INVESTIGATION ON THE EFFECT OF TRACE MINERAL SOURCE ON PARAMETERS OF BIOAVAILABILITY IN BROILER CHICKENS

Annika Mählmeyer[1], Jennifer Lindel[1], Alexandra Schlagheck[1], Bastian Hildebrand[1], Klaus Männer[2]

[1]Biochem Zusatzstoffe Handels- und Produktionsges. mbH, Küstermeyerstraße 16, D-49393 Lohne
[2]Freie Universität Berlin, Faculty of Veterinary Medicine, Institute of Animal Nutrition, Königin-Luise-Straße 49, D-14195 Berlin

Corresponding author: Annika Mählmeyer
e-mail: maehlmeyer@biochem.net; tel.: +494442 9289-0
Address: Küstermeyerstraße 16, D-49393 Lohne

Abstract. A study was conducted to investigate the effect of two sources of Cu, Fe, Mn, and Zn on zootechnical performance, as well as apparent total tract digestibility (ATTD) and accumulation of these trace minerals in liver, tibia bone (TB), breast muscle, and skin of broiler chickens by a depletion-repletion study design. Seventy-two 1-day-old male broiler chickens (Cobb 500) were divided in three feeding groups. A 14-days depletion period with feeding only native contents of trace minerals was followed by a 14-day repletion with two groups supplemented either with glycine-bound (Gly) or sulfate-bound (Sul) trace minerals. The third group received the repletion diet without trace mineral supplementation (Basal). The trace mineral supplementation improved the body weight gain ($P<0.05$) and food conversion ratio ($P<0.05$) compared to basal diet. ATTD of trace minerals was higher in the Gly-group for all elements and higher in the Sul-group for Mn compared to basal group ($P<0.05$). Significant differences in ATTD between Gly and Sul were detectable for Zn and Cu ($P<0.05$). Compared to the basal group retention of Mn and Fe was significantly higher in the liver of Gly-supplemented birds and in the TB of Gly- and Sul-treated animals ($P<0.05$). Retention of Cu in liver and TB, and of Zn in TB, tended to be higher in Gly-group than in basal group ($P<0.1$). The present study confirmed that the trace minerals Fe, Mn, Zn and Cu are available in broilers to a higher amount when supplemented in glycine-bound form instead of sulfates in a deficient situation. The results indicated that the measured parameter influences the conclusions made from bioavailability studies, comparing different trace mineral sources.

Keywords: Trace minerals, broiler chickens, performance, digestibility, retention

Introduction. Trace minerals are well known to play an important role in a variety of body functions, although their requirement is compared to other nutrients less than 100 mg per kg dry matter (Yatoo et al., 2013). Deficiency of trace minerals is difficult to detect under practical conditions. That makes it even more important to guarantee a sufficient supply of the animal. The steady improvements in animal performance on the one hand, and environmental concerns on the other hand, have led to a risen interest for efficient trace mineral sources in animal feeding. Hence diverse trace mineral sources are listed in the register of feed additives (Regulation (EC) No. 1831/2003; EU, 2003) accompanied by a reduction of maximum levels of application during the last years. Due to their chemical bonding and stability in the gastrointestinal tract (GIT), the bioavailability of various trace minerals is determined. Commonly inorganic sources like oxides and sulfates are used in animal production. But it is widely accepted, that trace minerals iron (Fe), manganese (Mn), zinc (Zn) and copper (Cu) can be absorbed to a higher extent when supplemented in organically bound forms compared to inorganic forms (Ammerman et al., 1995; Jongbloed et al., 2002). This can be confirmed by the results out of the experiment conducted by Abdallah et al. (2009). Chickens fed with organic trace minerals showed a significant higher body weight, better feed conversion and higher tibia ash compared to those who were fed with inorganic trace minerals. With a higher absorption of trace minerals, lower intake is needed for a sufficient trace mineral supply on the one side and less excretion of metals is assumed on the other side (Schlegel, 2006).

In order to evaluate bioavailability of trace minerals from different sources *in vivo*, studies using a depletion-repletion trial design have been performed, mostly in piglets. Respective data with broiler chickens as model animal are scarcely available (Männer et al., 2016). Moreover, the effect of trace mineral source on differences in accumulation of trace minerals in specific tissues and organs of broiler chickens after a depletion period is not fully clarified.

The aim of this research was to investigate the effect of two sources of Cu, Fe, Mn, and Zn (sulfate vs. glycine chelate) on zootechnical performance, as well as apparent total tract digestibility (ATTD) and accumulation of these trace minerals in several body tissues of broiler chickens by a depletion-repletion study design.

Materials and Methods

The study was carried out in seventy-two 1-day-old male broiler chickens (Cobb 500) at the Institute of Animal Nutrition at Freie Universität Berlin. All birds were fed a basal diet containing only native contents of trace minerals for the first 14 days (depletion period). The birds were housed in floor pens with bedding of softwood shavings and were given ad libitum access to water and feed in mash form. After the first 14 days of life, the birds were divided in three feeding groups to stainless steel metabolic cages with a base area of 0.19 m^2 (34 cm x 55 cm) with three birds per cage. In the repletion period from day 15 to 28 of age, two feeding groups were supplemented either with glycine-bound trace minerals (Gly, E.C.O.Trace®, Biochem Zusatzstoffe GmbH) or sulfate-bound trace minerals (Sul) according to official feeding recommendations (NRC, 1994), whereas the third feeding group received the repletion diet without trace mineral supplements (Basal diet). De- and repletion diets were manufactured in the institute (registration number: DE-BE-100001) and prepared without inclusion of enzymes, growth promotors, acidifiers, probiotics or coccidiostats. Diet composition is shown in Table 1. During the repletion period 3 g titanium(IV)-dioxide (TiO_2) per kg diet were added to all the diets as a marker for ATTD measurements. Analyzed nutrient contents in the de- and repletion diets are shown in Table 2.

Body weights per cage were recorded every week. Feed consumption per broiler chicken was estimated as the total feed supplied per cage and period corrected for dispersed/leftover feed and number of birds per cage. Feed conversion ratio (feed:gain) was calculated from average feed intake and body weight gain per cage. Excreta of all birds were collected at day 13 and day 14 of the repletion period and were pooled per pen (3 birds per pen), frozen and analysed for the trace mineral contents at the end of the trial. At the end of the repletion period all birds from each treatment were stunned and culled by bleeding (approval number A 0100/13). Liver, left breast muscle, left TB and overall breast skin were excised, whereby the tissues were pooled per cage. Samples of feeding stuffs as well as the pooled, freeze dried excreta samples and pooled samples of liver, breast-muscle, skin, and tibia bones were analyzed for dry matter, crude ash, iron, manganese, zinc and copper in accordance to the methods given by VDLUFA (dry matter: VDLUFA III 3.1; crude ash: VDLUFA III 8.1; iron: VDLUFA III 11.1.2; manganese: VDLUFA III 11.4.2, zinc: VDLUFA III 11.5.2, copper: VDLUFA III 11.3.2). Feed samples were additionally analyzed by the following methods: crude protein: VDLUFA III 4.1.1 modified according to macro-N determination (vario Max CN); crude fiber: VDLUFA III 6.1.4; ash: VDLUFA III 8.1; crude fat: VDLUFA III 5.1.1; starch: VDLUFA III 7.2.1; total sugars: VDLUFA III 7.1.1; calcium: VDLUFA VII 2.2.2.6; phosphorus: VDLUFA VII 2.2.2.6; sodium: VDLUFA VII 2.2.2.6. Trace minerals in bones were measured after defatting. TiO_2 content in diets and excreta were measured using a UV spectrophotometer following the method of Myers et al. (2004). Statistical analysis was carried out by one-way ANOVA. Multiple comparisons between groups were made by LSD Post-hoc test and significant differences were declared at $P<0.05$.

Results. Body weight (BW) at start of repletion period was not different between feeding groups and averaged 381.1±20.2 g. At trial end, significant differences in BW were detected between groups. The trace mineral supplementation improved the BWG well as the food conversion ratio (Table 3).

The ATTD was lowest in animals receiving the basal diet. The supplementation of the glycine-bound trace minerals improved ATTD of all trace minerals (Mn, Fe, Zn, Cu), whereas significant differences in ATTD between Sul-group and basal group were detectable only for Mn. ATTD values for all trace minerals were numerically higher in the Gly-group than in the Sul-group, on a significant level for the elements Zn and Cu (Table 4).

The differences in ATTD of trace minerals were partly reflected in metal concentrations in body tissues at the end of the trial (Table 5). In body tissues of Sul-group the retention of Mn and Fe in different tissue samples was numerically but not significantly higher than in basal group, except retention of Mn and Fe in TB.

Table 1. **Diet composition**

Ingredients		Depletion period[3)]	Repletion period[4)]
Corn	%	47.80	60.80
Soybean meal (49% CP)	%	35.80	24.00
Soybean-oil	%	8.20	6.50
Wheat bran	%	4.00	4.00
Limestone	%	1.68	1.60
Monocalcium-phosphate	%	0.80	0.65
Premix[1)]	%	1.20	1.20
Titanium(IV)-dioxide	%	-	0.30
DL-Methionine	%	0.27	0.25
L-Lysine–HCL	%	0.25	0.35
L-Threonin	%	-	0.07
Trace mineral mix[2)]	%	-	0.117

[1)] Contents per kg Premix: 600000 I.U. Vit. A (acetate); 120000 I.U. Vit. D_3; 6000 mg Vit. E (α-tocopherol acetate); 200 mg Vit. K_3 ; (MSB); 250 mg Vit. B_1 (mononitrate); 420 mg Vit. B_2 (cryst. riboflavin); 300 mg Vit. B_6 (pyridoxin-HCl); 1500 µg Vit. B_{12}; 3000 mg niacin (niacinamide); 12500 µg biotin (commercial, feed grade); 100 mg folic acid (cryst., commercial, feed grade); 1000 mg pantothenic acid (Ca d-pantothenate); 60000 mg choline (chloride); 45 mg iodine (calcium-iodate); 20 mg selenium (sodium-selenite); 140 g sodium (NaCl); 55 g magnesium (magnesium sulfate); carrier: calcium carbonate (calcium min 38%); [2)] per kg trace mineral premix: 29231 mg Fe, 20342 mg Zn, 37692 mg Mn, 2479 mg Cu from sulfates or glycine chelates; [3)] day 1 to 14 of feeding period; [4)] day 15 to 28 of feeding period

Table 2. **Analyzed nutrient contents in the diets**

Nutrients		Depletion period[1)]	Repletion period[2)]		
			Basal	Sul	Gly
Dry Matter	g/kg	940	911	908	912
Crude protein	g/kg	228	191	193	195
Crude fiber	g/kg	26.2	28.1	28.5	29.0
Crude fat	g/kg	105	98.5	99.2	98.3
Ash	g/kg	55.1	48.8	49.2	49.1
Starch	g/kg	319	386	384	387
Sugars	g/kg	43.8	37.9	37.9	37.5
Calcium	g/kg	9.0	8.1	7.9	8.0
Total phosphorus	g/kg	6.0	5.3	5.4	5.5
Sodium	g/kg	1.6	1.6	1.7	1.8
Iron	mg/kg	43.5	45.8	103.8	99.6
Manganese	mg/kg	14.2	16.9	61.6	62.0
Zinc	mg/kg	16.0	16.2	50.8	52.3
Copper	mg/kg	4.8	5.1	8.9	8.6
[1)] day 1 to 14 of feeding period; [2)] day 15 to 28 of feeding period					

Table 3. **Performance in broiler chickens during repletion period, body weight (BW, g), body weight gain (BWG, g) and food conversion ratio (FCR), (Means, ±SD, n=8)**

	Basal	Sul	Gly	P-value
BW day 15	381.7 ±19.6	381.2 ±19.4	382.4 ±24.2	0.996
BW day 28	1407 ±33.4^{a}	1501 ±32.7^{b}	1523 ±28.1^{b}	<0.001
BWG day 15 - 28	1025 ±26.5^{a}	1120 ±31.5^{b}	1140 ±35.9^{b}	0.001
FCR day 15 - 28	1.39 ±0.05^{a}	1.28 ±0.0^{b}	1.25 ±0.0^{b}	<0.001
P<0.05, ^{ab} significant differences between groups				

Table 4. **Apparent total-tract digestibility of complete diet (%) at day 28, (Means, ±SD, n=8)**

	Basal	Sul	Gly	P-value
Fe	14.2^{a} ±2.47	15.5^{a,b} ±1.63	17.3^{b} ±2.81	0.048
Mn	10.7^{a} ±3.48	14.3^{b} ±1.86	16.5^{b} ±1.42	<0.001
Zn	16.5^{a} ±3.04	17.9^{a} ±2.98	21.9^{b} ±2.89	0.004
Cu	13.6^{a} ±2.9	15.6^{a} ±2.76	18.7^{b} ±2.48	0.005
P<0.05, ^{ab} significant differences between groups				

Significant differences in retention of Mn and Fe were detected in liver and TB for Gly-group vs. basal-group. Zinc retention in liver, TB and skin was numerically higher in the Gly-group than in the basal- and Sul-group (P<0.1). The retention of Cu in all tissues was numerically higher in the supplemented groups whereas in liver and TB Cu tended to be higher in Gly-group compared to the Sul-group (P<0.1).

Discussion. It is well known that native trace mineral contents in feed aren't sufficient to support the performance of commercial broiler chickens (NRC, 1994; GfE, 1999). This was confirmed by the current study. Broilers supplemented with Zn, Mn, Cu and Fe showed a higher BW and a better FCR compared to non-supplemented birds. Regarding zootechnical performance, the Gly-group showed numerically higher BW and FCR compared to the Sul-group. Similarly, in another depletion-repletion study the use of a mixture of Zn-, Mn-, Cu-, and Fe-glycinates was significantly more efficient than respective sulfate forms (P<0.05) in improving BWG within 21-days repletion period (Männer et al, 2016). In line with these observations, De Marco (2017) reported a significantly improved BW of broilers after 35-days supplementation of glycine-bound trace minerals compared to the control group, which was supplemented with a mixture of sulfates and oxides, however without a preceding depletion phase. Moreover, numerical improvements in zootechnical performance of broilers by exchanging inorganic forms by glycine-bound forms are reported from studies investigating single elements (Ma et al., 2013; Kwiecien et al., 2015; Kwiecien et al. 2016). Controversial, other studies showed no effect of replacing inorganic by organic forms on the performance of broiler chickens (Nollet, 2007; Shi et al., 2015; Untea and Panaite et al., 2016).

Studies in other animal species have shown that glycine-bound trace minerals are more digestible than inorganically bound trace minerals (Spears et al., 2004; Ettle et al., 2007; Zhou et al., 2013; Hildebrand and Männer, 2013). The higher digestibility of organically bound trace minerals in broilers has also been demonstrated (Nollet et al., 2007; Aksu et al., 2011). According to the ATTD results of the present study, glycine-bound trace minerals are better absorbable than the inorganic sulfate forms. In line these findings, significantly higher apparent ileal digestibility of Zn, Mn and Cu after 21 days of repletion have been reported when using glycinates instead of sulfates (Männer et al. 2016).

Table 5. **Trace mineral concentration in liver, breast-muscle, skin and TB of broiler chickens at day 14 of the repletion, (Means, ±SD, n=8)**

		Basal	Sul	Gly	P-value
Liver					
Fe	mg/kg	428.1 ±18.5[a]	439.9 ±25.8[ab]	464.1 ±19.4[b]	0.004
Mn	mg/kg	9.60 ±0.94[a]	10.18 ±0.66[ab]	10.56 ±0.55[b]	0.001
Zn	mg/kg	67.00 ±3.90	67.41 ±3.36	69.84 ±3.87	0.167
Cu	mg/kg	11.22 ±0.97	12.02 ±1.66	12.41 ±1.52	0.378
Breast muscle					
Fe	mg/kg	16.78 ±0.58	17.74 ±1.70	18.23 ±1.74	0.172
Mn	mg/kg	0.46 ±0.05	0.52 ±0.03	0.53 ±0.10	0.187
Zn	mg/kg	19.41 ±1.00	20.38 ±1.80	19.35 ±1.41	0.541
Cu	mg/kg	1.69 ±0.32	1.77 ±0.48	1.87 ±0.56	0.436
Skin					
Fe	mg/kg	30.60 ±2.07	33.23 ±3.6	34.04 ±2.44	0.130
Mn	mg/kg	2.82 ±0.19	2.99 ±0.20	3.17 ±0.26	0.230
Zn	mg/kg	44.47 ±3.07	45.34 ±3.43	46.57 ±3.87	0.526
Cu	mg/kg	5.06 ±0.29	5.07 ±0.45	5.13 ±0.75	0.827
Tibia bone					
Fe	mg/kg	123.7 ±9.7[a]	132.5 ±6.0[b]	136.1 ±5.0[b]	0.001
Mn	mg/kg	3.37 ±0.31[a]	4.01 ±0.48[b]	4.19 ±0.6[b]	0.003
Zn	mg/kg	105.2 ±8.2	110.0 ±8.4	113.4 ±11.2	0.229
Cu	mg/kg	4.59 ±0.35	4.76 ±0.18	4.87 ±0.29	0.170
$P<0.05$, [ab] significant differences between groups					

As expected, the trace mineral concentration was lowest in the body tissues of non-supplemented animals. The use of glycine-bound trace minerals resulted on average in a higher retention of metals compared to the use of sulfate-bound trace minerals. This is in line with the ATTD data of the current study. Most studies examining the accumulation of trace minerals in broilers focused on the element Zn. In the present study Zn concentrations in skin, liver and TB were numerically highest in the Gly-group, followed by the Sul-group. In accordance with these results the Zn content of 42-days old broiler chickens has been shown to be significantly higher when 30 ppm Zn from Zn glycinate where used instead of 40 ppm Zn from Zn sulfate (Sridhar et al., 2015). In another depletion-repletion study in piglets, a significant increase in the Zn content of skin by using glycinate form instead of sulfate form has been demonstrated (Hildebrand and Männer, 2013). Effects of Zn supply level on TB are well documented. The use of glycine-bound Zn allows a higher Zn concentration in the TB of 42-days old broiler chickens compared to an inorganic Zn form and allows a reduction in Zn dosage (Kwiecien et al., 2016). Higher levels of Zn in liver have been reported when using a glycine-bound form instead of an inorganic one (Untea and Panaite, 2016), however it has to be considered that liver Zn concentration is not assumed as first choice criteria for evaluation of bioavailability of Zn sources (Jongbloed et al., 2002). The current study showed clear effects of Fe supply on Fe retention. As shown by Shi et al. (2015) replacing Fe sulfate with Fe glycinate can effectively improve hemoglobin status. In the studies of another working group, the use of Fe glycinate instead of sulfate form had a positive effect on the hematological and biochemical parameters of broilers (Kwiecien et al., 2015a), whereas no clear effects of Cu source (glycinate vs. sulfate) on blood parameters and Cu level in liver were detectable (Kwiecien et al., 2015b). Studies on body retention of manganese from glycinate forms in broiler chickens are scarcely available. No effect of Mn source (chelated forms vs. sulfate form) on bone Mn concentration was detected after 42 days, whereas concentration in the heart muscle tissue indicated a higher bioavailability of Mn from the organic sources (Li et al., 2004). Numerically increased levels of Mn in the muscle tissue were also reported by Hildebrand and Männer (2013) in piglets when glycine-bound Mn was supplemented instead of sulfate form. These observations are in contrast to the current study, where Mn content of TB was higher in the Gly-group than in the Sul-group, but no difference in breast muscle tissue were detectable. Basically, the study indicated that the number of replicates might have been too low to prove differences between the trial groups on a significant level, especially in body tissue accumulation.

Conclusion

The study demonstrated that the supplementation of glycine bound trace minerals showed a numerically improvement in zootechnical performance, ATTD and accumulation rates in tissues in comparison to inorganic salts. Present results confirmed that differences in ATTD are reflected to a higher extent in the accumulation of metals in TB and liver than in skin or breast muscle tissue. Due to the better ATTD, organically bound trace minerals may replace high dosages of inorganically bound trace minerals and this can reduce mineral excretion and consequently effects the environment.

References

1. Abdallah, A. G., El-Husseiny, O. M., Abdel-Latif, K. O. Influence of Some Dietary Organic Mineral Supplementations on Broiler Performance. 2009. International Journal of Poultry Science. No 8(3), P. 291-298.

2. Aksu, T., Özsoy, B., Saeipinar Aksu, D., Yörük, M. A., Gül, M. The Effects of Lower Levels of Organically Complexed Zinc, Copper and Manganese in Broiler Diets on Performance, Mineral Concentration of Tibia and Mineral Excretion. 2011. Kafkas Universitesi Veteriner Fakultesi Dergisi. No. 17(1), P. 141-146.

3. Ammerman, C. B., Baker, D. H., Lewis, A. J. Bioavailability of nutrients for animals. 1995. Academic Press. San Diego, California.

4. De Marco, M., Zoonb, M.V., Margetyala, C., Picarta, C., Ionescub, C. Dietary administration of glycine complexed trace minerals an improve performance and slaughter yield in broilers and reduces mineral excretion. 2017. Animal Feed Science and Technology. No. 232, P. 182–189.

5. Ettle, T., Schleger, P., Roth, F. X. Investigations on iron bioavailability of different sources and supply levels in piglets. Journal of Animal Physiology and Animal Nutrition. 2007. No. 92, P. 35-43.

6. EU. European Union. Register of Feed Additives, Regulation (EC) No 1831/2003. Annex II: List of additives subject to the provisions of Art. 10 § 2 of Reg. (EC) No 1831/2003 for which no application for reevaluation was submitted before the deadline of 8 November 2010. 2003. P. 24-29.

7. GfE, Society of Nutritional Physiology. Empfehlungen zur Energie- und Nährstoffversorgung der Legehennen und Masthühner. Ausschuss für Bedarfsnormen der Gesellschaft für Ernährungsphysiologie. 1991. DLG-Verlag GmbH, Germany.

8. Hildebrand, B., Männer, K. Investigations on the effect of trace mineral source on parameters of bioavailability in weaned piglets. 12. Tagung Schweine- und Geflügelernährung. 2013. Halle-Wittenberg, Germany. P. 167-169.

9. Jongbloed, A. W., Kemme, P. A., De Groote, G. Lippens, M., Meschy, F. Bioavailability of major and trace minerals. International Association of the European (EU) Manufacturers of Major, Trace and specific Fed mineral Materials (EMFEMA). 2002. Brussels, Belgium.

10. Kwiecien, M., Samolinska, W., Bujanowicz-Haras, B. Effects of iron–glycine chelate on growth, carcass characteristic, liver mineral concentrations and haematological and biochemical blood parameters in broilers. Journal of Animal Physiology and Animal Nutrition. 2015a. P. 1-13.

11. Kwiecien, M., Winiarska-Mieczan, A., Piedra, J. V., Bujanowics-Haras, B., Chalabis-Mazurek, A. Effects of copper glycine chelate on liver and faecal mineral concentrations,and blood parameters in broilers. Agricultural and Food Science. 2015b. No. 24, P. 92-103.

12. Kwiecien, M. Winiarska-Mieczan, Milczarek, A., Tomaszewska, E., Matras, J. Effects of zinc glycine chelate on growth performance, carcass characteristics, bone quality, and mineral content in bone of broiler chicken. Livestock Science. 2016. No. 191, P. 43-50.

13. Li, S., Lou, X., Liu, B., Crenshaw, T. D., Kuang, X., Shao, G., Yu, S. Use of chemical characteristics to predict the relative bioavailability of supplemental organic manganese sources for broiler. Journal of Animal Science. 2004. No. 82, P. 2352-2363.

14. Ma, W. Q., Sun, H., Zhou, Y., Wu, J., Feng, J. Effects of Iron Glycine Chelate on Growth, Tissue Mineral Concentrations, Fecal Mineral Excretion, and Liver Antioxidant Enzyme Activities in Broilers. Biological Trace Element Research. 2012. No. 149, P. 204-211.

15. Männer, K., Pelletier, W., Ader, P., Zentek, J. Effects of inorganic and organic dietary trace minerals (Fe, Mn, Zn, Cu) in male broiler chickens. 20th Congress of the European Society of Veterinary and Comparative Nutrition (ESVCN). 2016. Berlin, Germany. P. 110.

16. Myers, W. D., Ludden, P. A., Nayigihugu, V. and Hess, B. W. Technical Note: a procedure for the preparation and quantitative analysis of samples for titanium dioxide. Journal of Animal Science. 2004. No. 82, P. 179-183.

17. National Research Council (NRC). Nutrient Requirements of Poultry. Ninth Revised Edition, National Academy Press. Washington, USA, 1994. NRC 1994.

18. Nollet, L., van der Klis, J. D., Lensing, M., Springs, P. The Effect of Replacing Inorganic With Organic Trace Minerals in Broiler Diets on Productive Performance and Mineral Excretion. The Journal of Applied Poultry Research. 2007. No. 16, P. 592-597.

19. Schlegel, P. Experimental designs to study organic trace mineral sources in animal nutrition. In: Windisch, W., Plitzner, C. (Eds.): Experimentelle Modelle der Spurenelementforschung, 21. Jahrestagung

der Gesellschaft für Mineralstoffe und Spurenelemente. 2006. Wien, Herbert Utz Verlag, München.

20. Shi, R., Liu, D., Sun, J., Zhang, P. Effect of replacing dietary $FeSO_4$ with equal Fe-levelled iron glycine chelate on broiler chickens. Czech Journal of Animal Science. 2015. No. 60(5), P. 233-239.

21. Spears, J. W., Schlegel, P., Seal, M. C., Lloyd, K. E. Bioavailability of zinc from zinc sulfate and different organic zinc sources and their effects on ruminal volatile fatty acid proportions. Livestock Production Science. 2004. No. 90, P. 211-217.

22. Sridhar, K.., Nagalakshmi, D., Rama Rao, S. V. Effect of graded concentration of organic zinc (zinc glycinate) on skin quality, hematological and serum biochemical constituents in broiler chicken. Indian Journal of Animal Science. 2015. No. 85(6), P. 543-648.

23. Untea, A. E., Panaite, T. D. Effects of dietary symbiotics and organic acids on the mineral composition of boiler meat. Archiva Zootechnica. 2016. No. 19(2), P. 27-35.

24. Yatoo, M. I., Saxena, A., Deepa, P. M., Habeab, B. P., Devi, S., Jatav, R. S., Dimri, U. Role of trace elements in animals: a review. Veterinary World. 2013. No. 6(12), P. 963-967.

25. Zhou, Z., Fang, S., Yue, M., Feng, J. Iron Glycine Chelate on Meat Color, Iron Status and Myoglobin Gene Regulation of *M. longissimus dorsi* in Weaning Pigs. International Journal of Agriculture & Biology. 2013. No. 15, P. 983-987.

Received 14 May 2018
Accepted 5 June 2018