The new role of vitamins in broiler diets to improve immunity and gut health

Global broiler industry is committed to optimise animal health and welfare as prerequisite to improve animal performance and feed utilisation for producing quality meat and make animal farming more sustainable.

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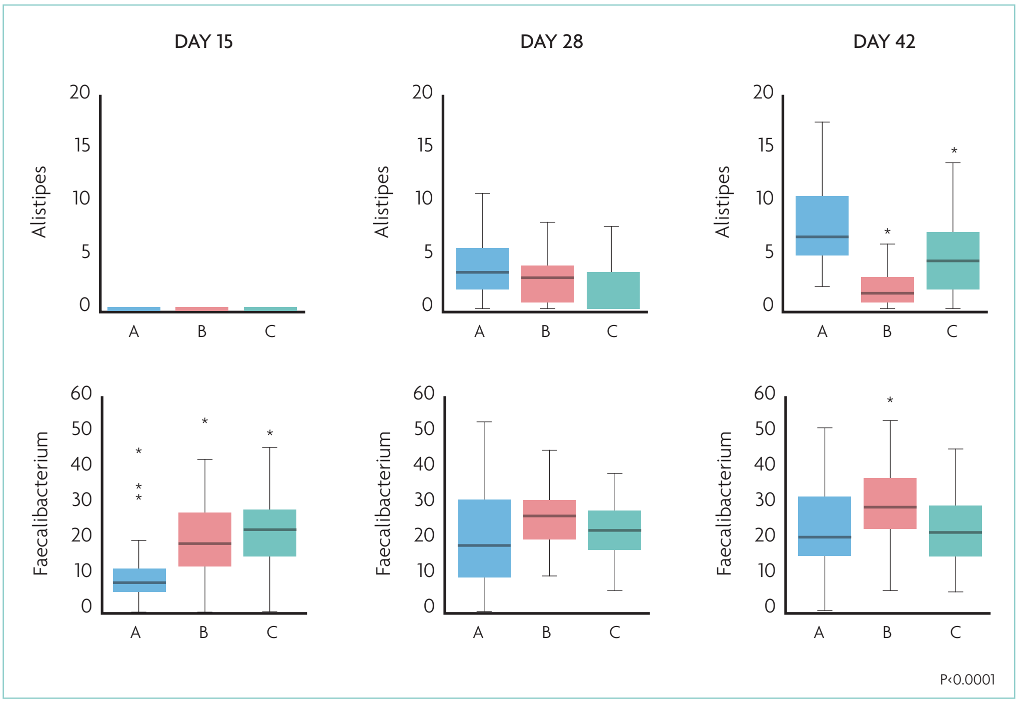

Fig. 1. Relative abundance distributions of Alistipes and Faecalibacterium in the caecal microbiota of broilers. Distributions of relative abundances (%) in all samples at day 15, 28 and 42. (from left to right) between the three groups (A, blue 5mg/kg B2; B, red 50mg/kg B2; C, green 100mg/kg B2) (Adapted from Biagi et al., 2021).

Vitamins play crucial roles in both human and animal nutrition. As organic catalysts they are essential for the normal functioning of metabolic and physiological processes such as growth, development, health and reproduction.

The requirements for vitamins in animals are not static: they vary according to new genotypes, levels of yield and production systems.

In this article, we aim to provide a specific overview about the effect of vitamins on improving immune competence and gut health and ultimately impacting on animal productivity.

Fat soluble vitamins

● Vitamin A (Retinol):
Vitamin A deficiency causes hyperproliferation of enterocytes, a decrease in the number of goblet cells, and other effects reducing the functionality of the small intestine. Vitamin A act as a cofactor in the formation of mucin within goblet cells which appears to require retinol. Epithelial cells and immune cells express the vitamin A receptor.

While a deficiency of vitamin A seems unlikely in practice because of routine supplementation, marginal levels are open to question, particularly when digestive issues occur with their supplemental esters and/or existing conditions that extend their oxidative loss.

Niu et al (2009) showed that the total antibody content in chickens depended on the dose of vitamin A in the diet. T-lymphocyte proliferative responses were decreased at low vitamin A intake and enhanced at high vitamin A intake.

In broiler chickens challenged with Newcastle disease virus (NDV), both humoral and cellular immune responses were modulated by dietary vitamin A.

Broiler chicks challenged with coccidiosis revealed that insufficient supply of vitamin A compromised local immune defences as reflected in lymphocyte profiles, oocyst shedding and interferon-gamma levels.

● Vitamin D3 (cholecalciferol) and 25OHD3 (25 hydroxy cholecalciferol):
The discovery of the vitamin D receptors (VDRs) in almost all immune cells, including T and B cells, neutrophils, macrophages and dendritic cells, strongly indicated that both vitamin D3 and its metabolite 25OHD3 play a role in immunity. The diverse immunological functions of vitamin D3 contribute to the creation of the first line of defence against pathogens without the induction of aberrant inflammatory responses. Morris et al. (2014) showed that 25OHD3, but not vitamin D3, improved immune response in LPS challenged birds and that it induces a specific immune response by reducing pro-inflammatory cytokines and stimulating nitric oxide production.

VDRs are present also in the intestinal tract and vitamin D3 plays a role in maintaining the integrity of intestinal barrier and controlling mucosal inflammation.

Chou et al. (2009) showed that supplemental 25OHD3 improves small intestinal morphology and protective humoral immunity to infection of broilers. Moreover, a recent review based on mice and small human populations supported the hypothesis that vitamin D3 influences the composition of the gastrointestinal microbiome.

● Vitamin E (alpha-tocopherol):
Vitamin E is one of the most effective nutrients known to modulate immune function, which is in part due to its protective effect against oxidation of polyunsaturated fatty acids (PUFAs).

In the gut, the antioxidant action of vitamin E (and vitamin C) may influence immunity by maintaining the functional and structural integrity of important immune cells and hence epithelial barrier functions. The disruption of the intestinal barrier function exposes the host to luminal antigens and bacteria, leading to oxidative stress and inflammation.

In fact, free radicals produced by oxidative stress protect against pathogens but their excessive production causes inflammation. Thus, it is of major importance for

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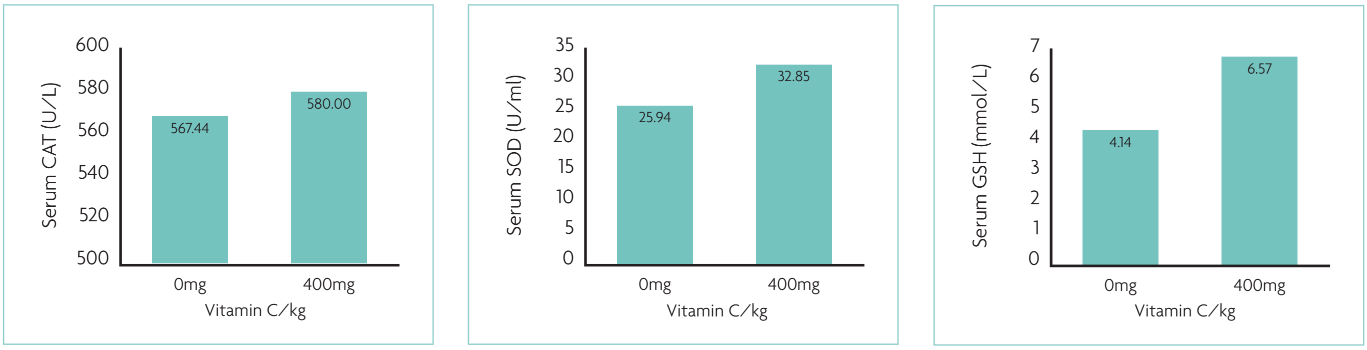

Fig. 2. Effect of vitamin C supplementation (400mg/kg) on antioxidant enzymes CAT: Catalase; SOD: Superoxide dismutase; GSH: Glutathione (Adapted from Amer et al., 2021).

Continued from page 13

gut function and animal health the control function of vitamin E and C on the production of oxygen species and inflammatory reactions.

Finally, heat stress can disrupt the intestinal barrier integrity through mechanisms that include inflammation and oxidative stress, and it has been observed that dietary vitamin E and selenium can maintain intestinal permeability in heat stressed birds.

Tengerdy et al. (1972) were the first group reporting a significantly increased immune response in chicks and hens supplemented with elevated dietary levels of vitamin E. Vitamin E was also demonstrated to induce a higher production of circulating antibodies against NDV and Pasteurella anatipestifer, following vaccination of broiler chicks against these antigens.

In broilers, immunised against NDV, the highest vitamin E level (300mg/kg) in combination with various selenium levels resulted in elevated body weight gain, favourable feed conversion ratio, significantly higher antibody titers and improved cellular immune responses.

In cockerels, fed diets containing either 10 or 300ppm of vitamin E the ratio heterophils/lymphocyte ratio was enhanced with higher level suggesting an improved phagocytic ability of the immune system.

In a study related to the immune response of broilers to coccidiosis-immunised chickens, dietary supplementation with Se or vitamin E reduced mortality and increased body weight gain of non-immunised chickens infected with E. tenella.

In a meta-analysis Pompeu et al. (2018) showed that the total Immunoglobulin production was significantly improved by dietary vitamin E supplementation with highest levels achieved with 200-250mg/kg feed.

Water soluble vitamins (Group B-Vitamins)

The vitamins of the B-complex, all belonging to the water-soluble group, have very important functions in the avian metabolism. Most of them represent coenzymes, which can fuse with larger enzyme molecules in order to accelerate various metabolic processes. Vitamin B1, vitamin B2, vitamin B6, niacin, pantothenic acid and biotin are all involved in the energy metabolism, while vitamin B12 and folic acid exert their activity on growth and cell maintenance.

It is however important to consider that those B-vitamins, related to a particular metabolic effect, are interacting with each other, which makes it difficult to determine individual requirements for each vitamin of the B-group.

It is relevant to notice that the exceptional performances of modern genetics require a very high ability to metabolise nutrients. Hence both the integrity of the gut as well as the metabolism must be very efficient for the expression of the genetic potential.

The chicken GIT harbours a very diverse microbiota, estimated to comprise over 900 species of bacteria, that aids in the breakdown and digestion of food. It is well-known that the gut microbiota synthesises not only B-vitamins like biotin, B12 (cobalamin), folic acid (and other folates), nicotinic acid, pantothenic acid, B6 (pyridoxine), B2 (riboflavin), and B1 (thiamine) but also vitamin K.

However, it must be considered that the major proportion of the microbial synthesised B-vitamins is utilised by other non-vitamin producing bacteria hence reducing the allowance of these vitamins for the broiler.

Moreover, most of B-vitamins production occurs in the hindgut, whereas absorption occurs predominantly in the first tract of the intestine. Finally, some B-vitamins are synthesised by bacteria in forms not easily available for the host.

Therefore, the old statement that group B vitamins supplementation could be less important than fat soluble vitamins must be revised in the light of the most updated knowledge and attention must be paid to their supplementation to poultry diets. This article will focus on some B-group vitamins playing a more specific function on gut health and immunity.

Vitamin B2 (Riboflavin):

There is evidence that vitamin B2 has prebiotic effects affecting its ability to regulate the innate immune system, reduce intestinal inflammation and apoptosis, and regulate gut protease activity.

Although not providing a direct substrate for microbial fermentation, riboflavin has been shown to influence the gastrointestinal redox state, ultimately modulating the composition of the intestinal microbiota towards an advantageous configuration.

In a study in broilers Biagi et al. (2021) showed that vitamin B2 supplementation at supra-nutritional dosages (50 and 100mg/kg) for ensuring a sufficient presence of riboflavin in the intestine, did not affect the ecosystem specificity of the microbial communities but was able to exert a specific effect on both caeca and ileum microbiota components, affecting different bacterial groups and influencing the caecal concentration of different metabolites, depending on the vitamin dosage.

More specifically, confirming previous reports both vitamin B2 dosages induced an increase of the well-known health-promoting bacteria belonging to the genus Faecalibacterium and reduced the progressive increase in Rikenellaceae avoiding the appearance of the species Alistipes finegoldii in the caecal core microbiota, bacterial species previously associated to low FCR in broilers, whereas Faecalibacterium was reported as positively correlated to FCR, as well as other productivity parameters (Fig. 1).

Vitamin B6 (Pyridoxine):

Vitamin B6 plays a central role in many metabolic processes in poultry, which are responsible for normal growth, the development of a strong skeleton and a functional nervous system as well as for disease resistance through an effective immune response.

Accordingly, chicks on a pyridoxine-deficient diet, develop characteristic vitamin B6 deficiency signs like increased mortality, decreased body weight gain, increased incidence of abnormal leg conformation and a significant reduction in antibody levels.

Vitamin B6 is considered critical to mucin formation because of its extensive involvement with many facets of oligosaccharide and protein synthesis. Its pivotal role relates in its use with glutamine to provide all glycosamines for its oligosaccharide component, while interconverting many frequently limiting amino acids to optimise access for inclusion in its protein.

Niacin (Nicotinic acid, Nicotinamide, Vitamin B3):

Nicotinic acid is active in the metabolism as nicotinamide and represents an indispensable component of hydrogen-carrying co-enzymes: NAD (nicotinamide adenine dinucleotide) and NADP (nicotinamide adenine dinucleotide phosphate).

Nicotinamide participates directly in the transfer of hydrogen, which is of utmost importance in the intermediary metabolism. These biochemical functions are important for normal tissue integrity, particularly for the skin, the gastrointestinal tract and the nervous system.

Vitamin C (Ascorbic acid):

Ascorbic acid is present in all living cells as an important antioxidant, essential for the formation of intercellular substances (connective tissue, bones, cartilage) and supports wound healing via its key function, the hydroxylation of proline to hydroxyproline.

Furthermore, due to its involvement in corticosteroid synthesis it has an antistress activity and it stimulates defensive mechanisms such as phagocytic activity of leukocytes and formation of antibodies. Despite poultry can endogenously produce ascorbic acid,

Continued on page 16

Continued from page 15
vitamin C supplementation has been demonstrated to beneficially influence performance and health of broilers, particularly under conditions of stress.

It has been reported that supplemental vitamin C improved disease resistance by strengthening the immune responses. Amer et al., (2021) showed that supplemental vitamin C (400mg/kg) increased significantly the antioxidant enzymes (SOD, CAT and GSH).

High levels of vitamin C, supplied to broiler chickens just prior to slaughter, reduced the stress response of starvation and transport to the abattoir indicating that ascorbic acid supplementation might improve the quality of broiler meat.

Supplemental vitamin C was demonstrated to positively affect the duodenum and jejunum villus height, width, surface area and lamina propria thickness, which is an indication of an improved gut morphology (Fig. 2).

Benefits of supra-nutritional levels of vitamins

Recently, researchers from Canada and Brazil reviewed the benefits of supra-nutritional levels of fat- and water-soluble vitamins focusing on their ability for improving poultry immunity and gut health, key factors to make animals more resilient to diseases and stress resulting in a better animal performance and productivity.

The extensive review published by Shojadoost et al in 2021 showed how vitamins A (15,000 IU/kg feed in broilers; until 35,000 IU/kg in broiler breeders), D (5,000 IU D3 + 69 mcg per kg feed of 25 OHD3), E (200ppm) and C (between 200-1,000ppm) have positive tangible effects on the poultry immune system. Their inclusion in poultry feed is not only essential for efficient growth and health but also for maintenance and enhancement of immune system function.

These effects include enhancement of innate responses against micro-organisms, more efficacious adaptive immune responses to infection and vaccination, and regulation of inflammatory responses.

Suckveris et al, 2020 concluded that the use of supra-nutritional levels of vitamin B2 (49ppm), B12 (0.119ppm), pantothenic acid (98ppm), niacin (297ppm) and folic acid (6.9ppm) resulted in improved body weight and feed intake for chickens that were raised in batteries and fed a corn and soybean-based diet formulated with low-dietary density.

It is known that vegetable diets are more aggressive to the intestinal mucosa of birds in relation to diets containing animal by-products.

Therefore, the results obtained in this study would allow us to conclude that under these conditions, the use of higher levels of B-vitamins might help birds reach their optimum production potential.

There are still many questions to answer and research to be done to better understand the potential benefit of supra-nutritional levels of vitamins, targeting specifically the optimum development of the immune system and gastrointestinal tract.

Certainly, this will contribute to make current and future birds with improved genetics healthier, more resilient and therefore more productive. ■

References are available from the authors on request

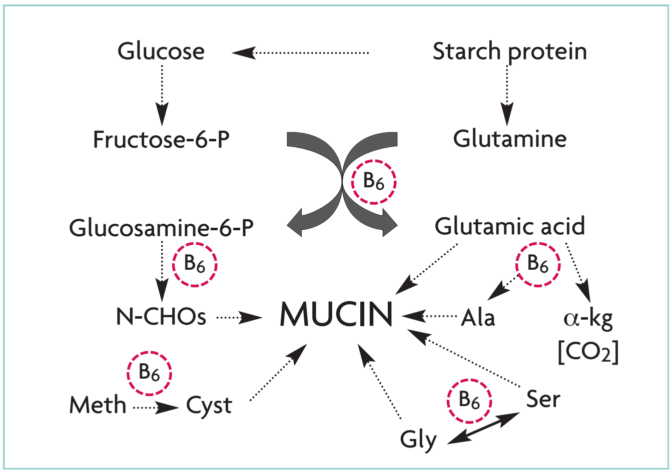

Fig. 3. Vitamin B6 is involved in many steps of mucin formation (adapted from Moran, 2017).