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VITAMIN AND MINERAL DEFICIENCIES: INFERENCE IN BONE INTEGRITY IN BIRDS

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Abstract: Vitamin and mineral deficiencies, as well as imbalanced ratios between them, have a significant impact on the bone and joint health of birds. These deficiencies can cause a series of osteo-articular diseases, affecting the development and functioning of the skeleton, having negative effects on the production and welfare of birds. Deficiencies in minerals such as calcium and phosphorus result in abnormal calcification and loss of bone density. In broilers, calcium/phosphorus deficiencies result in the cessation of bone mineralization, leading to insufficient mineralization, a condition called rickets. As for adult birds, these mineral deficiencies will lead to osteoporosis, characterized by reduced bone mineral density, compromised bone structure and bone predisposition to fracture. In young birds, calcium deficiency correlated with excess phosphorus can lead to tibial dyschondroplasia. The abnormal development of cartilage from the proximal tibial growth plate in detriment to normal ossification process, can induce fractures. Tibial dyschondroplasia can also be a result of electrolyte imbalance, the condition being more frequent when the diet contains excess sodium in detriment of potassium. Manganese and zinc deficiency is the main cause of perosis. This condition is characterized by short bones, enlargement and malformation of tibial-metatarsal joint and an uncharacteristic position of Achilles tendon. Birds with copper deficiency can express lameness, bone fragility and thickening of the epiphyses. Vitamin D3 deficiency leads to abnormal absorption of calcium and phosphorus, favoring the appearance of rickets or osteoporosis, even if the calcium/phosphorus level in the diet is normal. Vitamin B complex deficiency can be associated with polyneuritis, degeneration of the myelin sheath, instability of the tibiotarsal joint, changes in limb position (varus/valgus) and perosis. Vitamin A deficiency is usually encountered after 7 weeks of age and is manifested with anorexia, weakness, stiff and uncoordinated gait, without directly affecting the locomotor system. Difficult and ataxic movement also occurs in vitamin E deficiency, but this clinical sign is specific to encephalomalacia, not to osteo-articular diseases.

Introduction

In the context of intensive poultry farming, whether we are referring to birds raised for meat or eggs, maintaining health, productivity and well-being is closely linked to nutritional balance, especially concerning the intake of essential vitamins and minerals. Nutritional deficiencies in minerals and vitamins can sometimes occur accidentally, most commonly due to the absence of vitamin-mineral premixes or their degradation during feed storage, a phenomenon influenced by factors such as temperature, humidity and storage time (Lilburn et al., 2024, Waldenstedt et al., 2006, Fallah et al., 2020, Yamada et al., 2021, Valable et al., 2018, David et al., 2023, Singh et al., 2015, 39, 40).

Mineral deficiencies

Achieving high zootechnical performances in a short period of time implies a high nutritional requirement and a well-regulated metabolic balance. Minerals play an essential role in maintaining the body's homeostasis, bone development, neuromuscular activity, immunity and production (eggs, meat). Deficiencies in the intake of these elements, either through incorrect ration formulation, deficient absorption, or through antagonistic interactions between nutrients, can have serious consequences on the health and productivity of birds (39). The main osteo-articular diseases that are caused by mineral deficiencies are: rickets, osteoporosis, perosis, tibial dyschondroplasia, but also limbs defects (39).

1. Calcium/phosphorus deficiencies

Calcium is the mineral with the largest dispense in the body, contributing to a proper functioning of the nervous and muscular systems, as well as protein synthesis, being directly dependent on calcium. Around 99% of a bird's calcium is found in its bones, in a 2-1 ratio with phosphorus. Phosphorus contributes to bone mineralization, being an important component of ATP (adenosine triphosphate), necessary for the supply of cellular energy. Phosphorus enters the structure of nucleic acids and membrane phospholipids, being essential for the normal functioning of cells. Vitamin D3 maintains calcium/phosphorus homeostasis and promotes phosphorus absorption at the intestinal level (Valable et al., 2018, David et al., 2023, Uhl et al., 2018). For the efficient absorption of calcium/phosphorus, the rations must cover the needs of these minerals for the different categories of birds. For broilers, the calcium/phosphorus ratio recommended by Fallah et al. (2020) is 7.03/4.47 g/kg feed. For laying birds, during the peak production period, the mineral requirement in the diet should reach 3.6% calcium and 0.43% phosphorus (Cheng et al., 2025, Valable et al., 2018). It has been shown that increased levels of calcium in relation to low levels of phosphorus can lead to a decrease in the absorption of both minerals, as well as other minerals and nutrients in the digestive tract. Also, in farms where the calcium/phosphorus ratio was approximately equal, the performance of the birds was minimal, the increase in phosphorus in the diet registering an immediate increase in feed consumption but also in the average daily gain (Valable et al., 2018). Although administered at the appropriate rate, Newcastle disease virus infection appears to significantly decrease serum phosphorus, an event associated with decreased egg production (Igwe et al., 2017).

1.4 Perosis ("slipped tendon")

Deficiencies of manganese, zinc and B vitamins are involved in the development of perosis. The condition is characterized by swelling and deformation of the tibio-metatarsal joint, tibia shortening, and "slipping" of the Achilles tendon. All these osteo-articular changes are accompanied by pain and lameness. Manganese deficiency affects the development of the tibia growth plate. Manganese bioavailability is reduced when the diet contains excess of calcium/phosphorus. At the level of intestinal absorption, manganese interacts with iron and zinc, thus choosing the right type of manganese is essential for maximum absorption, which should be taken into account when designing the diet. The bioavailability of this mineral varies from 29% for manganese dioxide to 174% for manganese methionine (Goluch et al., 2023, David et al., 2022, Liu et al., 2023). Manganese requirements differ according various reasearchers between 30-40 mg/kg body weight/day, with the concentration of manganese in feed being between 70-120 mg manganese/kg of feed. (Noetzold et al., 2020, Liu et al., 2023). Manganese deficiency leads to a reduction of manganese at the intestinal level, therefore the ration should be supplemented with until 50 mg/kg. Was observed that in chickens ration to which zinc was added, doesn't seem to have notable effects, but increases the resistance to fracture of the tibia. In addition, it has a positive effect on the intestinal microbiome of chickens, favoring the development of *Lactobacillus* spp. and decreasing the total bacterial load at the cecal level. (Nguyen et al., 2021, Lilburn et al., 2024). Deficiency of B vitamins, especially B1, B2, B3, B6, B7, and B12, interferes with essential processes such as collagen, methionine and choline synthesis, affecting joint and neuromuscular integrity, which can lead to the development of pernicious anemia (40).

1.2 Rickets

Rickets in broiler chickens is a metabolic condition that affects bone development and is frequently manifested with limbs weakness, lameness and difficulty of moving. Two types of rickets have been identified: hypophosphatemic and hypocalcemic. Histopathologically, in both types of rickets, a proliferation of chondrocytes was observed. Only in hypophosphatemic type, the lesions included aggregations of hypertrophied chondrocytes at the metaphysis level (Dinev et al., 2011). Hypophosphatemic rickets can appear earliest in 3-day-old chicks whose ration is deficient in phosphorus. In one-week-old chicks, lameness is the clinical sign most frequently noted in the flock. The more phosphorus-deficient is the feed, the more serious the lameness is, affecting a high number of chickens in the flock. The differential diagnosis between the two forms of rickets can be established by determining the serum concentrations of calcium, phosphorus and magnesium (Dinev et al., 2011). The necropsy examination usually reveals lesions such as bone flexibility with a tendency to bend, chondrocostal osteodystrophy, ribs deformed in "S" and lack of calcification of the tibiotarsal growth plates. There were no notable lesions of the viscera. Also, drug treatments, including antibiotics, antivirals or natural supplements did not have notable therapeutic effects (Xu et al., 2021). The defective mineralization of the bones can be attributed to the deficient calcium/phosphorus ratio in detriment of phosphorus. Thus, insoluble calcium/phosphorus complexes will form, reducing the digestibility of calcium, favoring its elimination and increasing plasma phosphorus (Xu et al., 2021; Venäläinen et al., 2006). The lack of energy caused by insufficient phosphorus sources together with bone fragility and joint pain does not help the harmonious development of muscles, which is why chickens will have poor growth performance, developing various forms of myopathies (38).

1.3 Osteoporosis

Osteoporosis is a state of hypomineralization of the bones, which will lead to bone fragility and a predisposition to fractures. During laying, three mechanisms overlap that affect bone integrity, namely: the excessive increase in the concentration of estradiol, which seems to negatively influence calcium absorption, leading to bone demineralization; on the other hand, the lack of calcium will lead to a decrease in the concentration of estrogen hormones, thus the concentration of parathormone will increase and will produce osteoclast resorption, the effect being bone demineralization; the increase in the calcium requirements necessary for eggshell formation has the same effect, namely the mobilization of bone calcium. During the period of maximum production, calcium is mobilized from both trabecular and cortical bone. The effect of calcium mobilization is the loss of bone structure, leading to osteopenia and osteoporotic fractures, especially in birds older than 40 weeks of age (Fleming et al., 2008; Yamada et al., 2021; Whitehead et al., 2000). For this reason, it is desirable that the amount of 7-9 g calcium/kg feed administered to young birds to be increased to 35-36 g calcium/kg feed for birds older than 14 weeks of age. These amounts of calcium will minimize the calcium mobilized from bones during the laying period, maintaining the health and the integrity of bones (Rodriguez-Navarro et al., 2018). From a histopathological point of view, the thickness of bone trabeculae decreases with the age of the birds and the egg production. Computed tomography highlighted the fact that with advancing age thinning of the bone marrow a is noticeable, along with a high porosity of the bone cortex, which indicates a low density of the bone (Yamada et al., 2021). Supplements with vitamin D and vitamin K appear to have a beneficial effects on the development and maintenance of bone health, but the effects are inconsistent in birds. The 25-hydroxyvitamin D is commercially available in premixes at a concentration of 4 μ g/kg feed and has benefits for bone strength in young birds, but not with the same results in adult

1.5 Tibial dyschondroplasia This condition represents 30% of bone diseases in broilers. Thus, the chondrocytes of the tibiotarsal growth plate cannot reach the normal size, suffering premature necrosis processes. The extracellular proteins produced in reduced quantities by the chondrocytes, which are also insufficiently developed, will generate an immature cartilage (dischondroplastic), formed by a dense collagen network. These immature cartilages will be resistant to resorption, so their replacement with bone tissue is no longer achieved, thereby the immature cartilage remains in the metaphysis until the broiler stops growing, when is transformed into bone tissue. This mechanism, accompanied by incomplete vascularization of the growth plate, leads to bone deformations of the tibiotarsal (varus/valgus), manifested by difficult movement and lameness. Rapid growth and development of excessive body mass predispose to the development of dyschondroplasia (Liu et al., 2023, Waldenstedt et al., 2006, Orth et al., 1994, Jahejo et al., 2022). Many conditions can predispose to the development of tibial dyschondroplasia, including deficiencies of copper, calcium, phosphorus, vitamins D, C, E, A and B complex (Liu et al., 2023, Waldenstedt et al., 2006, Orth et al., 1994, Jahejo et al., 2022). Histopathological examination of the tibial growth plate of broilers revealed progressive disorganization of chondrocyte architecture. Chondrocytes showed degenerative changes, including rounding and clusters of pyknotic chondrocytes. Chondrocyte hyperplasia associated with vascular atrophy and angionecrosis leads to the formation of a rigid, avascular cartilaginous mass, characteristic of tibial dyschondroplasia (Jahejo et al., 2022). mass, characteristic of tibial dyschondroplasia (Jahejo et al., 2022).

1.6 Other vitamins and minerals

The recommended dose of vitamin D3 for broilers is 200 IU/kg. Supplementing the ration with vitamin D3 up to 3000 IU/kg leads to increasing the average daily weight gain of birds, as well as to increasing serum calcium and phosphorus concentrations, which favor good bone mineralization. Supplementation with D3 has shown an increase in the length, weight and mineral content of the tibia and femur (Wei et al., 2024, Sakkas et al., 2019). In broilers with yarus/valgus, the serum vitamin D concentration was up to 28% lower. (Newbrey et al., 1988). In broilers with D3 deficiency, conditions such as sternum deformation, support on the hocks, soft and flexible claws and beak, as well as other skeletal malformations have been recorded (40). In humans, increased intake of vitamin B6 is correlated with better bone mineralization, being the substrate of alkaline phosphatase in bone formation. The deficiency affects collagen formation and thus bone integrity. Supplementation with vitamin B6 has positive effects in the case of osteoporotic fractures, stimulating the collagen, a component of the bone matrix. (Welan et al., 2023). Dai Z et al., 2015). In broilers whose ration included only 2.5 mg/kg riboflavin, decreases in feed consumption and bird weight were observed, which was remedied by supplementing the ration with up to 4.0 mg/kg riboflavin (B2) (Shastak et al., 2023). The fracture resistance of the chickens' bones with B6 deficiency is lower compared to thosewith adequate vitamin intake. Although the bones have normal mineralization, they are more fragile, due to changes in collagen. This fact shows that B6-deficient bones are osteopenic (Masse et al., 1996). Thiamine (B1) deficiencies are noticed in birds by the appearance of polyneuritis. (Cai et al., 2009). The lack of thiamine causes a decrease in ATP synthesis in nerve cells, affecting neuronal energy metabolism and leading to axonal degeneration and demyelination, which explains the appearance of polyneuritis. Clinical manifestations of this deficiency on the h nutritional myopathy, clinically manifested by lethargy, difficulty moving, muscle weakness predominantly in the chest and limb muscles (39, 40).

Conclusions

Osteoarticular diseases in poultry, and especially in broilers, are a multifactorial problem, with a major impact on bird welfare and economic performance. Bacterial infections with Staphylococcus spp., Mycoplasma synoviae and avian reoviruses are some of the potential infectious causes of osteoarticular diseases in poultry. To ensure the optimal intake of vitamins and minerals has a significant impact on the bone and joint integrity of birds, a fact demonstrated by the multitude of diseases installed in birds with nutritional deficiencies. Also, environmental factors such as high density, poor ventilation and inadequate feed management contribute to the increased incidence of these diseases, especially in breeds selected for accelerated growth. Effective prophylaxis involves correcting the nutritional balance, controlling infectious agents and optimizing environmental conditions, in order to maintain osteoarticular health, bird well-being and production profitability.

