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Metabolic limits of calcium and phosphorus digestibility

Several recent articles have been making the case for standardizing nutrient requirements based on nutrient availability in feed ingredients. Nutrient digestibility is also a factor that makes the argument for standardizing requirements. Many changes have occurred over the years in the poultry industry. Animal physiology and development results in larger birds that grow faster, produce more eggs, and utilize nutrients more efficiently than in the past. In addition to the changes in animals, nutrient availability in the diet is one factor. Another factor is the amount of space for exercise and available sunlight. Increasing consumer demand, industry changes, environmental concern and pending legislative and policy interests are converging to significantly influence the role of nutrient management in livestock production.

The nutrients of interest are calcium, phosphorus and vitamin-D_3. To understand consistent terminology and generate reliable requirements, it is imperative to understand factors affecting metabolism and how to control them to ensure efficient nutrient utilization.

Absorbed calcium and phosphorus in birds is used for bone formation. Calcium is found in three forms: hydroyzapatite in a ratio of 2:1 with phosphorus; ionized calcium in the extracellular matrix, and bound calcium attached to proteins or anions.

There are two ways calcium is absorbed across the intestinal wall: through transcellular and paracellular routes. Transport through transcellular activity occurs mainly in the upper section of the intestine and vitamin D_3 is involved in the absorption process. The paracellular pathway is nonsaturable, so calcium absorption occurs throughout the entire small intestinal tract with vitamin D_3 involvement. This pathway is a process of facilitative diffusion, dependent on vitamin D_3. While there are many transporters in the upper section (duodenum and upper jejunum) of the intestine for transcelluar absorption, the fact that paracellular routes are present throughout the entire, longer intestinal tract allows for greater surface area of nutrient absorption through the vitamin D_3 dependent route. The length of digestion time in lower portions of the intestine may also account for significant absorption of calcium and phosphorus through paracellular routes. It is important to note that these statements indicate calcium *absorption* is regulated by physiology and

Continued on p. 2

Metabolic limits of calcium and phosphorus digestibility

(Continued from p.1)

nutrition, with vitamin D_3 playing a major role. Calcium *metabolism* is regulated by vitamin D_3, calcitonin and parathyroid hormone.

Vitamin D_3 is well researched as a steroid hormone required in poultry diets and has a clearly established role in calcium and phosphorus metabolism along with other vitamin D metabolites. Vitamin D_3 acts on genes that are responsible for the intestinal active phosphate transport. Vitamin D_3 stimulates intestinal calcium absorption through a process involving a vitamin D_3 metabolite. Vitamin D_3 from an animal source is more available (60-70%) than vitamin D_3 from a plant source to poultry.

Phosphate is found in inorganic forms or bound to protein, lipid, DNA or RNA (organic compounds). Cellular and extracellular levels of phosphate are maintained within a tight range to prevent negative effects in many biochemical processes. Intestinal phosphate absorption is a component of phosphate homeostasis within that tight range. Phosphate homeostasis in the body is the result of intestinal uptake of dietary phosphate, renal phosphate reabsorption and excretion and the exchange of phosphate between the extracellular and bone storage pool (Marks et al., 2010 and Wideman, 1987). Phosphate transport across the intestinal wall is sodium dependent and independent, depending on the transporters involved. Along with animal age and source of calcium, phosphate transport may also influence the rate of calcium absorption in the small intestine of chickens (Walk, et al., 2012). As an example of this, a large amount of calcium would be absorbed in the duodenum and upper jejunum if the birds were fed a diet with the majority of calcium coming from calcium carbonate processed from limestone or mono- or dicalcium phosphate. If the majority of dietary calcium comes from a source that is not highly available in the gastrointestinal tract a lesser concentration of calcium may be absorbed in the duodenum.

Obviously, calcium absorption and metabolism is regulated by many factors. Birds' calcium status at any point in development influences their calcium absorption level. That level is regulated by the parathyroid hormone and vitamin D_3. The interaction of calcium, phosphorus and vitamin D_3 must be understood to understand calcium and phosphorus metabolism. In addition, this knowledge should give consideration to different breeds or strains of poultry, age, feed ingredients, and any phytase inclusion levels. Another indicator of calcium and phosphorus metabolism is the amount of these nutrients that is excreted. This indicator results from many factors, the source of the nutrients (which effects digestibility or availability), the dietary concentration of nutrients, the parathyroid hormone status, reproductive status and blood pH. As an example in healthy birds in normal conditions, domestic chickens excrete under 2% of filtered calcium in urine, due to the role of the parathyroid hormone's stimulation of the kidneys to reabsorb most of the filtered calcium.

The ratio of calcium to phosphorus may also influence digestion and absorption of the nutrients in the poultry gastrointestinal tract. Physiological responses can be initiated dependent on the ratios. This means that potential detrimental interactions between calcium and phosphorus should be minimized, i.e. high dietary calcium may result in high digesta pH and thus negatively affect phytase efficacy and mineral absorption. When comparing ratios, consistent terminology is necessary and other *Mineral Writes* articles have made this argument. Available phosphorus and non phytate phosphorus are used interchangeably. Since poultry have the ability to release some phytate-bound phosphorus in cereal and oilseeds, the 2:1 ratio of calcium available may underestimate the amount of available phosphorus. It is also likely that the amount of calcium in those diets may also be underrepresented. Again it may make more sense to use digestible or retainable calcium or phosphorus and move away from comparing the calcium to phosphorus ratio.

Calcium and phosphorus metabolism is also affected by other factors, including dietary, physiological or animal factors. Feed ingredients from plant sources have low levels of calcium and phosphorus. Significant amounts of phosphorus are unavailable to birds from grains, since it is in the form of phytate and the birds lack an adequate endogenous phytase enzyme. This phytate-

Continued on p. 4

Increasing vitamin D_3 concentrations through feeding

Many foods in the human diet are fortified with vitamin D_3, including milk, cereal, and processed foods. Eggs are a natural source of D_3 and research finds that D_3 supplementation in hen diets increases nutrient content without affecting the taste or functional properties of eggs (Mattila et al., 1999, 2003). Research conducted in 2013 (Persia, et al.) at Iowa State University attempted to determine how production, egg quality and hen mortality is affected by higher concentrations of D_3 in laying hen diets over a 40 week feeding period (19-58 wk of bird age) . ILC Resources provided calcium carbonate for this study and is always happy to support research efforts. In this study, there were no significant findings from calcium carbonate.

The Experiment

Vitamin-D_3 was added to basal diets in varying concentrations between 7,500 and 22,500 IU of D_3/kg of diet and at a higher addition of 100,000 IU/kg. The higher addition was made to illustrate the effects of higher than normal feeding levels in order to define dosage tolerance in laying hens. Five treatment diets were fed to 360 Hy-Line W36 laying hens. The diets were formulated to meet or exceed NRC (1994) requirements with the following D_3 supplementation: 1) control diet (containing 2,200 IU of D_3/kg of diet), 2) control diet plus 7,500 IU of D_3/kg of diet (9,700 IU of D_3/kg of diet total), 3) control diet plus 15,000 IU of D_3/kg of diet (17,200 IU of D_3/kg of diet total), 4) control diet plus 22,500 IU of D_3/kg of diet (24,700 IU of D_3/kg of diet total) and 5) control diet plus 100,000 IU of D_3/kg of diet (102,200 IU of D_3/kg of diet total).

Egg production was recorded daily and expressed as a weekly average for each replicate group. Eggs were collected and stored daily for the first four weeks of the experiment and then for three consecutive days every other week of the experiment. Eggs collected during the three consecutive days were pooled, and weighed by replicate, then divided for analysis. One egg from each of these five replicates was selected and used to determine individual egg weight, shell weight, yolk weight, albumen weight, shell thickness and Haugh units. (Haugh units are the measure of egg protein based on the height of the egg white or albumen). Egg yolks were also analyzed for yolk color. Fifteen additional eggs were randomly selected to determine egg mass. Feed efficiency was calculated as grams of egg mass produced per hen per day divided by daily feed intake resulting in grams of egg mass per kilogram of feed. Hens were weighed at the start of the experimental period (week 19) and every four weeks after the start of each dietary treatment. Fat-free tibia ash was sampled from 3 hens per replicate group at the end of the experiment.

Findings

Two articles report on the results of this study. Persia et al. (2013) reported performance and egg quality data. Yao et al. (2013) reported on D_3 transfer and egg components. The cholecalciferol (D_3) transfer increased linearly up to 24,700 IU/kg of diet. The research also showed that the egg yolk lipid profile, physical properties and sensory quality of eggs were not altered by including D_3.

The highest level of D_3 inclusion (102,200 IU of D_3/kg of diet did not consistently affect laying hen performance or egg quality over the course of the experiment (weeks 19-58 of age). Egg mass and feed efficiency were not different between the diets. These findings are further support of other previous reports that up to 15,000 IU of D_3/kg of diet had no consistent negative effects on feed intake, egg production or feed efficiency (Mattila et al., 2004). Within this study, the level of 102,200 IU of D_3/kg of diet was well tolerated by the birds; the resulting egg production, feed intake, feed efficiency and mortality were similar to the hens fed the control diet.

Egg weights averaged 46.9 g per egg over the first four-week collection period and increased to the ending average weight of 61.1 g per egg. There were no significant differences among the treatment diets. Shell quality, as measured by specific gravity also did not differ significantly among treatments over the entire trial and averaged 1.089 ± 0009. These results support previous reports of no differences in egg performance with high-concentrations of D_3 supplementation (Mattila et al., 2004).

Continued on p. 4

Increasing vitamin D_3 concentrations

(continued from p. 3)

What this means for the industry
Studies of high concentrations of D_3 supplementation have not shown adverse effects on the egg production, feed intake, feed efficiency, mortality, or egg quality. The resulting significant improvements in these qualities and the efficient transfer of dietary D_3 to the egg yolk are convincing arguments for utilizing high concentrations of D_3 supplementation in laying hen diets. The eggs produced are value-added to the consumer without affecting performance or egg quality negatively. Also, a higher tolerance dose of D_3 can be fed, increasing from 40,000 up to 102,200 IU of D_3/kg of diet. This study is a great example of how we are developing into a **complete food system** with the continued interest in vitamin D supplementation into popular foods due to decreased UV light exposure in the winter or the use of sunscreen protection for UV light in the summer.

Information for this article taken from:
Mattila, P., K. Lehikoiment, T. Kiiskimen, and V. Piironen. 1999. Cholecalciferol and 25-hydroxycholecalciferol content of chicken egg yolk as affected by the cholecalciferol content of feed. *Journal of Agricultural and Food Chemistry.* 47: 4089-4092.

Mattila, P., T. Rokka, K. Konko, J. Valaja, L. Rossow, and E.L. Ryhanen. 2003. Effect of cholecalciferol-enriched hen feed on egg quality. *Journal of Agricultural and Food Chemistry.* 51: 283-287.

Mattilla, P., J. Valaja, L. Rossow, E. Venalainen, and T. Tupasela. 2004. Effect of vitamin D_2 and D_3-enriched diets on egg vitamin D content, production and bird condition during an entire production period. *Poultry Science.* 83: 433-440.

Persia, M.E., M. Higgins, T. Wang, D. Trample, and E.A. Bobeck. 2013. Effects of long-term supplementation of laying hens with high concentrations of cholecalciferol on performance and egg quality. *Poultry Science* 92:2930-2937. http://dx.doi.org/10.3382/ps.2013-03243.

Yao, L., T. Wang, M. Persia, R.L. Horst, and M. Higgins. 2013. Effect of vitamin D(3) –enriched diet on egg yolk vitamin D(3) content and yolk quality. *Journal of Food Science.* 78:C178-C183.

Metabolic limits

(continued from p. 2)

phosphorus is generally about 2.5 to 4.0 g/kg in conventional poultry diets, with less than one-half available to the bird (Selle and Ravindran, 2007). If diets were formulated on digestible calcium and phosphorus, the negative effects of extreme concentrations of calcium relative to phosphorus would be minimized and may lead to precipitation in the gut. When phytase is supplemented to the diet, diet formulation is based on digestible calcium and phosphorus. To minimize problems with this, a consistent protocol to estimate endogenous nutrient losses needs to be developed.

Calcium and phosphate transport in the gastrointestinal tract is influenced by gut health. Inflammation, passage rate and digesta viscosity all affect nutrient digestion and absorption. Bird age, sex, production and genetics also influence nutrient metabolism.

All these factors affecting calcium and phosphate utilization and the role of vitamin D_3 warrant re-evaluation and new recommendations for different poultry classes. This re-evaluation may improve nutrient metabolism and produce healthier and stronger bones ultimately, improving growth, performance and bird husbandry in conjunction with proper environmental stewardship.

Information for this article taken from:
Adedokun, S.A. and O. Adeola. 2013. Calcium and phosphorus digestibility: Metabolic limits. *Journal of Applied Poultry Research.* 22:600-608.

Marks, J., E.E.S. Debnam, and R.J. Unwin. 2010. Phosphate homeostasis and the renal-gastrointestinal axis. *American Journal of Physiology- Renal Physiology.* 299:F285-F296.

Selle, P.H., and V. Ravindran. 2007. Microbial phytase in poultry nutrition. *Animal Feed Science and Technology.* 135:1-41.

Walk, C.L., M.R. Bedford and A.P. McElroy. 2012. Influence of limestone and phytase on broiler performance, gastrointestinal pH , and apparent ileal nutrient digestibility. *Poultry Science.* 91:1371-1378.

Wideman, R.F., Jr. 1987. Renal regulation of avian calcium and phosphorus metabolism. *Journal of Nutrition.* 117:808-815.