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Effects of dietary symbiotics and organic acids on the mineral composition of broiler meat

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SUMMARY

The purpose of this study was to evaluate the effects of symbiotics and organic acids supplements, in the presence of chelated Zn, on the retention of essential minerals (Cu, Fe, Mn Zn) in broiler muscle tissues (breast, thigh) and in organs (liver). The trial was conducted on 150 Ross chicks aged 2 days, assigned to 5 experimental groups, with 30 chicks/group. The birds were housed in metabolic cages (6 chicks/cage) in the poultry experimental hall of the Laboratory of Nutrition Physiology within IBNA Balotesti. In the end of the experiment 6 chicks/group were slaughtered and samples of breast, thigh and liver were collected. The experimental diets differed from the control diet (C) (conventional diet) by the replacement of the inorganic Zn with an organic source (chelated Zn). Group E2 received 1% symbiotic supplement, group E3 received 0.15% organic acids supplement, while group E4 received a mixture of 1% symbiotic and 0.15% organic acids. Unlike the broiler breast samples, the broiler thigh samples displayed significant ($P \leq 0.05$) increases of the copper concentrations in groups E3 and E4 compared to group E1 (E1: 1.14 ± 0.19 ppm; E3: 1.74 ± 0.14 ppm; E4: 1.72 ± 0.58 ppm). The groups treated both with symbiotic and with organic acids mixture (E2-E4) showed significant ($P \leq 0.05$) increases of zinc concentration, compared to group C (C: 55.22 ± 2.36 ppm; E2: 60.83 ± 2.19 ppm; E3: 59.11 ± 3.41 ppm; E4: 62.06 ± 4.19 ppm). Liver sample analyses showed that groups E1, E2 and E4 recorded significant increases of Zn concentrations compared to group C (C: 86.90 ± 6.75 ppm; E1: 94.54 ± 4.17 ppm; E2: 93.93 ± 2.91 ppm; E4: 98.73 ± 5.20 ppm). The results of the study indicate that symbiotics and organic acids supplements improve mineral parameters of broiler meat quality. The mechanisms underlying these effects are not fully understood and more research is necessary to evaluate the consistency of these results.

Keywords: symbiotics, organic acids, chelated zinc, broiler, trace minerals.

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INTRODUCTION

Probiotics are microorganisms that change host microflora, having thus beneficial effects on the health status of the host (Schrezenmeir, and de Vrese, 2001). *Prebiotics* are indigestible feed ingredients which have a beneficial effect on the host by the selective stimulation of the growth and/or activity of a limited number of bacteria (probiotics) within the gastrointestinal tract, thus promoting health (Roberfroid, 2000). The term of "*symbiotic*" is used when a product contains both probiotics, and prebiotics. As the word hints towards synergism, it should be used only when the prebiotic compound selectively favours the probiotic compound (Schrezenmeir, and de Vrese, 2001).

The literature mentions several factors contributing to the mechanism by which the symbiotics favour mineral absorption. The positive effects are due to the increased solubility of the minerals due to the high bacterial production of fatty acids, to the larger absorption surface due to enterocytes proliferation mediated by the products of bacterial fermentation (Scholz-Ahrens et al., 2007).

Organic acids have been successfully used to replace antibiotics, after EU the acceptance of using non-therapeutic products in animal feeding (Li et al., 2008, Choct, 2001; Dibner et al., 2007). The organic acids supplementation of the drinking water improved the growth performance and decreased E. coli count in the intestine of weaned piglets. The probable action of the organic acids included the decrease of gastrointestinal tract digesta pH, which regulates the balance of the microbial populations in the intestine, stimulating the secretion of digestive enzymes (Thaela et al., 1998) and favouring the growth and recovery of the intestinal morphology (Galfi and Bokori, 1990). In some situations, feeding organic acids had no effect on the growth performance (Omogbenigun et al., 2003; Sacakli et al., 2006) and microbial populations (Risley et al., 1992).

The use of organometallic compounds in farm animal diets started from the fact that minerals can be naturally found, both in plants and in animals, as chelates of proteins. They accomplish their role within the organism through these protein chelates. The inorganic compounds currently used as minerals supplements (feed additives) must by hydrolysed by the digestive juice. Once solubilized, the metallic ion becomes very sensitive to any oxidation-reductive reaction that may occur within the reaction medium and may be chemically bonded by any ligand existing within the stomach fluid before they enter the duodenum (Vieira, 2008).

The purpose of our study was to evaluate the effects of symbiotics and organic acids supplements in the presence of chelated Zn on the retention of essential minerals (Cu, Fe, Mn Zn) in chicken muscle (breast, thigh) and organ (liver) tissues.

MATERIAL AND METHODS

The experiment was performed comply with Directive 2010/63/EU on the protection of animals used for scientific purposes and all procedures described, were approved by Ethical Commission of National Research and Development Institute for Biology and Animal Nutrition.

Experimental design

The experiment was conducted on 150, Ross 308 chicks aged 2 days assigned to 5 experimental groups with 30 chicks per group. The chicks were housed in metabolic cages (6 chicks/cage) in the poultry experimental hall of the Laboratory of Nutrition Physiology within IBNA Balotesti. The birds had free access to the feed and water. In the end of the experiment 6 chicks/group were slaughtered and samples of breast, thigh and liver were collected.

Diets

The following supplements were considered for utilization:

- A commercial product, BiotronicR SE Forte, produced by BIOMIN, GmbH Austria. According to the manufacturer, it is a synergic combination of organic acids, inorganic acids and their salts; the combination with plant extracts aims to prepare the digestive tract of the pigs or poultry, improving digestibility and inhibiting the proliferation of gram-negative bacteria. According to the technical specification, the product contains formic acid (19.2%); propionic acid (19.2%); lactic acid; citric acid; sorbic acid; it has a pH of 4.0.
- A commercial product, BiominR IMBO Pro/prebiotic, produced by BIOMIN, GmbH Austria. According to the manufacturer, the essential concept is the principle of competitive exclusion, being an alternative to antibiotics and growth promotors. The product is a combination of 4 products with synergic action: “Enterococcus faecium” probiotic; “fructo-oligosaccharide – inulin” prebiotic; cell walls fragments; phycophytic substances (derived from sea algae).
- A commercial product, chelated Zn, E.C.O.Trace® Trace minerals, produced by Biochem Zusatzstoffe Handels- und Produktionsgesellschaft mbH Küstermeyerstr, Germany.

Tables 1-3 show the compound feeds given to the broilers.

Chemical analysis

The samples (feed, faeces, meat, organ) were dried at 65^{0}C using a stove BMT model ECOCELL Blueline Comfort (Nuremberg, Germany) and grounded using laboratory mill Grindomix GM 200 (Retsch, Germany).

Table 1. Compound feeds formulation (phase I – starter), %

Specification	C	E1	E 2	E 3	E 4
Corn	53.2	53.2	52.2	53.05	52.05
Soybean meal	38.7	38.7	38.7	38.7	38.7
Oil	3.06	3.06	3.06	3.06	3.06
Monocalcium phosphate	1.9	1.9	1.9	1.9	1.9
Calcium carbonate	0.92	0.92	0.92	0.92	0.92
Salt	0.37	0.37	0.37	0.37	0.37
Methionine	0.37	0.37	0.37	0.37	0.37
Lysine	0.25	0.25	0.25	0.25	0.25
Choline	0.06	0.06	0.06	0.06	0.06
Threonine	0.12	0.12	0.12	0.12	0.12
Coccidiostats	0.05	0.05	0.05	0.05	0.05
ZOOFORT A1 (inorganic Zn)*	1	-	-	-	-
ZOOFORT A1 (organic Zn)**	-	1	1	1	1
Symbiotic	-	-	1	-	1
Organic acids mix	-	-	-	0.15	0.15

*1 kg IBNA premix (A1) contains: = 1100000 IU/kg vit. A; 200000 IU/kg vit. D3; 2700 IU/kg vit. E; 300 mg/kg Vit. K; 200 mg/kg Vit. B1; 400 mg/kg Vit. B2; 1485 mg/kg pantothenic acid; 2700 mg/kg nicotinic acid; 300 mg/kg Vit. B6; 4 mg/kg Vit. B7; 100 mg/kg Vit. B9; 1.8 mg/kg Vit. B12; 2000 mg/kg Vit. C; 8000 mg/kg manganese; 8000 mg/kg iron; 500 mg/kg copper; 6000 mg/kg zinc; 37 mg/kg cobalt; 152 mg/kg iodine; 18 mg/kg selenium; 6000 mg/kg antioxidant.

**source of organic zinc

Table 2. Compound feeds formulation (phase II – grower)

Specification	C	E1	E 2	E 3	E 4
Corn	54.78	54.78	53.78	54.63	53.63
Soybean meal	36.79	36.79	36.79	36.79	36.79
Oil	4.34	4.34	4.34	4.34	4.34
Monocalcium phosphate	1.65	1.65	1.65	1.65	1.65
Calcium carbonate	0.60	0.60	0.60	0.60	0.60
Salt	0.29	0.29	0.29	0.29	0.29
Methionine	0.29	0.29	0.29	0.29	0.29
Lysine	0.11	0.11	0.11	0.11	0.11
Choline	0.06	0.06	0.06	0.06	0.06
Threonine	0.04	0.04	0.04	0.04	0.04
ZOOFORT A1 (inorganic Zn)*	1	-	-	-	-
ZOOFORT A1 (organic Zn)**	-	1	1	1	1
Symbiotic	-	-	1	-	1
Organic acids mix	-	-	-	0.15	0.15

1 kg IBNA premix (A1) contains: = 1100000 IU/kg vit. A; 200000 IU/kg vit. D3; 2700 IU/kg vit. E; 300 mg/kg Vit. K; 200 mg/kg Vit. B1; 400 mg/kg Vit. B2; 1485 mg/kg pantothenic acid; 2700 mg/kg nicotinic acid; 300 mg/kg Vit. B6; 4 mg/kg Vit. B7; 100 mg/kg Vit. B9; 1.8 mg/kg Vit. B12; 2000 mg/kg Vit. C; 8000 mg/kg manganese; 8000 mg/kg iron; 500 mg/kg copper; 6000 mg/kg zinc; 37 mg/kg cobalt; 152 mg/kg iodine; 18 mg/kg selenium; 6000 mg/kg antioxidant.

**source of organic zinc

Trace mineral concentrations were determined in feed, faeces, meat and organ samples applying flame atomic absorption spectrometry (FAAS) as described by Untea et al., (2012) after microwave digestion. The used equipment was as follows: Atomic absorption spectrometer Thermo Electron – SOLAAR M6 Dual Zeeman Comfort (Cambridge, UK), with deuterium lamp for background correction and air-acetylene flame and microwave digestion system with remote temperature measurement, BERGHOF, Speedwave MWS-2 Comfort (Eningen, Germany). Stock solutions of Cu, Fe, Mn, Zn, 1000 ppm traceable to SRM from NIST, were used to standardize the flame atomic absorption spectrometer. Class A glassware was used for transvasation, dilution and storage.

Table 3. Compound feeds formulation (phase II – finisher)

Specification	C	E1	E 2	E 3	E 4
Corn	60.31	53.2	52.2	53.05	52.05
Soybean meal	30.61	38.7	38.7	38.7	38.7
Oil	5.01	3.06	3.06	3.06	3.06
Monocalcium phosphate	1.61	1.9	1.9	1.9	1.9
Calcium carbonate	0.72	0.92	0.92	0.92	0.92
Salt	0.38	0.37	0.37	0.37	0.37
Methionine	0.22	0.37	0.37	0.37	0.37
Lysine	0.09	0.25	0.25	0.25	0.25
Choline	0.05	0.06	0.06	0.06	0.06
ZOOFORT A1 (inorganic Zn)*	1	-	-	-	-
ZOOFORT A1 (organic Zn)**	-	1	1	1	1
Symbiotic	-	-	1	-	1
Organic acids mix	-	-	-	0.15	0.15

*1 kg IBNA premix (A1) contains: = 1100000 IU/kg vit. A; 200000 IU/kg vit. D3; 2700 IU/kg vit. E; 300 mg/kg Vit. K; 200 mg/kg Vit. B1; 400 mg/kg Vit. B2; 1485 mg/kg pantothenic acid; 2700 mg/kg nicotinic acid; 300 mg/kg Vit. B6; 4 mg/kg Vit. B7; 100 mg/kg Vit. B9; 1.8 mg/kg Vit. B12; 2000 mg/kg Vit. C; 8000 mg/kg manganese; 8000 mg/kg iron; 500 mg/kg copper; 6000 mg/kg zinc; 37 mg/kg cobalt; 152 mg/kg iodine; 18 mg/kg selenium; 6000 mg/kg antioxidant.

**source of organic zinc

Statistics

The analytical results have been compared with the variance analysis (ANOVA), with WINDOWS StatView (SAS, version 6.0). The differences of the average values were considered significant for $P < 0.05$. The results have been expressed as mean ± SD for all measurements.

RESULTS AND DISCUSSION

The following parameters have been monitored throughout the experimental period: body weight (g), total gain (kg); average daily feed intake (g CF/chick/day); feed conversion ratio (kg CF/kg gain). Table 4 shows that there we no significant (P≤0.05) differences in the production parameters, except for the body weight at 42 days, where group E3 had significantly (P≤0.05) higher results than group E4. The use of the mixture of organic, inorganic acids and their salts for group E3 had a positive effect on the dietary nutrients digestibility, which yielded better performances: significantly higher (P≤0.05) average body weight at 42 days (2333.55± 314.01 g) and average daily weight gain (56.03 ± 3.31 g gain/chick) compared to the other experimental groups.

Table 4. Chick performance throughout the experimental period

	C	E1	E2	E3	E4
Initial weight (g)	39.08 ± 1.16	39.49 ± 0.98	39.35 ± 0.95	40.22 ± 2.63	39.29 ± 1.09
Final weight (g)	2226.45± 378.54	2184.52± 354.20	2174.52± 326.14	2333.55± 314.01 ^{e}	2094.84± 272.91 ^{d}
Average daily feed intake (g/chick/day)	91.13 ± 7.18	89.97 ± 5.65	91.35 ± 3.65	95.01 ± 4.33	87.60 ± 9.16
Average daily weight gain (g/chick/day)	53.48 ± 4.10^{d}	52.32 ± 2.46^{e}	52.12 ± 2.39	56.03 ± 3.31^{a}	50.13 ± 3.21^{b}
Feed conversion ratio (g CF/g gain/period	1.70 ± 0.09	1.72 ± 0.07	1.75 ± 0.02	1.70 ± 0.08	1.75 ± 0.19

Where: a – significantly different from C; b – significantly different from E1; c – significantly different from E2; d – significantly different from E3; e – significantly different from E4; (P ≤ 0.05)

At the end of the experiment, according to the working protocol approved by the ethics commission from IBNA Balotesti, we slaughtered 6 chicks/group to collect breast, thigh and liver samples. The samples were dried in a drying cabinet for 48 hours at 65^{0}C. These samples were analysed for the essential trace elements Cu, Fe and Zn. Tables 5-7 show the results of these determinations.

Table 5. Essential trace elements composition of the breast samples, mg/kg

Group	Cu	Fe	Zn
C	0.84 ± 0.15	22.01 ± 2.92	26.47 ± 1.50
E1	0.86 ± 0.04	22.56 ± 1.40	26.35 ± 1.39
E2	0.74 ± 0.05	23.20 ± 1.68	25.31 ± 2.07
E3	0.81 ± 0.20	22.85 ± 2.35	26.69 ± 2.21
E4	0.76 ± 0.25	23.96 ± 5.08	25.18 ± 0.80

No significant (P > 0.05) differences were noticed between the studied groups, for any of the surveyed metals. We may thus conclude that the dietary supplements (symbiotic and mixture of organic acids) didn't have a positive effect on the feeding quality of the chick breast.

Table 6. Essential trace elements composition of the thigh samples, mg/kg

Group	Cu	Fe	Zn
C	1.54 ± 0.38	34.54 ± 8.03	55.22 ± 2.36^{cde}
E1	1.14 ± 0.19^{de}	33.06 ± 3.39	57.59 ± 2.56^{e}
E2	1.34 ± 0.24	34.15 ± 2.98	60.83 ± 2.19^{a}
E3	1.74 ± 0.14^{b}	35.21 ± 2.37	59.11 ± 3.41^{a}
E4	1.72 ± 0.58^{b}	33.09 ± 2.56	62.06 ± 4.19^{ab}

Where (within the column): a – significantly different from C; b – significantly different from E1; c – significantly different from E2; d – significantly different from E3; e – significantly different from E4; (P ≤ 0.05)

Unlike the chicken breast samples, significant (P ≤ 0.05) increases of the copper concentrations were noticed in groups E3 and E4 (supplemented with organic acids and symbiotic, organic acids and organic zinc, respectively) compared to group E1 (no supplements, just organic zinc). The groups treated both with symbiotic and with the mixture of organic acids (E2-E4) recorded significant increases (P ≤ 0.05) of zinc concentrations, compared to group C.

Table 7. Essential trace elements composition of the liver samples, mg/kg

Group	Cu	Fe	Mn	Zn
C	13.61 ± 1.94	385.97 ± 67.05^{e}	11.60 ± 0.56	86.90 ± 6.75^{bce}
E1	13.98 ± 1.00	411.51 ± 62.46^{e}	12.92 ± 0.65^{c}	94.54 ± 4.17^{a}
E2	12.86 ± 0.77	364.72 ± 81.492	10.79 ± 1.31^{b}	93.93 ± 2.91^{a}
E3	13.21 ± 1.30	401.58 ± 78.61^{e}	11.88 ± 2.40	90.19 ± 6.43^{e}
E4	13.19 ± 0.99	316.12 ± 36.42^{abd}	11.79 ± 0.64	98.73 ± 5.20^{ad}

Where (within the column): a – significantly different from C; b – significantly different from E1; c – significantly different from E2; d – significantly different from E3; e – significantly different from E4; (P ≤ 0.05)

No significant variations (P ≤ 0.05) of the copper concentrations were noticed in the studied groups. The highest iron concentrations were noticed for group E3, same as for the chicken thigh samples, although the differences were not statistically significant. Groups E1, E2 and E4 displayed significant increases of zinc concentrations compared to group C, the situation being almost similar for the thigh samples, where the groups treated with symbiotic and organic acids displayed significant increases compared to group C. we may presume that this is due to the joint effect of the organic zinc and of the supplements. Group E3 was not significantly different from C, having the lowest zinc concentrations among the experimental groups. Since the lowest iron concentration was also noticed in this group, we may explain this by the Fe-Zn antagonism within the chicken liver, the main place of mineral retention.

While the liver is the main deposit of minerals, the chicken breast is the poorest deposit of minerals, while it has considerable amounts of protein. The corroborated results for the three anatomic parts show that the presence of organic zinc and of the symbiotic supplement enriched the chicken thigh and liver in zinc.

The literature showed that the oligosaccharides increase mineral (Ca, Mg, P) and oligo-elements (Cu, Fe, Zn) absorption (Scholz-Ahrens and Schrezenmeir, 2007). The mechanisms by which inulin-type fructans mediate this effect include the acidification of the intestinal lumen via short-chain fatty acids, which increase mineral solubility in the intestine and increase the contact surface. Vanhoof and DeSchrijver (1996) tested the effect of inulin of Fe and Zn absorption in pigs and noticed that Zn absorption was significantly higher in the experimental group. Yalçınkaya et al. (2012) showed that manon-oligosaccharide prebiotics facilitate Fe and Zn absorption and ease Cu retention in broilers.

CONCLUSIONS

The results of the study indicate that symbiotics and organic acids supplements improve mineral parameters of broiler meat quality. The concentrations of Cu and Zn in leg meat samples and Fe and Zn in liver samples were improved, indicating the symbiotic and organic acids potential in developing functional foods. The mechanisms underlying these effects are not fully understood and more research is necessary to evaluate the consistency of these results.

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