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Utilization of certain unconventional feeds in poultry as natural alternatives to curb antimicrobial resistance

NEETI LAKHANI[1✉] and MANISH CHATLI[2]

College of Veterinary Science, Rampura Phul, Bathinda, Punjab 151 103 India

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ABSTRACT

Poultry market has been demonstrating emerging potential as an efficient producer of meat with consumption occupying 40.6% of market globally. The requirement of animal protein is anticipated to grow continuously for meeting the requirements by ever-growing human population. To fulfill this demand, the major challenge encountered by poultry breeders was to fasten production in most efficient and economical way. Use of antibiotic growth promoter in feed help in augmenting poultry growth and alter gut microbiota. However, the growing concern of its likely fatal impacts on animal, food safety and on humans for developing microbial resistance; their use was restricted and banned in several countries. With insinuation of ban, several alternatives were explored for having potential growth promoting benefits without hampering the normal gut microbiota. Various phytobiotics, prebiotics, probiotics, organic acids and unconventional feed have positive effects on feed intake, efficiency and play a role as antimicrobial. However, the meager knowledge on availability and potential use of unconventional feeds as potent antimicrobial restricts its application. This review focuses on certain non-conventional feeds rich in specific bioactive compounds, which are attributed to modulate intestinal microbiota, their effects on growth performance and overall health status of poultry. The review aims to serve as a reference for young researchers and poultry industry to recognize alternative feed ingredients to be used as antimicrobial growth promoters minimizing competition between human and animal consumption.

Keywords: Antibiotic, Antimicrobial, Microbial resistance, Microbiota, Non-conventional

The peculiarity of antimicrobials in fighting against bacterial infections of human and livestock in varying dosage has gained tremendous attention among production sector. Incorporation of substantial dose of antimicrobials as non therapeutic drug promotes growth in livestock (Jagtap *et al.* 2019) and poultry (Rahmann *et al*. 2022). The ability of antimicrobials in suppressing release and absorption of harmful toxins by bacteria in the intestine can be postulated as one of the factors in promoting growth of broilers. Many authors postulated benefits of dietary antibiotics on growth rate by 1–10% (Chattopadhyay 2014), efficient feed conversion and better meat quality (Hughes and Heritage 2002) in poultry. However, exploitation of antimicrobials as a part of feed to stimulate growth changed the dynamics of feed industry leading to its indiscriminate use in sub therapeutic doses (Chattopadhyay 2014). The rising concern on food safety was reiterated in form of bans by European Union on use of antimicrobials.

Exploitation of antibiotics as growth promoters seems to have developed antimicrobial resistance within poultry. Antimicrobial resistance remains a regularly swelling menace for poultry health, minimizing the ability to treat bacterial infections and enhancing risk of morbidity and mortality (Hedmann 2020). In addition to the emergence of resistant bacteria from poultry production, the critical factors that prohibit their inclusion in poultry diet were rising human health apprehensions on presence of antimicrobial residues in egg (Goetting *et al*. 2011) and meat. The emergence, spread and diligence of antimicrobial resistance remains a critical global health threat (WHO 2014) (Fig. 1). There is an urgent need to identify effective, economical alternatives to antimicrobials for animal growth promotion, elevating the need for deeper understanding of microflora physiology and interaction with host. Phytobiotics or phytogenic feed additives come as a saviour in this regard. Phytobiotics are rich in plant derived products such as essential oils, resins imparting them growth promoting effects (Lakhani and Lakhani 2018), antimicrobial, antioxidant (Lakhani *et al*. 2019) and anti-inflammation activity (Lakhani *et al*. 2019). However, contradictory results have been published regarding the benefits of phytogenic feed additives (PFA) in animal health. Non-conventional feed resources have recently emerged as potentially non-toxic additives improving the balance

Present address: [1]College of Veterinary Science, Rampura Phul, Bathinda, Punjab (Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab). [2]ICAR-Central Institute on Goats, Makhdoom, Mathura, Uttar Pradesh. ✉Corresponding author email: lakhani10aishi@gmail.com

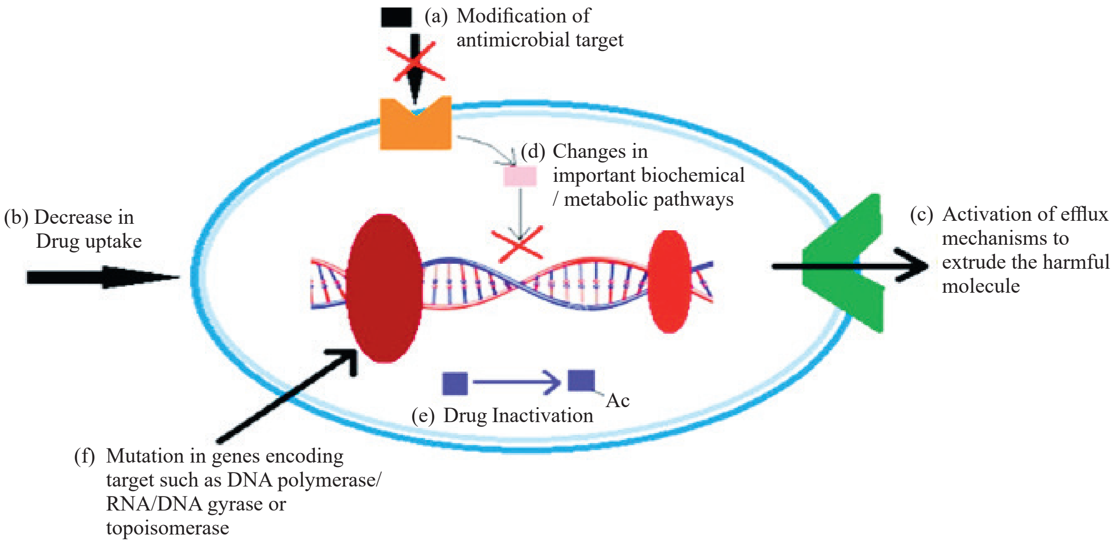

Fig. 1. Antimicrobial resistance mechanism through different biochemical routes inside the body.

of beneficial bacteria in gut of poultry. Utilization of non conventional feeds as poultry growth promoter is because of the presence of secondary metabolites such as essential oils, saponins, tannins (Lakhani *et al*. 2019) modulating the gut environment and morphology of intestine in poultry. The present review attempts to highlight the prospects of some newer unconventional feeds as potential natural antimicrobials in broiler chicken diet being rich in bioactive compounds. The focus of the present review is to furnish safer antimicrobial level having an achievable choice for lower mortality rate, good feed efficiency and safeguarding environment and consumer health in poultry industry.

Ginger as a natural antimicrobial

Zingiber officinale, a monocotyledonous herb, familiar as ginger; is conventionally used for its feed flavouring property (Wang *et al*. 2005). Numerous pharmacological properties of ginger include anti-inflammatory, gastrointestinal modulating agent, antimicrobial and as an antioxidant. Ginger is known to have broad pharmacological impacts because of presence of numerous chemical constituents in basic ginger oil (Table 1).

Table 1. Chemical constituents of ginger oil

Component	Effect	Metabolite	Reference
Ginger roots	Antifungal	Zingiberene (28.62%) Camphene (9.32%) Curcumene (9.09%)	Singh *et al*. (2005)
Roots	Antimicrobials	Sesquiterpenes (66.66%) Monoterpenes (17.28%)	Sharma *et al*. (2015)
Rhizomes	Antioxidant and antimicrobial	β-sesquiphellandrene (27.16%) Caryophyllene (15.29%) Zingiberene (13.97%)	El-Baroty *et al.* (2010)

Ginger oil primarily includes large characters of monoterpenes as sesquiterpenes, and have critical antimicrobial action against most pathogenic microorganisms (Sharma *et al.* 2015). El Baroty *et al.* (2010) reported that inclusion of 20 to 120 µg/mL of ginger oil is effective in suppressing the population of bacteria and fungi in the intestine. Increasing the dose level may further be fruitful in controlling the *E coli* population. The efficacy of ginger oil is more than turmeric oil for their bacteriostatic and bactericidal impact, and serves as an important substitute governing *Salmonella* infection (Majolo 2014) (Table 2).The incorporation of ginger powder in poultry diet may further aid to minimize cost-benefit ratio and improve economic feasibility.

The antibacterial activity of ginger oil and ginger powder is attributed to their potent cytotoxic potential . The cytotoxic potential is due to presence of saponins, steroids, alkaloids in ginger (Ezez *et al.* 2021) imparting the AMR activity. The presence of phenolic ketone derivatives also imparts antioxidant activities to ginger oil. The enhanced serum antioxidants may also be partially attributed to the slowing of the process of oxidation of the feed by ginger powder supplementation (Zhao *et al*. 2011) (Fig. 2).

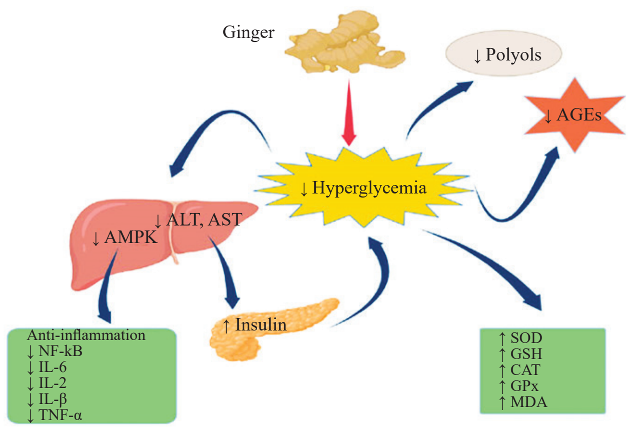

Fig. 2. Mechanism of action of ginger inside the body.

Table 2. Effective dose of ginger in diet and its effect on poultry

Component	Dose	Poultry	Effects	References
Ginger powder	0.6% of diet	Broiler	Better feed conversion efficiency	Hassan *et al.* (2019)
Ginger extract	100g per tonne	Layer	Increased egg weight and egg quality and antioxidant status	Wen *et al.* (2019)
Ginger oil	0.4% of diet	Broiler	Increase FCE, increase *Lactobacillus* and decrease Salmonella spp in caecum	Oluwafemi *et al.* (2021)
Ginger powder	15g/kg diet	Broiler	Increase antioxidant indices	Al-Khalaifah *et al.* (2022)

Impact of marigold supplementation as potent antimicrobial

Marigolds (*T.erecta*) are medicinal plants popular for their high therapeutic values. It contains active components such as alkalonoids, terpenes, flavanoids and phenolic compounds (Regaswamy and Koilpillai 2014) of which flavonoids exert mild anti-inflammatory action and vitamin C/carotenoids improve immune function (Fig. 3).

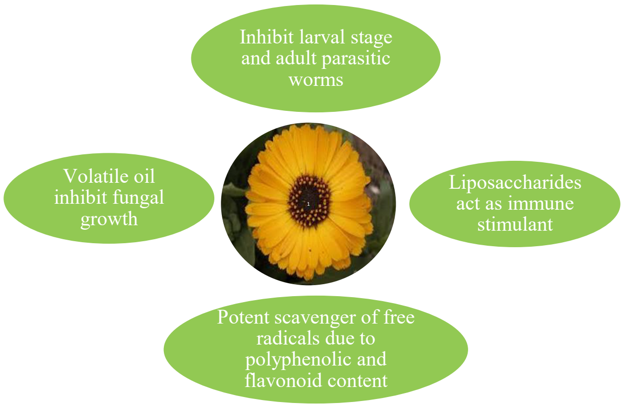

Fig. 3. Mechanism of action of marigold.

Flavonoids impart bright yellow colour to egg yolk improving its marketing rate by Rs 2 kg/quintal. Feeding 1% and 3% of marigold to laying birds stimulated proliferation of total leukocytes as well as T and B lymphocytes at 3% level of supplementation (Balenovic *et al*. 2018). Frankic *et al*. (2009) reported antioxidant effects of marigold petals and flower-top extracts which was comparable to those of vitamin E supplementation in laying birds. Agiang *et al.* (2011) added 0, 2.5%, 5, 7.5 and 10% marigold leaf in quail diet and reported significant effect on final weight, daily feed intake and mortality rate at 5% level. However, the presence of certain toxic factors inherent in leaf products have been implicated for the depression in feed intake when supplemented at higher doses.

Uchewa *et al*. (2012) demonstrated that inclusion of 250 ml/2 litres of marigold leaf extract showed highest feed intake attributing to the active phytochemical ingredients such as high crude protein and sterol. The active ingredients have beneficial effects on gastrointestinal ecosystem by inhibiting growth of pathogenic microorganism and improving health status of digestive system thereupon reducing exposure of birds to microbiological toxins, stress situations and increment in the absorption of essential nutrients. However, many studies have reported (Alcicek *et al*. 2003) non-significant effect on body weight, FCR and carcass traits with marigold supplementation which might be due to difference in species, dose rate and mode of administration.

Moringa oleifera as a potential antibacterial, antifungal and coccidiostat

M oleifera is popularly known as a miracle tree owing to its wealthy resource of numerous nutrients and high biological value (Table 3). Moringa is used as a feed additive in poultry diet for its coccidiostat and antibiotic growth promoter effects. It perpetuates intestinal integrity and improves nutrient utilization efficiency and ultimately animal health.

Table 3. Active compounds in *M. oleifera* and their properties

Ingredient	Property	Reference
Vitamin B complex, Vitamin C, pro-Vitamin A, Vitamin K	Highly nutritious	Chandran *et al.* (2022)
Acetone	Antibacterial	
Phytochemicals, PUFA	Anti-septic and detergent	
Saponin	Coccidiostat, antifungal, antibacterial	

The antimicrobial activities of the *Moringa oleifera* may be due to the presence of lipophilic compounds and metabolites (carboxylic acid and chitinases) in plant cell walls (Abd El-Hack *et al.* 2022) (Fig. 4). Moreover, the saponins present in Moringa are also known to modify the cell membrane structure, interrupting interaction of cell membrane and pathogens (Table 4).

Coccidiosis is one of the most common diseases in poultry. Plant extract having 3.24% saponin was responsible for damage of 36% *Eimeria* oocytes by entering inside the egg and disrupting their sporocyst. The results were similar to that reported by Hassan *et al.*

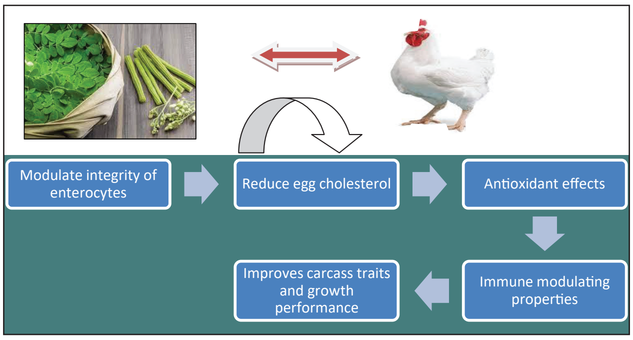

Fig. 4. Mechanism of action of *Moringa oleifera.*

Table 4. Effective dose of *Moringa oleifera* in diet and its effect on poultry

Component	Dose	Result	Reference
Moringa oleifera leaf	3, 6, 9 g/kg	Increased egg production and quality, decrease cholesterol	Ahmed *et al*. (2021)
Moringa oleifera leaf	4-6% diet	Increased egg production, feed efficiencies, shell thickness, β-carotene, Mg and Ca contents	Bidura *et al.* (2020)
Moringa oleifera seed	1 g/kg	Urea decrease, increase egg production	Ashour *et al*. (2020)
Moringa meal	0.2%	Increase SOD, GPx and FCR	Yang *et al*. (2023)
M. oleifera extract	12%	Decrease *E. coli* infection	Ullah *et al*. (2022)
M. oleifera extract	3%	Inactivate *B. cereus* and *E. coli*	Sharma *et al*. (2020)

(2016) when fed 5% saponin in diet of poultry. Reduction in incidence of coccidiosis was confirmed by reduced haemorrhage in caecum. Inclusion of *M. oleifera* leaf meal at 25% had no negative effect on body weight and FCR of poultry when used as supplemental diet for soyabean meal (Gadzirayi *et al.* 2012). *Moringa oleifera* is known as a potential antioxidant due to the presence of vitamin C and E, carotenoids, flavonoids, and selenium (Moyo *et al*. 2015). Supplementing 1.2% *Moringa* leaf powder in diet of poultry resulted in better absorption of nutrients, modulating the intestinal structure and acidic mucin production (Khan *et al.* 2017).

Seaweeds as potent alternative to antibiotics

Seaweeds serve as a rich source of carbohydrates, protein, minerals, vitamins and dietary fibers, having well-balanced amino acid profiles and unique blend of bioactive compounds. Chemical composition of different seaweeds are variable with brown seaweed being rich in mineral and iodine (Misurcova *et al*. 2011), red seaweed containing highest protein level (10-50%) and green seaweed having protein content not more than 15% (Overland *et al*. 2018). Green seaweed (*Ulva spp*.) improved breast muscle yield and dressing percentage in broilers at 3% level (Abudabos *et al.* 2013) attributed to availability of S containing amino acid and crude fibre whereas inclusion of green seaweed at level of 4% and 6% serve as prebiotic supplement in diet of poultry. Brown algae contain functional polysaccharides alginates and fucoidans possessing anti-coagulant, anti-inflammatory, anti-viral properties. Red seaweeds are considered highly nutritious for poultry diet and considered as direct source of protein to growing chickens. Seaweeds are known to increase the lactic and acetic acid concentration in gut, altering the microbiome of caecum and improving the overall performance of broilers (Choi *et al*. 2014). Supplementing *Sargassum* spp in diet of broilers led to high FCR, blood low density lipoprotein (LDL), which might be attributed to bountiful of vitamin, mineral, amino acids, fatty acid content. However, effect of seaweed as prebiotic, antibiotic growth promoter or as additive varies with inclusion level, extraction method, its purification and difference in the method for its preparation. Table 5 enlists few important beneficial aspects of seaweed inclusion in diet of broilers.

Aloe vera as feed additive in broiler diets

Aloe vera is one of the oldest drug having traditional medicinal properties. The most important part of aloe vera is its leaves containing gel having 99% water (Femenia *et al.* 1999) and rich in active ingredients possessing anti-inflammatory, immunomodulatory and antioxidant properties (Christaki and Florou-Paneri 2010). The well known effect of aloe vera on intestinal microflora, is attributed to the polysaccharide 'aceman-nan' in the dry matter of aloe vera gel (Choi and Chung 2003). Inclusion of 1.5%, 2%, and 2.5% of aloe vera extract in broiler diet lead to increase *Lactobacillus* and decrease in *E. coli* count (Darabighane *et al.* 2012). Similarly, decline in *E. coli* count was observed when acemannan (0.1% and 0.05%), and aloe vera gel (0.1%) were added separately to broiler feed (Dai *et al*. 2007). Many other studies have been reported on the antibacterial properties of aloe vera extract (Pandey and Mishra 2010). The exact mechanism by which aloe vera extract exhibits its antibacterial property is not clear but it is most likely that they possess properties similar to prebiotics (Guo *et al*. 2003) or presence of fumaric acid.

Moreover, it has been reported that diet treated with 2% aloe vera gel caused an increase in the antibody titre of broilers and improve humoral immune response (Valle-Paraso *et al.* 2005). Similar results were reported

Table 5. Inclusion level and beneficial effects of seaweeds

Seaweed	Inclusion level	Function	Reference
Sargassum spp	2%, 4% and 6%	No effects on carcass traits Reduced total cholesterol, elevated HDL	El-Deek (2011)
Sargassum spp	1%, 2%	Reduced both blood plasma cholesterol and globulins	Kumar *et al.* (2018)
Fucoxanthin	100 mg/kg	Increased CAT, SOD activities and glutathione (GSH) levels	Gumus (2018)
Polysiphonia spp.	3%	Elevated protein and mineral level	El Deek *et al.* (2009)
Kappaphycus alvarezii	1.25%	Improve performance, immunity and breast yield	Qadri *et al*. (2019)
P. palmate	0.15%	Reduce pathogenic (*E. coli*) and increase beneficial (*Lactobacillus*) bacteria	Gumus (2018)

in broilers on inclusion of 0.5%, 0.75% and 1% aloe vera gel (Alemi *et al*. 2012) and 0.1% and 0.05% (Jiang *et al*. 2005) acemannan in broiler diet. Besharatian *et al.* (2012) reported an increase in total immunoglobulin of 35-day-old broilers offered aloe vera leaf powder (0.5% and 1% mixed with feed) and aqueous extract of aloe vera leaf (15 and 30 ml/l, added to drinking water).

Cabbage and its benefits as an antimicrobial

Cabbage (*Brassica oleracea*) contains a plethora of active ingredients providing it its antimicrobial status (Table 6). These bioactive constituents include glycosides, alkaloids, flavonoids and saponin, as well as tannin, terpenes, steroids (Chauhan *et al*. 2016). Cabbage extract has highest antimicrobial activity against Gram negative bacteria. The antimicrobial activity in cabbage is imparted by the various sulphur compounds present in them (sinigrin, allyl isothiocyanate, S-methyl-L-cysteine sulfoxide, dimethyl disulfide, methyl methanethiosulfinate, dimethyl sulfide and methyl methane thiosulfonate) (Buttery *et al.* 1976).

Table 6. Active constituents in cabbage

Ingredient	Function	Reference
Sulforaphane	Anti inflammation, oxidative stress and cholesterolemia	Leja *et al*. (2010)
Indole-3-carbionol	Cancer prevention	Guerrero-Beltrán *et al*. (2012)
Flavonoids	Cancer prevention	Guerrero-Beltrán *et al*. (2012)

Body weight of chicken increased significantly when fed 3% cabbage whereas body weight decreased when levels were increased upto 6% cabbage in diet (Adesina and Toye 2012). Addition of cabbage tree extract rich in fructose (5-10g/kg) in poultry diet resulted in reduction of *Clostridium perfringes* in caeca of birds. Similarly, the number of Lactobacilli increased in ileum and caeca of birds with the plant extract (Vidanarachchi *et al*. 2010) indicating that the plant extract beneficially modulated the microflora of gut. Mustafa and Baurhoo (2017) also reported similar decrease in body weight of broilers with increasing level of dried cabbage residue from 3 to 6% in diet. Adding 20g/kg of cabbage slurry in broiler diet improved intestinal development in broilers causing growth of beneficial bacteria and increasing length of duodenum villi, villus:crypt ratio (Jianping *et al*. 2018). Addition of 12% dietary dried cabbage residue in layer diet showed marked improvement in nutrient utilization suggesting that bioactive constituents in cabbage had no harmful effect on gut microbiota of layers (Mustafa and Baurhoo 2018) (Table 7).

Table 7. Inclusion level and beneficial effects of cabbage

Component	Dose	Effect	Reference
Cabbabge slurry	20g/kg	Enhanced growth and intestine development	Jianping *et al.* (2018)
Cabbage leaf residue	12%	Increased PUFA, linolenic acid in eggyolk, increased nutrient utilization	Mustafa and Baurhoo (2018)
Cabbage leaf	10g/kg	Increased number of caecal lactobacilli	Vidanarachchi *et al*. (2010)

Conclusion

Major public health challenges now-a-days are the expanding foodborne infections emerging from indiscriminate use of antibiotics in food animals. This has paved way to sought for alternatives to antibiotic application in food animals responsible for many related infections. Incorporation of available alternatives to antibiotics (probiotics, prebiotics, plant extract and organic acids) has a potential role in lowering dependency on the existing antimicrobial substances. Moreover, unconventional feeds have now emerged as potent antimicrobials to fight against antimicrobial resistance. The active ingredients present in these non-conventional feeds have paved way for future research to utilize these metabolites in proper dose and help improve poultry performance and health.

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