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Mineral nutrition in broilers: Where are we at?

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ABSTRACT

Mineral nutrition remains an essential consideration for broiler nutritionists. The complex interactions between different mineral sources, other ingredients, and the broiler itself, coupled with their relatively low costs, have led to the current status of elevated levels of minerals in broiler diets. New perspectives on sustainability have brought about a rethink of the way we formulate practical diets. There are considerable opportunities for reducing macro minerals such as calcium and phosphorus, particularly for older birds. Our understanding of attaining an ideal dietary cation-anion balance is confused by the lack of clarity in measuring the balance and the bird's ability to perform over a wide range of relative values. The use of 'organic' mineral sources, with a higher mineral availability and uptake, will reduce the levels of trace minerals used in broiler diets. The reduction of mineral levels will lead to more cost-effective feeds, with a concomitant reduction in the size of the environmental footprint of broiler production.

Keywords: mineral, organic, DCAB, calcium, phosphorus

Introduction

An essential component of the overall health and well-being of the broiler chicken is ensuring that mineral nutrition, in all its facets, is adequate. As growth rates increase and production systems intensify, mineral nutrition will become more critical. Most minerals in the bird occur in the skeleton, so skeletal integrity is perhaps the critical issue. However, minerals play a role in all aspects of metabolism, both as cofactors of enzymes and controlling free radicals within the body (Goff, 2018). There is an increasing awareness of how trace minerals impact aspects as diverse as the immune system, gut health and carcass quality. Coupled with this is the drive to produce broilers more sustainably. Demand for scarce resources such as mineral phosphate and environmental pollution caused by animal production systems is becoming critical. Nutritionists will have to meet the increased demands of society, together with the potentially higher nutrient requirements of broilers, while minimizing the residual amount of minerals in their manure. Research into trace mineral nutrition has lagged behind other areas due to the relatively low cost of inclusion in premixes (Bao and Choct, 2009). These low costs also explain why high levels of minerals are often included in broiler diets. Increasingly, mineral levels in broiler diets will come under scrutiny, and providing excess to requirements will have to be curtailed (Leeson, 2018). The formulation of diets that target precision nutrition will eventually demand extensive use of alternative mineral sources, which could be added to feeds to promote specific responses (Vieira, 2008).

Nutritionists will need to redefine the mineral levels used in broiler diets for several reasons. These include decreasing feed costs while improving performance; enhancing the sustainability of broiler production; reducing antagonistic interactions with other nutrients; a better understanding of the impact of using the phytase enzyme and other additives; the possible formulation of diets on an 'available' nutrient basis; and the potential for using alternative mineral sources. This paper aims to give a broad overview of the current status of mineral nutrition as currently understood and practised and will consider some of the aspects that need to be modified or improved upon. It will deal with mineral uptake and how this impacts the determination of requirements, together with a discussion on how to supply broilers with optimal rather than adequate macro and micro minerals levels.

Availability, Absorption and Requirements

Much of the research on the mineral requirements of chickens is limited, outdated, or has not remained abreast of modern production systems. As mentioned, this has much to do with

the low impact mineral inclusion has on broiler profitability. Reassessment will entail reappraising requirements for current genotypes and production systems. Any study of mineral nutrition and metabolism is complicated by the interactions between the various elements themselves, with other feed components and the bird itself. Understanding of the mineral requirements of broilers is far from complete. Several factors actively interfere with mineral absorption and should be considered when setting out to meet the bird's requirements (Ashmead, 1993; Vieira, 2008; Linares, 2018; Goff, 2018).

The bird relies on homeostatic mechanisms to control the uptake and excretion of essential minerals, both at the cellular and systemic levels. This ability maintains mineral concentrations within a narrow range. The efficiency of mineral uptake is enhanced by the up-regulation of mineral transporters when a mineral is marginally bioavailable in the diet. When levels exceed requirements, these transporters are down-regulated, making them less efficient (Hu et al., 2018).

Absorption occurs mainly in the proximal small intestine. The mechanism of mineral uptake is complex, with both paracellular and transcellular absorption taking place. Paracellular absorption occurs when the concentration of a mineral in the digesta adjacent to a tight junction exceeds the concentration in the extracellular fluid. In addition, minerals complexed with various substances in the diet (water, amino acids (AA), peptides, volatile fatty acids) may move through the tight junctions employing solvent drag, which accounts for a substantial portion of the minerals absorbed by the animal (Goff, 2018).

When dietary mineral concentrations are low, the body relies on transcellular absorption. The phospholipid bilayer that makes up the cell membrane of the enterocytes of the gut is impervious to both water and minerals. Hence, some form of specialized transcellular transport mechanism is required (Goff, 2018). This process occurs in stages. In step one, specialized transport proteins (mainly the ZIP4 transporter) in the cell membrane facilitate the diffusion of mineral ions with a specific valence or charge associated with them. This pathway is highly regulated, and the bird can only utilize metallic ions once taken up by a transporter (Linares, 2018). Thus, minerals may compete for uptake pathways, and excess of a single mineral may impede the uptake of others and may limit bird performance.

In some cases, transport proteins may be regulated to allow minerals to enter the enterocytes only if the body needs that particular mineral. Once within the cell, the mineral needs to move from the apical membrane to the basolateral membrane. This may occur through simple diffusion, but most minerals require specialized proteins or transport vesicles to carry out this process. The final step involves removing the mineral from the cytosol of the

epithelial cell into the interstitial space below the tight junctions. In many cases, this involves moving minerals against their concentration gradients, requiring ATP powered protein mineral pumps (Goff, 2018).

The availability of minerals differs between different sources, making it impossible to establish a single absorption rate that would fit all sources of different minerals. Carbonates and sulphates are the most widely used feeding sources. The oxides of most minerals are poorly available. Organic trace minerals tend to be more available than inorganic sources, but this depends on their ligand and the mineral status of the animal (Suttle, 2010). Regardless of a minerals source, solubilization of all inorganic minerals is required in order for absorption to proceed. This process is pH-dependent; therefore, an acidic environment in the proventriculus improves their solubilization. By contrast, the neutral or alkaline pH of the small intestine reduces mineral solubility. Further complexity of mineral availability is demonstrated in the calcium (Ca) digestibility of limestone. The material's underlying geology impacts calcium digestibility, but absorption is higher when particle size is larger (Ravindran 2017, Li et al., 2021).

Absorption and utilization of minerals are not linear, and generally, low dietary levels are utilized with greater efficiency (Shuttle, 2010). In addition, other physiological responses may impede mineral absorption. For example, inflammation, which is a component of digestion and forms a specific component of any immune response, damages or retards the function of the tight junctions, which affects the dynamic of trace minerals within the body (Klasing, 2007). Chronic inflammation blocks the absorption of trace minerals and increases their excretion, inducing a nutrient-deficient state. Zn and Fe are sequestered by the liver and the spleen from circulation during septicaemia while Cu is released. Nutritionists need to anticipate immune challenges and ensure that the bird has adequate mineral reserves.

All bacteria require minerals for replication. They are, in essence, the only 'essential' component of the diet as bacteria can synthesize all the organic molecules they require. High levels of dietary trace minerals, specifically Zn and Fe, may well promote microbial growth. Thus, the routine use of high trace mineral levels in the diet needs to be reconsidered (Leeson, 2018).

It is unlikely that the underlying physiological mechanisms for mineral uptake will have changed as broiler genotypes have evolved; however, as the growth rate potential of broilers increases, the birds will produce more meat from less food. Thus, fast-growing broilers may exceed standard mineral absorption mechanisms. The only way to achieve constant mineral supply at a cellular level is by increasing dietary levels or using sources with higher relative

bioavailability. One such option is metal amino acid complexes (organic minerals), which will be discussed below.

Calcium and Phosphorus

The situation regarding the Ca and phosphorus (**P**) requirement of broilers is not clear cut. The widespread introduction of exogenous phytase into broiler diets has introduced a new dynamic to mineral nutrition. The skeleton contains about 99% of the Ca and about 80% of the P in the body (Korver, 2020). The mineral crystal of bone (hydroxyapatite) contains both Ca and P, which interact before and after absorption from the digestive tract (Rawlinson et al., 2009).

When considering the Ca and P nutrition of the broiler chicken and the impact that this is likely to have on skeletal integrity, the entire life cycle should be considered. Mineral nutrition begins 10 to 12 days before the egg is laid when the follicle matures, and vitamins and minerals are deposited in the yolk. Skeletal development begins during incubation, with embryonic bone growth reaching a plateau after 19 days of incubation (Angel, 2007). The embryonic growth rate of fast-growing broilers may exceed its capacity to deposit bone minerals, leaving a porous bone structure not fully mineralized at hatch; Torres et al., 2018). Egg Ca level would appear to be adequate due to Ca from the shell being transferred to the embryo, whereas towards the end of incubation, P and most other mineral reserves in the yolk are reduced (Angel, 2007; Li et al., 2016). Trace mineral nutrition of the breeder hen influences the bone development of the embryo, and any deficiency that occurs during egg formation can have significant repercussions (Nasir and Peebles, 2018). However, increasing embryo skeletal development by increasing egg mineral content (increasing dietary levels in the breeder diet) has limited success. It would appear that the use of highly available organic sources of trace minerals will boost the mineral status of the embryo and the chick that ultimately hatches (Surai, 2013; Vieira, 2020). Younger hens produce eggs with lower levels of critical minerals, due to which the bones of their embryos are poorly mineralized (Torres and Korver, 2018).

The broiler skeleton is poorly mineralized at hatch, but skeletal growth and bone mineralization occur rapidly during the first two weeks of life (Angel, 2007). Rapid bone formation occurs in the period 4–18 days of age, and mineralization from 4–11 days of age, regardless of the bird's growth potential. However, the cortical bone in rapidly growing genotypes is less mineralized (Williams et al., 2004).

An appreciation of phytate and commercial phytases and their mode of action is essential if Ca and P nutrition are to be understood. Amerah et al. (2014) demonstrated that the

inclusion of phytase had a positive effect on phytate hydrolysis and that high dietary Ca intake suppresses growth. Faridi et al. (2015) conducted a meta-analysis of the effects of non-phytate phosphorus on broiler performance and tibia ash concentration, both with and without phytase inclusion. They demonstrated that the higher the Ca level of the diet, the higher the P level needs to be and that the addition of phytase to the diets allows for lower levels of P, and thus Ca, to be used. Many workers have shown conclusively that reduced dietary Ca and P levels, particularly in the presence of the phytase enzyme, lead to improved broiler performance without necessarily reducing skeletal integrity (Li et al. 2016; Kim et al., 2017; Cheng et al., 2018; Moss et al. 2019; Walk et al. 2012). The current trend is for nutritionists to use higher levels of phytase (1000 FTU/kg or more), further complicating the situation. Kim et al. (2017) fed diets containing 3.5 g/NPP and 1000 FTU phytase at calculated Ca levels of 6, 7, 8, 9, or 10 g/kg. An increase in the level of Ca resulted in a linear decrease in gain, feed intake (**FI**) and FCR in Ross 308 broilers up to 21 days of age (Table 1). The percentage ash, Ca and P in the bone and tibia breaking strength were not affected by the Ca level.

The question which needs to be addressed here is what levels should be used in commercial practice? Table 2 carries a summary of both the primary breeder recommendations and the values used commercially in the USA. As our understanding of the use of the phytase enzyme increases, research organizations have published recommendations for Ca and P in the diets of broiler chickens (Table 3). Before any comparison can be considered, it needs to be appreciated that different systems for measuring P were used in their compilation. The values used by the primary breeding companies and those of commercial practice are expressed as Available Phosphorus (AP). The values of Khaksar et al. (2017), the CVB (2018) and Angel (2020) are expressed as digestible phosphorus (DP). Plumstead et al. (2007) presented data showing that AP values are 11% higher than DP values. Applying this correction, it is possible to conclude that the primary breeding companies' values proposals for the starter phase and those proposed by the research community largely overlap. Commercial practices match these recommendations, but there has been a noticeable alteration of the Ca:P ratio over the past five years. Even though research shows that the Ca and P levels may be reduced during the grower and finisher phases, current practices do not reflect this. Perhaps there is an opportunity to adjust both Ca and P specs downwards, particularly if higher doses of the phytase enzyme are being used.

Any discussion on Ca requirements in broilers would be incomplete without a mention of digestibility. Although nutritionists have tended to use total Ca for diet formulation, the move

towards more sophisticated P digestibility measures necessitates a closer look at Ca digestibility and its impact on P uptake (Angel, 2017; Ravindran, 2017; Walk et al. 2021). It is widely assumed that all sources of Ca are equally digestible, but the data detailed in Table 5 indicates that this is not the case. The degree of variability can also be seen in Table 5. The challenge nutritionists face, is not the absence of suitable data for feed ingredients, but rather the variation resulting from different methods of determination values. This variation arises from inherent ingredient variability, the impact of the physical form of the ingredient, particle size, for example, and the methodology used for the determination (Walk et al., 2021). It is possible that this variability that has limited the adoption of formulating diets on an available basis.

Ignoring Ca digestibility could lead to an oversupply of Ca, which will negatively affect P uptake. It may also negatively impact the digestibility of protein, fat and even energy, with a consequent worsening of broiler performance (Li et al. 2021; Walk et al., 2021). Angel (2020) published values for the requirement of digestible Ca for all phases of broiler production (Table 4). These data will likely require refinement, but they represent a valuable starting point. Advances in understanding are not possible if measurements are not made, however rudimentary the values appear.

Digestibility of limestone, the most critical source of Ca in broiler diets, is dependent on its innate solubility (primarily determined by the underlying geology and hence chemical structure of the material), as well as on particle size. The average ileal Ca digestibility coefficient for limestone is 0.52 ± 0.12 (Walk et al., 2021). Ravindran (2017) found that the Ca digestibility of different limestone sources ranged from 52–62%, while the digestibility of coarse particles was higher than that of fine particles (71% versus 43%). Sources that are too fine lead to rapid Ca digestion, which is likely to retard phytase efficacy (Li et al. 2021). From a practical point of view, it is probably safe to assume that the Ca spared through the use of the phytase enzyme is all available to the bird (it is in its elemental form). If this assumption is correct, the amount of digestible Ca made accessible through phytase use is significant. Assuming a typical corn/soy diet contains 7 g/kg of digestible Ca (Angel 2020). If a high dose of phytase was used (1000 FTU/kg), a sparing of at least 1.9 g/kg of Ca could be expected, meaning that phytase supplies nearly 30% of the bird's requirements for Ca.

The Acid-Base Balance

The acid-base homeostasis of an animal plays a crucial role in its health and production. Homeostasis entails maintaining intracellular and extracellular protons (H⁻) at a constant level (Quiniou and Narcy, 2019). Diet can influence the body's acid-base balance, and the dietary

cation-anion balance (**DCAB**) may play a role in this regard. The DCAB is the difference between total dietary cations and anions. Electrolytes are classified either as cations that carry a positive charge or anions that are negatively charged. Sodium (**Na**), potassium (**K**), sulphur (**S**), and chlorine (**Cl**), referred to as ‘strong ions’, exert the strongest ionic effects on the DCAB. Excessive cations in the diet lead to alkaline diets, while excessive anion inclusion leads to acidic feeds. The calculation is carried out in milli-equivalents because it is necessary to define the balance of charges present in the diet. Equation 1 is what should be used to determine the DCAB of the diet. The constants used represent the molecular weight of each element. The exclusion of S from the generalized equation used for poultry is the norm. This is unfortunate, as S levels in broiler diets have increased by feeding ingredients such as lysine sulphate, sodium sulphate, DDGS and canola.

Equation 1:

$$\text{DCAB (mEq)} = (\text{mg/kg Na} \div 23) + (\text{mg/kg K} \div 38.1) - (\text{mg/kg Cl} \div 35.5) - (\text{mg/kg S} \div 16)$$

In reality, electrolyte imbalance seldom occurs because the buffering systems within the body ensure near-normal physiological pH. In extreme conditions, the need for maintaining buffering capacity seems to harm other physiological conditions (which have the first call on the electrolytes), thereby producing or accentuating potentially debilitating conditions. An electrolyte imbalance can cause tibial dyschondroplasia in broilers and affects the metabolism of several essential amino acids, particularly lysine and arginine. In most situations, it seems as though an overall diet balance of 250 mEq/kg is optimal for normal physiological function, although the range of published figures varies from 200–300 mEq/kg (Borges et al., 2011; Quiniou and Narcy, 2019). In a recent experiment, Chrystal et al. (2020) demonstrated that maintaining a constant DCAB or not made no difference to broiler performance when offered low crude protein diets, indicating that the bird can self-regulate its electrolyte balance. When dietary Cl is elevated, adjustment of the diet cations may be required to maintain overall balance. Chloride levels can be reduced, although most species have requirements of around 1.2–1.5 g/kg of the diet, below which deficiency symptoms will develop. Bear in mind that DCAB can impact microbial fermentation. It may be true that the bird can self-regulate its DCAB, but it cannot alter the DCAB of the digesta in the gut to the same extent. Feed changes, along with changes in the DCAB, may negatively impact gut microflora and gut health.

Understanding Na usage in broiler chicks is less clear. The NRC (1994) suggests that the requirement of the broiler may be as low as (1.2 g/kg). However, far higher levels are used in commercial practice, as Na is often viewed as a growth promotor because it stimulates water and FI intake. Commercial dietary Na levels depend on the ingredients used in the manufacture of the diet (maize versus wheat), how the litter is managed and the personal preferences of the nutritionist formulating the diet.

The NRC (1994) lists the toxic level of S as 14 g/kg; however, this is based on data from 1960. Leeson and Summers (2005) demonstrated that broiler performance was retarded when high levels of dietary S were present in the feed. Bobeck et al. (2013) showed that FI intake in broilers was significantly reduced when the dietary S level reached 3.6 g/kg using copper and zinc sulphate at supplemental levels. However, broiler performance was not affected at these levels. The use of sodium sulphate (Na_2SO_4) as an alternative to sodium bicarbonate needs further consideration. The S it contains will change the DCAB to some extent, and the impacts of these changes need to be better understood. The current understanding is that the bicarbonate ion helps the bird overcome the adverse effects of heat stress, rather than the fact that it does contain Cl.

Trace Minerals

Trace minerals are essential in dietary terms and have traditionally been supplemented in broiler feeds using inorganic mineral salts. This is relatively inexpensive because of the low levels of trace minerals used in broiler diets, resulting in high levels being included for safety reasons. For example, Aviagen (2019) suggest that 110 mg/kg of Zn be included in all diets, while Cobb-Vantress (2018) recommend 100 mg/kg. Bao et al. (2009) conclude that the total Zn requirement for broiler is around 60 mg/kg until 14 days of age, increasing to 70 mg/kg after that. An additional consideration is that the use of the phytase enzyme frees Zn bound to vegetable matter for use by the broiler (Schlegel et al., 2013). Mineral salts often escape scrutiny concerning their quality and purity. Variable availability of the mineral and the presence of contaminants (heavy metals) are essential considerations that are often ignored (Vieira, 2008). Heavy metals are widely present in the earth's crust, soil, water and, almost all plant and animal tissue but occur in tiny amounts. They are generally classified as cadmium, lead, mercury, molybdenum and arsenic and as such, heavy metal pollution in animal feed is unavoidable (Korish and Attia, 2020).

The organic minerals (a misnomer because all minerals are inorganic by definition, rather they should be termed organically complexed minerals) comprise minerals bound to organic

compounds. These include compounds such as protein (proteinates), organic acid (propionate, butyrate), AA (amino acid complexes), methionine hydroxy analogue (MHA) and carbohydrates (alginates) (Vieira,2008). Organic minerals are chemically inert due to the covalent and ionic bonds between the mineral and a ligand and employ uptake mechanisms that differ from mineral salts. A molecule containing a single AA (1:1) is called a mineral complex, while molecules with two or more AA (1:2) are mineral chelates. Chelates have reduced conformation and stability, thus decreasing solubility and, ultimately, mineral availability. Also, they are not affected by factors that lead to precipitation, as happens to minerals ionized after salt solubilization (Boa et al.,2009). Organic mineral inclusion in broiler diets allows for a reduction in the total mineral inclusion, reducing the potential for environmental contamination. An additional benefit is that organic minerals are less likely to be contaminated with heavy metals (Vieira, 2019; Leeson 2018).

Differences in stability, solubility and absorption pathways occur, depending on the type of bond and the ligand used. However, due to their stability and small size, most complexed minerals are not altered during their passage through the digestive tract. Small amounts of minerals are bonded to proteins and peptides but will dissociate when the proteins are hydrolyzed. On the other hand, AA have a much higher binding affinity for minerals. Their ligands have been shown to be absorbed through the enterocyte, utilizing the amino acid transporter system (Gao et al., 2014). Once a metal is bound to an AA, both its size and charge change. Consequently, there are differences in which AA can better deliver the metal to the animal. After absorption, organic minerals may present physiological effects, which improve specific metabolic responses such as the immune response (Linares, 2018).

Many studies have demonstrated the benefits of organic minerals, both on animal metabolism and performance, but the detection of positive effects on live performance is less consistent (Vieira, 2008). The real advantage of using organic minerals is that they can be supplemented at lower concentrations than mineral salts without impacting bird performance (M'Sadeq et al., 2018). Zinc (**Zn**) uptake in broilers supplemented with Zn AA complexes, compared to those supplemented with Zn sulfate, was shown to be higher. Higher Zn uptake significantly impacted villus length, gastric microbiota, and a lower FCR in young broilers (de Grande et al., 2020).

The nutrient status of the embryo and the chick that subsequently hatches depends on the nutrient status of the breeder hen. A sound eggshell ensures hatchability but also prevents penetration by harmful bacteria and avoid contamination of the embryo. The importance of elevated mineral levels on embryo survival and chick vitality has already been addressed.

Studies show that organic minerals in breeder diets improve eggshell quality and hatchability (Hudson et al., 2004; Favero et al., 2013; Ebbing et al., 2019; Vieira, 2020) and the oxidative and immune status of the day-old chick (Surai, 2013).

Organic minerals are considerably more expensive than inorganic salts, and their benefits have yet to be fully elucidated. Regardless, it is probably worth considering the use of organic minerals in all starter diets. This is when the demand for minerals is greatest (skeletal development, immunocompetence and development of the gastrointestinal tract). The starter phase also represents a relatively small proportion of the total feed consumed. However, experience has shown benefits to feeding organic minerals throughout the broiler product cycle. For example, the use of organic Zn can achieve enhanced Zn status of the carcass, with a concomitant reduction in footpad dermatitis and skin tearing in carcasses (Saenmahayak et al., 2010). For this reason, a phased approach to the introduction and evaluation of organic minerals into any series of broiler diets is advised.

Knowing how to formulate diets using organic minerals can be confusing as different suppliers of commercial products use different approaches. Total replacement technology is the preferred method of some suppliers, with (Araujo et al. 2019) finding that the removal of inorganic supplementation leads to an enhanced breeder and chick performance. These results are in agreement with those of De Grande et al. (2020). On the other hand, (Hudson et al. 2014) demonstrated that the best performance was achieved using a 50:50 inorganic to organic ratio. From a practical perspective, it would perhaps be best to use various mineral sources, thus offering the broiler several different uptake mechanisms for each mineral. Understanding of this aspect will be enhanced by the ongoing research effort currently in progress.

Conclusions

In conclusion, the broiler industry continues to overfeed most minerals. Real opportunities exist for reducing mineral levels in broiler diets, particularly in the case of older birds. This will improve the sustainability of broiler production. The advent of exogenous phytase enzymes has given nutritionists more latitude regarding the levels of Ca and P we use in commercial diets and will allow for a reduction of the levels of both minerals. This will alleviate the potential adverse effect of high levels of dietary Ca. A better understanding of the digestibility of Ca in the ingredients used will enable this relationship to be fine-tuned. What construes an ideal DCAB is a little vague, as no clarity exists as to how it should be measured. In addition, it may be of lesser importance, as the bird would appear to be able to

self-regulate ionic balance within a narrow range. In contrast to the macro-minerals, providing the broiler embryo and young chick with adequate levels of trace minerals would appear to be paramount. The use of highly digestible organic mineral sources in the diets of breeding hens has been shown to boost the mineral status of the embryo and the broilers that subsequently hatch. Organic mineral use in broiler diets has been demonstrated to enhance performance and carcass quality whilst reducing overall trace mineral inclusion and subsequent polluting effects.

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Table 1. Effects of dietary calcium concentrations in low non-phytate phosphorus diets (3.5 g/kg) containing phytase (1000 FTU/kg) on selected growth performance and tibia characteristics of growing broiler chickens (after Kim et al., 2017).

	Analyzed dietary Ca concentrations (g/kg)					
	5.2	6.6	7.4	8.6	9.2	9.7
35 d Weight (kg)	1.931	1.964	1.912	1.954	1.888	1.889
FCR*	1.733	1.686	1.757	1.73	1.798	1.825
Tibia Ca%	42.5 ^c	46.4 ^b	49.7 ^a	47.6 ^{ab}	48.4 ^{ab}	48.4 ^{ab}
Tibia P %	26.0 ^c	27.8 ^b	28.8 ^{ab}	28.8 ^{ab}	29.2 ^a	29.0 ^{ab}
Breaking strength (kg/cm ³)	20.4	19.5	22.3	20.1	23.7	22

* Significant on linear basis

Means within columns not sharing common superscripts are significantly different (P < 0.05)

Table 2. A summary of the recommendations for calcium and available phosphorus made by the primary breeding companies, together with levels used in commercial practice

	Cobb 500 (Cobb- Vantress, 2018)	Ross 308 (Aviagen, 2019)	3rd Party Industry Data (Reported by Angel 2017)	3rd Party Industry Data (Reported by Jasek 2021)
Starter				
Calcium (g/kg)	9.0	9.6	10.0	9.35
Available P (g/kg)	4.5	4.8	4.8	4.95
Ca:AvlP ratio	2.0	2.0	2.1	1.89
Grower				
Calcium (g/kg)	8.4	8.7	10	8.64
Available P (g/kg)	4.2	4.35	4.4	4.6
Ca:AvlP ratio	2.0	2.0	2.3	1.88
Finisher				
Calcium (g/kg)	7.6	8.1	8.5	7.93
Available P (g/kg)	3.8	4.05	4.0	4.2
Ca:AvlP ratio	2.0	2.0	2.1	1.88

Table 3. : A summary of the recommended levels for calcium and digestible phosphorus in broiler diets (after Khaksar et al., 2017; Angel 2020; CVB 2018)

	INRA (2017)	Angel (2020)	CVB (2018)
Starter			
Age (days)	0-10	0-10	0 -10
Calcium (g/kg)	9.0	10.0	8.8 – 9.2
Digestible Ca (g/kg) ¹		6.1	
Digestible P (g/kg)	4.3	5.3	4.0
Ca:DigP ratio	2.10	1.88	2.20
Grower			
Age (days)	11-20	10–16	10-30
Calcium (g/kg)	7.6	8.0	6.8 – 7.1
Digestible Ca (g/kg)		7.0	
Digestible P (g/kg)	3.75	3.9	3.1
Ca:DigP ratio	2.02	2.05	2.19
Finisher			
Age (days)	21-30	16-25	
Calcium (g/kg)	6.4	7.0	
Digestible Ca (g/kg)		6.0	
Digestible P (g/kg)	3.0	3.1	
Ca:DigP ratio	2.10	2.26	
Post Finisher			
Age (days)	31-40	25-31	30 - 40
Calcium (g/kg)	5.4	6.0	6.2 – 6.4
Digestible Ca (g/kg)		5.0	
Digestible P (g/kg)	2.5	2.4	2.8
Ca:DigP ratio	2.16	2.5	2.21

1. Angel (2020) is the only worker to propose a digestible Ca recommendation.

Table 4. Calcium digestibility of feed ingredients (after Anwar et al., 2015; Ravindran, 2017; Stein, 2017; Angel, 2017; Trairatapiwan et al., 2018; Walk et al. 2021).

Ingredient	Digestibility %	Range
Limestone	53	41–70
Coarse limestone (850 nm GMD)	70 ¹	
Fine limestone (150 nm GMD)	55 ¹	
Meat and Bone meal	50	41-56
Fish meal	46	
Poultry by-product meal	29	
Dicalcium phosphate	42	28-45
Monocalcium phosphate	35	32-34
Soya bean meal ³	54	46 – 66
Maize	<60	46 – 70
Canola meal ³	70	31–70 ²

1. Estimated values

2. These values derived from swine

3. These values assume the use of phytase which