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DECREASE OF Cu AND Fe CONCENTRATIONS FROM BROILER DROPPINGS USING DIETARY ORGANOMETALLIC COMPOUNDS

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Abstract: The purpose of this study was to determine the effect of the partial or total replacement of the inorganic Cu and Fe salts by organic forms of these elements on broiler performance and on the elimination of minerals through broiler droppings. A study was conducted for 42 days on 240 broiler chicks treated with trace elements chelates with amino acids: B-TRAXIMRTEC Cu-I30; B-TRAXIMRTEC Fe-120. The broiler chicks were housed in cages (10 chicks per cage, 6 cages per group) and assigned to 4 groups (C, E1, E3 and E3) fed on the same corn-soybean meal-based diet. Phased-feeding was used according to the developmental stages of the broilers (starter, grower and finisher). Feed intake and the amount of droppings were recorded daily. The daily records of droppings and their chemical analysis was used to determine the trace elements load of the droppings. The total or partial replacement of the inorganic Cu and Fe salts by chelates of these minerals with amino acids in broiler diets didn't affect broiler performance. The inclusion of Cu chelates in broiler diets at the level recommended by the manufacturer of by NRC decreased by at least 34% the level of Cu (environmentally toxic element) in the droppings. The corresponding decrease was of 5-21% for Fe.

Key words: chelates, broiler, faeces, environmental pollution

Introduction

The diets used in the intensive system of poultry production are supplemented with minerals in order to avoid the deficiencies that might occur due to a variety of clinical and pathological problems *(Underwood and Suttle, 1999)*. After absorption, in the organism, the mineral elements, under different ionic forms, fulfil multiple roles: they maintain the osmotic pressure within the intra and extra cellular spaces, they intervene in the neuromuscular excitability, they activate enzymatic systems through mechanisms of hormonal regulation *(Sandulescu et al., 2000)*. Animal requirements of minerals are difficult to determine, being influenced by several factors (species, hybrid, category of production). For the essential minerals there are maintenance and production requirements clearly set for almost all species *(NRC, 1998)* and therefore, the nutritionists are concerned to ensure animal requirements by including mixes of salts of these minerals in the compound feeds.

Because of the low concentration of minerals, particularly in the cereal grains, and due to the presence of nutrients with antagonist action (the phytic acid, for instance), the basic premises when the dietary supplements are calculated is that the bioavailability of the dietary minerals can be considered null *(Cao et al., 2000)*. Traditionally, the inorganic salts (oxides, sulphates) have been used in poultry diets due to their advantageous price and because they meet the requirements *(Bao et al., 2007)*, but the organic sources with a higher bioavailability increased in importance *(Creech et al., 2004; Revy et al., 2003)*. This means that less mineral supplements must be added to the diet *(Jondreville et al., 2003)*, compared to the usual inorganic salts, without affecting poultry performance, which decreases the amount of excreted minerals. The efficiency of these products in which the mineral is bound to an organic compound, leaves place for debates, economic particularly.

Currently, the animal diets have a surplus of minerals in order to maximize animal performance. These amounts are 3-4 times higher than animal requirement. For instance, the inorganic Cu salts, an environmentally toxic heavy metal, is the acceptation of EU documents, are used in excess due to its role of growth promoter *(Hill et al., 2000)* but also because of the practice of adding higher amounts of chemical premixes in order to have a wide margin of "economic" safety. In conclusion, the droppings have very high concentrations of these elements, in a direct relation with their dietary level *(Zielinska et al., 2009)*. The toxic elements, once accumulated in the soil, have adverse effects on the plants and microorganisms, becoming an environmental problem in the areas with intensive animal production *(Kingery et al., 1994)*. Thus, the high mineral concentrations in the droppings will accumulate in the soil and in the long-term, about 50 years, they will degrade the soil severely.

The purpose of our study was to determine the effect of the partial or total replacement of the inorganic salts by organic forms of Cu and Fe on broiler performance and on the mineral concentrations in the droppings.

Materials and Methods

A 42 days trial on 240 broilers was organised in order to evaluate the potential of Cu and Fe chelates with amino acids to decrease the level of Cu and Fe in broiler droppings. The diets were treated with Cu and Fe chelates with amino acids as follows: B-TRAXIMRTEC Cu-I30 (manufacturer recommendation: 80-120 g/t DM compound feed); B-TRAXIMRTEC Fe-120 (manufacturer recommendation: 160-180 g/t DM compound feed).

The broilers, housed in cages (10 chicks per cage, 6 cages per group) were assigned to 4 groups (C, E1, E2 and E3) which received the same basal diet. Table 1 shows diet formulation. The feeds were manufactured in the Pilot Station of IBNA (National Research Development Institute for Biology and Animal Nutrition – Balotesti) and assayed chemically. The broilers were phase-fed according to the developmental stages of the broilers (starter, grower and finisher).

Table 1. Diet formulation

Ingredients	%
Corn	42.74
Rice	10
Soybean meal	20
Gluten	4
Wheat	10
Soybean oil	1
Monocalcium phosphate	1.4
CaCO3	9.2
Salt	0.3
Methionine	0.16
Lysine	0.15
Choline	0.05
Vit-min. premix	1
TOTAL diet	100

The diets differed by the mineral treatment as follows: conventional mineral premix for C ; manufacturer recommendation provided through chelates for E1; 0.5% inorganic salts + 0.5% chelates for E2; NRC requirements ensured with chelates for E3.

Table 2. Mineral (Cu, Fe) supply from the premix

	C (g/kg premix)	E1 (g/kg premix)	E2 (g/kg premix)	E3 (g/kg premix)
Fe sulphate	21.5	-	10.8	-
Cu sulphate	2.4	-	1.2	-
Fe chelate	-	18	9	46.7
Cu chelate	-	5	2.5	3.1

The amount of dietary chelates was calculated on the basis of NRC requirements for trace elements in broiler chicks. Table 3 shows the dietary amounts of Cu and Fe.

Table 3. Dietary amounts of Cu and Fe

	C	E1	E2	E3
	COPPER (mg/kg)			
Phase 1	23.53	17.57	20.55	16.14
Phase 2	22.79	16.64	19.58	15.36
Phase 3	19.03	13.15	16.15	11.72
Average	21.78	15.79	18.76	14.41
% from C	100	72.50	86.14	66.17
NRC requirement	5	5	5	5
	IRON (mg/kg)			
Phase 1	165.27	137.18	151.24	191.18
Phase 2	155.90	127.62	141.94	181.73
Phase 3	117.04	85.52	99.83	139.62
Average	146.07	116.77	131.00	170.84
% from C	100	79.95	89.69	116.96
NRC requirement	75	75	75	75

The treatments produced diets with a lower load of Cu (27, 14 and 34% lower than in C). The Fe load was decreased by 20% and 10% for groups E1 and E2, respectively, while in the group where NRC1994 recommendation was observed, the dietary Fe level exceeded the control diet by about 17%

Feed intake was recorded daily. After each batch of forage was manufactured, in the second week of feeding, the amounts of droppings were collected and recorded on a daily basis, and weekly average samples of droppings/cage were formed. Bothe the feed and the droppings samples were analysed chemically. The load of minerals was calculated from the daily amounts of droppings and from their chemical analysis.

The standardization of the atomic flame spectrophotometer was done with stock solutions of 1000 ppm Traceable to SRM from NIST, $Fe(NO_3)_2$ in HNO3 0,5 mol / L; $Cu(NO_3)_2$ in HNO_3 0,5 mol / L. We only used double-distilled and deionised water (Milli-Q Millipore, 18.2 MΩ/cm). We also used: HNO_3, Merck 65%, density 1.39 kg/L; H_2O_2 30%, Merck. The analytical determinations were performed with an atomic absorption spectrophotometer Thermo Electron – SOLAAR M6 Dual Zeeman Comfort (Cambridge, UK), fitted with deuterium lamp for background correction. The samples were prepared with a microwave digester, with remote temperature measurement, BERGHOF, model Speedwave MWS-2 Comfort (Eningen, Germany); analytical scales Sartorius (Gottingen, Germany); drying stove BMT model ECOCELL Blueline Comfort (Neuremberg, Germany); water distiller Milli-Q Ultrapure Water Purification System, Millipore (Billerica, USA). Only class A glassware was used for solution transfer, dilution and storage.

For the statistical analysis, each cage in the trial was considered an experimental unit. The bioproductive parameters, the data on mineral ingesta and excreta were analysed with ANOVA, program STATVIEW. The differences between groups were considered significant for $P \leq 0.05$.

Results and Discussions

Broiler parameters are the first stage in the evaluation of the dietary treatment effect *(Jongbloed and Kemme, 2007)*. Table 4 shows broiler characteristics throughout the trial.

Table 4. Broiler parameters

	C	E1	E2	E3
Initial weight (g)	45.79 ± 1.02	44.13 ± 0.46	45.35 ± 1.19	45.67 ± 1.2
Final weight (g)	1928.16 ± 81.04	1902.37 ± 57.85	2029.41 ± 39.96	2018.11 ± 90.09
Average daily feed intake (g/broiler/day)	94.26 ± 3.25	93.57 ± 5.72	93.63 ± 4.26	92.01 ± 6.25
Average daily gain (g/ broiler/day)	44.84 ± 1.23	44.24 ± 4.36	47.24 ±2,27	46.26 ± 3.76
Feed conversion ratio	2.10	2.11	1.98	1.99

Data processing showed an increase of the average daily gain for groups E2 and E3, but no significant differences were detected between groups for any studied parameter. The chelate treatment didn't affect broiler performance, as also shown by *Nollet et al. (2007)* and *Liotta et al. (2009)*.

Relating the dietary mineral levels to the feed intake during the experimental period and to the Cu and Fe load of broiler droppings, we obtained the following data on the ingestion and excretion of minerals (Cu and Fe).

Table 5. Cu and Fe ingestion and excretion

	C	E1	E2	E3
	Ingesta			
Cu (mg/day/broiler)	1.96	1.42	1.68	1.27
% of C	100	72.35	85.53	64.72
Fe (mg/day/broiler)	20.85	18.20	19.39	22.58
% of C	100	87.30	92.98	108.29
	Excreta			
Cu (mg/day/broiler)	1.31	0.86	1.24	0.75
% of C	100	65.84	95.06	57.37
Fe (mg/day/broiler)	12.76	10.07	11.31	12.09
% of C	100	78.93	88.64	94.81

The 14-34% decrease of the dietary Cu compared to C (Table 3) decreased by about 5-35% the amount of ingested Cu and by 5-43% the amount of Cu excreted in the droppings, compared to C (Table 5). When NRC recommendations were observed and Fe chelates with amino acids were added into an amount sufficient to meet NRC recommendations (supposing zero bioavailability of the minerals from the basic diet), the dietary Fe level in the diet for E3 exceeded the dietary level of group C, therefore Fe ingesta also was higher than in group C (108.29%). Despite this situation, the amount of Fe in the droppings from group E3 was slightly lower than in the control group. The graphical representation of the correlation between the Cu and Fe concentrations in the ingesta and excreta, show a direct proportionality between the ingested level and the excreted level for all the studied groups (Figure 1), as also reported by *Zielinska et al. (2009)*. The correlation factors exceed the value of 0.93.

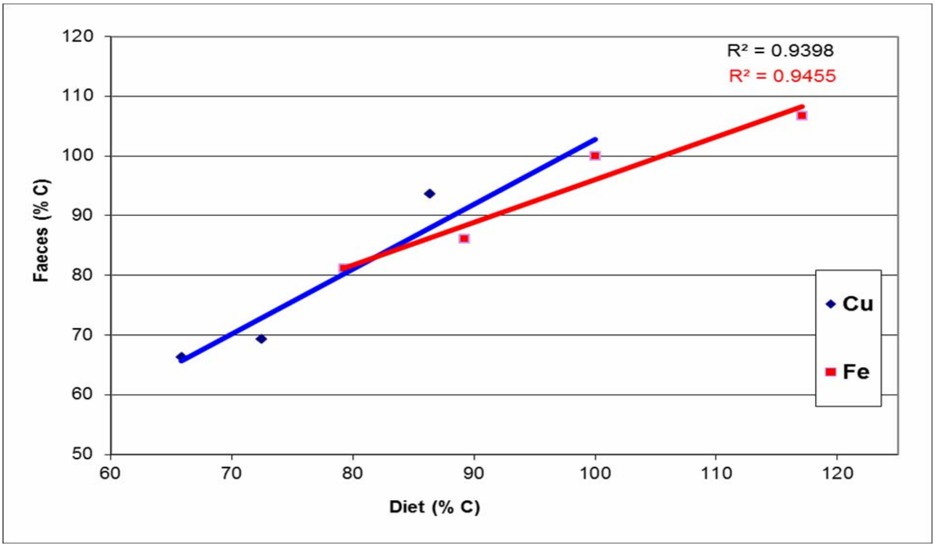

Figure 1. Correlations between Cu and Fe concentrations in the diets and droppings

The amounts of trace elements (Cu and Fe) eliminated in the droppings follow an increasing slope, in agreement with the values for the intake and excretion recorded during the trial. Figure 2 shows mineral excretion in time.

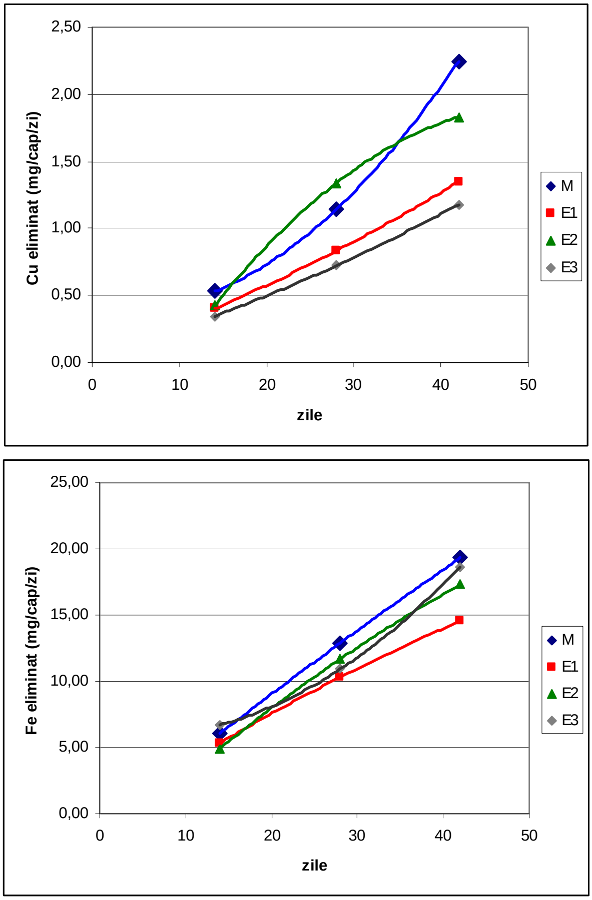

Figure 2. Cu and Fe excretion in time

The results for Cu, both from E1 and from E3, differed significantly from C, but the amounts excreted by group E3 (NRC requirements) are the lowest. However, since the differences were not significant between E1 and E3, we may consider that Cu excreta can be reduced by more than 34% if the diets for E1 an E3 are used, in comparison with a amounts of Cu excreted using a conventional diet. The chart shows that the lowest Fe excretion was obtained when the recommendations of the manufacturer were observed – the diet for E1 (11% less than C).

Conclusion

The partial or total replacement of the inorganic Cu and Fe salts in broiler diets by Cu and Fe chelates with amino acids didn't affect significantly broiler performance.

The inclusion of Cu chelates with amino acids in the amount recommended by the manufacturer or at the level recommended by NRC (5 mg/kg) produced a decrease of up to 34% of the excreted Cu (potentially toxic for the environment). Regarding the Fe, the excreted amounts were 5-11% lower than in group C, the highest decrease being obtained when the level recommended by manufacturer was used.

The decrease of the amounts of excreted Cu and Fe compared to the conventional diets has both environmental and economic positive impact.

Smanjenje koncentracije Cu i Fe u fecesu upotrebom organskih oblika ovih elemenata u ishrani brojlera

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Rezime

Cilj ovih istraživanja je determinacija efekta delimične i totalne zamene neorganskih soli Cu i Fe organskim oblicima ovih elemenata na performanse brojlera i eliminaciju ovih minerala fecesom.. Istraživanja su rađena na 240 brojlerskih pilića u trajanju od 42 dana koji su tretirani helatnim oblicima Cu i Fe sa aminokiselinama: B-TRAXIMRTEC Cu-I30; B-TRAXIMRTEC Fe-120. Brojleri su smešteni u kaveze (10 pilića po kavezu, 6 kaveza po grupi) i podeljeni u četiri grupe (C, E1, E2 i E3), hranjeni su istim smešama na bazi kukuruz/soja. Ishrandbene faze su primenjivane u skladu sa razvojem brojlera (starter, grover i finišer). Konzumacija hrane i količina fecesa su evidentirani svakodnevno.. Dnevna merenja fecesa i analiza hemijskog sastava su korišćeni za determinaciju sadržaja Cu i Fe u fecesu. Totalna i delimična zamena neorganskih soli Cu i Fe helatima ovih minerala sa aminokiselinama u smešama za ishranu brojlera nije uticala na njihove performanse. Uključivanjem helata Cu u isranu brojlera prema preporukama industrije ili NRC, smanajuje nivo Cu (ekološki toksičan element) u fecesu za najmanje 34%. Odgovarajuće smanjenje za Fe je 5-21%.

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