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Review article

Plant based feed additives (phytogenic) as a primary solution to an antibiotic free nutritional program and feeding strategy in rabbit production

Never Assan*
Department of Agriculture Management, Faculty of Science and Technology, Zimbabwe Open University, Zimbabwe

*Corresponding author: neverassan@gmail.com

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ABSTRACT

Previously, before their restriction and/or ban, sub-therapeutic antibiotic growth promoters had been extensively used to improve growth rate and efficiency of feed utilization, in addition to curtailing morbidity and mortality in rabbit production. Due to the public health risk associated with the irrational and irresponsible use of low and sub-therapeutic prescription of antibiotics in food animals have raised great concern. For the last few years the focal point of various nutritional studies has aimed at searching for safe, natural and ecological products to eliminate the use of antibiotics in rabbit nutrition. The argument is that the use of sub-therapeutic antimicrobial drugs in food animals has increased the transmission and the proliferation of resistant bacteria via the food chain hence their adverse effect on human health and environment. The aftermath is the heightened advancement of resistance of diversified pathogenic microbial populations, especially bacteria against more antibiotics that are ultimately missing for the therapy in human medicine. In response to the elimination of antibiotics as growth promoters' rabbit nutritional studies have refocused on finding alternative organic compounds (natural) feed additives to replace the sub-therapeutic antimicrobial drugs. Testing of herbs, spices and other extracts (botanicals) as alternative feed additives have shown their effectiveness as appetizers, digestive and physiological stimulants, colorants, antioxidants, and for the prevention and treatment of certain pathological conditions. In this

regard, use of a wide range of phytogenic feed additives in rabbit nutrition has recently become a common management practice, since plant based feed additives have demonstrated to productively enhance rabbit growth performance, improve animal oxidative status, prevention and treatment of certain pathological conditions and acting as rabbit product quality enhancers. This has confirmed that plant based feed additives can be bestowed as a conceivable option to enhance a variety of critical processes in food animal physiology without adverse effects on human health and environment. Plant based feed additives may comprise among supplements that could positively influence feed quality, improve animal health besides animal products decidedly of their categorically efficacious elements. This is on the backdrop that food safety and identifiable high quality animal products are recommended to sustain consumer confidence and consumption. The present review attempt to assess the prospects of phytogenic feed additives as a major solution to an antibiotic free nutritional programs and feeding patterns in rabbit production.

1. Introduction

Low and sub-therapeutic prescription of antibiotics in animal feed played a crucial role for promotion of animal growth, enhance feed efficiency utilization and prevention and treatment of certain pathological conditions (Dibner and Richards, 2005; Niewold, 2007). However, due to the restriction and/or ban of sub-therapeutic antimicrobial drugs in food animals as growth promoters has forced rabbit nutrition specialists to find alternative feeding procedures or strategies targeting the use of rich bioactive feed additive compounds and/or mixtures which guarantee animal health and improve performance (Gidenne and Garcia, 2006; Maertens et al., 2006; N'Guessan et al., 2015; Nassar et al., 2013). Since remote past it has been widely acknowledged that plant based feed additives could be used for their antimicrobial properties, however their characterization has been possible only recently. Public health associated with antimicrobial resistance has accelerated focus on search for alternative plant and their derivatives based feed additives seen as safer and more environmentally friendly (Steiner, 2009). In this respect, phyto-additives and their extracts have provided an alternative feasible option to improve welfare and health in rabbit production (Szaboova et al., 2008) without harmful side effects for consumers. Several nutritional studies in rabbits, as well as pigs and poultry have confirmed efficacy of a variety of biologically active phytogenic feed additives on metabolic changes and immunological status of rabbits (Nosal et al., 2014; Nader et al., 2010; Meineri et al., 2010). Biologically active compounds found in onion, garlic, oregano and bananas (El-Wafa et al., 2002; Ismail et al., 2003; Matekaire et al., 2005; Rahman and Nada, 2006; Gugolek et al., 2008; Simonova et al., 2008) and herbal additives (Das and Bora, 2004; Gugolek et al., 2006; Zhuang et al., 2007) have been effectively characterized for the purpose of inclusion as rabbit feed additives. The existence of a variety of biologically active elements point to the fact that phytogenic feed additives may influence different metabolic pathways, different enzyme systems action and the mode of action of immunological responses together with performance parameters. Feed additives/supplements are a class of feed factors that can cause a covet animal response in a non-nutrient role for instance metabolic modifier, pH regulation or growth (Hutjens, 1991). Due to their mode of action the different plant based feed additives are classified into: nutritional feed additives (vitamins, minerals, plant enzymes etc.), sensory additives (influencing sensory attributes of animal products), zootechnical additives (immnomodulators, digestive stimulants, growth promoters of non-microbial origin, performance and animal product quality enhancers, etc.) and technological additives (antioxidants, substance curtailing mycotoxins contamination of feeds, etc.) (Karaskova et al., 2015). Feed processing, storage and packaging conditions may reduce utilization of some of the biological active compounds in feed additives (Gerencser et al., 2014). However, it is important to note that use of plant based feed additives is limited until their biological potential and efficacy,

as well as their impact on human health safety are ascertained (Hashem et al., 2017). Feed processing, storage and packaging conditions may reduce utilization of some of the biological active compounds in feed additives (Gerencser et al., 2014). This is on the backdrop that food safety and identifiable high quality animal products advance consumer confidence and promote consumption behavior. The present review attempt to assess the prospects of phytogenic feed additives as a major solution to an antibiotic free nutritional programs and feeding patterns in rabbit production.

2. Phytogenic feed additives and rabbit performance

Body weight gains observations were encouraging in growing rabbits fed herbal extracts, which also reduced faecal coccidian oocyst counts (Nosal et al., 2014). The use of garlic and oregano feed additives had a positive bearing on the level and nature of coccidia communicability, while sustaining an acceptable level of animal performance. Garlic and oregano feed additives showed a potential value in coccidiosis prophylaxy. Provision of intact onion bulbs provided an enriching stimulus that exerted a satisfactory biological effect on growth and reproduction, in addition to growing rabbits having improved final market weights (Gugolek et al., 2008). El-Wafa et al. (2002) observed that inclusion of dried powdered onion to feed improved body weight and daily gains of New Zealand White rabbits. 8% of linseed inclusion in rabbit diet had an adverse effect on daily weight gain besides final live weight as compared to rabbits without linseed supplementation (Bianchi et al., 2006). Inclusion of live yeast (*Saccharomyces cerevisiae*) in rabbit diet did not influence growth parameters, however it induced a short lived increase in weight gain and slightly modified caecal microbiota composition after weaning (Belhassen et al., 2016). This confirmed the results by Chaudhary et al. (1995) and Kimsé et al. (2012), where no improvement on growth parameters were registered in fattening rabbits due to live yeast inclusion in diet. However, this is in contrary with the observation made by Onifade et al. (1999) and Shanmuganathan et al. (2004) who reported that feed intake, feed conversion and body weight gain were improved with the inclusion of live yeast in rabbit diet. Lapiñski et al. (2017) demonstrated that herbal feed additives promoted faster weight gain due to kits willingness to eat, while having a positive effect on meat properties. In the same study, phytogenic additives of oregano, garlic and rosemary improved body weight as compared to rabbits fed coccidiostat. However, herbal water extract of garlic and oregano did not influence weight gain. Supplementing 3% microalgae (Arthrospira platensis) and/or 2.5% thyme (Thymus vulgaris) leaves in 7-week old dwarf rabbits did not influence growth performance and energy or nutrient digestibility (Dalle Zotte et al., 2013). Application of ethanol extract of propolis and an herbal mixture (*Rumexcrispus, Potentilla* and *Polygonumaviculare*) in drinking water significantly improved feed intake and final body weight, in addition to stabilization of acid base homeostais and hematological attributes (Kupczynski et al., 2016). The application of these herbal mixture extracts in drinking water point towards a significant impact on the welfare and performance of growing rabbits. Live weight gains were improved by inclusion of 0.5% chestnut tannins extracts in rabbit diet (Maertens and Struklec, 2006), while 0.45% inclusion of chestnut tannins extracts enhanced average daily feed intake and average daily gain in a similar study (Zoccarato et al., 2008). The chestnut tannins extracts efficacy is derived from its ability to protect intestinal mucosa against oxidative damage and pathogenic infection, while limiting peristaltic action in digestive malady reducing occurrence of diarrhea (Kermauner and Laurencic, 2008). Liu et al. (2009) observed that tannins improve the endogenous synthesis of palmitic acid, which could enhance dietary utilization of carbohydrates. In the same study, chestnut tannins improved the activity of α-amylase in small intestines in rabbits. Gondret (1999) argued that the increase in palmitic acid probably was a result of elevated availability of simple sugars because it has been confirmed that adipocytes of rabbits prefer to use glucose rather than acetate to synthesize fatty acids.

3. Phytogenic feed additives and rabbit carcass and meat quality

Carcass weight, dressing and liver percentage were improved using herbal feed additive (digestarom) in rabbits (Abd-El-Hady, 2014). In a similar study, there was no influence of plant based feed additive on most carcass attribute, except for acknowledged decline of abdominal fat content implying hypolipidemic effect of the additives (Hashem et al., 2017). This was in agreement with several researchers who observed that there was non-significant effect on carcass properties of fattening rabbits (Coloni et al., 2007; Eiben et al., 2011; Attia et al., 2014). Peiretti and Meineri (2008) observed that inclusion of 10 or 15% of *Salvia hispanica* seed in rabbit diets did not influence the proportion hind legs, four legs loin and abdominal wall, breast and ribs. Hashem et al. (2017) assessing the

potential contribution of two phytogenic feed additives compared with vitamin E on metabolism and growth in rabbits, all dietary formulations did not influence most carcass attributes except abdominal fat. On the other hand, plant based feed additives promoted relative weights of excratory organs, lungs and kidneys. The increase in excretory organs might be a sign of compensatory process due to dealing with an increase in elimination in catabolites of different phytogenic compounds from the blood system. Bernardini et al. (1999) observed that inclusion of 160g of ground linseed in rabbit diet significantly improved meat quality in terms of decreasing the n-6/n-3 PUFA ratio as compared to fish oil diet. Fermented wheat powder supplementation decreased the n-6/n-3 PUFA ratio displayed by a significant reduction of saturated fatty acids and a notable increase in polyunsaturated fatty acids, which favorably enhanced the rabbit nutritional quality as expected for health maintenance on consumers (Casamassima et al., 2016). PUFA content in *longissimus dorsi* muscle and peripheral fat improved with enrichment of 15% of *Salvia hispanica* seed in rabbit diets, while saturated fatty acids decreased. This influenced the decline of the n-6/n-3 ratio of rabbit meat from 4.55 to 1.03 (Peiretti and Meineri, 2008). In a similar study, inclusion of linseed in rabbit diet increased the long chain n-3 PUFA levels in meat and subsequently reducing the n-6/n-3 PUFA ratio (Kouba et al., 2008). The supplementation of dried artichoke bracts in rabbit diet imparted acceptable degree of unsaturation and low saturation, even if the n-6/n-3 ratio was slightly not good (Dabbou et al., 2014). Replacing palm oil with maize oil and Curcuma longa supplementation improved α-linolenic acid and PUFA n-3 contents and lowered the vaccenic acid levels, while reducing the n-6/n-3 ratio (Peiretti et al., 2011). Apart from improved blood serum lipid profile and antioxidant activity, linseed oil fortified with either organic selenium or ginger enhanced the composition of meat lipid component by means of increasing the level of n-3 fatty acids (Zeweil et al., 2016). Rabbit meat has been described as health meat for consumers and feeding has been a feasible option to elevate the content of n-3 PUFA, conjugated linoleic acid or vitamin E (Hernandez, 2008). The supplementation of fresh alfalfa and flux sprout to rabbit diet improved the fat content and fatty acids profiles, however, flux sprout alone compromised the oxidative status of meat (Dal Bosso et al., 2015). The variation in fatty acids composition apart from being influenced by feeding, likewise age, sex and breed have also been implicated (Cambero et al., 1991).

Modern consumers are progressively worried about food safety and animal welfare, linked to type of production systems, that have overwhelmingly dictated the supply of animal products, since they are known to define quality, healthy and safe food (Hernandez, 2008; Preziuso et al., 2009; Gerencser et al., 2013; Matics et al., 2014). Saturated fatty acids have been associated with adverse human health conditions, when in fact the unsaturated fatty acids, especially monounsaturated and n-3 polyunsaturated fatty acids, are beneficial to human health. Likewise, trans fatty acids have proved to have a negative effect on human health, whereas conjugated fatty acids being beneficial (Tvrzicka et al., 2011). Current rabbit nutritional research drive has targeted enhancing the n-3 polyunsaturated fatty acids content in meat through manipulation of dietary sources (Zeweil et al., 2016). Dietary modification has been a primary management practice to improve fatty acid profiles in order to address the reduction of triglycerides and food related disorders in consumers (Simopoulos, 1991).

4. Phytogenic feed additives and rabbit welfare and health

Various herbal plants possess pharmacological bioactive compounds which make them beneficial as curatives for numerous health conditions. The use of herbs, plants, fruits and their derivatives have shown potential to sustain animal health and improve performance, in addition to favorably succeed coccidiostats in rabbits (Ibrahim et al., 2000; Rohilla and Bujarbaruah, 2003; Gugolek et al., 2006; Rahman and Nada, 2006). Recently, many medicinal and aromatic herbals and their extracts have been characterized for their antimicrobial attributes in animal production, this is despite their use since ancient times (Costa et al., 2013). In pigs, supplemented herbal extracts have enhanced digestive secretion, improved digestibility and nutrient absorption, modification of intestinal microflora, excite immune system and promote antibacterial activities, coccidiostatc, anthelmintic, antiviral or anti-inflammatory and antioxidant activities (Costa et al., 2007; 2011). *Castaneasative* extract modified the stability of the gastrointestinal microflora possibly resulting in efficient microbial synthesis (Gai et al., 2011). It was noted that efficacy of *Castaneasative* extract was dependent on age of rabbit, being more favorable in growing kits than older rabbits. Kupczynski et al. (2016) demonstrated that inclusion of ethanol extract of propolis and an herbal mixture (*Rumexcrispus, Potentilla* and *Polygonumaviculare*) in drinking water had a positive influence on the reduction of time of recovery of chronic diarrhea in rabbits. In a similar study, which used propolis as a feed additive, it was noted that the effect of propolis is distinguished by its favorable antibacterial attributes

regards to gram-positive bacteria strain (e.g. S. aureus) (Umthong et al., 2009). There was no inhibitory influence on *E. coli*, despite propolis and herbal extracts showing bacteriostatic properties (Kupczynski et al., 2013). The influence of propolis was also dependent on origin and composition (Farnesi et al., 2009). Nader et al. (2010) observed that ethanol extract of propolis had a protective effect against atherosclerosis development in growing rabbits supplemented with a high cholesterol diet. However, in a similar study additives did not influence blood serum cholesterol (Kupczynski et al., 2016). Degree of coccidiostat infection were comparable in New Zealand White females and Flemish Giant males fed diets supplemented with coccidiostat and finely ground white mustard (Sinapis alba) seeds (Gugolek et al., 2011). Elsewhere, administration of ginseng extract in rabbits significantly reduced oocyst counts from 1435 to 217 in 1 gram of feacal samples collected from 10-week old rabbits (Simonov et al., 2008). It has been noted that coccidial infection rates might not be correlated with performance outcome. It seems rabbits in good health, challenged with high oocyst fecal count may not succumb to reduced performance (Gugolek et al., 2011). Improved rabbit health and reduced risk of mortality was registered as a result of feeding vegetable supplements (Gugolek et al., 2006). Soultos et al. (2009) registered a significant decrease in *Pseudomonas* spp and *Enterobacteriaceae* due to inclusion of oregano essential oils as compared to a control standard without its supplementation.

5. Phytogenic feed additives and rabbit meat antioxidant status

Lipid oxidation is the primary form of chemical deterioration in meat (Medina-Meza et al., 2014; Shah et al., 2014) and their products. This process is the major contributor to imparting poor quality meat and their products as a result influencing negatively meat products acceptability to the consumer. Meat and their products should maintain a certain degree of rancidity to be acceptable to consumers (Sammet et al., 2006). Lipid oxidation has an adverse effect on meat flavor (Faustman et al., 2010) and it encourages formation of rancid odors (Fernandez et al., 1997), discoloration (Juncher et al., 2001) and formation of potentially poisonous compounds (Richards et al., 2002) in meat. The lipid oxidation activities result in formation of various compounds which are associated with off flavors and compromised meat quality. In this regard, oxidation of unsaturated fatty acids has some economic implications through reduced nutritional composition of meat, poor flavor, safety and storage problems (Kanner, 2007). The hydroxyperoxides produced during lipid oxidation will give out secondary oxidation products imparting adverse sensory meat properties (Frankel, 2005). Dietary antioxidants protect cells structures (proteins, lipids and DNA) against oxidative damage. Non-antioxidant status induces physiological and pathological processes associated with poor performance and pathological disorders that also impinge on meat quality status (Abdel-Khalek, 2013). The sensitivity of muscle tissue to lipid oxidation can be lessened by use of antioxidants, despite the process being influenced by various factors. Herbal mixture of ten different herbs and spices rich in flavonoids enhanced meat antioxidant activities in rabbit meat (Mattioli et al., 2016). Hashem et al. (2017) demonstrated the potential of vitamin E and propolis as feed additives for improving rabbit performance and antioxidant status of growing kits. In a similar study, Popovic et al. (2017) observed that inclusion of wormwood in rabbit diet improved antioxidant status and performance, as well as reducing the oocysts count. 3 to 6% of linseed plus 200mg of vitamin E/kg imparted sufficient oxidative stability on rabbit meat, in addition to enhancing the proportion of alpha linolenic acid (Petracci et al., 2009). These results were in agreement with Bianchi et al. (2009) using 35% alfalfa plus 200mg vitamin E/kg feed. Dietary antioxidant vitamins had inverse, dose response associated with gamma-glutamyl transferase content based on reference range (Makhlouf and Makhlouf, 2012). Plant based extracts on *Lippiacitriodora*, horseradish and lycopene improved plasma oxidative status in rabbits (Casamassima et al., 2017). Castillo-Lopez et al. (2017) working with *Echinacea purpurrea* extract supplementation to pregnant and weaned rabbits observed an increase in antioxidant activities and enhanced immunological function, in addition to curtailed mortality rate after weaning. In contrary, Dalla Zotte and Cossu (2009) observed an ineffective antioxidant status influence on meat rabbit. The inclusion of dietary alfalfa flavonoids enhanced muscle oxidation stability without any negative effects on growth performance in rabbits, while a slight impact on meat color was registered (Dabbou et al., 2018). Spirula supplementation improved IgG production without influencing the animal antioxidant status in rabbits (Kovacs et al., 2016). However, Deng and Chow (2010) argued that Spirula is effective in preventing diseases that are related to oxidative stress or inflammation. Inclusion of phytogenic feed additives may promote antioxidant efficacy and reduce the circulation of free radicals as a result protecting cells from damage, while minimizing free radical induced disorders (Ghoneim et al., 2016). Free radicals induced cell damage

has been the prime source of pathogenesis of most diseases. Herbal feed additives supplementation in this regard assist in immunity modulation and curtail free radical induced disorders.

6. Implications

Based on the literature reviewed, the several rabbit nutritional studies suggest that plant based feed additives and their derivatives can be effectively utilized without adverse effects, enhance performance and animal welfare. Supplementation of various natural feed additives have resulted in improved animal antioxidant status and performance, sustained health and reduced risk of mortality. This proffer an essential justification for an alternative antibiotic free rabbit nutritional programs and feeding strategies which improve growth traits, reproductive performance, curtail mortality and produce acceptable meat quality. Significant decrease in the fecal oocyst count has been registered with some herbal supplements.

It would be reasonable to suggest that due to the anatomical and physiological nature of the rabbit digestive system which is sedo-ruminants (rabbits are mono-gastric non-ruminant herbivores) the efficacy of phytogenic feed additives might be different in rabbits as compared to pigs and poultry. This is on the backdrop that modern intensive pig and poultry production systems have realized exceptional gains in the economics and efficiency in production of high quality and safe products by way of use of feed additives. This is probably as a result of the specific peculiarities in the digestive system of a rabbit as compared to monogastric and polygastric might interfere or influence the physiological limit to achievable and/or expected responses in efficacy of phytogenic feed additives utilization. Nutritional strategies which attempt to break this physiological limit on the efficacy of phytogenic feed additive utilization need to be exploited in rabbits' nutritional science. Probably the potential target to improve phytogenic feed additives utilization in rabbits digestive system is to synchronize the functionality of the cecum, which might lower the digestive system physiological limit on the expected response in performance. Manipulation of the digestive ecosystem to enhance the efficacy of phytogenic feed additives should be a priority in rabbit nutritional studies. It seems the digestive microbiota composition in the caecum constitutes potential core species and satellite species of which some have not yet been characterized and are unique to the rabbit. Therefore, a comprehensive rabbit digestive ecosystem evaluation on its interrelated functionality in terms of physiological roles of hydrolysis and fermentation might provide new insights on the effective utilization of phytogenic feed additives.

Generally, the key challenge in use of plant based feed additives is to identify the presence and concentration of biological active compounds. The characterization of the biologically active ingredients and their combination is dependent on the quality of plant raw material. This is on the premise that the presence and concentration of the biologically active ingredients in plant based feed additives will be influenced by genetics, agro-ecological conditions, extraction technology, and also how the raw material is processed and stored. Therefore, the choice of plant based material which takes into account farming condition is crucial for comprehensive quality of plant based feed additives. This implies that consistency and standardization of concentration and the quality of the biological active compounds is an essential benchmark to assert the efficacy of plant based feed additives. Given that the list of characterized plant based feed additives could be expanded, further and more detailed studies need to be carried out to ascertain their safety on consumers and environmental impact.

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