

The New Progresses in Trace Mineral Requirements of Broilers, a Review

Review Article

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ABSTRACT

Since 1994 till today a few up to date have been made on the NRC (1994) recommendations for trace mineral requirements of broilers. The high prevalence of skeletal disorders in modern broilers could be a consequence of the uncoordinated growth rate and mineral nutrition. The commercial companies of broiler production often use a huge safety margin of trace minerals in feed formulation, which will result in trace mineral excretion into the environment. There are differences in the commercially recommended levels of dietary trace element between commercial strains of broilers and all of them are different from NRC (1994) recommendations.

KEY WORDS broiler chickens, NRC, trace mineral requirements.

INTRODUCTION

A significant progress has been achieved in broilers growth rate since 1994, when the latest version of nutrient requirements of poultry (NRC, 1994) was published. In fact, a comparison between the last version of NRC publication and commercial strains recommendation for trace mineral concentrations in broiler diets, reveals that the industry has been often used a large safety margin in feed formulation. Table 1 shows the NRC (1994) recommendations for trace element requirements of broiler chickens and Tables 2 and 3 demonstrate the trace element requirements of the last two recommendations for Ross 308 and Cobb 500 commercial broiler strains, respectively (Aviagen, 2007; Cobb, 2008). An unfavorable consequence of this dietary safety margin is the higher mineral excretion into the environment. Introducing the organic trace mineral complexes, allowed the modern broilers to reach their genetic potential with a lower dietary trace mineral supplementation. The nano particles of trace minerals are a more recent genera-

tion of supplements which are under investigations. This review tries to summarize the more recent findings on the trace mineral requirements of broiler chickens.

Iron (Fe)

Iron has numerous essential roles in animal cells and is especially involved in blood hemoglobin and muscle myoglobin composition, energy metabolism, neurotransmitter synthesis, phagocyte antimicrobial activity, and also, the synthesis of DNA, collagen, and bile acids (Jia *et al.* 2015). Sulfates, oxides, and carbonate salts are common Fe supplements in poultry rations, however, organic components of Fe with higher availability are also accessible now (Bao *et al.* 2007). The first symptom of iron deficiency is anemia, which decreases physical activity because of reduced hemoglobin, myoglobin levels and as well as a lower activity of iron-dependent cytochromes which reduce cellular ATP and also immune system performance (Van paemel *et al.* 2010). The growth rate, meat quality, and immune status of chickens determine their Fe requirements (Yang *et al.*

2011). Unlike the majority of the trace elements, the Fe requirement recommended for commercial strains of broilers in almost half of the NRC (1994) values. A level of 80 mg Fe/kg dry matter of broiler's diet has been recommended by NRC (1994) which has been re-suggested by Bureau of Indian Standards (BIS, 2007). However, the *Gesellschaft für Ernährungsphysiologie* (1999) suggested 100 mg dietary Fe to kg feed.

Shinde *et al.* (2011), found no differences in the effects of the organic (ferrous methionine chelate 10% Fe) and inorganic ($\text{FeSO}_4 \cdot \text{H}_2\text{O}$, 28% Fe) sources of iron supplementation at 80 mg/kg on improving performance, nutrient digestibilities and hematological parameters of broilers. They suggested that organic sources of Fe were more efficient in depositing Fe in the hemopoietic organs (liver and tibia) of birds fed low Fe diets. In a comparable report, Arnaudova-Matey *et al.* (2013), found that iron methionate was more effective than iron sulfate for better Fe deposition in the liver (40 to 60% more) and considerably lower amount of Fe excretion than that in the control ones.

The commercial companies have downward updated the Fe requirements of broilers. Fe requirement of Ross 308 have been declined from 40 mg/kg in 2007 to 20 mg/kg in the 2014 manual. Comparison of Cobb 500 requirements shows no differences in supplementary levels between the 2008 and 2013 manuals for all growing periods. The recommended Fe requirements values for Cobb 500, Arbor Acres and Hubbard were 40, 20 and 60 mg/kg diet, respectively. Maximum tolerance limit of 500 ppm Fe has been reported in poultry by NRC (2005). In commercial broiler diets, Fe is found in the range of 150 to 400 mg/kg, which is less than the toxic level (Yang *et al.* 2011). There are also reports that high dietary Fe supplement, up to 200 mg/kg or more had no adverse effect on birds performance and health. In conclusion, in recent decade there was a trend to reducing dietary concentration of Fe for broiler chickens and a 20-40 mg/kg is a good recommendation for modern broiler strains.

Copper (Cu)

Copper is a well-known cofactor for vital enzymes in physiological processes of poultry. The Cu is a key element in mitochondrial oxidative phosphorylation, neurotransmitter metabolism, pigment synthesis and formation of connective tissue and red blood cell (Underwood and Suttle, 1999). It's important to use more bioavailable sources of copper in broiler diets to minimize the Cu excretion into the environment. Copper sulfate pentahydrate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) is the common Cu supplement in poultry diets because of its low cost and easy accessibility. However, organic Cu and nano-sized Cu have been revealed to be more bioavailable than Cu sulfate pentahydrate (Gonzales-Eguia *et al.* 2009).

In a study, cupric citrate supplementation on the grower phase increased feed conversion ratio of broiler chickens as compared to the group receiving cupric sulfate. Copper excretion also decreased in broilers fed cupric citrate, as compared to cupric sulfate. Pesti and Bakalli (1996) reported that cupric citrate was a more efficient growth promoter than cupric sulfate. In two experiments, Ewing *et al.* (1998) reported an improved weight gain and feed conversion ratio in broilers supplemented with cupric citrate than those fed copper sulfate.

The last edition of the NRC, mentioned 8 mg/kg of diet as the minimum Cu requirement of broilers (NRC, 1994). The required dietary Cu can differ due to the severity, extent and form of stress experienced by an animal. It has been reported that dietary copper supplementation over the minimum proposed requirement could improve poultry performance (Pesti and Bakalli, 1996; Zhang *et al.* 2009a).

The potential of growth promoting effect of dietary supplemented copper in broiler chicks fed up to 300 mg Cu/kg of the diet has been reported. The other organizations have recommended rather different dietary Cu needs for broiler chickens. The 7 mg Cu/kg diet was recommended by GFE (1999) which was slightly lower than the value proposed in NRC (1994), while in BIS (2007) the proposed level was far 12 mg per kg diet. Between the commercial broiler chicken strains, the recommended dietary Cu level for Hubbard strain is 10 mg/kg, which is closest to the NRC value, however the suggestions for other strains are at higher levels, such that the Cobb 500, Ross 308 and Arbor Acres were suggested 15, 16 and 16 mg Cu/kg, respectively. Because of improving effects of Cu on poultry health and immunity, its dietary supplementation rate deserves renewed attention. Dietary supplementation of copper at 125-250 ppm is a common commercial practice in poultry rations (Pesti and Bakalli, 1996; Zhang *et al.* 2009b), which improve body weight and feed conversion ratio (Fisher *et al.* 1973; Hoda and Maha, 1995). Arias and Koutos (2006) reported a better body weight gain in broiler chicken fed diets that supplemented with 150 ppm Cu compared to the control group. The species, age, dietary inclusion rate, feed type and microbial population of the intestinal tract may affect the antimicrobial characteristics of Cu in a given situation (Arias and Koutsos, 2006).

It has been reported that broiler chickens under stress benefits more from higher dietary copper supplementation and it seems that the level of required Cu for optimal performance becomes increasingly higher with increased stress (Hooge *et al.* 2000). Broilers are always challenged with different stresses that may last for a few hours (e.g. catching, crating and transport) or for almost the whole rearing period (e.g. heat stress, immune challenges) (Lin *et al.* 2004).

Table 1 The NRC (1994) recommendations for trace element requirements of broiler chickens (mg/kg)

Item	Growth phase (weeks)		
	0-3	3-6	6-8
Iron	80	80	80
Copper	8	8	8
Manganese	60	60	60
Zinc	40	40	40
Selenium	0.15	0.15	0.15
Iodine	0.35	0.35	0.35

Table 2 The Ross (308) broilers trace element recommendations in years 2007 and 2014 versions (mg/kg)

Trace element	Starter (0-10 d)		Grower (11-24 d)		Finisher (24 d-market)	
	2007	2014	2007	2014	2007	2014
Iron	40	20	40	20	40	20
Copper	16	16	16	16	16	16
Manganese	120	120	120	120	120	120
Zinc	100	110	100	110	100	110
Selenium	0.30	0.30	0.30	0.30	0.30	0.30
Iodine	1.25	1.25	1.25	1.25	1.25	1.25

Table 3 The Cobb 500 broilers trace element recommendations in years 2008 and 2013 versions (mg/kg)

Trace element	Starter (0-10 d)		Grower (11-22 d)		Finisher (23-42 d)	
	2008	2013	2008	2013	2008	2013
Iron	40	40	40	40	40	40
Copper	15	15	15	15	15	15
Manganese	100	100	100	100	100	100
Zinc	100	100	100	100	100	100
Selenium	0.3	0.35	0.3	0.35	0.3	0.35
Iodine	1	1	1	1	1	1

The Cu requirement of young chickens is more than adults and they are more likely to show a deficiency (NRC, 1994), particularly while they are experiencing an immune challenge (Koh *et al.* 1996).

Extra Cu beyond immediate needs is deposited as metallothionein, mainly in the hepatocytes. Most plasma Cu is found as a constituent of ceruloplasmin and does not originate from newly consumed feeds, but from the Cu fed in the past weeks or months (Brody, 1993). The growth-suppressive effects of higher levels of Cu supplementation (>300 ppm) have even been reported (Persia *et al.* 2004; Luo *et al.* 2005).

In the study of Chen *et al.* (1997), the body weight gain significantly reduced in the broilers fed 500 mg Cu/kg diet. Cu ion's capacity to promote oxidative damage is usually attributed to the production of very reactive hydroxyl radical (OH⁻) from hydrogen peroxide (H₂O₂) via the Haber-Weiss reaction (Bremner, 1998; Kadiiska *et al.* 1993). This situation may lead to lipid peroxidation and creation of reactive products which may be involved in severe damage of cell molecules and structures (Videla *et al.* 2003). As a conclusion, it seems that dietary supplementation of copper up to 250 mg to kg diet can promote growth without any adverse effect on production performance of broiler chickens.

Zinc (Zn)

Zinc, acts as a co-factor in more than 300 metalloenzymes and plays an essential role in several metabolic routes, including protein synthesis (Salim *et al.* 2008). Zinc deficiency negatively affects the protein and carbohydrate metabolism in broilers and decrease feed intake and growth rate, and results in poor FCR, immunological, reproductive, skeletal and skin problems (Underwood and Suttle, 1999). In birds under heat stress, using zinc compounds in the diet could be useful due to its antioxidant and anti-stress effect (Nollet *et al.* 2008; Kucuk, 2008). The source of Zn could determine the dietary inclusion level of Zn. Huang *et al.* (2009), compared the relative bioavailability of Zn in 3 organic zinc sources with different chelation strength and ZnSO and found that the bioavailability of organic Zn sources was closely related to their chelation strength. Lina *et al.* (2009), reported that dietary supplementation of protein-bound zinc and nano-zinc oxide improved antioxidant function in broilers, strengthen the activities of antioxidant enzymes and decrease the concentrations of free radicals. The optimum dietary inclusion levels of nano-zinc oxide were 40-80 mg/kg. In the study of Star *et al.* (2012), there were no differences in production performance of broilers fed inorganic zinc sulfate (biological value 1.00, 0-40 mg of Zn/kg of feed) and organic Availa-Zn (biological value 1.64, 0-15

mg of Zn/kg of feed), until 21 d of age. They also reported a linear relationship between tibia zinc content and dietary Zn concentration up to 20 mg/kg zinc sulfate.

A level of 40 mg Zn/kg diet dry matter has been recommended by NRC for broiler chickens. The results of the subsequent researches were suggesting a higher dietary Zn requirement for broilers. *El-Wahab et al. (2013)*, reported the optimistic effect of a mixture of organic Zn-methionine (150 g/kg) and high dietary biotin on skin quality, resulting in considerably reduced severity of foot pad dermatitis in broilers exposed to wet litters. There are some reports that the nano particles of zinc supplementation could reduce the dietary inclusion rate and even at the same level as usual Zn supplement, the nano counterparts may result in adverse effects. In the study of *Zhao et al. (2014)*, broilers fed a basal diet supplemented with 60 mg/kg zinc oxide, or 20, 60 and 100 mg/kg ZnO nanoparticles. At 14 d, broilers fed diets containing 20 and 60 mg/kg nano-ZnO had higher weight gains and better feed conversion ratios than the control birds. Conversely, the body weight of broilers fed 100 mg/kg nano-ZnO was noticeably decreased after 28 days.

Mohammadi et al. (2015), supplemented broiler diets with 80 mg/kg zinc-nano-methionine and zinc-nano-max and observed an improvement in growth performance. On the other hand, the zinc-nano-sulfate supplement reduced growth performance in broilers. *Zhao et al. (2014)*, suggested that the 20 mg/kg nano-ZnO is the dietary inclusion level for broilers which is particularly lower than the dietary inclusion level of ordinary Zn supplements.

Taken together, the new research outcomes data was resulted in a revision in broiler Zn requirements. The 40 mg Zn to kg diet dry matter which has been recommended by NRC for broiler chickens, was increased in other resources so that *GFE (1999)* and *BIS (2007)* were suggested 50 and 80 mg/kg, respectively. The recommended dietary Zn level for commercial strains of broilers are 2-3 times more than *NRC (1994)* recommendation, as follow: 100, 110, 110 and 80 mg/kg for *Cobb (2013)*, *Ross 308 (Aviagen, 2014)*, *Arbor Acres (2014)*, and *Hubbard (2016)*, respectively. In conclusions, the 100-110 mg Zn/kg diet recommended by the commercial strains is more acceptable than the *NRC (1994)* recommendation; however, some new findings suggest that even slightly more Zn supplementation up to 150 mg/kg could still be useful in the health protection of broiler chickens (*El-Wahab et al. 2013*). The new researches also suggest that using nano-Zn supplements could reduce the dietary Zn inclusion rate (*Navidshad et al. 2016*).

Selenium (Se)

Selenium is a component of at least 25 selenoproteins (*Zhou et al. 2013; Naziuroglu et al. 2012*), with a seleno-

cysteine at the active site (*Naziuroglu and Yurekli, 2013*). Selenium-dependent iodothyronine deiodinases convert the inactive precursor to active thyroid hormone (*Naziuroglu et al. 2012*).

It seems that dietary Se supplementation protects the small intestine against oxidative stress. Selenium is also involved in pancreatic tissue protection against oxidative damage (*Surai, 2000*). *Placha et al. (2014)*, showed that the blood and duodenal mucosa Se concentrations and also the activities of thioredoxin reductase and glutathione peroxidase in the duodenal mucosa significantly increased in the broilers fed 0.4 mg Se/kg diet. Selenium deficiency decrease appetite and growth rate (*Fischer et al. 2008*) and also affects the elasticity of bones (*Turan et al. 1997*). Selenium is usually supplemented to poultry diets, because common feed ingredients are slightly selenium deficient. The most common selenium supplements in poultry rations are sodium selenite and sodium selenate. *Yoon et al. (2007)*, reported a higher bioavailability of Se from the organic source than inorganic sodium selenite. Recent findings have demonstrated the more bioavailability of organic forms of selenium, selenomethionine and selenium-yeast, than the inorganic forms of selenium in chicken diets (*Choct et al. 2004; Yang et al. 2014*).

Payne and Southern (2005) and *Rajashree et al. (2014)* supplemented broiler feeds with 0.30 or 0.50 ppm Se from Se-enriched yeast respectively, and found that organic Se increased tissue Se level, but it does not improve growth performance, compared with inorganic form of Se. The *NRC (1994)*, *GFE (1999)* and *BIS (2007)* recommended the same value which is 0.15 mg/kg as the required selenium for broiler chickens, while the suggested requirement for commercial strains is almost the twice of this value as follow: 0.35, 0.30, 0.30 and 0.20 for Cobb 500, Ross 308, Arbor Acres and Hubbard strains, respectively. Some more recent publications confirm that Se supplementation of broiler chickens beyond the *NRC (1994)* recommendation has been useful to birds. Some of these reports are as follow. *Yoon et al. (2007)*, fed broilers 0 (negative control), 0.1, 0.2, or 0.3 ppm of supplemental Se from SelenoSource AF (Se yeast A, Diamond V Mills, Cedar Rapids, IA), 0.3 ppm of Se from Sel-Plex (Se yeast B, Alltech, Nicholasville, KY), or 0.3 ppm of Se from sodium selenite. Their results demonstrated that Se retention had an inverse relationship with the Se supplementation level and was influenced by the source of Se. *Wang (2009)*, compared the effects of sodium selenite (0.2 mg/kg) and nano-Se (0.2 and 0.5 mg/kg) on growth rate, tissue Se retention and glutathione peroxidase (GSH-Px) activity of avian broiler. All the dietary selenium supplementation treatments significantly improved daily weight gain, feed efficiency, survival rate and GSH-Px activity.

Zhou and Wang (2011), supplemented broiler diets with 0.00, 0.10, 0.30, and 0.50 mg/kg of Nano-Se and found that supplementing diets with 0.30 mg/kg of Nano-Se was more effectively improved growth rate, feed efficiency, the Se content of tissues, and the quality of the meat of broilers. Cai *et al.* (2012), used nano-Se as a dietary supplement at 0.0, 0.3, 0.5, 1.0, or 2.0 mg/kg diet of broilers. They suggested that 0.3 to 0.5 mg/kg is the most favorable level of supplementation of nano-Se, and the maximum inclusion rate of nano-Se could not be more than 1.0 mg/kg in broiler diets. Rao *et al.* (2013), supplemented 0, 100, 200, 300, or 400 µg organic Se/kg broiler diet, under tropical climatic conditions. The results showed that the supplementation of Se did not influence body weight and feed efficiency, however, antioxidant status and lymphocyte proliferation were improved.

A negative interaction has been reported between dietary selenium and copper levels. Torki *et al.* (2015) reported a decrease in feed intake and body weight gain and increased feed conversion ratio in broilers fed diet supplemented with 1 mg/kg sodium selenite and 200 mg/kg copper sulfate, however no adverse effects were found when selenium or copper supplements were used alone.

In conclusion, it seems that dietary Se supplementation at 0.2 to 0.5 mg/kg level could be beneficial to broilers however the nature of the supplement is a determining factor. The organic Se supplements and newly introduced nano-Se supplements are apparently more effective than inorganic Se supplements.

Manganese (Mn)

Manganese is needed for normal homeostatic processes, regulation of reproduction, connective tissue and bone formation, lipid and carbohydrate metabolism, and brain function (Grandjean and Landrigan, 2014). Liu *et al.* (2015), studied the effects of Mn deficiency on the microstructure of the proximal tibia and OPG/RANKL gene expression in chicks. The results showed that Mn deficiency can affect the development of tibia in broiler chickens, leading to metaphyseal osteoporosis, which may be due to decreased OPG/RANKL mRNA expression. The chicks fed 60 mg Mn/kg had the best development of tibia.

The NRC (1994) recommended 60 mg/kg as Mn requirement of broiler chickens. A maximum tolerable level for manganese in poultry is 2000 mg/kg DM and toxic symptoms can be seen in more than 3000 mg per kg of diet (NRC, 2005). Since 1994, the organic Mn supplements become more available commercially and there are reports that confirm their more effectiveness than mineral Mn. Berta *et al.* (2004), in a study on cockerel chicks, supplemented a basal corn-soybean diet containing 23 mg/kg Mn,

with levels of 0, 30, 60 and 240 ppm Mn from inorganic (MnO) or organic (Mn fumarate) Mn sources.

The Mn source did not affect body weight, feed efficiency or the mortality rate. Li *et al.* (2005), used a basal diet containing high calcium level (the Control, containing 18.5 g Ca/kg and 20 mg Mn/kg) and 60, 120, or 180 mg Mn/kg supplement as Mn sulfate, Mn methionine E with weak chelation strength, Mn amino acid B with moderate chelation strength, or Mn amino acid with strong chelation strength. The results showed higher relative bioavailability for organic Mn sources with moderate or strong chelation strength which present acceptable resistance to interference from high dietary calcium during digestion.

Mn accumulation in the tibia was higher with Mn supplemented diets compared to the control group. Accumulation in the liver increased significantly only with supplements of 60 and 240 ppm independently of the Mn source compared to the control. At the same dosage of supplementation of the organic and inorganic Mn sources, there were no major differences between the Mn concentrations of organs and tissues.

Lu *et al.* (2007), reported that the organic Mn was more available than inorganic Mn for reducing lipoprotein lipase (LPL) activity in abdominal fat of broilers. They also found that dietary Mn could decrease abdominal fat deposition through diminishing LPL and malate dehydrogenase activities or increasing hormone-sensitive lipase activity in abdominal fat tissue. They also reported that dietary Mn upregulated muscle MnSOD gene expression which was resulted in a lower malondialdehyde level in leg muscle.

Bao *et al.* (2007), studied the effect of organically complexed Mn on broiler performance, mineral excretion, and accumulation in tissues and showed that 40 mg/kg dietary Mn supplement from organic source could be adequate for normal broiler growth to 29 d of age. Singh *et al.* (2015), supplemented broiler diets with methionine chelates or yeast proteinate forms of Mn and found an improved body weight and feed conversion ratio (FCR) and significantly reduced excretion of Mn.

The 60 mg/kg recommended as Mn requirement of broiler chickens by NRC (1994), has been updated such that the GFE (1999) has suggested 50 mg Mn/kg and the value recommended by BIS (2007) is far higher, 100 mg/kg.

The commercial strains recommendations are more near to the latter suggested requirement as follows: 100, 120, 120 and 80 mg/kg for Cobb 500, Ross 308, Arbor Acres and Hubbard strains, respectively. It seems that broilers Mn requirement is not more than 60 mg/kg diet and the additional amounts used in commercial diets are for ensuring of poultry Mn consumption.

Iodine (I)

The vital biological role of iodine (I) is in thyroid gland functions (Van Middlesworth, 1996), particularly as a component of triiodothyronine and tetraiodothyronine hormones, which are involved in metabolism regulation, intermediary cell activity, and cellular oxidation processes (Lewis, 2004). Moreover, reproduction, performance (Travnicek *et al.* 1997), circulation and muscular systems, cell development, nervous system performance, and pituitary gland action indirectly depends on the supply of iodine (Delange, 2002; Liu *et al.* 2001).

Compounds such as KI, NaI, $\text{Ca}(\text{IO}_3)_2$, and $\text{Ca}(\text{IO}_3)_2 \times 6\text{H}_2\text{O}$ are common as iodine sources (Slupczynska *et al.* 2014).

The recommended iodine requirement of broiler chickens in NRC (1994) is significantly lower than other resources and commercial recommendations. The value recorded in NRC (1994), GFE (1999) and BIS (2007) are 0.35, 0.50 and 1.2 mg/kg, respectively. The recommended iodine requirement for commercial strains of broilers are almost three times more than NRC (1994) and more near to the BIS (2007) values. The recommended values are 1, 1.25, 1.25 and 1.0 mg/kg for Cobb 500, Ross 308, Arbor Acres and Hubbard strains, respectively.

In recent years there were attempts for enrichment of poultry products with iodine. Certainly in this situation the dietary inclusion rate of iodine will be more than the chicken iodine requirement itself. In one of the such studies, Rottger *et al.* (2011), determined the influence of iodine (I) supplementation of feed, within the range of the European guidelines, on the performance of broiler chickens and iodine transfer into different organs and tissues, especially meat. They found that iodine enrichment of broiler meat using dietary iodine supplements was not so effective.

CONCLUSION

There are differences in the recommended levels of dietary trace elements between commercial strains of broilers and all of them are different from NRC (1994) recommendations. In brief, the iron requirements of broilers have been increased and the requirements of copper, manganese, zinc, selenium and iodine have decreased compared with the NRC (1994) recommendations.

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