Feeding layers for longer laying cycles and optimized production

Conference report

At the recent EW Nutrition Poultry Academy in Jakarta Indonesia, Dr Steve Leeson, Professor Emeritus, University of Guelph, Canada, commented that “genetic progress in layer breeding has been substantial in recent decades. Since 1995, the yearly change has included +1 egg, -0.01 feed/dozen eggs, -10g final bodyweight, 0.02% mortality, and +1 week at >90% egg production. This improved persistency of commercial laying hens enables egg producers to keep flocks longer in production, provided egg shell quality can be maintained.”

He noted that “the increase in hen-housed egg production is mainly due to longer clutch length and improved uniformity of layer flocks. No doubt, there is a trend in cage layers to longer production cycles. A popular commercial goal is 500 eggs in one cycle with no moult, although this has already been surpassed in many flocks. The modern layer is capable of laying 150 eggs per clutch.”

Dr Leeson, however, stressed that “genetic progress and longer laying cycles have consequences. Long laying cycle programmes start during pullet rearing – you can’t make decisions at 72 weeks of age. Instead, you must start with your end goals, such as persistency, egg size and shell quality, in mind. You can then develop a life-cycle approach to feeding, lighting, nutrition, and general management.” Important issues to manage include:

Body weight control – early and late

Mature body weight dictates subsequent egg size. In the past, the common goal was being at, or above, management guide weight recommendations. For extended lay, a larger body weight results in too large an egg past 70 weeks of age, and so it is more difficult to maintain egg shell quality. Now the goal is to grow a slightly smaller pullet, and emphasis changes to achieving adequate early egg size from this smaller bird. This makes pre-lay nutrition for these slightly smaller pullets even more important.

The scheduling of rearing diets is more important than diet formulation. Dr Leeson's guidelines are:

- Starter diet – 19-20% CP, 2,850-2,900 kcal ME/kg from day old to target pullet body weight
- Grower diet – 17-18% CP, 2,800-2,900 kcal ME/kg from target body weight to mature body size
- Pre-lay diet (or layer diet?) – 16-18% CP, 2,800-2,900 ME/kg, mature body size to first egg

All nutrients are important, but energy is usually limiting for egg number, whereas protein/amino acids influence egg size (and feathering).

There is now even more emphasis on pullet growing to ensure adequate fat reserves through peak production, so birds are in a positive energy balance. The establishment of an energy reserve occurs during the rearing phase and has a significant effect on the bird's body composition at point of lay.

Egg size control – early and late

The obvious solution to manage body weight (and egg size) is to light-stimulate a smaller pullet, or at least to not light-stimulate a heavy pullet. This achieves a balance between accepting reduced early egg size, versus limiting an increase in egg size late in the production cycle.

Egg size can be increased in smaller early-lay pullets by:

- Reducing environmental temperature, if possible, to stimulate feed intake
- Midnight feeding 19-29 weeks
- Adequate amino acid nutrition intake, tailored to feed intake, especially methionine
- Increased number of feedings/day and increased feed particle size (pellets)

Shell strength is negatively correlated with egg size. To temper egg size late in the cycle, Dr Leeson recommended:

- Body weight control
- Controlled day length: longer day length = increased feed intake, 14 hours maximum day length in controlled-environment houses
- Warmer temperature – 26°C is ideal
- Reduce number of feedings and particle size
- Temper amino acid nutrition (with caution). Low crude protein/high amino acid diets limit the increase in egg size.

Midnight feeding provides about 1-hour extra light per day and therefore stimulating feed consumption in the middle of the dark period. Having access to feed during this period improves eggshell quality via the supply of calcium during the time when shell calcification takes place. The extra light period is perceived by the bird to be part of the night. The dark period after the light period must be longer than the initial dark period, as the bird perceives the start of the day is the end of the longest period of darkness. Removing midnight feeding should be done gradually – 15 minutes per week, advised Dr Leeson.

Preventing calcium depletion

Also known as cage layer fatigue, calcium depletion is becoming more common in all strains due to high sustained egg output. Calcium deficiency in the feed leads to loss of medullary or long bone (a reservoir of

about 4g of calcium) and increased bone fragility. It is commonly seen at 35-40 weeks of age, with a 1-2% occurrence. If the incidence is more than 2%, seek advice for your pre-lay nutrition.

The development of the medullary bones takes about 10 days and requires additional calcium. Pre-lay rations support a smooth transition from developer feed to layer feed, with 2-2.5% calcium, while the other nutrients are similar to a layer feed. Pre-lay rations help the birds to adapt to the high calcium content of layer feed and to maintain sufficient daily feed intake.

To prevent calcium depletion, Dr Leeson suggested:

- Optimise pre-lay calcium (Ca) and phosphorous (P) nutrition
- Intake of 1.5g Ca, 350-450mg available P/day for at least 7 days prior to first egg
- During early lay, ensure 3.5-4 g Ca and 420 mg available P/day
- Consider vitamin D_3 water treatment (150 IU/day, twice weekly)

Pre-lay diets provide the bird with the opportunity to deposit medullary bone. This bone deposition coincides with follicular maturation and is under the control of both estrogens and androgens. The latter hormone seems essential for medullary bone growth, and its presence is manifested in the growth and reddening of the comb and wattles. Consequently, there will be little medullary deposition, regardless of diet calcium level, if the birds are not showing comb and wattle development and this stage of maturity should be the cue for increasing the bird's calcium intake.

Liver health

Excess energy relative to needs results in excess fat accumulation that is prone to oxidation. This is why you never see fatty liver haemorrhagic syndrome (FLHS) in poor-producing flocks. Layers normally have a very fatty liver, as 100% of egg yolk synthesis occurs in the liver.

The lower the fat content of the diet, the greater the stress/need to fat synthesis in the liver. With a low energy/low fat/carbohydrate diet FLHS is almost universal to varying degrees. One treatment is to add fat to the diet! Haemorrhage (not always FLHS) is inevitable with dietary omega-3s that are very prone to oxidation.

Dr Leeson recommended prevention/control for FLHS, which usually starts about weeks 36-40, including:

- +1.0 kg choline
- +0.5 kg methionine
- +100 IU vitamin E
- +30% does Hy-D because of impaired liver metabolism of vitamin D_3 (that can also impact calcium absorption)
- Add 2% dietary fat without change in diet energy level

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EW Nutrition's Poultry Academy took place in Jakarta and Manila in early September 2023. Dr. Steve Leeson, an expert in Poultry Nutrition & Production with nearly 50 years' experience in the industry, was the distinguished keynote speaker.

Dr. Leeson had his Ph.D. in Poultry Nutrition in 1974 from the University of Nottingham. Over a span of 38 years, he was a Professor in the Department of Animal &Poultry Science at the University of Guelph, Canada. Since 2014, he has been Professor Emeritus at the same University. As an eminent author, he has more than 400 papers in refereed journals and 6 books on various aspects of Poultry Nutrition & Management. He also won the American Feed Manufacturer's Association Nutrition Research Award (1981), the Canadian Society of Animal Science Fellowship Award (2001), and Novus Lifetime Achievement Award in Poultry Nutrition (2011).

Nutritional considerations for immunity and gut health

Conference report

At the recent EW Nutrition Poultry Academy in Jakarta, Indonesia, Dr Steve Leeson, Professor Emeritus, University of Guelph, Canada, opened his presentation by stating that “it is obvious that any nutrient deficiency will impact bird health, but not so obvious is that nutrition *per se* can positively impact immunity and health in an otherwise healthy and high-producing bird.”

Modern high-performing broilers are characterized by extremely high feed intake. This puts a lot of stress on the physiology of the entire gastrointestinal tract, but particularly so on the absorptive epithelial cells of the small intestine. Any organism requires a nutrient source for survival and reproduction. Dr Leeson asked “can we significantly reduce nutrient supply to pathogens, while sustaining bird productivity?”

He reminded the audience that no cellular function comes for free: so there is always a “cost”. A general conclusion is that 10% of nutrients can be used for immune function during disease challenge, and always get priority. Therefore, you don’t want to overstimulate the immune system, which in extreme situations leads to an inflammatory response. In his presentation, Dr Leeson considered factors determining gut health and nutritional tools which are available to support gut health.

Gut microflora

Gut pathogens impact the bird and/or the consumer. *Clostridia* and *E. coli* are the major concerns regarding bird health and productivity, whereas *Salmonella* and *Campylobacter* are major pathogens important for human health.

The chick hatches with a gut virtually devoid of microbes, so early colonizers tend to predominate quite quickly. Microbial species present on the hatching tray, during delivery and during the first few days at the farm will likely dictate early gut colonization. In some instances, the chick's microflora may be established by the time it gets to the farm, so the probiotic faces more of a challenge to establish itself as the predominant species.

Antibiotic alternatives

Gut villi development matures at around 10-15 days of age. The broiler pre-starter diet therefore is a target for feed additives that positively impact gut structure and development.

- Among the **short chain fatty acids**, butyric acid is considered the prime energy source for enterocytes and it is also necessary for the correct development of the gut-associated lymphoid tissue (GALT). Butyric acid can also be added indirectly via fermentation of judicious levels of soluble fiber to encourage optimal gut villi development. Dr Leeson added that he is a big believer in butyric acid, encouraging a good gut structure at 10 days, which can be worth about 50 kcal.
- **Exogenous enzymes** should also be considered in an attempt to maximize digestion and limit the flow of nutrients to the large intestine and ceca. Protease enzymes have great potential in this regard, since they allow nutritionists to reduce dietary crude protein and hopefully reduce the supply of nitrogen that fuels proteolytic Clostridia bacteria in the large intestine and ceca.
- **Amino acids**, particularly threonine, play a critical role in the maintenance of intestinal mucosal integrity and barrier function, especially for mucin synthesis, which protects enterocytes from adherence by pathogenic bacteria, and from attack by endogenous enzymes and acids.
- **Polyunsaturated fatty acids** (PUFAs) – Omega-3s and especially DHA from fish oil help to reduce inflammatory response (overstimulation). Omega-3s are poorly converted to DHA by the chicken, so conventional sources such as flax are of limited application for immunity.
- **Blood plasma** from pigs or cattle is a complex spray-dried mixture of proteins and amino acids, many of which are immunoglobulins that "temper" the immune system, much like PUFAs.
- **Vitamins A, D, E and C** have vital roles in the normal function of the immune system and have antioxidant capacity.
- Certain **complex carbohydrates**, such as ß-glucans, influence gut health due to their fermentation, leading to the production of short-chain fatty acids, such as butyrate.
- **Antioxidants** – to firstly control oxidation of fats and fat-soluble vitamins in feed, and secondly to optimize birds' cellular oxidative capacity, to prevent cell damage, therefore maintaining healthy cellular and immune function.
- **Betaine** increases intracellular water retention, reducing "dehydration" of microvilli and increasing their volume/surface area.
- **Fiber** – moderate levels (1-2%) of soluble (fermentable) and insoluble fiber can be beneficial to early gut development by stimulating gizzard development and endogenous enzyme production.
- **Phytogenics** are becoming very common in combination with acidifiers (upper tract) and probiotics. Essential oils are becoming more mainstream the more we know about them.

Recommendations for optimizing gut health and immunity

Fast growth rate and high egg output are negatively correlated with immune response. Consequently, nutrient-dense diets are not optimal for immunity. With bacteria, it's a numbers game – but these numbers quickly multiply. The first 7 days are important, therefore probiotics must be established early. Consider

the role of targeted feed additives, such as butyrate, phytogenics, antioxidants, PUFAs etc.

Also, maximize feed particle size – the limit is usually pellet quality. Mitigate nutrient transition at any diet change. Review the supply of trace minerals, as there is a trend to lower levels of organic minerals. With all the factors that weigh into production performance, any support that can be rallied through nutrition needs to be considered.

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Phytomolecules: A tool against antibiotic-resistant E. coli

Diseases caused by E. coli entail use of antibiotics in animal production

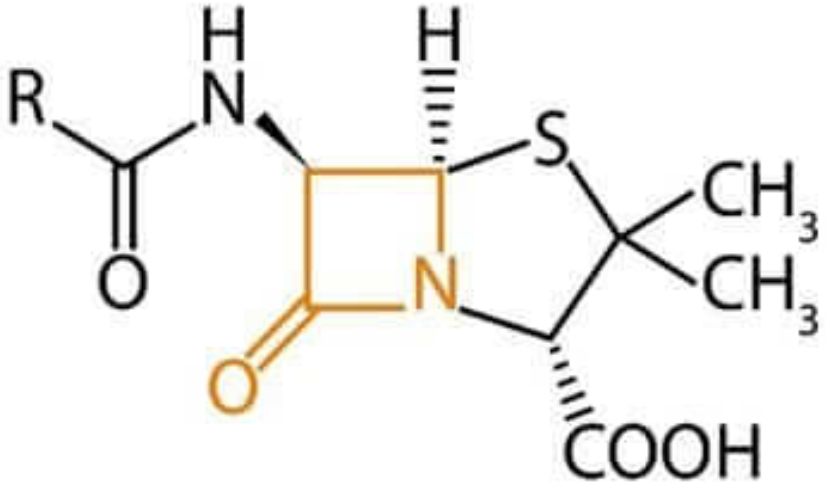
E. coli infections are a major problem in pig production. Especially young animals with an incompletely developed immune system are often unable to cope with the cavalcade of pathogens. In poultry, E. coli are responsible for oedema, but also for respiratory diseases. In young piglets, E. coli cause diarrhoea , oedema, endotoxic shock and death. In order to cure the animals, antibiotics often must be applied. Besides this curative application, antibiotics were and in many countries still are used prophylactically and as growth promoters.

The excessive use of antibiotics, however, leads to the occurrence of antimicrobial resistance (AMR): due to mutations, resistance genes are created which enable enterobacteria such as Salmonella, Klebsiella and E. coli to produce enzymes (ß-lactamases) in order to withstand ß-lactam antibiotics. In case of an antibiotic treatment, the resistant bacteria survive whereas the other bacteria die.
The major problem here is that these resistance genes can be transferred to other bacteria. Harmless bacteria can thus transfer resistance genes to dangerous pathogens, which then cannot be combatted with antibiotics anymore. In this article we explore in detail how AMR happens and how phytomolecules, which have antimicrobial properties, could be a key tool to reduce the need for antibiotics in animal production.

How ß-lactam antibiotics work

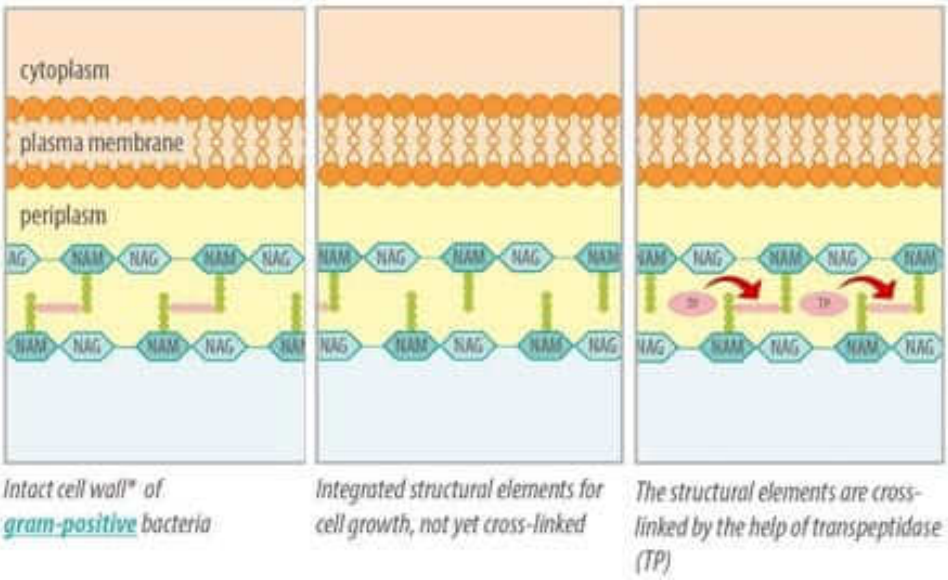
The group of ß-lactam antibiotics consists of penicillins, cephalosporins, monobactams, and carbapenems. These antibiotics are characterised by their lactam ring (Figure 1).

Figure 1: An antibiotic with a ß-lactam ring (in orange)



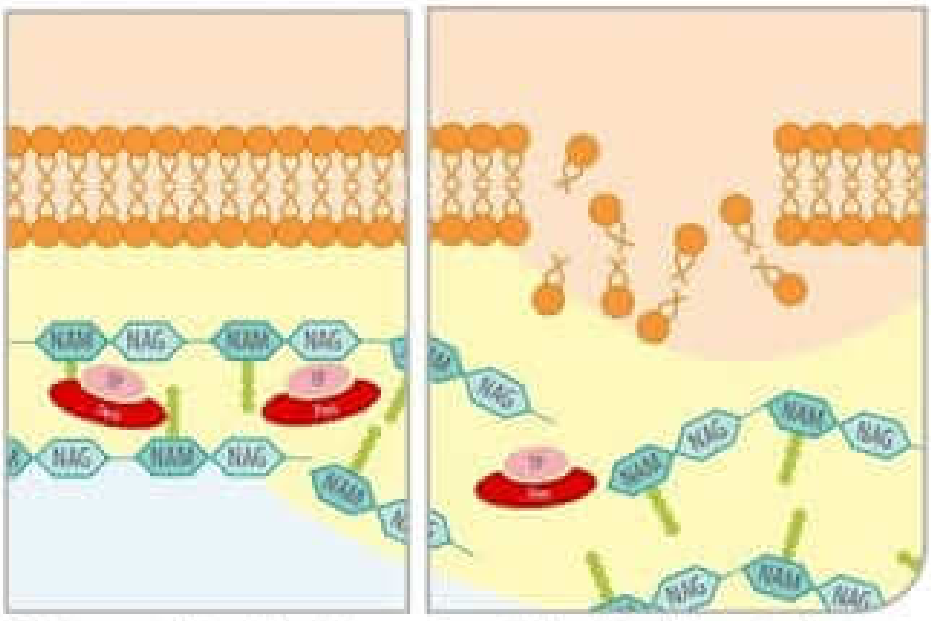
If bacteria are growing, the cell wall also has to grow. For this purpose existing conjunctions are cracked and new components are inserted. In order for the cell wall to remain a solid barrier, the new components must be interconnected by crosslinks. For the creation of these crosslinks an enzyme is essential, the transpeptidase (figure 2).

Figure 2: building up a stable cell wall with the help of transpeptidase



Due to their structure, ß-lactam-antibiotics also fit as binding partner for transpeptidase. They bind to the enzyme and block it (Kohanski et al., 2010). The crosslinks cannot be created and the stabilization of the cell wall is prevented. Disturbance of cell wall stability leads to the death of the bacterial cell, hence ß-lactam antibiotics act bactericidal.

Figure 3: blocked by ß-lactam antibiotics, transpeptidase cannot serve as enzyme for building the cell wall



The challenge: E. coli producing ß-lactamases

Resistant bacteria, which are able to produce ß-lactamases – enzymes that destroy the ß-lactam ring – prevent their own destruction. Divers point mutations within the ß-lactamase genes lead to the occurrence of “extended-spectrum-beta-lactamases“ (ESBL). ESBL are able to inactivate most of the ß-Lactam-antibiotics.

Another mutation leads to so-called AmpC (aminopenicillin and cephalosporin) ß-lactamases. They enable the *E. coli* to express a resistance against penicillins, cephalosporins of the second and third generation as well as against cephamycins.

Phytomolecules – an alternative?

One approach to reduce the use of antibiotics is the utilization of phytomolecules. These secondary metabolites are produced by plants to protect themselves from moulds, yeasts, bacteria and other harmful organisms.

The use of plants and their extracts in human and veterinary medicine is well-established for centuries. Besides digestive and antioxidant characteristics they are well known for their bacteriostatic and bactericidal effects.

Consisting of a high number of chemical compounds, they attack at diverse points and their antimicrobial effect is not caused by only one single specific mechanism. This is crucial because it is therefore very unlikely that bacteria can develop resistances to phytomolecules like they do to antibiotics.

How phytomolecules work

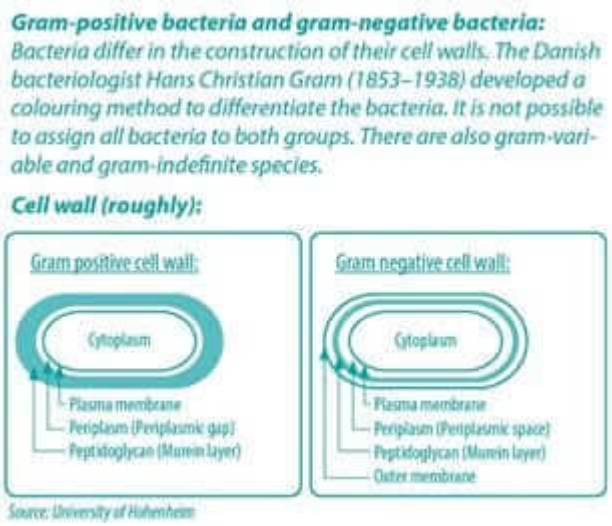
Mostly, phytomolecules act at the cell wall and the cytoplasm membrane level. Sometimes they change the whole morphology of the cell. This mode of action has been studied extensively for thymol and carvacrol, the major components of the oils of thyme and oregano.

They are able to incorporate into the bacterial membrane and to disrupt its integrity. This increases the permeability of the cell membrane for ions and other small molecules such as the energy carrier ATP

(Adenosin-tri-phosphate). It leads to the decrease of the electrochemical gradient above the cell membrane and to the loss of energy equivalents of the cell.

A special challenge: gram-negative bacteria

Gram-negative bacteria such as *E. coli* and *Salmonella* pose a special challenge. The presence of lipopolysaccharides in the outer membrane (OM) provides the gram-negative bacteria with a hydrophilic surface (Nikaido, 2003; Nazarro et al., 2013) (*see also blue infobox*).



The cell wall therefore only allows the passage of small hydrophilic solutes and is a barrier against macromolecules and hydrophobic compounds such as hydrophobic antibiotics and toxic drugs. The bypassing of the OM therefore is a prerequisite for any solute to exert bactericidal activity toward gram-negative bacteria (Helander et al., 1998).

Based on their trial results Helander et al. (1998) (1998) concluded that trans-cinnamaldehyde and partly also thymol and carvacrol gain access to the periplasm and to the deeper parts of the cell. Nikaido (1996) also concluded that OM-traversing porin proteins allow the penetration of lipophilic probes at significant rates.

Evaluating phytomolecules I – in vitro trial, Scotland

A trial conducted in Scotland evaluated the effects of Activo Liquid, a mixture of selected phytomolecules and citric acid, on ESBL-producing *E. coli* as well as on *E. coli* that generate AmpC.

Material and methods

For the trial two strains for each group were isolated from the field, a non-resistant strain of *E. coli* served as control. Suspensions of the strains with 1×10^4 CFU/ml were incubated for 6-7 h at 37°C (98.6°F) together with diverse concentrations of Activo Liquid or with cefotaxime, a cephalosporin. The cefotaxime group saved as a control for differentiating resistant and non-resistant *E. coli*.

The suspensions were put on LB agar plates and bacteria colonies were counted after further 18-22h incubation at 37°C.

Results

The antimicrobial efficacy of the blend of phytomolecules depended on the concentration at which they were used (see table 1). A bacteriostatic effect could be shown at dilutions up to 0.1 %, a bactericidal effect at higher concentrations.

Table 1: Effect of phytomolecules against resistant E. coli producing ESBL and AmpC in poultry

Poultry Microbiology Laboratory, Edinburgh, Scottland	Cefotaxime	Phytomolecules (Activo® Liquid)			
	30 µg / ml	0.1 %	0.2 %	0.4 %	0.5 %
E. coli ESBL 1 (Poultry)	-	+	++	++	++
E. coli ESBL 2 (Poultry)	-	+	++	++	++
E. coli AmpC 1 (Poultry)	-	+	++	++	++
E. coli AmpC 2 (Poultry)	-	++	++	++	++
E. coli non-resistant	+	+	++	++	++
- no effect + growth inhibiting (bacteriostatic) ++ bactericidal					

Evaluating phytomolecules II – in vitro trial, Germany

A further trial was conducted in Germany (Vaxxinova, Münster), confirming the preceding results.

Material and methods

Four ESBL producing *E. coli* all isolated from farms and a non-resistant reference strain as control were tested concerning their sensitivity against Activo Liquid. Every bacteria strain (Conc.:1×10^4 CFU/ml) was subjected to a bacterial inhibition assay in an appropriate medium at 37°C for 6-7 hours.

Results

In this trial Activo Liquid also showed a dose-dependent efficacy, with no or just a bacteriostatic effect up to a concentration of 0.1 %, but bactericidal effects at a concentration of ≥0.2 % (table 2).

Table 2: Effect of phytomolecules against resistant ESBL producing E. coli in pig and in poultry

Vaxxinova GmbH, Münster	Phytomolecules (Activo® Liquid)			
	0.1 %	0.2 %	0.4 %	1 %
E. coli ATCC25922	+	++	++	++
E. coli ESBL 1 (Pig)	-	++	++	++
E. coli ESBL 2 (Pig)	+	++	++	++
E. coli ESBL 3 (Poultry)	+	++	++	++
E. coli ESBL 4 (Poultry)	-	++	++	++
- no effect + growth inhibiting (bacteriostatic) ++ bactericidal				

Phytomolecules: a promising outlook

E. coli infections have devastating effects on animals, from diarrhea to edema, enterotoxic shock and even death. Antibiotic treatments have long been the only practicable answer. However, their excessive use – for instance, the metaphylactic application to thousands of animals in a flock – has led to the development of resistant strains. There is evidence that a reduction of antibiotic use reduces the occurrence of resistances (Dutil et al., 2010).

The results of the two in vitro trials in Scotland and Germany demonstrate the bactericidal effects of phytomolecules on *E. coli* that produce ESBL and AmpC. Using phytomolecules could thus reduce the use of antibiotics and therefore also the occurrence of AMR.

While it is theoretically possible for bacteria to also become resistant against phytomolecules, the probability of this happening is very low: unlike antibiotics, phytomolecules contain hundreds of chemical components with different modes of action. This makes it exceedingly difficult for bacteria to adapt and develop resistance. To tackle the problem of antibiotic-resistant *E. coli*, antimicrobial phytomolecules therefore offer a promising, sustainable and long-term solution.

By Dr. Inge Heinzl, Editor, EW Nutrition

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Secondary plant compounds are the new frontier in poultry nutrition

Why should you read another story about phytogenics? Or, is it botanicals, spices, herbs, and extracts? No matter what we call them, scientists have named them "secondary plant compounds", and if we are to follow the American tradition we can call them SPC. Then, here is the first interesting thing we can discuss about this plant-derived class of active compounds. They are "secondary" in nature, but not insignificant. They play no role in normal metabolism, but they help plants (and now animals) survive under adverse conditions. Perhaps, this is why some experts consider them as the next frontier in poultry nutrition. With poultry that are raised in less than ideal conditions, especially when we consider the movement towards antibiotic reduction (for growth promoting reasons, not complete removal of all medicines), we understand that such natural compounds can be of significant help.

As it happens, the majority of poultry specialists in Europe and increasingly in the Americas consider SPC as an almost-essential element in diets for broilers and layers (and turkeys, ducks, and all poultry for that matter) when birds are raised without antibiotics. Some go even further and use them along with antibiotics because, as we all know, antibiotics are never 100% efficient as bacteria sooner or later develop some form of resistance. Such resistance has not yet been observed with SPC. So if one is to use SPC in poultry feds, which ones to buy? A quick glance at the market will reveal more commercial products than can possibly be imagined. Some must be better than the rest, but how can we separate the wheat from the chaff? Price alone is not always a good indicator. A high quality product must be expensive – for there is no such thing as a free lunch – but all expensive products are not always of the highest possible quality!

There are three basic criteria, which we can mention briefly here:

1. **SPC are volatile** – at least most of them. As such, unprotected products will soon evaporate if left in the open air as it happens with feed prepared in commercial farms. So, some form of protecting SPC is essential.
2. **SPC are innumerable** – so finding the right mix for the job required is important. You cannot get the same results with any kind of mix. So, in designing an SPC mix, the manufacturer must declare and have knowledge of the target to be accomplished.
3. **SPC are powerful** – meaning you cannot just keep adding as much as possible. Here finding the exact dosage for the right purpose is a difficult balancing exercise. So, the right mix and the right dosage must be combined, otherwise animals will refuse the feed (worst case scenario) or just fail to benefit from SPC inclusion.

There is so much more to learn about this exciting class of compounds that can replace the growth promoting action of antibiotics that it is worth spending time learning more about them.