Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
www.entomoljournal.com
JEZS 2020; 8(4): 1643-1649
© 2020 JEZS
Received: 14-05-2020
Accepted: 16-06-2020

Sonali Namdeo
M.V.Sc. Scholar, NDVSU, Jabalpur, Madhya Pradesh, India

RPS Baghel
Dean Faculty, NDVSU, Jabalpur, Madhya Pradesh, India

Sunil Nayak
Professor & Head, NDVSU, Jabalpur, Madhya Pradesh, India

Ankur Khare
Assistant Professor, NDVSU, Jabalpur, Madhya Pradesh, India

Ravi Prakash Pal
Teaching Associate, NDVSU, Jabalpur, Madhya Pradesh, India

Amit Chaurasiya
Ph.D. Scholar Jabalpur, Madhya Pradesh, India

Sahil Thakur
M.V.Sc. Scholar, NDVSU, Jabalpur, Madhya Pradesh, India

BVV Reddy
M.V.Sc. Scholar, NDVSU, Jabalpur, Madhya Pradesh, India

Corresponding Author:
Sonali Namdeo
M.V.Sc. Scholar, NDVSU, Jabalpur, Madhya Pradesh, India

Essential oils: an potential substitute to antibiotics growth promoter in broiler diet

Sonali Namdeo, RPS Baghel, Sunil Nayak, Ankur Khare, Ravi Prakash Pal, Amit Chaurasiya, Sahil Thakur and BVV Reddy

Abstract
Essential oil (EO) is a complex mixture of fragrant volatile compounds obtained from plants and isolated through various methods like fermentation, extraction and steam distillation. Essential oils consist of two classes of compounds, i.e. terpenes and phenylpropenes. Differences between essential oils mainly depend on several variables, such as plant species, physical and chemical soil conditions, harvest time, degree of plant maturity, technology of drying, duration of storage and extraction processes. In broiler production, EO has potential for possible therapeutic exploitation in variety of conditions. These have antimicrobial, antifungal, antiparasitic, and antiviral properties. Besides, other beneficial effects of EOs include appetite stimulation, improvement of enzyme secretion related to food digestion, and immune response activation. Along with acting as an antioxidant for animal it also extends storage time of feed and meat in which it is incorporated due to its antioxidant property. Hence, many researches have been carried out across the world with possible cocktail and combinations of these essential oils or crude extracts of their bioactive principles to explore the multifaceted eminence of these feed additives. This review briefly elucidates the role of essential oils on the development of poultry industry.

Keywords: Antibiotic, antioxidant, broiler, essential oils, growth

Introduction
Antibiotic at sub-therapeutic levels has been utilized at a very large extent in broiler industry to improve growth performance as well as to decrease morbidity and mortality. Due to residues in poultry products and cross resistance against pathogens antibiotics are banned by European Union. So the alternate strategy is required to enhance the growth performance without any resistance of antibiotic in poultry and residues in meat. Phytogenic feed supplements like the essential oils (EO), are suitable alternatives in broilers production for improving growth performance and the characteristic properties of meat products [1]. Essential oils are composite mixture of fragrant volatile compounds derived from plants and isolated through various methods like fermentation, extraction and steam distillation. As they are mixture of volatiles, there effect is sum of all the components and their interaction. Differences between essential oils, mainly depend on several variables, such as plant genotype, physical and chemical soil conditions, harvest time, degree of plant maturity, technology of drying, duration of storage and extraction processes [2]. Source of variability also, depend on type and origin of the EO, level of EO that is incorporated in poultry diet, composition and digestibility of basal diet, level of feed intake, hygiene and environmental conditions [3].
In broiler production, EO has potential for viable restorative exploitation in variety of conditions. They enhance production performance by stimulating various digestive enzymes, alleviate the amount of fermentation products, lowers the level of pathogenic microorganism, amplify precaecal nutrient digestion, ameliorate intestinal availability of vital nutrients for absorption, exert antioxidant status and immune responses, hypocholesterolemic and act as coccidiostat, Some of the major essential oils used in broiler diet are anise, oregano, cinnamon, garlic, thyme and turmeric.
Essential oils are categorised into two classes, i.e. terpenes and phenyl propenes [4]. Depending on the number of isoprene units (5-carbon building blocks), terpenes are further sub-divided into mono-, sesqui-, and di-terpenes with 2, 3 and 4 isoprene units respectively. Phenylpropenes consist of six carbon aromatic ring with three carbon side chain [5].

Biological effects of essential oils

Eos are potent digestive stimulant, growth promoter, hypolipidemic agent, immunomodulator, antioxidant, anti–microbial and antiparasitic agents.

Effect of EOs on growth performance in chicken

Studies on chicken indicated that inclusion of essential oils in diet improves their growth performance as it stimulates secretion of digestive enzymes resulting into improved nutrient digestion, rate of passage of digesta [6]. Further, its observed that incorporation of EOs in animal potentially minimise occurrence of intestinal diseases caused by undesirable bacteria and thus favor beneficial gut microbiota growth which support growth performance [7].

Factors such as temperature, viscosity, visual appearance, saliva production, nutritive value of feed, particle size, toxicity of feed components and social interaction influence feed intake [8]. Many studies confirmed the beneficial effect of feeding essential oils on intake in poultry. Elevated feed intake was observed when broiler diet supplemented with EO mixture containing clove, oregano and anise@ 200 and 400 mg/kg for 3 weeks. Presence of bioactive compounds like thymol, anetole, eugenol and carvacrol in respective EO are responsible for appetizing effect which ultimately increases feed intake [9]. Similarly, improved overall feed intake in broilers fed 18 mg/kg each of cinnamon, oregano, thyme and eucalyptus EO was observed due to balancing of gut microbiota [10]. Further researchers studied the effects of oregano essential oil on broilers growth performance for a period of fourty two days. Dietary treatments were as follows: basal diet, basal diet + 8 mg/kg avilamycin (AVI), basal diet + 300 mg/kg oregano essential oil (OEO300), and basal diet + 600 mg/kg oregano essential oil (OEO600). They concluded that on 42nd day of feeding, group's supplemented with 300 mg/kg and 600 mg/kg oregano essential oil showed significant increase ($p<0.05$) in average daily feed intake and average daily gain as compared to the control group [11].

Body weight gain increases on essential oil supplementation, this is associated with its antimicrobial action and stimulation of various digestive enzymes which improve utilizations of nutrients [12]. Live weight gain of poultry increased in 200 ppm EO supplemented group by approximately 8% and 16% over antibiotic group and control group respectively. Thus showing growth enhancing property of EO in poultry [9].

Feed conversion ratio is the efficiency to convert feed mass to body mass over a specific period of time. Cinnamon powder which contains cinnamaldehyde helps in improvement of FCR in broilers [12] and they also revealed the favourable effect of diet fed with cinnamaldehyde over harmful bacteria in gut by decreasing their number which ultimately leads to higher digestibility of nutrients. Also, supplementation of essential oil mixture (EOM) @ 200 ppm significantly improved FCR by 6% and 12% over the antibiotic group and the control group respectively in broilers feed [9]. Likewise, it is reported that broilers fed EO (myrtle leaf oil, oregano oil, sage leaf oil, laurel leaf oil, citrus peel oil and fennel seed oil) showed significant improvement in FCR [13]. Recent studies [14] showed that supplementation of EO (thyme extract) in broiler diet increased secretion of digestive enzymes i.e. amylase and chymotrypsin which results into increased rate of absorption in intestine and consequently improves feed utilization.

Essential oil effect on feed intake, body weight gain, and feed conversion ratio

Source of EO	Dose	Feed intake	Body wt. gain	FCR	References
Clove	600mg/kg	Improved	Improved	Improved	[15]
EO blend (clove, cinnamon)	300mg/kg	Improved	Improved	Improved	[16]
EO blend (thyme, carvacol)	200mg/kg	No Effect	Improved	Improved	[17]
Cinnamon	5%	Improved	Improved	Improved	[18]
Cinnamon	1%	Improved	Improved	Improved	[19]
Clove	450 ppm	Improved	Improved	Improved	[20]
Cinnamon	200 ppm	Improved	Improved	Improved	[21]
EO blend (cinnamon, turmeric)	0.50%	Improved	Improved	Improved	[22]
Thyme	1g/kg	Improved	Improved	Improved	[23]
Clove	1%	Improved	Improved	Improved	[24]

EO inclusion in broiler diet decrease the growth of undesirable bacteria and enhances beneficial gut microbiota growth, which ultimately improve the growth performance of broilers as shown by [25], they revealed that supplementation of blend of essential oils (EO) consisting of 5 g/tone cinnamaldehyde and 15 g/ton thymol on the performance of broilers. Similar study was also conducted by [26] who revealed the effect of mixture of essential oils from caraway, basil, lemon, laurel, sage, oregano, thyme and tea on growth performance of broiler. Also, birds allocated Tecnaroma Herbal Mix at 0,,100, 200,.300, 400, and 500 g/t of feed showed significantly ($p<0.05$) increased ADG and heavier body weight and had improved ($p< 0.05$) FCR compared to the control group. Likewise, significantly improved body weight was observed in broilers supplemented with essential oil blend at dose of 300 and 600 g/kg of feed [11].

Antioxidant activity of essential oils

Antimicrobial mechanisms of EO includes various activities, such as metal chelation by flavonoids and phenols, disruption of membrane by phenolics and terpenoids and damage of genetic material by alkaloids and coumarin that possibly impede growth of microorganisms [27]. Essential oils are hydrophobic in nature; this enables it to partition lipids in bacterial cell wall and mitochondria thus accumulate in lipid layer, disrupting cell membrane integrity and ion transport which results into subsequent loss of cellular components and lysis afterwards. Moreover, it also cause acidification inside cell which block production of cellular energy, collapse proton pump and reduce membrane potential [28].

As compared to gram negative bacteria EOs are more active against gram positive bacteria [28] as EO are lipophillic in nature. However, molecules of EO with low molecular weight results in membrane disruption by either crossing cell wall of bacteria either through lipopolysaccharides layer or by diffusion through membrane proteins [29]. The effects of garlic and oregano essential oils (OEO) on intestinal microflora of broilers. Four diets were formulated: basal diet, basal diet+

OEO at 300 mg/kg, basal diet+ garlic essential oil at 300 mg/kg and basal diet + oregano and garlic essential oil at 150 mg/kg. Total organism i.e. *Streptococcus, Lactobacillus spp.,* and *Coliform* counts in the ileum were unaffected by treatments however a significant ($p< 0.05$) decrease in Clostridium count was observed in birds supplemented with garlic oil, oregano oil and garlic oil + oregano oil compared to control [30].

EOs such as cinnamaldehyde and thymol have selective antibacterial properties and also inhibit yeast and fungi growth [7]. It beneficially affect the ecosystem of gastrointestinal microflora by controlling potential pathogens, alleviating the oxidative stress caused by them and stabilizing gut microbiota. Result of experiment convoyed by [31] is also in agreement with above findings. They revealed the effect of 2 herbal extracts and virginiamycin on intestinal bacterial population in broilers. Four diets were formulated as: basal diet, basal diet + 15 ppm of virginiamycin, basal diets + 0.1% dose of either garlic, thyme or blend of 2. Ileo-cecum digesta of supplemented groups showed a significant reduction in colony forming units of *E. coli* as compared with control, however a significant increase in lactic acid bacteria count is observed in the thyme group as compared to other groups. Similarly, supplementation of cinnamon oil (CNO) revealed detrimental changes in cell wall of treated *E. coli* [32].

An experimental trial [33] was conducted to study the effect of ajwain oil (AJO), clove oil (CLO) and Cinnamon oil (CNO) @ 400, 600, 300 mg/kg of diet respectively was studied as an substitute to antibiotic to estimate their effect on intestine of broiler chickens. The author found that the counts of *E. coli* in pre-ceacal contents reduced in antibiotic and CNO group and the count of *Clostridium* spp. decreased in the antibiotic group compared with the control. However, population of lactobacilli spp. was not affected by any of the diet.

Effect of essential oils on intestinal morphology

First tissue that comes in contact with dietary constituents is the gastrointestinal mucosa. It is acknowledged that villus height/crypt depth ratio is a important criterion for evaluating the digestive capacity of small intestine and a decrease in this ratio is not acceptable in terms of digestion and absorption, and vice versa [34]. The introduction of feed antibiotics into diets of chickens leads to decreased thickness of walls, reduced weight and increased length of intestine [35]. Increased villus height due to supplementation of EO has been reported by several scientists [36, 37]. The essential oil induced increased villus height might be correlated to their antioxidant property [38]. Oxygen radicals released during digestive process attack superficial mucous of intestine and can shorten intestine villi. Antioxidant enzymes like catalase, glutathione peroxidase and superoxide dismutase can attract the oxygen radicals and quench the free radicals. Components like cinnamaldehyde present in EO prevent oxidative damage of villi by enhancing the activity of antioxidant enzymes, also the phenolic group in the cinnamon EO act as a hydrogen donor [38].

Thymol supplementation increased intestine length as well as width and depth of the villi, thus creating better conditions for absorption of nutrients [39]. Similarly, combination of carvacol (5.4%), cinnamaldehyde (2.9%), capsicum oleoresin (2.18%) showed increased villus length and intestinal diameter in the treated group in comparison to other birds ($P<0.05$) [40]. Furthermore, studies showed that feeding of leaves of *M. piperita* partially improves histomorphological structure of mucosa of small intestine in broiler chickens [41]. Also, [33] revealed that supplementation of cinnamon oil @0.3mg/gm of diet results in increased height of villi in duodenum, jejunum and ileum as compared to control group.

Effect of essential oil on antioxidant activity

Oxidation of lipids (lipid peroxidation) and free radical production are natural processes that destroy the membrane structure, disturb transport processes and cause cell organelles dysfunctioning. In cell membrane phospholipids are particularly more susceptible to damage due to oxidation that corresponds with the degree of unsaturation of fatty acids. Polyunsaturated fatty acids (PUFAS) are responsible for aliementation of major cell membrane properties including permeability and fluidity. [42]. In order to retard effect of lipid peroxidation by scavenging peroxyl radical, synthetic antioxidants (butylated hydroxyl anisole, butylated hydroxyl toluene, tert-butyl hydroquino, gallates etc.) are used but their use has come into question due to their carcinogenic potential. All this resulted into increased inclination of consumer for natural antioxidant sources. Due to their chemical structure and high redox propertie. Eos have ability to neutralize free radicals, quench singlet and triplet oxygen and chelate transitional metals. Also, theyare rich source of natural antioxidants, such as the phenolic compounds [32]. However, basedon dosage andtype, EO can act as prooxidants by deranging cell and organelle integrity, with further cytotoxic effects on living cells [2]. Also, EOs exert beneficial effects on antioxidant enzymes (superoxide dismutase, glutathione peroxidase and catalase) and prevent production of reactive oxygen species (ROS) and off-flavors derived from polyunsaturated fatty acids oxidation [43, 44]. Thus, dietary supplementation with EO is highly effective in retarding lipid oxidation of animal products compared to postmortem addition [45].

In several studies a significant improvement of antioxidant activity have been observed on EO supplementation in poultry. On supplementing equal mixture of carvacol and thymol to broilers @ 0, 60, 100 and 200 mg/kg, it was reported that SOD activity linearly increased ($P<0.05$) [17]. Research showed that on supplementation ofginger essential oil @150 mg/kg, increases total superoxide dismutase (TSOD) activity and decreases malondialdehyde (MDA) concentrations in liver which could be due to presence of several antioxidant compounds such as shogaol, gingerol, zingerone and diaryl heptanoids in ginger root[46]. Likewise, Chowdhury *et al*. (2018) [33] also showed that cinnamon bark oil (300 mg/kg diet) as an substitute to antibiotic (50 mg/kg diet) has improved antioxidant status in broilers as superoxide dismutase activity was increased significantly in cinnamon bark oil supplemented birds as compared with the antibiotic group.

Effect of essential oils on lipid metabolism

Supplementation of EO can decrease cholesterol level. Bioactive compounds present in essential oils such as cineole, borneol, citral, menthone, geraniol, menthol, fenchyl alcohol, fenchone & ionone are important to inhibit HMG-CoA (Hepatic 3-hydroxy-3-methylglutaryl coenzyme a) reductase activity [47], a key regulatory enzyme in synthesis of cholesterol. Study conducted by [48] observed 2% reduction in serum cholesterol on inhibition of 5% HMG-CoA reductase in poultry. Correlation exist between LDL or total cholesterol and HMG-CoA reductase activity in chicken, but not with HDL cholesterol [49].

A study was conducted to know the potential of essential oil as growth promoting agents and as a potential hypocholesterolemic agent. They had selected broilers which are 240 day-old and divided them into 3 equal parts which are as follows: antibiotic or positive control (100 ppm oxytetracycline), negative control (no antibiotic or essential oil) and essential oil (125 ppm which includes essential oil originated from anis, oregano and citrus peel).The results showed reduced serum levels of cholesterol by both the treatment with antibiotic or essential oil, although essential oil supplementation received by birds found to have higher total polyphenolic compounds, lower VLDL levels and higher total flavonoids [50]. Recently, a study conducted by Chowdhury *et al.* (2018) [33] revealed that in broiler chickens significant reduction of serum cholesterol level can be achieved by supplementing them with cinnamon essential oil at 300mg/kg. Essential oil down regulate HMG-CoA reductase enzyme post-transcriptionally without altering mRNA levels of the enzyme [51]. The inhibitory action of essential oils on hepatic HMG-CoA reductase is independent of the diurnal cycle of the enzyme, and of hormones such as glucocorticoids, insulin, glucagon and triiodothyronine [52]. The complete inhibition of cholesterol synthesis requires two regulators, i.e. Cholesterol derived from LDL and a non-sterol products derived from mevalonate, both of which modulate HMG-CoA reductase activity [53]. Thymol, ionone and carvacrol might induce a putative regulatory non-sterol product [54].

Immunomodulatory activity of essential oils

Several researchers have examined the immunomodulatory potential of various EO. It wasreported that incorporation of garlic essential oil in chicken diet significantly increased the antibody production against *Pasteurella multocida*, *Leptospira Pomona* and *Salmonella enteritidis* [55]. Study conducted by [56] revealed that garlic has immunomodulating effect as it enhances production of interferon (INF-γ), interleukins, tumor necrosis factor (TNF-α). Also, it increases phagocytosis of antigen presenting cells (APC) and macrophages. Another study by [31] showed that relative weight of bursa of fabricius in the garlic supplemented group significantly ($p<0.05$) increased in compared to other groups, however no significant effect was seen on relative weight of spleen. Awaad *et al*. (2010) [57] studied immunostimulant effects of eucalyptus and peppermint essential oils on humoral and / or cell mediated immunity in birds vaccinated against Avian Influenza (AI) and Newcastle disease (ND). Volatile oils treated group showed higher HI titers against both AI and ND vaccines as compared to control. Specific antibody response against influenza vaccine virus showed significant increase due to supplementation of 0.2% thyme extract [58].

Yang *et al*. (2018) [59] conducted an experiment to study the the effect of feeding thymol essential oil (EOA) on immunity of broiler chickens. Dietary treatments were as follows: base diet (CON) or diets with 0.15 g/kg enramycin during the grower period (AG), or 0.30 g/kg EOA during the grower period (EG), or 0.30 g/kg EOA during the finisher period (EF). They concluded that birds of grower and finisher phase fed 0.30g/kg essential oil showed a higher ($p< 0.05$) spleen index than birds of control group. In duodenum and ileum mucosa level of secretory immunoglobulin A significantly increased ($p<0.05$) in finisher group in comparison to other groups.

Anticoccidial effect of essential oils

EOs are potent botanical products which either interfere directly or indirectly with parasitic metabolism by amplyfing host immunity and antioxidant status for the effective control and eradication of parasitic invasion, especially for controlling coccidiosis [60]. Direct anticoccidial actions include inhibition of glycoprotein synthesis [61], ultra-structural changes in mitochondrial membrane [62] and cysteine protease enzyme inhibition [63]. The indirect antiparasitic actions of essential oil include enhancing immunity of host [60] and show antioxidant effect by scavenging of reactive oxygen species [2]. Plants such as *Allium cepa, Origanumvulgare,* [64] *Allium sativum, Echinacea purpure,* [65] *and Chenopodium ambrosioides, Mentha spp.* are efficacious against intestinal parasites including the *Eimeria* species. Giannenas *et al*. (2004) [66] revealed that on feeding oregano essential oil (300 mg/kg) in experimentally infected (*Eimeria tenella)* birds resulted into reduced number of oocysts per gram of excreta.

The EO acquired from *O. Vulgare* have widely been investigated for anticoccidial activity against *E. Tenella,* [67] mixed *Eimeria* spp. Infection [68], *E. Maxima* [69] and *E. Acervulina.* For controlling avian coccidiosis supplementation EOs along with coccidial vaccination have been reported as an effective alternative by various researchers [70]. Moreover, many researchers have shown the oocysticidal activity of phenols against *E. Tenella* [71]. For estimating level of coccidial infection parameter which is generally considered is number of *Eimeria* oocysts shedding in the birds excreta. Bioactive components [66] present in essential oils [70] shown to have constructive effect on reduction of faecal oocyst shedding and also minimise damage of intestinal epithelium in affected birds [72].

Effect of essential oils on carcass traits

Improvement in production of poultry meat depends mainly on the increase in muscle proportion, the enhancements in growth and reduction in abdominal fat [73]. Alcicek *et al.* (2003) [39] found that there is improvement in the carcass yeild with the presence of an essential oil in combination of 48 mg/kg and 72 mg/kg. Khattak *et al.* (2014) [26] found that there is increase in carcass weight, breast weight and breast meat with the supplementation of a natural blend of Eo (essential oil) (basil, lemon, caraway, oregano, laurel, sage, thyme and tea). These results indicated that there are beneficial effect on carcass traits of broilers with dietary supplementation of essential oils. However, dietary supplementation of OEO @ 300 mg/kg showed non-significant effect on characteristics of carcass [74].

Sang-oh *et al*. (2013) [75] also noted that the dressing percentage and the thigh and breast muscle were significantly ($P<0.05$) higher in the cinnamon powder groups. Eltazi (2014) [18] studied the effect of cinnamon powder in broiler chicks where graded levels of this powder was added (0,3,5 and 7%) along with basal diet and showed that there is highest dressing percentage and commercial cuts (breast, drumstick and thigh) in the diet with 5% cinnamon powder. Likewise, oregano essential oil supplementation @ 600mg/kg exhibited high breast muscle percentage along with increased dressing percentage, eviscerated rate, and leg muscle percentage as compared to the control [11].

Effect of essential oils on economics of broiler production

The broiler production economics have been found variable on incorporation of Eos in diet. Recently, Wade *et al.* (2018) [14] conducted his study by forming 4 different treatment group of feed named as A, B, C, D where group A control group, group B supplemented with thyme essential oil, 100mg/kg diet, group C supplemented with thyme essential oil,

200mg/kg diet and group D supplemented with thyme essential oil, 300mg/kg diet. And found out that the feed supplemented with thyme essential oil at 100mg/kg resulted in significantly higher body weight gain, improved feed conversion ratio, livability and profit in broiler production. Finally it concluded that the highest net profit per kg live weight is observed in group B Rs.13.30, followed by group C Rs.11.22, then group D Rs.10.71 and least in group A Rs.9.94.

Conclusion

The beneficial effects of Eos is recognised since ancient times and their properties reported in foods and experimental animals. Ability of essential oil to enhance flavour and stimulating various digestive process is of great importance to improve poultry performance, however, further studies required in this area. Few properties of these oils like antioxidant, hypolipidemic and immunomodulatory are gaining more interest among the poultry industrialists. During the elongated low temperature storage, these oils with their antioxidant property provides effective shielding against drip loss which increased its acceptance among the consumers and reduces the loss for the meat processors. Thus, to enhance the growth performance in the poultry diet essential oil could be used as replacement to antibiotics and it can also helps in creating value added products with low cholesterol meat, tenderness etc. Its also gaining prominence as it helps in improving the keeping quality, durability of both raw and processed meat and nutraceutical property. In the near future, it is expected that essential oils will have a prodigious role in the poultry industry development.

References

1. Simitzis PE, Deligeorgis SG. The effects of natural antioxidants dietary supplementation on the properties of farm animal products.Journal of Nutrition. 2011; 40:155-168.
2. Bakkali F, Averbeck S, Averbeck D, Idaomar M. Biological effects of essential oils-A review. Food Chemistry Toxicology. 2008; 46:446-475.
3. Brenes A, Roura E. Essential oils in poultry nutrition: Main effects and modes of action. Animal Feed Science Technology. 2010; 158:1-14.
4. Lee KW, Everts H, Beynen AC. Essential oils in Broiler Nutrition. International Journal of Poultry science. 2004; 3(12):738-752.
5. Gopi M, Karthik K, Manjunathachar HV, Tamilmahan P, Kesavan M, Dashprakash M *et al*. Essential oils as a feed additive in poultry Nutrition. Advance in Animal Veterinary Sciences. 2014; 2(1):1-7.
6. Jamroz D, Wiliczkiewicz A, Wertelecki T, Orda J, Skorupinska J. Use of active substances of plant origin in chicken diets based on maize and locally grown cereals. British Poultry Science. 2005; 46:485-493.
7. Bento M, Ouwehand AC, Tiihonen KS, Nurminen P, Saarinen MT, Schulze H *et al*. Essential oils and their use in animal feeds for monogastric animals- Effects on feed quality, gut microbiota, growth performance and food safety: a review. Veterinarni Medicina. 2013; 58(9): 449-458.
8. Blair R. Effect of dietary thyme oil and vitamin E on growth, lipid oxidation, meat fatty acid composition and serum lipoproteins of broilers. South African Journal of Animal Science. 2008; 36:189-196.
9. Ertas ON, Guler T, Ciftci M, DalkIlIc B, Simsek UG. The effect of an essential oil mix derived from oregano, clove and anise on broiler performance. International Journal of Poultry Science. 2005; 4(11):879-884.
10. Ulfah M. Essential oils as multi-function feed additive (MFA) to improve broilers performance, metabolism, dung consistency and efficiency of production. Journal of Agriculture Rural Development. 2006; 88:50-55.
11. Peng QY, Li JD, Li Z, Duan ZY, Wu YP. Effects of dietary supplementation with oregano essential oil on growth performance, carcass traits and jejunal morphology in broiler chickens. Animal Feed Science and Technology. 2016; 214:148-153.
12. Hernandez F, Madrid J, Garcia V, Orengo J, Megias M. Influence of two plant extractson broilers performance, digestibility, and digestive organ size. Poultry Science Journal. 2004; 83:169-175.
13. Cabuk M, Bozkurt M, Alcicek A, Akbas Y, Kucukyılmaz K. Effect of a herbal essential oil mixture on growth and internal organ weight of broilers from young and old breeder flocks. South African Journal of Animal Science. 2006; 36:135-141.
14. Wade MR, Manwar SJ, Kuralkar SV, Waghmare SP, Ingle VC, Hajare SW. Effect of thyme essential oil on performance of broiler chicken. Journal of Entomolology and Zoology. 2018; 6(3):25-28.
15. Mukhtar MA. The effect of dietary clove oil on broiler performance. Australian Journal of Basic and Applied Sciences. 2011; 5(7):49-51.
16. Chalghoumi R, Belgacem A, Trabelsi I, Bouatour Y, Bergaoui R. Effect of dietary supplementation with probiotic or essential oils on growth performance of broiler chickens. International Journal of Poultry Science. 2013; 12(9):538-544.
17. Hashemipour H, Kermanshahi H, Golian A, Veldkamp T. Effect of thymol and carvacrol feed supplementation on performance, antioxidant enzyme activities, fatty acid composition, digestive enzyme activities, and immune response in broiler chickens. Poultry science. 2013; 92(8):2059-2069.
18. Eltazi S. Effect of using cinnamon powder as natural feed additive on performance and carcass quality of broiler chicks. International Journal of Innovative Agriculture and Biology Research. 2014; 2(3):1-8.
19. Shirzadegan K. Reactions of modern broiler chickens to administration of cinnamon powder in the diet. Iranian Journal of Applied Animal Science. 2014; 4(2):367-371.
20. Mehr MA, Hassanabadi A, Moghaddam HN, Kermanshahi H. Supplementation of clove essential oils and probiotic to the broiler's diet on performance, carcass traits and blood components. Iranian Journal of Applied Animal Science. 2014; 4(1):117-122.
21. Qotbi AAA. The Effect of cinnamon powder and cinnamon extract on performance, blood parameters and microbial population of broiler chicks. Journal of University of Babylon. 2016; 24(9):2654-2660.
22. Kanani PB, Daneshyar M, Najafi R. Effects of cinnamon (*Cinnamomum zeylanicum*) and turmeric (Curcuma longa) powders on performance, enzyme activity, and blood parameters of broiler chickens under heat stress. Poultry Science Journal, 2016; 4(1):47-53.
23. Attia YA, Bakhashwain AA, Bertu NK. Thyme oil (*Thyme vulgaris*) as a natural growth promoter for broiler chickens reared under hot climate. Italian Journal of

Animal Science. 2017; 16(2):275-282.
24. AL-Tabari AS, AL-Zuhairi ZA, Abdulrazzaq M. Study the effect of adding aqueous extract of clove (*Eugenia caryophyllus*) to drinking water in productivity and physiological efficiency of broiler chicken. Basrah Journal of Veterinary Research. 2018; 17(1):165-175.
25. Tiihonen K, Kettunen H, Bento MHL, Saarinen M, Lahtinen S, Ouwehand AC *et al*. The effect of feeding essential oils on broiler performance and gut microbiota. British poultry science. 2010; 51(3):381-392.
26. Khattak F, Ronchi A, Castelli P, Sparks N. Effects of natural blend of essential oil on growth performance, blood biochemistry, cecal morphology, and carcass quality of broiler chickens. Poultry science. 2013; 93(1):132-137.
27. Cowan MM. Plant products as antimicrobial agents. Clinical Microbiology.1999; 12:564-582.
28. Burt S. Essential oils: Their antibacterial properties and potential applications in foods-A review. International Journal of Food Microbiology. 2004; 94:223-253.
29. Dorman HJD, Deans SG. Antimicrobial agents from plants: Antibacterial activity of plant volatile oils. Journal of Applied Microbiology. 2000; 88:308-316.
30. Kirkpinar F, Unlu HB, Ozdemir G. Effects of oregano and garlic essential oils on performance, carcass, organ and blood characteristics and intestinal microflora of broilers. Livestock Science. 2011; 25:137-219.
31. Zheng W, Wang SY. Antioxidant activity and phenolic compounds in selected herbs. Journal of Agriculture Food Chemistry. 2001; 49:5165-5170.
32. Rahimi S, Zadeh ZT, Torshizi M, Omidbaigi R, Rokni H. Effect of the three herbal extracts on growth performance, immune system, blood factors and intestinal selected bacterial population in broiler chickens. Journal of Agriculture Science Technology. 2011; 13:527-530.
33. Chowdhury S, Mandal GP, Patra AK, Kumar P, Samanta I, Pradhan S *et al*. Different essential oils in diets of broiler chickens. Animal Feed Science and Technology. 2018; 36:39-47.
34. Pluske JR, Hampson DJ, Williams IH. Factors influencing the structure and function of the small intestine in the weaned pig - a review. Livestock Production Science. 1997; 51:215-236.
35. Hedemann MS, Hojsgaard S, Jensen BB. Small intestine morphology and activity of intestinal peptidases in piglets around weaning. Journal of Animal Physiology and Animal Nutrition. 2003; 87:32-41.
36. Garcia V, Gregori P, Fernandez F, Megias MD, Madrid J. Effect of formic acid and plant extracts on growth, nutrient digestibility, intestine mucosa morphology and meat yield of broilers. Journal of Applied Poultry Research. 2007; 16:555-562.
37. Samanta S, Haldar S, Ghosh TK. Comparative efficacy of an organic acid blend and bacitracin disalicylate as growth promoters in broiler chickens: effects on performance, gut histology and small intestinal milieu. Veterinary Medicine International. 2009; 4:651-654.
38. EI-Baroty GS, El-Baky HH, Farag RS, Saleh MA. Characterization of antioxidant and antimicrobial compounds of cinnamon and ginger essential oils. African Journal of Biochemistry. 2010; 4:167-174.
39. Alcicek AHMET, Bozkurt M, Cabuk M. The effect of an essential oil combination derived from selected herbs growing wild in Turkey on broiler performance. South African Journal of Animal Science. 2003; 33(2):89-94.
40. Awaad MHH, Elmenawey M, Ahmed KA. Effect of a specific combination of carvacrol, cinnamaldehyde and on the growth performance, carcass quality and gut integrity of broiler chickens. Veterinary World. 2014; 7(5):200-210.
41. Hamedi S, Shomali T, Ghaderi H. Effect of dietary inclusion of *Mentha piperita* L. on histological and histomorphometrical parameters of the small intestine in broiler chickens. Organic Agriculture. 2017; 7(2):105-110.
42. Pisoschi AM, Pop A. The role of antioxidants in the chemistry of oxidative stress: A review. Europian Journal of Medical Chemistry. 2015; 97:55-74.
43. Marcincak S, Cabadaj R, Popelka P, Soltysova L. Antioxidative effect of oregano supplemented to broilers on oxidative stability of poultry meat. Slovinian Veterinary Research. 2008; 45(2):61-66.
44. Miguel MG. Antioxidant and anti-inflammatory activities of essential oils: A short review. Molecules. 2010; 15:9252-9287.
45. Decker EA, Park Y. Healthier meat products as functional foods. Journal of Meat Science, 2010; 86:49-55.
46. Habibi R, Sadeghi GH, Karimi A. Effect of different concentrations of ginger root powder and its essential oil on growth performance, serum metabolites and antioxidant status in broiler chicks under heat stress. British Poultry Science. 2014; 55(2):228-237.
47. Yu SG, Abuirmeileh NM, Qureshi AA, Elson CE. Dietary-ionone suppresses hepatic 3- hydroxy-3-methylglutaryl coenzyme A reductase activity. Journal Agriculture and Food Chemistry. 1994; 42:1493-1496.
48. Case GL, Mo H, Elson CE. Induction of geranyl pyrophosphate pyrophosphatase activity by cholesterol-suppressive isoprenoids. Animal Feed Science and Technology. 1995; 30:357-359.
49. Qureshi AA, Lehmann JW, Peterson DM. Amaranth and its oil inhibit cholesterol biosynthesis in 6-week-old female chickens. Journal of Nutrition. 1996; 126:1972-1978.
50. Hong JC, Steiner T, Aufy A, Lien TF. Effects of supplemental essential oil on growth performance, lipid metabolites and immunity, intestinal characteristics, microbiota and carcass traits in broilers. Livestock science. 2012; 144(3):253-262.
51. Qureshi AA, Din ZZ, Abuirmeileh N, Burger WC, Ahmad Y, Elson CE. Suppression of avian hepatic lipid metabolism by solvent extracts of garlic and Impact on serum lipids. Journal of Nutrition. 1983; 113:1746-1755.
52. Middleton B, Hui KP. Inhibition of hepatic S-3-hydroxy-3-methylglutaryl- CoA reductase and *in vivo* rates of lipogenesis by a mixture of pure cyclic monoterpenes. Biochemical Pharmacology. 1982; 31:2897-2901.
53. Goldstein JL, Brown MS. Regulation of the mevalonate pathway. Advances in Experimental Medicine and Biology. 1990; 43:425-430.
54. Elson CE. Novel lipids and cancer. Isoprenoids and other phytochemicals. Advances in Experimental Medicine and Biology. 1996; 399:71-86.
55. Szigeti G, Palfi V, Nagy B, Ine E, Nagy G. New type of immune stimulant to increase antibody production

generated by viral and bacterial vaccines. Magyar Allatorvosok Lapja. 1998; 120:719-721.
56. Hanieh H, Narabara K, Piao M, Gerile C, Abe A. Modulatory effects of two levels of dietary Alliums on immune response and certain immunological variables following immunization in White Leghorn chickens. Animal Science Journal. 2010; 81:673-680.
57. Awaad MH, Abdel-Alim GA, Sayed KS, Ahmed A, Nada AA, Metwalli ASZ *et al*. Immunostimulant effects of essential oils of peppermint and eucalyptus in chickens. Pakistan Veterinary Journal. 2010; 30(2):67-71.
58. Talazadeh F, Mayahi M. Immune response of broiler chickens supplemented with pediatric cough syrup including thyme exteract in drinking water against influenza vaccine. Journal of Herbal medicine and Pharmacology. 2017; 6:522-526.
59. Yang X, Xin H, Yang C, Yang X. Impact of essential oils and organic acids on the growth performance, digestive functions and immunity of broiler chickens. Animal nutrition. 2018; 4(4):388-393.
60. Machado M, Dinis AM, Salgueiro L, Cavaleiro C, Custodio JBA, Sousa M. Anti-Giardia activity of phenolic-rich essential oils: effects of *Tymbracapitata*, *Origanumvirens*, *Tymuszygis* subsp. *sylvestris*, and *Lippiagraveolens* on trophozoites growth, viability, adherence and ultrastructure. Parasitology Research. 2010; 106:1205-1215.
61. Rodrigues GH, Kimura EA, Peres VJ, Couto AS, Aquino DFA, Katzin AM. Terpenes arrest parasitic development and inhibit biosynthesis of isoprenoids in *Plasmodium falciparum*. Antimicrobial Agents and Chemotherapy. 2004; 48:2502-2509.
62. Ueda-nakamura T, Mendonca-filho RR, Morgado-diaz JA, Korehisa P, Prado B, Aparicio D *et al*. Antileishmanial activity of eugenol-rich essential oil from Ocimumgratissimum. Parasitology International. 2006; 55:99-105
63. Ogungbe IV, Setzer WN. Cruzain inhibition by terpenoids: A molecular docking analysis. Natural Product Communications. 2008; 3:865-868.
64. Milhau G, Valentin A, Benoit F, Mallie M, Bastide JM, Pelissier Y *et al*. *In vitro* antimalarial activity of eight essential oils. Journal of Essential Oil Research. 1997; 9:329-333.
65. Zhai Z, Yi L, Wu L, Senchina DS, Wurtele ES, Murphy, PA *et al*. Enhancement of innate and adaptive immune functions by multiple *Echinacea* species. Journal of Medicinal Food. 2007; 10:423-434.
66. Giannenas I, Florou-paneri P, Papazahariadou M, Botsoglou NA, Christaki E, Spais AB. Effect of diet supplementation with ground oregano on performance of broiler chickens challenged with *Eimeriatenella*. International Journal of Poultry Science. 2004; 68:247-252.
67. Da silva MA, Pessotti BMS, Zanini SF, Colnago GL, Rodrigues MRA, Nunes LC *et al*. Intestinal mucosa structure of broiler chickens infected experimentally with *Eimeriatenella* and treated with essential oil of oregano. Cienc Rural Santa Maria. 2009; 39:1471-1477.
68. Hume ME, Clemente-Hernandez S, Oviedo-Rondon EO. Effects of feed additives and mixed *Eimeria* species infection on intestinal microbial ecology of broilers. Poultry Science Journal. 2006; 85:2106-2111.
69. Tsinas A, Giannenas I, Voidarou C, Tzora A, Skoufos J. Effects of oregano based dietary supplement on performance of broiler chickens experimentally infected with *Eimeria acervulina* and *Eimeria maxima*. Journal of Poultry Science. 2011; 48:194-200.
70. Kucukyilmaz K, Bozkurt M, Selek N, Guven E, Eren H, Atasever A *et al*. Effects of vaccination against coccidiosis, with and without a specific herbal essential oil blend, on performance, oocyst excretion and serum IBD titers of broilers reared on litter. Italian Journal of Animal Science. 2012; 11:1-8.
71. Williams RB. Laboratory tests of phenolic disinfectants as oocysticides against the chicken coccidium *Eimeria tenella*. Veterinary Records. 1997; 141:447-448.
72. Bozkurt M, Giannenas I, Kucukyilmaz K, Christaki E, Florou-paneri P. An update on approaches to controlling coccidia in poultry using botanical extracts. British Poultry Science. 2013; 54:713-727.
73. Musa HH, Chen GH, Cheng JH, Li BC, Mekki DM. Study on carcass characteristics of chicken breeds raised under the intensive condition. International Journal of Poultry Science. 2006; 5:530-533.
74. Alp M, Midilli M, Kocabagli N, Yilmaz H, Turan N, Gargili A *et al*. The effects of dietary oregano essential oil on live performance, carcass yield, serum immunoglobulin G level, and oocyst count in broilers. Journal of Applied Poultry Research. 2012; 21:630-636.
75. Sang-oh P, Min RC, Byung-sung P, Jong H. The meat quality and growth performance in broiler chickens fed diet with cinnamon powder. Journal of Environment Biology. 2013; 34:127-133.