



Dietary Effect of Antibiotic Growth Promoter and Essential Oil on Growth Performance, Carcass Traits and Some Physiological Indicators in Broilers

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Abstract

In this bioassay, we assessed the comparative effect of antibiotic growth promoter (AGP) and essential oil (EO) addition on the growth performance, carcass traits, and some physiological indicators in the male Ross 308 broilers. A total of 90 healthy chicks (1 day of age) were randomly allocated to three groups, each comprising 6 replicates of 5 chicks. The first group was a control without any additive in water or feed, the second one was offered a drinking water with 0.5 g/l/day of antibiotic, and the third one was supplied a drinking water with 0.5 ml/liter of EO. In this study, neither AGP nor EO supplementation affected BW, BWG, FI, and FCR ($p < 0.05$) compared with those fed the control diet group. AGP and EO supplemented diets had no significant effect on the carcass, breast and thigh muscles, liver, gizzard, heart spleen, and abdominal fat weight ($p < 0.05$). All the blood plasma concentrations under this study were not affected ($p < 0.05$) by AGP and EO supplementations compared to the untreated control group. In conclusion, it seems that there were no significant differences between the use of AGP or EO supplementation compared with the control group. Therefore, finding from this study demonstrated that essential oil (EO) could be used as a substitute for colistin antibiotic growth promoters (AGP) for broilers, and it's the best strategic way to achieve healthy hygiene meat production free of antibiotic residues.

Keywords: broilers; antibiotic growth promoters (AGP); essential oil (EO); growth performance; physiological indicators

1. Introduction

For decades, antibiotics have played an important role as a growth and health enhancer in animal and poultry production (Attia et al., 2017a, 2017b). Colistin is one of the most often used antibiotics in the poultry industry and the last kind of antibiotic used to treat multidrug-resistant bacterial infections (Kumar et al., 2020). On the one hand, the most important benefits of adding colistin antibiotic to broiler diets at sub-therapeutic levels appear to be an increase in growth performance and gut function, as well as enhancing gut morphology and health (Hung, 2020). In contrast, colistin use by animals and poultry may result in huge amounts of colistin and resistant bacteria being excreted into the environment via feces, leading to the spread of colistin resistance among people (Peng et al., 2021). After learning about the negative consequences of antibiotic residues inside an animal's body and the increased resistance of pathogens to these residues, people began to be concerned about their use. (Attia et al., 2018; 2019). It was first banned by the European Union in 2006, with a request to investigate suitable alternatives to antibiotics for use in animal feed (Barug et al., 2006; Yang et al., 2019).

As a result, poultry production faced numerous challenges, including decreased growth performance, disease resistance, and financial loss, increasing the demand to create safe and effective techniques for maintaining animal intestinal health. Additionally, this ban of antibiotic use in poultry prompted researchers to look for alternative substances such as organic acidifiers, probiotics, prebiotics, enzymes, medicinal herbs, and associated EOs (Pandey et al., 2019; Attia and Al-Harhi, 2015; Diarra and Malouin, 2014). Accordingly, the poultry industry has set a goal of replacing antibiotic performance enhancers with other safe and natural chemicals that have multiple properties and uses and can be used as

growth promoters (Adaszyńska-Skwirzyńska and Szczerbińska., 2017).

Today, prove with evidence-based studies show that the use of EOs worldwide is increasingly focused on poultry for improvement in production and modulation of antioxidant status (Dhama et al., 2015; Attia et al., 2019). Bozkurt et al., (2009) displayed those certain effective supplements, such as medicinal herbs and associated EOs, were gradually used in antibiotic substitution diets.

EOs are colorless liquids composed mostly of aromatic and volatile particles found in the flowers, seeds, stems, bark, husks, or the entire plant (Attia et al., 2019; Sánchez-González et al., 2011). Distillation, fermentation, extraction with solvents or steam are some of the methods used to isolate the EOs from plants. It is noteworthy that EOs contain two types of compounds terpenes and phenylpropenes. Moreover, factors influencing EOs variations include plant species, physical and chemical soil conditions, harvest timing, plant maturity, drying technology, storage duration, and extraction techniques (Namdeo et al., 2020). Medicinal plants and their EOs, known as phytobiotics, are suitable substitutes for growth-promoting antibiotics in broiler diets or drinking water (Grashorn. 2010; Alali et al., 2013; Attia and Al-Harhi, 2015; Galal et al., 2016). Phytobiotics in addition to the growth and production performance of poultry can cause positive changes in other areas such as health status (Willis et al., 2007; Grashorn, 2010; Adaszyńska-Skwirzyńska and Szczerbińska., 2017), intestinal microbial population (Nazzaro et al., 2013; Patel, 2015), serum concentration of antibodies to viral diseases, biochemical parameters and hematological (Demir et al., 2008; El-Ghousein and AL-Beitawi., 2009).

Given the inconsistency of results from previous studies and because of limited studies of the comparison between AGP and

EO impact on growth performance, carcass characteristics, and physiological indices in the broiler, further research is necessary. Adding EOs may have beneficial effects on the anti-pathogenic bacteria and antioxidant status of broilers, nevertheless, analysis of production performance, carcass characteristics and some physiological indicators are important tools also for assessing the health status of broilers.

The main objective of our research study, thus, was to assess the effect of antibiotic growth promoters (AGP) and essential oil (EO) in comparison with AGP on growth performance, carcass traits and some physiological indicators in broilers.

2. Material and methods

The current research study was carried out under the approval of the Local Experimental Animals Care Ethics Committee and performed in compliance with the guidelines of Kafrelsheikh University, Egypt (Number 4/2016EC).

2.1. Supplements preparation:

Colistin antibiotic (Colistin Sulfate 4800000 IU) proved from INTERMEDICAVET Company for animal health products, Makram Eubeed St., Cairo, EGYPT. The recommended dose in drinking water is 0.5 g/l/day. Essential oil (mentocin plus) was proved from Asterpharma company, Egypt. Each 1 Liter contains 10 g Thymol oil, 83 g Menthol oil, and 41 g Eucalyptus oil). The recommendation doses in 0.5 ml/liter drinking water.

2.2. Birds, diets, and management:

A total of 90 chicks (1 day old) were randomly allocated to three groups, each comprising 6 replicates of 5 chicks. All the birds were fed commercial pellet diets, (23, 21, and 19% CP by 3000, 3040 and 3140 kcal/kg, respectively) formulated to meet the nutrient requirements recommended by Ross 308 broiler nutrition specifications guide (Aviagen, 2019). The first group control without any additive in water or feed, the second one drinking water with antibiotic, and the third one drinking water with EO.

The experimental diet composition during starter, grower and finisher periods, respectively is presented in Table 1. At the start of each age period of the experiment, the pellet diets were manufactured in the feed factory and stored at standard environmental conditions according to the firm's guidelines. The diets were provided to the birds' ad libitum and were reared in an environmentally controlled room. All birds had the same environmental management (temperature, moisture, ventilation and light).

2.3. Growth performance:

Feed intake (FI) was recorded by subtracting the weight of the leftover feed from the total feed, body weight gain (BWG) was recorded in each replicate unit at the beginning and end of each experimental period. Also, the body weight gain (BWG) was recorded body weight at the end from the beginning period and a calculation of the feed conversion ratio (FCR) on the whole, and periodically gain was calculated by using the following formula: $FCR = \text{total feed taken/whole live body weight gained}$. Throughout the experimental phase, the health status and mortalities were measured daily on a regular basis.

2.4. Carcass organs traits:

At the end of the experiment, five birds from each treatment were randomly chosen and slaughtered. Eviscerated weight, liver, gizzard, heart, carcass, and abdominal fat were weighted as carcass traits. Based on live body weight, the percentage yield of each part was calculated.

2.5. Biochemical parameters analysis:

At slaughtering time, the blood samples (5ml) were collected and centrifuged at 3000 rpm for 20 minutes. The plasma produced was frozen at -20°C till the time of chemical analysis. Blood parameters including total cholesterol, triglyceride, aspartate aminotransferase (AST) and alanine aminotransferase (ALT), total protein (TP), albumin, and glucose concentrations were measured colorimetrically using

Table 1. The ingredients and chemical compositions of the commercial basal diets (1-34 d).

Ingredient, g/kg	Starter	Grower	Finisher
	1-10 days	11-25 days	26-34 days
Yellow corn, 8.5%	532	592	633
Soybean meal, 46%	370	317	275
Corn gluten meal, 62%	25	26	20
Soybean oil	28	24	34
Dicalcium phosphate	18.0	15.5	14.0
DL- Methionine, 99%	2.7	2.0	1.9
L-Lysine HCL, 98%	2.5	2.3	2.2
Threonine, 99%	1.1	0.7	0.6
Limestone	13.6	13.3	12.0
NaCl	2.3	2.6	2.7
Premix*	3	3	3
Sodium bicarbonate	1.8	1.6	1.6
Total	1000	1000	1000
Calculated analysis**			
Crude protein, %	23	21	19
TME, Kcal/kg diet	3001	3043	3142
Calcium, %	1.036	0.955	0.861
Total phosphorus, %	0.735	0.671	0.625
Na	0.17	0.17	0.17
Cl	0.18	0.20	0.20
L-Lysine, %	1.47	1.31	1.17
DL- Methionine, %	0.66	0.57	0.53

*Each 3 kg of vitamin-mineral premix contain: 6000000IU vit A, 900000 IU vit D3, 40000mg vit E, 2000mg vit K, 2000mg vit.B1, 4000mg vit B2, 2000mg vit B6, 10mg vit B12, 50000mg Niacin, 10000 mg pantothenic acid, 50mg Biotin, 3000mg Folic acid, 250000 mg choline, 8500mg Mn, 50000mg Fe, 50000mg Cu, 200mg I, 100mg Se and 100mg Co.

commercial kits (Diamond Diagnostics, Egypt) according to the procedure outlined by the manufacturer.

2.6. Statistical analysis:

The collected data were analyzed according to a completely randomized design (CRD) using the general linear model (GLM) procedure of the SAS version 9.4 (2016). The significance of means' differences was checked using Tukey's multiple comparison test bases on $p < 0.05$.

3. Results

3.1. Growth performance:

The effects of AGP and EO supplementation on growth performance in broilers were displayed in Table 2. In this study, neither AGP nor EO supplementation affected growth performance parameters, including BW, BWG, FI and FCR ($p < 0.05$) in the birds on AGP and EO treatments

compared the overall experimental period.

3.2. Carcass traits:

The results of carcass organ traits are exposed in Table 3. AGP and EO supplemented groups did not affect carcass traits, including carcass, breast and thigh muscles, liver, gizzard, heart spleen, and abdominal fat weight ($p < 0.05$).

3.3. Plasma parameters:

Table 4 displays the effect of AGP and EO supplementation on the plasma total protein, albumin, globulin, glucose, total cholesterol, triglyceride, GPT and GOT concentrations. All the plasma parameters concentrations under this study such as total protein, albumin, globulin, glucose, total cholesterol, triglyceride, GPT and GOT were not affected ($p < 0.05$) by AGP and EO supplementation compared to the untreated control group.

Table 2. Dietary effect of antibiotic growth promoters (AGP) and Essential oil (EO) on growth performance in broilers.

Item	Diet treatments			SEM	P-Tukey
	Control	AGP	EO		
Initial Body weight, g/b	39.93	39.87	39.93	0.13	0.704
Final body weight, g/35 d	1858.33	1940.00	1877.67	20.72	0.071
Body weight gain, g/b	1818.47	1900.23	1837.73	20.81	0.072
Feed intake, g/35 d	3171.33	3170.00	3157.67	44.57	0.972
Feed conversion ratio, g/g	1.71	1.64	1.68	0.02	0.168

^{a-b} The means placed at the rows with different superscript letters are significantly different ($P > 0.05$); SEM, standard error of the means; Control, a basal diet without supplements; AGP, antibiotic growth promoters; EO, essential oil.

Table 3. Dietary effect of antibiotic growth promoters (AGP) and Essential oil (EO) on carcass traits in broilers.

Item	Diet treatments			SEM	P-Tukey
	Control	AGP	EO		
Carcass weight, g/100g BW	66.22	66.53	66.34	0.56	0.927
Breast muscle weight, g/100g BW	23.56	23.28	23.11	0.62	0.877
Thigh muscle weight, g/100g BW	16.17	16.88	16.35	0.21	0.328
Liver weight, g/100g BW	2.80	2.82	2.81	0.21	0.998
Gizzard weight, g/100g BW	1.27	1.25	1.27	0.06	0.971
Heart weight, g/100g BW	0.58	0.59	0.62	0.02	0.422
Spleen weight, g/100g BW	0.07	0.06	0.06	0.02	0.824
Abdominal fat weight, g/100g BW	1.39	1.34	1.32	0.08	0.836

^{a-b} The means placed at the rows with different superscript letters are significantly different ($P > 0.05$); SEM, standard error of the means; Control, a basal diet without supplements; AGP, antibiotic growth promoters; EO, essential oil.

Table 4. Dietary effect of antibiotic growth promoters (AGP) and Essential oil (EO) on plasma parameters in broilers.

Item	Diet treatments			SEM	P-Tukey
	Control	AGP	EO		
Total protein, g/dL	4.47	4.60	4.03	0.26	0.213
Albumin, g/dL	1.44	1.54	1.56	0.03	0.095
Globulin, g/dL	3.03	3.06	3.47	0.19	0.274
Glucose, mg/dL	152.33	154.67	151.33	9.21	0.862
Total cholesterol, mg/dL	122.00	125.33	122.33	5.75	0.905
Triglyceride, mg/dL	6.23	5.40	4.30	0.66	0.198
GPT, mg/dL	6.27	6.66	5.83	0.58	0.619
GOT, mg/dL	249.33	241.33	250.67	21.17	0.945

^{a-b} The means placed at the rows with different superscript letters are significantly different ($P > 0.05$); SEM, standard error of the means; Control, a basal diet without supplements; AGP, antibiotic growth promoters; EO, essential oil; GPT, glutamic pyruvic transaminase; GOT, glutamic oxaloacetic transaminase.

4. Discussion

4.1. Growth performance:

The present study aimed to evaluate the comparative effect of antibiotic growth promoters (AGP) and essential oils (EO) and whether substituting antibiotics with EOs would have the same beneficial effects over antibiotics on growth performance, carcass traits, and some physiological indicators in male Ross 308. There were no significant

differences ($p < 0.05$) in all growth performance parameters between treated groups compared with the control group. Except, the APG and EO groups numerically increased FBW and BWG than the control group. Similar results were observed by Botsoglou et al., (2002) who reported that dietary oregano EO supplementation to broilers had no positive effect on growth performance. Hernandez-Coronado et al.

(2019) also found that the addition of dietary oregano EO had no major effects on broilers' growth and that differential BWG effects could be determined by differences in EO levels and ingredients in diets. Another study found that adding thyme oil did not affect broiler BWG (Cross et al., 2003). According to Lee et al. (2003), 200 mg/kg of thymol in the diet did not affect the BWG, FI, or FCR of female broilers. In contrast to our findings, treatment with 1000 mg/kg thyme EO enhanced BWG while lowering FI by about 10% (Cross et al., 2007; Hashemipour et al., 2013). Studies have shown that EO supplementation in the diet improves their growth performance by encouraging digestive enzyme secretion that results in improved nutrient digestion, digestive transmission rate (Attia et al., 2017a; Jamroz et al., 2005). The antibacterial effect of EO and the stimulation of several digestive enzymes, which improve nutrition utilization, lead to an increase in body weight growth (Attia et al., 2019; Hernandez et al., 2004). However, Tiihonen et al. (2010) found that include EO in broiler diets reduces the growth of pathogenic bacteria while increasing the growth of beneficial gut microbiota, which improves broiler growth performance. The stability of EOs and AGP during feed processing where researchers reported that significant loss of activity of EOs was caused by a pelleting temperature of 58 °C (Maenner et al., 2011; Cromwell., 2002; Attia and Al-Harthi, 2015). The lack of performance effects of AGP and EO could be explained by levels of the supplemental in the diet, environmental, and feed processing conditions.

4.2. Carcass traits:

A positive relationship has been established between improving poultry meat quality by increasing muscle and growth while reducing abdominal fat content (Musa et al., 2006). The current findings on the effect of AGP and EO on carcass organs were consistent with the findings of Alp et al., (2012) they reported that the addition of 300 mg/kg of oregano EO did not have a

significant impact on the carcass characteristics. On the other hand, several studies showed the positive effects of EOs supplementation on carcass traits in broilers (Sang-oh et al. 2013; Khattak et al., 2014; Peng et al., 2016). Whatever the case, the low concentrations of additives or the reduced effectiveness of AGP and EO due to the high temperature during the feed processing production possibly explain why the additives did not influence carcass attributes.

4.3. Plasma parameters:

Supplementation of AGP and EO did not affect total protein, albumin, globulin, glucose, total cholesterol, triglyceride, GPT, and GOT in broilers during the experimental period. These findings were comparable to those of Lee et al., (2003, 2004) and Bampidis et al., (2005), who showed no significant variations in serum cholesterol, HDL, or triglyceride levels across groups. Meanwhile, other studies showed a positive effect on reducing the plasma total protein and globulins due to supplementation of Eos to broiler's diets (Attia et al., 2018; Ghazalah and Ali., 2008). There are contradictory results regarding the effect of dietary EO to broiler diets on plasma biochemistry parameters, due to the differences between the types of essential oils according to the type of plants extracted, as well as the concentration of active substances in them and the extent of the effect of the feed processing temperature.

5. Conclusions

In conclusion, this study demonstrated that no differences in the parameters including growth performance, carcass traits and physiological indicators between all the treatment groups because of the use of 0.5 g/l/day of AGP or 0.5 mg/l of EO in broilers' diets. Although the APG and EO groups had higher FBW and BWG than the control group, the differences were not statistically significant. Nevertheless, no negative effect was observed when the AGP or EO supplementation was in the broilers diet. Therefore, finding from this study

demonstrated that essential oil (EO) could be used as a replacement for Colistin antibiotic growth promoters (AGP) in broilers commercial diets, and it's the best strategic way to achieve healthy hygiene meat production free of antibiotic residues.

Conflict of interest statement

This manuscript has no conflicts of interest.

Data availability statement:

All data sets collected and analyzed during the current study are available from the corresponding author on reasonable request.

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