

## **Impacts of organic acids and nutrient density of basal diets on broiler growth and gut histomorphology**

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### **Abstract**

Day-old male 240 chicks of a Ross 308 strain were used to test the effects of an commercial organic acid (OA), Biotronic SE Forte, on broiler performance and gut histomorphology. OA preparation was supplemented at 3 g/kg level into starter and finisher broiler diets of standard (NRC, 1994) or lower (approximately 10%) nutrient density (ND). Thus, the experiment was set up according to 2x2 factorial design with 6 reps of 10 birds allocated in each.

BWG and FCR of broiler chicks both at 21 and 35 d experimental periods were significantly ( $P<0.001$ ) improved by OA supplementation regardless of the diets ND. OA supplementation increased BWG by approximately 20% in 21 and 12% in 35 d experimental periods. FCR value dropped from 1.276 to 1.105 and from 1.263 to 1.129 in the two group fed with standart or lower ND, respectively. Proventriculus and heart weights were significantly decreased by dietary treatments too. Ileal viscosity was significantly ( $P<0.028$ ) affected by ND while the viscosity in feed was significantly ( $P<0.035$ ) effected by OA supplementation. Villi height was significantly ( $P<0.015$ ) increased by OA supplementation both in the standart or lower ND groups. This effect was better pronounced in lower ND fed group and the rate of the increase was 20%. In the case of villi depth interaction effect was observed, while significant drop in the group fed with standart ND, only slight increase was seen in the group fed with lower ND. Organic acid supplementation consistently decreased crypt depth (from 120.1 to 113.1 and from 117.3 to 96.3  $\mu\text{m}$ ) irrespective of ND. No significant effect could be observed by dietary treatments with respect to thickness of ileal muscle layer.

### **Introduction**

Antibiotics have been supplemented into some farm animal feeds including poultry mainly to control the growth of pathogenic bacteria. However, their use is rapidly disappearing world-wide. The effort to replace the antibiotics as growth promoters is gaining more and more attention, and therefore an appreciable amount of research particularly on probiotics, prebiotics, essential oils as well as organic acids is being carried out. In a recent review (Dibner and Buttin, 2002) it was very well documented that OA have long been used for feed preservation and their activities against gut microflora have been reported to be very similar. In addition to their positive effects on growth and feed utilization beneficial effects on protein and ME digestibility, gut flora, significant reduction in gastrointestinal pH, ammonia in stomach, small intestine and cecum of weaned piglets fed 1.25% formic acid reduction of biogenic amines which proved to have exerted growth depressing effect in the gut of animals fed by 0.6% formic acid (Vogt and Matthes, 1981; Eidelsburger et al., 1992b; Eckel et al., 1992; Runho et al., 1997) Additionally, increase in intestinal mucosa growth in rats in response to short-chain fatty acid infusion has been reported (Sakata and Engelhardt, 1983; Frankel et al., 1994. Moreover, it was reported that high protein diets cause low pepsin activity in the stomach due to their high buffer capacity (Koch, 2005). Most of these studies have been carried out considering normal requirement of single stomached animals, particularly pigs, however, this area of research with chicks remains to be done.

Therefore the aim of this study was to examine the influence of an OA preparation consisted of formic and propionic acids based on an inorganic phyllo-silicate carrier on broiler growth performance and gut histomorphology.

## Material and Methods

**Bird husbandry:** Day-old male 240 chicks of a Ross 308 strain were used to test the effects of an commercial organic acid (OA), Biotronic SE Forte (Biomim GmbH, Austria), on broiler performance and gut histomorphology. OA preparation was supplemented at 3 g/kg level into starter and finisher broiler diets of standard (NRC, 1994) or lower (approximately 10%) nutrient density (ND). Thus, the experiment was set up according to 2x2 factorial design with 6 reps of 10 birds allocated in each. Feed and water were provided *ad libitum*. Diets were fed to chicks as starter for the first period (0-21 day) and as finisher for the next (21-35 day). Lighting regime was 23h/d. Chickens were weighed individually at days 1, 21 and 35. The experiment was terminated at the end of 35 days of age.

**Gut histomorphology:** At 21 day of age, birds were weighed by pen and feed consumption determined. Two birds were taken randomly from each pen, and were killed by cervical dislocation, and weighed individually. Four centimetre segments of ileum (from Meckel's diverticulum to distal) were removed, rinsed, cut into pieces and placed into buffered formalin until further processing. For each sampling, tissues were cut into 5 mm sections and put into tissue cassettes. The tissues were processed, embedded in paraffin, and subsequently cut and placed onto slide with 5µm thickness. Then they were exposed to hematoxylin and eosin fixation stain. Pictures of villus height, depth and crypt depth and muscle thickness were taken by a digital camera and evaluated visually by treatments.

**Statistics:** Analysis of variance was used to measure the effects of dietary treatments and differences between means were identified by Duncan's Test according to SAS (1990).

## Results and Discussion

Data presented in Table 2 and Table 3 indicates that, weight gain of broiler chicks both at 21 and 35 d experimental periods were significantly ( $P < 0.001$ ) improved by OA supplementation regardless of feed nutrient density. Dramatic increase in weight gain obtained by OA was averagely 20% in 21 and 12% in 35 d periods. Significant ( $P < 0.001$ ) increase in FCR by OA supplementation was also evident in 21 and 35 d periods. FCR value dropped from 1.276 to 1.105 in the group fed with standard ND. Similar drop (from 1.263 to 1.129) was also seen in lower ND group. The overall improvement by OA supplementation at 21 d period was calculated to be 14%. At 35 d period, however, improvement in FCR tended to increase at lower rate. Decrease in response to OA supplementation could be related to very high feed intake recorded with the group fed on lower ND + OA. Feed intake was also increased by OA supplementation at about 6% and 11%, at 21 and 35 d periods, respectively. Similar improvement in BWG and FCR by OA supplementation to broiler diets were also reported by other authors in previous studies (Vogt and Matthes, 1981, Versteegh and Jongbloed, 1999; Eidelsburger et al., 1992b). Data obtained concerning to some of the organ weights of the digestive system indicated that except gizzard weight, proventriculus and heart weights were significantly affected by OA supplementation or ND. Significant ( $P < 0.001$ ) decrease in heart weight by ND was in contrast to the increase in BWG. Ileal viscosity was significantly ( $P < 0.028$ ) affected by nutrient density while the viscosity in feed was significantly ( $P < 0.035$ ) effected by organic acid supplementation (Table 4).

Villi height was significantly ( $P < 0.015$ ) increased by OA supplementation both in the groups fed with standard or lower ND. This effect was better pronounced in lower ND fed group with the increase of 20%. In the case of villi depth interaction effect was observed, while significant drop in the group fed with standard ND, only slight increase was seen in the group fed with lower ND. OA supplementation consistently decreased crypt depth (from 120.1 to 113.1 and from 117.3 to 96.3 µm) irrespective of nutrient density. No significant effect could be observed by dietary treatments with respect to thickness of muscle layer.

Increase in intestinal mucosa growth, villus height, surface area, and crypt depth accompanied by increase in gastrin were observed in rats in response to short-chain fatty acid infusion have also been reported (Sakata and Engelhardt, 1983; Frankel et al., 1994).

### Consequences

Consequently, the supplementation of the commercial organic acid preparation into broiler diets positively affected performance and altered ileal histomorphology. These effects can be summarised as; 1) dramatical improvement in growth and FCR of broilers, 2) decrease in proventriculus and heart weights, as well as in ileal viscosity and the viscosity in feed, and 3) increase in villi height and decrease in crypt depth.

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Table 1- Ingredients and chemical compositions of the basal diets

Ingredients, %	Standard ND		Lower ND	
	Starter	Grower	Starter	Grower
Corn	32.36	33.15	14.99	18.27
Soybean meal (46% CP)	27.05	14.47	18.66	11.78
Heated Soybeans	15.00	20,00	15,00	15,00
White wheat	10.00	15,00	20,00	25,00
Barley	5.00	7.80	20.00	20.50
Soybean oil	6.00	6.00	6.00	6.00
Others	4.59	3.58	5.35	3.45
<b>Nutrients</b>				
ME, MJ/kg	12.98	13.50	12.77	13.19
Crude protein, %	22.00	19.00	20.00	17.50
Lysine, %	1.40	1.06	1.10	0.91
Met + Cys, %	1.00	0.78	0.82	0.72
Methionine , %	0.63	0.45	0.48	0.42
Calcium, %	1.10	0.90	1.10	0.85
Nonphytate P, %	0.50	0.45	0.50	0.43

Table 2- Effects of organic acid supplementation on broiler performance

Dietary treatments	21 day			35 day		
	BWG g/bird	FI g/bird	FCR	BWG g/bird	FI g/bird	FCR
Normal nutrient density	540 <b>b</b>	689 <b>b</b>	1,276 <b>a</b>	1563 <b>b</b>	2419 <b>c</b>	1,548 <b>a</b>
Normal nutrient density +OA	637 <b>a</b>	704 <b>ab</b>	1,105 <b>b</b>	1750 <b>a</b>	2547 <b>b</b>	1,455 <b>b</b>
Low nutrient density	539 <b>b</b>	681 <b>ab</b>	1,263 <b>a</b>	1517 <b>b</b>	2315 <b>c</b>	1,526 <b>ab</b>
Low nutrient density +OA	657 <b>a</b>	742 <b>a</b>	1,129 <b>b</b>	1705 <b>a</b>	2688 <b>a</b>	1,576 <b>a</b>
SEM	13,70	10,16	0,025	26,65	34,46	0,017
Source of Variation	Probability					
Nutrient density	0,593	0,429	0,838	0,241	0,632	0,082
Organic acid	<0,001	0,060	<0,001	<0,001	<0,001	0,449
Nutrient density x Organic acid	0,529	0,235	0,706	0,976	<0,01	0,029

<sup>a-c</sup> Means in the same column with different letters differ significantly.

Table 3- Effects of organic acid supplementation on digestive parameters and intestinal morphology (35 d)

Dietary treatments	Proventriculus %	Heart %	Ileal viscosity cP	Villi height $\mu\text{m}$	Villi depth $\mu\text{m}$	Crypt depth $\mu\text{m}$
Normal nutrient density	0,47 <b>a</b>	0,56 <b>a</b>	2,45 <b>a</b>	720,2 <b>ab</b>	162,3 <b>a</b>	120,1 <b>a</b>
Normal nutrient density +OA	0,37 <b>b</b>	0,52 <b>ab</b>	2,29 <b>a</b>	824,5 <b>a</b>	111,8 <b>b</b>	113,1 <b>ab</b>
Low nutrient density	0,46 <b>a</b>	0,55 <b>a</b>	2,03 <b>ab</b>	638,6 <b>b</b>	140,9 <b>ab</b>	117,3 <b>ab</b>
Low nutrient density +OA	0,47 <b>a</b>	0,49 <b>b</b>	1,48 <b>b</b>	768,4 <b>ab</b>	153,8 <b>ab</b>	96,3 <b>b</b>
SEM	0,015	0,011	0,149			
Source of Variation	Probability					
Nutrient density	0,784	<0,001	0,028	0,130	0,473	0,180
Organic acid	0,899	0,598	0,159	0,015	0,196	0,063
Nutrient density x Organic acid	0,074	0,620	0,413	0,772	0,037	0,333

<sup>a-b</sup> Means in the same column with different letters differ significantly