



## Phytobiotics in Poultry Industry as Growth Promoters, Antimicrobials and Immunomodulators – A Review

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### ABSTRACT

Due to the hazardous use of antimicrobials in poultry production sector, development of drug resistance have become a worldwide problem. Therefore, using biotic or natural products, such as phytobiotics (phytogenics or botanicals) have received a great attention as antibiotic substitutes. The use of phytobiotics or their constituents have been considered as a relatively new class of natural herbs that gained popularity and acceptability among poultry farmers. The incorporation of several types of phytobiotic additives in poultry feed have proved their ability to enhance the productive performance of broilers as well as layers. Moreover, phytobiotics presented great efficacy in counteracting intestinal pathogenic microorganism while maintaining the population of normal inhabitant beneficial microflora. Immunostimulatory effect on both humoral and cellular immunity as well as antioxidant properties were recorded as characters of phytobiotics. Therefore, this review article aimed to give a spotlight on the uses of different types of phytobiotics as poultry dietary additives to improve the productive parameters, reduce the pathogenic intestinal bacteria, and potentiate the immune response, especially after vaccination processes.

**Keywords:** Antimicrobial, Immunity, Performance, Plants, Poultry

### INTRODUCTION

From several decades till now, poultry industry has been recognized as an important subsector of agricultural and veterinary fields due to increasing demand for meat and eggs as low cost protein sources. Continues feeding by sub-therapeutic levels of antimicrobials as growth promoter agents or antimicrobial compounds have had a negative impact on the balance of normal inhabitants of gut microflora, accumulation of antibiotic tissues residues as well as developing new strains of drug-resistant pathogenic bacteria (Castanon, 2007). Therefore, in 2006, the European Union Commission banned using of antibiotics in animal feeds as a growth promoter in different countries (European Union Commission, 2005).

Phytobiotics are also termed as phytogenics or botanicals. They are defined as natural, less toxic, and residue free plant-derived compounds that have been used as feed additives for livestock production (Wang et al., 2008). Phytobiotics are composed of natural bioactive components or substances of plant origin including terpenoids, alkaloids, glycosides and phenolics (Shad et

al., 2014). Phytobiotics could be classified as herbs from flowering, non-woody and non-persistent plants, botanicals or spices from non-leaf parts like seeds, fruits, bark or root, essential oils or extracts and oleoresins (Bote, 2004).

Numerous studies have been conducted to demonstrate the effect of phytobiotics as growth promoter feed additives like prebiotics and probiotics to enhance overall performance parameters as well as health conditions of poultry (Yasodha et al., 2019; Özbudak, 2019). Phytobiotics have been also used as antimicrobial, antiparasitic, anticoccidial as well as immunostimulant agents in poultry field (Manafi, 2015; Gilani et al., 2018; Hafeez et al., 2020). Phytogenic substances were extremely studied in different species of monogastrics (Gheisar and Kim, 2018), rabbits (Alagawany et al., 2018; Al-Sagheer et al., 2019) and fish (Naiel et al., 2019).

So, the purpose of current article review was to spotlight on phytogenic compounds that used in poultry field and their effects on the productive performance, antimicrobial activities and immuno-stimulatory properties.

### Production parameters

Improved growth parameters were detected in birds fed on different kinds of herbs, polysaccharides or essential oils components (Yasodha *et al.*, 2019). The enhancement of the growth performance parameters after supplementation of phytobiotics may depend on the synergistic mechanism among their active molecular complex (Hussein *et al.*, 2020a). Phytobiotics could maintain or improve normal intestinal architecture, increase the villus length and consequently increase the surface of intestinal absorption (Tabatabaei, 2016). It has been demonstrated that phytobiotics are able to stimulate saliva production, secretion of digestible enzymes and bile production resulting in improving the performance and digestibility (Alloui *et al.*, 2014). Moreover, phytobiotics enhance the digestion and utilization of protein in the intestine (El-Gendi, 1996), decrease the gut pathogens (Kubkomawa *et al.*, 2013) and increase *Lactobacillus* spp. count (Windisch *et al.*, 2008). It has been observed that supplementation with herbal feed additives can alter the histological structure of the intestine and indicated elevation of the intestinal villi by deepening of its crypts (Murali *et al.*, 2012), increasing the dendritic cells absorption capacity in the intestinal lumen, stimulation of toll-like receptors and activation of epithelial to release the mucosal cytokines. Alcicek *et al.* (2004) assumed that feeding of broilers on phytobiotics stimulated the secretion of high amount of intestinal mucus and consequently reducing the pathogens adhesion and establishment of gut microbial eubiosis.

Supplementing broiler feed or water with essential oil mixtures of thymol and cinnamaldehyde (Tiihonen *et al.*, 2010), thymol and star anise (Kim *et al.*, 2016), clove and cinnamaldehyde (Chalghoumi *et al.*, 2013), coriander (Ghazanfari *et al.*, 2015; Hady *et al.*, 2016), oregano (Hashemipour *et al.*, 2014), a mixture of oregano, anise, and citrus peel (Abdelnaser *et al.*, 2019), carvacrol (Jamroz *et al.*, 2006), a blend of carvacrol, cinnamaldehyde and capsicum oleoresin (Bravo and Ionescu, 2008) and ginger extract (Olaiifa *et al.*, 2019) had been detected for improving all performance parameters including; feed intake, feed conversion rate and body weight. Moreover, supplementation of broilers diet with garlic (Elagib *et al.*, 2013), a mixture of garlic, mushroom and propolis (Daneshmand *et al.*, 2012), turmeric powder (Ahmadi, 2010), guggul resin (Iranparast *et al.*, 2014), dried ground leaves of stevia (Atteh *et al.*, 2008) and black cumin seeds (Khalaji *et al.*, 2011) presented the enhancement of broilers performance. Studies on the effects of herbal compounds on the production of broilers

Japanese quail's revealed improvement of all performances (Manafi *et al.*, 2016).

The laying hens represented improvement in egg production, eggshell strength and thickness as well as internal egg quality after treatment with mixture of plant extracts and essential oils (Bölükbaşı *et al.*, 2008; Radwan *et al.*, 2008; Kaya *et al.*, 2013).

### Antimicrobial effect

It has been demonstrated that phytochemical compounds of phytobiotics have a strong antimicrobial activity against Gram-positive and Gram-negative bacteria either *in vivo* (Al-Kassie, 2010; Daka, 2013) or *in vitro* environment (Al-Mariri and Safi, 2014). Some phytochemical compounds as alkaloid inhibit DNA synthesis (Karou *et al.*, 2005) and form saponin complexes with the cell membrane (sterols) which leading to cells damage and collapse (Morrissey and Osbourn, 1999). The antimicrobial effect of essential oils could be refer their ability to penetrate through the bacterial membrane (Helander *et al.*, 1998), their chemical structure (Frag *et al.*, 1989) and their aromaticity (Bowles and Miller, 1993).

Modulation of the gut microflora by phytochemical compounds plays an important role in maintaining host health (Tollba *et al.*, 2012). Several studies revealed that phytochemical compounds or their extracts reduce the population of intestinal pathogenic organisms and their metabolites, but increase the number of intestinal normal and helpful microflora which relief the intestinal challenge and immune stress and consequently increase intestinal performance (Liu *et al.*, 2014). It has been speculated that organic acids of some phytobiotic feed additives may lower the intestinal pH that led to inhibiting the pathogenicity of local pathogens and lowering the level of their toxic products (Manafi *et al.*, 2016).

### Antibacterial effect

Essential oils could reduce the growth of common intestinal poultry pathogens. Inoculation of thyme and cinnamon in the broiler ration reduces the total bacterial as well as coliform count in the intestinal tract (Karangiya *et al.*, 2016). An *in vitro* study revealed that flowers of *Calendula* presented growth inhibitory effects on *Escherichia coli* (*E. coli*) (Arora *et al.*, 2013). However, some reports indicated the inhibitory effects of dietary phytobiotics or plant extracts on *E. coli* activity *in vivo* (Abd El-Ghany and Ismail, 2014; Diaz-Sanchez *et al.*, 2015; Elmenawey *et al.*, 2019). Diet containing thyme essential oil helped for increasing in *Lactobacillus* and decreasing in *E. coli* counts in the intestine of Japanese

quail (Khaksar et al., 2012). Essential oils of oregano revealed antimicrobial properties on broiler carcasses through reduction of the total bacterial count especially *Salmonella* spp. (Aksit et al., 2006). Short chain fatty acids of phytobiotics revealed growth promoting effect on the intestinal beneficial microbiota as well as controlling the growth of *Salmonella enteritidis* (*S. enteritidis*) (Hansen et al., 1997). Quail's diet containing phytobiotic feed additives had significant ( $P \leq 0.05$ ) increase in the number of *Lactobacilli*, decrease in *E. coli* population and inhibit the growth of *Salmonella* in the intestinal tract (Dorman and Deans, 2000).

The efficacy of a mixture of seven essential oils that inhibited the growth of *Clostridium perfringens* (*C. perfringens*) *in vitro* has been reported by Si et al. (2009). In the field studies of Mitsch et al. (2004), Siragusa et al. (2008), McReynolds et al. (2009), Abudabos et al. (2018), El-Sheikh et al. (2018) and Hussein et al. (2020b), different phytobiotic feed additives caused reduction of intestinal colonization and proliferation of *C. perfringens*, lesion score and mortalities as well as improvement in performance of broilers and their carcass quality.

The reduction in *C. perfringens* virulence after supplementation with essential oil compounds may be related to the stimulation of some digestible enzymes like trypsin which inactivates  $\alpha$  toxin of type A and  $\beta$  toxin of type C strains of *C. perfringens*, stabilization of natural resident gut microflora like *Lactobacillus* spp. and consequently inhibition of the organism pathogenicity (Cho et al., 2014).

Moreover, essential oils of lemon, green tea and turmeric blend proved great efficacy in reducing the count of *S. enteritidis* and *Campylobacter jejuni* on the surface of chicken's carcass (Murali et al., 2012). The Eucalyptus volatile oils have been found to have the ability to relief broilers complicated respiratory distress caused by *Mycoplasma gallisepticum* (Abd El-Ghany, 2008).

#### **Antifungal and detoxifying effect**

Phytogetic compound have antifungal properties. Akgul and Kivanc (1988) reported about the inhibitory effects of some spices and oregano components on some foodborne fungi. Essential oil of marjoram reduced the *in vitro* growth of *Aspergillus flavus*, *A. niger*, *A. ochraceus* and *A. parasiticus* up to 89% (Deans and Waterman, 1993). The oil of lemon and orange can reduce the formation of *A. flavus* (Hasan et al., 2005). It has been demonstrated that *Allium sativum* has a great (60-80%) *in vitro* antifungal activity against *Aspergillus* and *Penicillium* spp. (Afzal et al., 2010). Onion, garlic and

ginger revealed antifungal activity against *A. flavus*, *A. niger* and *Cladosporium herbaru*, *Cinnamomum verum* and *Piper nigrum* which was studied with successful results by Tago et al. (2011). Phytobiotics like *Ocimum gratissimum*, *Cymbopogon citratus*, *Xylopi aethiopica*, *Monodora myristica*, *Syzygium aromaticum* proved their ability to inhibit the formation of non sorbic acid which is a precursor in the pathway of aflatoxin synthesis (Awuah, 1996). Neem extract represented inhibitory effects on biosynthesis of aflatoxins (B and G) (Bhatnagar et al., 1990). Also, neem leaf extract and its oil have been found to inhibit the growth and morphology of *Penicillium* spp. and consequently prevent the production of ochratoxin A (Bhatnagar et al., 1990).

#### **Anticoccidial effect**

The anticoccidial activity of some herbal plants have been documented (Willis et al., 2013). Phytogetic compounds have been found to reduce the severity of *Eimeria* spp. infection in broilers by alleviation of droppings score, intestinal lesions score and also reducing oocyst shedding (Zyan et al., 2017). Numerous phytobiotics as *Atemesia annua* (Allen et al., 1997), *Astragalus membranaceus* and *Sophora flavescens* (Youn and Noh, 2001), green tea (Jang et al., 2007), *Ageratum conyzoid* (Nweze and Obiwulu, 2009), *Musa paradisiaca* (Anosa and Okoro, 2010), olive tree (De Pablos et al., 2010), oregano essential oil (Tsinas et al., 2011), *Carica papaya* leaf extract (Nghonjuyi et al., 2015) and coconut oil (Hafeez et al., 2020) have indicated an excellent anticoccidial activity against different types of *Eimeria* spp. in birds.

Some essential oils presented similar drug efficacy in prevention and control of coccidiosis in broilers. The oregano oil and other mixtures of oils were similar to ionophores lasalocid (Giannenas et al., 2003) and monensin (Oviedo-Rondón et al., 2006), while a mixture of carvacrol, camphor, cineole and thymol was similar to salinomycin (Bozkurt et al., 2014) in terms of reduction of shedding and lesions of different *Eimeria* spp. in broilers.

Combined experimental infection of *C. perfringens* and *E. maxima* has been ameliorated after dietary treatment of three breeds of broilers with Capsicum, *Curcuma longa* oleoresins (Kim et al., 2015) and *Allium hookeri* root (Lee et al., 2018).

#### **Immunomodulatory effect**

In poultry production, reduction of infection as well as improvement of production by stimulation of the immune system after using phytogetic substances were

investigated previously (Zaki *et al.*, 2016). The immunomodulatory mechanism of phytoactive substances in poultry have been studied (Hashemi and Davoodi, 2012). Polysaccharide are very important immunoactive components of phytobiotics (Xue and Meng, 1996). Phytoactive compounds also induce their immunomodulatory effects through increasing immune cells proliferation, arising cytokines expression and elevation of antibody titers (Lee *et al.*, 2010; Park *et al.*, 2011; Pourhossein *et al.*, 2015). The immunogenicity of phytobiotics could be manifested as increasing macrophages, lymphocytes and natural killer cells activities as well as stimulation of interferon production (Hashemi and Davoodi, 2010; Kumar *et al.*, 2014). Plants contain flavonoids, vitamin C and carotenoids are able to enhance the immune system (Craig, 1999). Some herbs and species like garlic, echinacea and liquoric have immunostimulatory properties due to their composition of vitamin C, carotenoids and flavonoids as well as their abilities to stimulate macrophages, lymphocytes and natural killer cells activities and interferon production (Frankic *et al.*, 2009). Detecting *in vitro* immunostimulatory effect of dandelion, mustard and safflower either on lymphocytes and macrophages of chickens was performed by Lee *et al.* 2007. The results indicated inhibition of tumor cell growth, antioxidant effects, stimulation of lymphocyte proliferation and nitric oxide production by macrophages. The immunostimulant effect of some essential oils extracts of phytobiotics may be due to the presence of certain compounds that may bind to Immunoglobulin G (Ig G) receptors which led to stimulation of immune response (Ahmed *et al.*, 2013). In the study of Placha *et al.* (2014), inoculation of thymol in the broiler diet increased the trans-epithelial electrical resistance of duodenal mucosa.

It has been found that mushroom and plant polysaccharides have immunomodulatory effects in chickens infested with *Eimeria tenella* (Guo *et al.*, 2004).

Potential antiviral activity of plant seeds was recorded (Yaseen, 2003). Different types of herbs mix, spices, plant extracts and essential oils presented enhancement of immune response of birds (Huang *et al.*, 2007; Pourali *et al.*, 2010; Kavyani *et al.*, 2012; Abou-Elkhair *et al.*, 2014; Awaad *et al.*, 2016).

A significant ( $P < 0.05$ ) elevation of antibody Enzyme-linked immunosorbent assay (ELISA) titer after vaccination with Newcastle Disease Virus (NDV) vaccine (Chowdhury *et al.*, 2018) was recorded after feeding on clove bud and cinnamon plant, while peppermint essential oil helped in significant ( $P < 0.05$ ) rising in

Haemagglutination Inhibition (HI) antibody titer against Avian Influenza (AI) virus vaccines (H9N2) in broiler chicks (Sultan *et al.*, 2017). In addition, significant ( $P < 0.05$ ) increase in HI titers against NDV vaccine and sheep red blood cells were observed in broiler chicks treated with Aloe vera gel (1%) in the drinking water for six weeks (Darabighane *et al.*, 2017)

Laying hens presented significant ( $P > 0.05$ ) increase in ELISA titer after vaccination with ND, Infectious Bronchitis (IB) and Infectious Bursal Disease (IBD) vaccines (Özek *et al.*, 2011). Landy *et al.* (2011) observed that inoculation of broiler ration with neem powder for six weeks resulting in an elevation of HI antibody titer against AI vaccine, but not against ND vaccine. Barbour *et al.* (2008) evaluated the effect of using Eucalyptus and peppermint essential oils during vaccination of ND and IBD. The results indicated improved health conditions of the birds associated with elevation of ELISA titers against the used virus vaccines. Moreover, an increase in IgG and IgM ELISA titers has been observed in chickens fed on oregano essential oils for three weeks period (Malayoglu *et al.*, 2010). Recently, Abdelnaser *et al.* (2019) indicated that treatment with essential oils of oregano, anise, and citrus peel at level of 125 gm/ton induced positive effects on the immune response of *C. perfringens* challenged broilers after vaccination against ND, IB, AI and IBD viruses as well as increasing in relative spleen weight.

## CONCLUSION

Using of phytobiotics in poultry diet as a feed additive and considering them as an antimicrobial substitute has become a very essential and critical issue currently. Phytobiotic compounds could replace antibiotic growth promoters to improve the productive performance of chickens, act as antibacterial, antifungal and antiparasitic agents as well as it has been considered as potential immunostimulants especially after routine vaccination programs of chickens. This review spotlight on the significant using of phytobiotics in poultry field and industry as natural antibiotic alternatives to avoid the emerged problem of antibiotic resistance,

### Competing interests

The author have not declared any conflict of interest.

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