]
Large intestine 2	9.59[a]	9.65[a]	9.27[a]

Table 4: Content of acids in the various gastro-intestinal sections (mmol/kg) (8 pigs per group).

Group	1	2	3
	Control	0.7% lactic acid 0.7% formic acid	2.0/1.0% Benzo-ic acid
Lactic acid			
Stomach	17.1	12.7	15.6
Small intestine 1	19.3	14.6	14.0
Small intestine 2	27.4	17.4	14.7
Small intestine 3	43.7	35.3	17.5
Caecum	0.6	1.4	2.1
Large intestine 1	0.0	1.3	2.0
Large intestine 2	0.4	1.4	1.4
Large intestine 3	0.0	0.0	1.7
Formic acid			
Stomach	0.0	9.9	0.0
Small intestine 1	0.3	0.8	0.0
Small intestine 2	0.6	3.4	0.6
Small intestine 3	8.6	13.0	13.1
Caecum	0.0	0.0	0.0
Large intestine 1	0.0	0.0	0.0
Large intestine 2	0.0	0.0	0.0
Large intestine 3	0.0	0.0	0.0
Benzoic acid			
Stomach	-	-	8.5
Small intestine 1	-	-	0.8
Small intestine 2	-	-	1.3
Small intestine 3	-	-	1.9
Caecum	-	-	-
Large intestine 1	-	-	-
Large intestine 2	-	-	-
Large intestine 3	-	-	-
VFA (volatile fatty acids)			
Stomach	5.9	4.1	2.6
Small intestine 1	1.7	2.0	0.8
Small intestine 2	3.1	5.8	2.4
Small intestine 3	12.6	16.9	16.1
Caecum	144.8	140.7	145.4
Large intestine 1	135.2	126.3	132.8
Large intestine 2	124.9	112.1	115.8
Large intestine 3	114.2	90.5	95.8