PEER REVIEWED

The Potential of Feed Additives in Reducing Antibiotic Usage

Over the last years, feed additives that support gut health have gained special attention from animal producers worldwide. Especially during the last decade, in many regions interest has been further nourished by regulatory ambitions to restrict the use of antibiotics, either as antimicrobial growth promoter (AGP) or as veterinary drugs. Attention should be given to the similarities and differences between the gut health promoting feed additives and antibiotics, if we want to understand the potential of these additives to reduce antimicrobial usage.

Microorganisms, such as bacteria, fungi and parasites, can develop resistance against antimicrobial substances, such as antibiotics, antifungals and anthelminthics. As a result, these antimicrobial substances become ineffective in limiting the growth of resistant microorganisms. The occurrence of antimicrobial resistance (AMR) is not a new phenomenon; in fact, it is a natural selection process that has been going on for billions of years. In recent years, however, microorganisms have been isolated that have developed AMR against a wide range of medically important drugs. As such, these 'superbugs' have become an increasing threat to human healthcare.

Antimicrobial Resistance and Feed Additives

The misuse and overuse of antimicrobials is accelerating this process. While it is recognised that the human use of antibiotics is the largest contributor to AMR in human pathogens, the over-use in intensively-produced farm animals is now believed to play a considerable role in this global problem as well. For this reason, governmental administrations and food animal producing organisations worldwide are looking into the possibility of reducing antimicrobial use, either as veterinary drug or as antimicrobial growth promoter (AGP), in animal production.

In this context, special attention is given to several strategies aiming to minimise the need to apply antibiotics to food animals, such as an increased focus on veterinary support, biosecurity measures, production management and nutritional interventions like the use of functional feed additives. Indeed, certain feed additives can have a beneficial effect on gut health in animals, and can mitigate the damage caused by intestinal pathogens, thereby reducing the need to routinely use veterinary drugs applied to tackle enteric infections. In addition, gut health will stimulate animal performance in general, and additives that strengthen the digestive tract can therefore be employed as part of a strategy replacing AGPs.

Gut Health Additives and Antibiotics: Similarities and Differences

However, in order to correctly evaluate the potential of gut health promoting additives as alternatives to antibiotics, it should be pointed out that the characteristics of both groups of active molecules are very different, which has consequences to their proper application. Often, when animal producers are faced with the need or the wish to reduce AGP supplementation for the first time, they are looking for ways to remove the antibiotics in the feed, just to replace them by other active ingredients which they hope will have the same effect. Similarly, some producers who want to minimise veterinary interventions, might try to apply feed additives as a way to cure an upcoming disease, expecting they will act in similar ways as drug. These approaches, however, deny the fact that additives have a different mode of action compared to AGPs/antibiotics, and that knowledge of these mechanisms is needed to correctly use those additives, as well as to have realistic expectations on their potential.

To illustrate this, in Figure 1, some of the modes of action of AGPs and gut health promoting feed additives are listed. Both classes of compounds can be said to have the same goal: improving intestinal health and function, thereby increasing animal performance. AGPs are hypothesised to do so by directly affecting the intestinal microbiota, and perhaps by having anti-inflammatory properties. The additives, on the other hand, can have different working mechanisms; typically, some of them are overlapping with those of AGPs, while others are distinctive. Some coated butyrate products, for instance, will have a direct or indirect effect on intestinal microbial species (see below), but often without directly killing them, or inhibiting their growth, as antibiotics do. Butyrate has also strong anti-inflammatory properties, while it also has the potential to trigger several physiological pathways, when it is delivered throughout the entire intestinal tract by means of a coating that sets butyrate free gradually after ingestion (ADIMIX®Precision, a Precision Delivery Coated Butyrate, PDCB). Intestinally-delivered butyrate can trigger signalling pathways which are not (or less) activated by antibiotics: for instance, it can fortify the epithelial lining of the digestive tract, it can increase the secretion of digestive fluids, and it might be used by other organs, such as the liver, as energy source and as modulator of metabolic and detoxification processes.

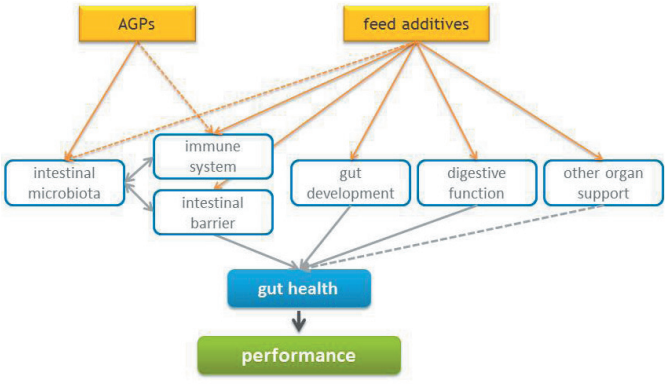

Figure 1: AGP's and gut health promoting feed additives have modes of action that are partially similar, and partially different

The fact that AGPs and PDCB have underlying modes of action that are partially similar and partially distinct, means that PDCB can be used to reduce dependency on AGPs: both products can be used in combination with each other, or, when the use of AGPs is forbidden or needs to be restricted, PDCB will be needed to fill this void, for example by mitigating the effect of pathogenic intestinal bacteria, albeit via a different route.

Below, some examples are given on how researching different application of PDCB can shed light on how this product can be employed to support animal health and production in a production context where less antibiotics are used.

Mitigating the Effects of Necrotic Enteritis

Necrotic enteritis (NE) is an important enteric disease in poultry, caused by the overgrowth of certain *Clostridium perfringens* strains, which evokes intestinal tissue damage. In its acute clinical form, the disease is characterised by a sudden increase in flock mortality, and gross lesions that are usually, but not always, restricted to the small intestine. In the sub-clinical form, no peak mortality is observed, but intestinal damage will lead to decreased nutrient digestion and absorption, thereby negatively affecting performance parameters such as growth-rate and feed conversion ratio. Especially since the ban of antibiotic growth promoters (AGPs), the increased incidence of this sub-clinical form is estimated to have a substantial impact on animal welfare and on the profitability of poultry production.

In a model for necrotic enteritis in broilers, 800 broilers were allocated to four treatment groups in a 2x2 factorial design: birds were supplemented with PDCB or not, and were either orally infected with a NE-associated *C. perfringens* strain right after IBD-vaccination (day 14), or were left unchallenged. After 35 days, birds that were infected with C. perfringens were significantly lighter than the control group, while their FCR was numerically higher[1]. In the challenged group, PDCB-fed birds outperformed the control group, with zootechnical characteristics comparable to those of the unchallenged birds. Importantly, also in unchallenged birds, supplementing feed with PDCB resulted in a significant increase in final weight and a reduction of FCR.

Interestingly, supplementing challenged birds with PDCB had limited effects on the enumeration of *Clostridium perfringens* in the intestinal tract. However, PDCB-treatment birds had lower necrotic lesion scores (Figure 2). These results are in line with the hypothesis that PDCB doesn't have a direct bacteriostatic effect against *Clostridium*, but that it prevents or repairs intestinal epithelial damage, thereby limiting nutrient loss and inflammation and, as such, restricting conditions that would favour the intestinal overgrowth of *Clostridium* in later life stages.

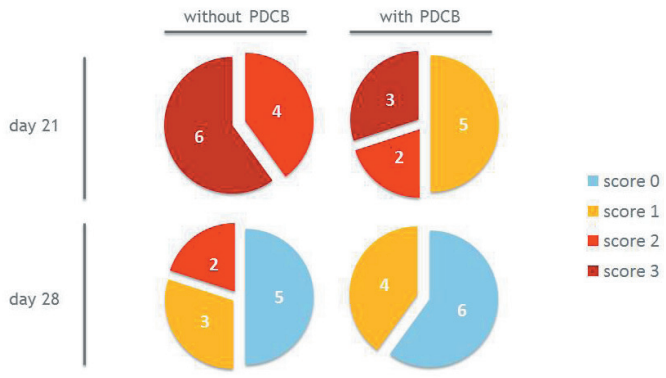

Figure 2: Lesion score in NE-challenged birds

Restricting Hindgut *Colonisation* of *Salmonella* and *Campylobacter*

Similarly, PDCB can restrict the colonisation of *Campylobacter* and *Salmonella* in the intestinal tract of production animals[2]. This effect is most likely not the result of a direct bacteriostatic effect of butyrate on *Salmonella* in the digestive tract. Rather, butyrate is documented to downregulate genes in *Salmonella* that are important for epithelial invasion and colonisation[3]. To have any significant effect *in vivo*, butyrate therefore needs to be delivered in the animal hindgut, where *Salmonella* resides. As butyrate is a small molecule that is readily absorbed in the first part of the digestive tract, this means that the gastric bypass and intestinal delivery characteristics of PDCB are critical to exert any anti-*Salmonella* effect inside the animal[2].

Likewise, butyrate has been shown to protect intestinal cell from Campylobacter invasion and translocation *in vitro*[4]. Recently, an extensive *in vivo* study was set up, in which broilers were orally infected with Campylobacter *jejuni* and were fed one of 24 feed additives, including (monoglycerides of) short- and medium-chain fatty acids and phytogenic substances (Fig. 3)[5,6]. Strikingly, the compounds that could have been predicted to be most efficacious based on their antimicrobial properties *in vitro* (monoglycerides of short/medium-chain fatty acids and essential oil components), weren't the groups that outperformed the others in the *in vivo* test. In fact, only two treatments were linked to a significant reduction in mean caeca *Campylobacter* counts at all sampling moments: a PDCB, included at 3 kg/T, and only one out of the seven monoglyceride treatments tested, included at 8 kg/T. These results suggest that when evaluating effective feed additives to be used in a production programme where the use of antibiotics is minimised, it is imperative to look beyond direct antimicrobial effects.

Similarly, it should be clear that gut health feed additives

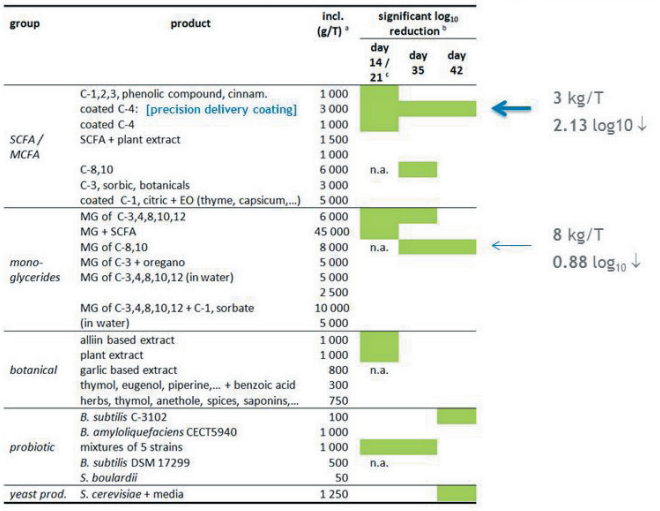

Figure 3: Overview of caecal Campylobacter reduction result in a study evaluating the effect of 24 feed additives

cannot be used as an antibiotic drug to be applied in the final production stage, to eradicate zoonotic pathogens just before the animals are slaughtered. This is demonstrated by an experiment, where the effect of PDCB on caecal *Campylobacter* colonisation was evaluated in more detail. Zero or three kg/T of PDCB was added to several feeds of broilers infected with *Campylobacter jejuni* in the growing stage (Figure 4). When PDCB was added to all feeds, or from the moment onwards that birds were orally infected, caecal *Campylobacter* loads were significantly reduced compared to birds from the control treatment. However, inclusion of PDCB in only the starter feed and/or the finisher feed had a less profound impact on caecal *Campylobacter* counts. These results demonstrate that PDCB, in addition to its effects on animal performance, can be of value in a programme reducing zoonotic pathogens, but that it is not a 'quick fix' to be applied during the end of production, exerting a specific antibacterial effect.

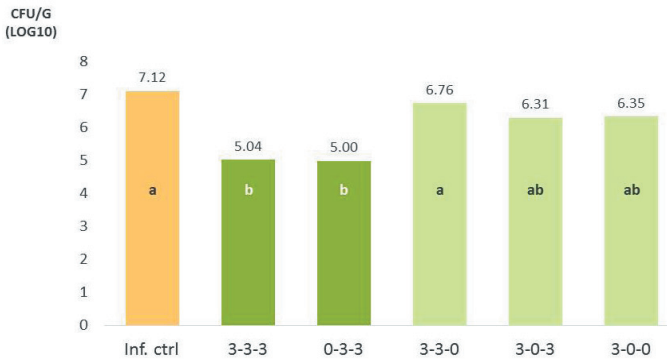

Figure 4: Mean Campylobacter counts (day 39) in caeca from infected broilers supplemented with 0 or 3 kg/T PDCB in the starter-grower-finisher diets

Conclusion

In conclusion, antibiotics, whether applied as drugs or AGPs, most likely exert a specific and direct effect on enteric bacteria. The downside of this approach is that bacteria can build up resistance against certain drugs, jeopardising future treatment of humans and animals with antibiotics. Functional feed additives such as PDCB, on the other hand, do not cure acute bacterial challenges, but prepare the gastro-intestinal tract of animals as well as possible to overcome negative consequences of certain diseases in a later life stage. As many of the effects of feed additives on bacteria are indirect, or modulate their virulence pathways, rather than having a specific bacteriostatic effect, the chances of building up resistance against these components is much less likely to happen. As such, feed additives can be a safer way of raising animals without AGPs, while they can also optimise intestinal integrity, thereby helping in reducing the need for veterinary application of antibiotics.

These examples also demonstrate that additives can never be a 'quick-fix', as replacer of antibiotics without careful consideration as to how the application of these products should be implemented in a production context that acknowledges that reducing dependency on antibiotics requires a multifaceted management approach.

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