

# The Effect of *Myrtus Communis* Oil Extract on Growth Performance and Immune Responses in Ross and Cobb Strain Broilers

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**Abstract**—This experiment was carried out to compare the performance and immune responses in Ross and Cobb broilers fed diets containing *Myrtus communis* oil extract (MCE). A total of 224 one-d-old broiler chickens randomly allocated to each of the 4 treatment groups, each with 4 replicate pens of 14 chicks. The experimental diets included two treatments for Ross strain – control (RC) and the diet contain 500mg/Kg MCE (RM) - and two treatments for Cobb strain –control (CC) and the diet contain 500mg/Kg MCE (CM). The performance parameters were measured during the experimental period and antibody titers against Newcastle disease virus were determined. The using of MCE in diet decreased feed intake and feed conversion ratio (FCR) but had not significant effect on daily body weight gain and live body weight. The lowest amount of feed intake and FCR were related to RM group. The relative weight of bursa of Fabricius increased significantly by using of MCE in both strains. In general, the results indicate that dietary inclusion of 500 mg/kg MCE can improve performance and immune system in Ross and Cobb broiler strains.

**Index Terms**—broiler, Cobb, immune system, *Myrtus communis*, performance, Ross

## I. INTRODUCTION

For more than fifty years antibiotic growth promoters have been used in poultry diets to improve growth performance and protect flock health. But the using of antibiotics has had some problem such as generation of bacterial resistant strains and the entrance of drug residues into the food [1]. For this reasons many of countries have banned the use of antibiotics in animal diets. In the last decade, many efforts have been done to replace antibiotics by natural materials in Medicinal plants. In recent years, the use of phytogenic compounds and herbal products have been given considerable attention due to antimicrobial properties and their potential role as natural substitutes to antibiotic growth promoters in animal nutrition [2]. Essential oils may specifically enhance the activities of digestive enzymes and nutrient absorption, which may improve the values of feed [3].

Myrtle (*Myrtus communis* L., Myrtaceae) is an evergreen scrub which grows spontaneously and is used

in many countries as medicinal plant [4]. It is believed that the main biologically active components in this herb are semimyrto-commulone, 1,8-cineole, arepolyphenols,  $\alpha$ -pinene, myrtucommulone, myrtenyl acetate, limonene, linalool and  $\alpha$ -terpinolene [5]. Various parts of this herb such as its berries, leaves and fruits have been used extensively as a traditional medicine for a number of centuries. The herb is used traditionally for the therapy of disorders such as hemorrhoid, inflammation, diarrhea, peptic ulcer, skin diseases and pulmonary [6]. It is well known for its therapeutic values as anantiseptic, disinfectant drug and hypoglycaemic agent [7]. The experimental and clinical studies demonstrate that it has a wide range of pharmaceutical and health effects such as anti-bacterial [8], antioxidant [9], anti-cancer [10], anti-diabetic [11], anti-viral, anti-fungal [12], neuroprotective and hepatoprotective [13] activity.

Therefore, the purpose of the present study was determine the effect of *Myrtus communis* oil extract (MCE) on performance parameters, immune organs weights and antibody titers against Newcastle disease virus in Ross and Cobb strain broilers.

## II. MATERIALS AND METHODS

### A. Animals and Diets

A total of 224 one-d-old broiler chickens (mean initial weight:  $37.5 \pm 1$  g) of mixed sex (Ross-308 and Cobb-500) were raised over 42 days. They were arranged in a completely randomized design in four treatments and four replicates. The experimental diets included two treatments for Ross strain – control (RC) and the diet contain 500mg/Kg MCE (RM) - and two treatments for Cobb strain –control (CC) and the diet contain 500mg/Kg MCE (CM). The basal diet (Table I) was formulated to meet the nutrient requirements of broilers base on digestible amino acids. The myrtle essential oils were prepared from a local market. The chicks were raised on floor pens (120 × 120 × 80 cm) for 6 weeks and the feed and water were supplied ad libitum. The chicks were reared under an illumination program which included of 23 hours of light and 1 hour of darkness. The ambient temperature was 34°C throughout of the first week, and it was reduced three degrees per week in following weeks until it was fixed at 22°C.

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TABLE I. COMPOSITION OF EXPERIMENTAL DIETS

| Item                                    | Starter<br>(0-14) | Grower<br>(15-28) | Finisher<br>(29-42) |
|---|-------------------|-------------------|---------------------|
| Ingredient, g/kg                        |                   |                   |                     |
| Corn                                    | 554.6             | 591.0             | 629.2               |
| Soybean meal (43.8%)                    | 363.0             | 319.2             | 275.1               |
| Fish meal (65%)                         | 25.0              | 25.0              | 25.0                |
| Soybean oil                             | 20.0              | 29.3              | 38.2                |
| Mono calcium phosphate                  | 12.7              | 12.1              | 11.0                |
| CaCO <sub>3</sub>                       | 12.0              | 11.1              | 9.8                 |
| NaHCO <sub>3</sub>                      | 0.1               | 0.1               | 0.1                 |
| NaCl                                    | 2.1               | 1.9               | 1.8                 |
| Mineral premix <sup>1</sup>             | 2.5               | 2.5               | 2.5                 |
| Vitamin premix <sup>2</sup>             | 2.5               | 2.5               | 2.5                 |
| DL-Methionine                           | 2.6               | 2.5               | 2.1                 |
| L-Lysine                                | 2.1               | 2.0               | 1.8                 |
| L- Threonine                            | 0.3               | 0.3               | 0.4                 |
| Choline                                 | 0.5               | 0.5               | 0.5                 |
| Apparent Metabolizable energy (kcal/kg) | 2925              | 3025              | 3125                |
| Crude protein (g/kg)                    | 223               | 203               | 186                 |
| Calcium (g/kg)                          | 9.8               | 9.3               | 8.6                 |
| Available phosphorus (g/kg)             | 4.8               | 4.5               | 4.2                 |
| Digestible Methionine (g/kg)            | 5.5               | 5.1               | 4.5                 |
| Digestible Lysine (g/kg)                | 11.5              | 1.5               | 9.4                 |
| Digestible Methionine + Cysteine (g/kg) | 8.5               | 7.9               | 7.25                |
| Digestible Threonine (g/kg)             | 7.3               | 6.8               | 6.3                 |

<sup>1</sup>-Ingredients per kg: Mg, 60 g; Fe, 80 g; Cu, 10 g; Zn, 50 g; Co, 2 g; I, 1 g,

<sup>2</sup>- Ingredients per kg : vitamin A, 1000,000 IU; D3, 1500000 IU; E, 15000 IU; K, 3g; B1 2g; B2, 4 g; B6, 3g; B12, 0.015 g; pantothenic acid, 10 g; nicotinic acid, 2 g; folic acid, 1 g; choline, 250g ; Se, 100 g

### B. Measurement of Parameters

The performance parameters included of daily body weight gain(DBWG), live body weight(LBW) and daily feed intake(DFI) were measured weekly and feed conversion ratio (FCR) was calculated as weight gain divided by feed intake. Mortality was also recorded daily. At the end of the experiment, two birds per treatment were randomly selected to study carcass characteristics. Chicks were fasted for an overnight period of 10 h and then individually weighed, slaughtered, feathered and eviscerated. Weights of the breast, thigh, heart, liver, spleen and some other organs were recorded and their relative weight was calculated as a percentage of live body weight.

### C. Immunity Parameters

The chicks were vaccinated against Newcastle disease (Iasota strain) at 10 and 22 days of age. At the age of 30 days, two birds from each repetition were randomly selected and blood samples were collected from the brachial vein and centrifuged for 10 min at 1500 rpm to obtain serum. Antibody titers against Newcastle virus were measured using hemagglutination inhibition test [14].

### D. Statistical Analysis

All data were analyzed by one-way ANOVA using the GLM procedure of SAS for Windows version 9.1 (SAS Institute Inc., Cary, NC). The significance of differences

between the means was compared by using of the Duncan's multiple range tests of SAS.

## III. RESULTS AND DISCUSSION

### A. Growth Parameters

The experimental data about performance have been shown in Table II. The impact of experimental treatment on feed intake was significant ( $p \leq 0.05$ ) in starter, grower, finisher and total period. The using of MCE in diet resulted in decreasing feed intake for Cobb and Ross strains in all of experimental periods. In total period, the reduction in feed intake was greater for Ross strain than Cobb strain. This reduction in feed consumption may be due to unpalatability and adaptation with MCE. In our previous experiment we observed that the feed intake decreased in starter period in chickens consumed savory essential oils in diet [15]. Ref. [16] also reported a decrease in feed intake when the broiler chicks fed by a diet containing Myrtle essential oils. Ref. [17] reported that even though fowl have very low number of taste buds than mammals, they do have an acute sense of taste and they clearly recognize a change in taste. In contrast to our finding, Ref. [18] reported that the using of myrtle oil in amount of 500, 1000, 2000 and 5000 mg/Kg feed had not significant effect on feed intake of laying quails.

The ADWG only was affected significantly ( $p \leq 0.05$ ) in starter period by using MCE in diet. The differences between treatments mean were not significant in grower,

finisher and total periods. Similar to ADWG, the effect of MCE on live body weight was not significant in days 42. Broilers receiving MCE had a lower FCR compared to controls groups in different experimental periods. The effect of MCE on FCR was greater for Ross strain, so the best FCR was related to RM treatment. No differences because of treatment effects were observed on mortality.

The better FCR can be the consequence of improving the balance of gastrointestinal microflora and absorption properties of intestine. The antimicrobial activity of Myrtle on *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *P. vulgaris*, *P. mirabilis*, *Klebsiella aerogenes*, *Salmonella typhi* and *S. shigella* was determined by Ref. [8].

TABLE II. EFFECT OF TREATMENTS ON PERFORMANCE PARAMETERS OF BROILERS

| Performance parameters | Diets               |                      |                     |                      |       |
|------------------------|---------------------|----------------------|---------------------|----------------------|-------|
|                        | RC                  | RM                   | CM                  | CC                   | SEM   |
| DFI <sup>1</sup>       |                     |                      |                     |                      |       |
| 0-14 d                 | 29.94 <sup>ab</sup> | 27.38 <sup>c</sup>   | 28.72 <sup>bc</sup> | 30.81 <sup>a</sup>   | 0.420 |
| 14-28 d                | 107.59 <sup>a</sup> | 98.14 <sup>c</sup>   | 103.45 <sup>b</sup> | 104.25 <sup>b</sup>  | 0.984 |
| 28-42 d                | 159.80 <sup>a</sup> | 153.27 <sup>bc</sup> | 151.22 <sup>c</sup> | 157.66 <sup>ab</sup> | 1.042 |
| 0-42d                  | 99.12 <sup>a</sup>  | 91.49 <sup>c</sup>   | 94.62 <sup>b</sup>  | 97.99 <sup>a</sup>   | 0.864 |
| DWG <sup>2</sup>       |                     |                      |                     |                      |       |
| 0-14d                  | 22.16 <sup>a</sup>  | 20.70 <sup>b</sup>   | 21.05 <sup>ab</sup> | 21.30 <sup>ab</sup>  | 0.224 |
| 14-28d                 | 61.6                | 60.04                | 62.38               | 63.31                | 0.602 |
| 28-42d                 | 73.56               | 75.80                | 73.16               | 73.62                | 0.454 |
| 0-42d                  | 52.40               | 52.18                | 52.20               | 52.74                | 0.327 |
| FCR <sup>3</sup>       |                     |                      |                     |                      |       |
| 0-14 d                 | 1.35 <sup>b</sup>   | 1.32 <sup>b</sup>    | 1.37 <sup>b</sup>   | 1.45 <sup>a</sup>    | 0.015 |
| 14-28 d                | 1.75 <sup>a</sup>   | 1.64 <sup>b</sup>    | 1.66 <sup>b</sup>   | 1.65 <sup>b</sup>    | 0.014 |
| 28-42 d                | 2.17 <sup>a</sup>   | 2.02 <sup>c</sup>    | 2.07 <sup>bc</sup>  | 2.14 <sup>ab</sup>   | 0.021 |
| 0-42d                  | 1.89 <sup>a</sup>   | 1.75 <sup>b</sup>    | 1.81 <sup>ab</sup>  | 1.86 <sup>a</sup>    | 0.018 |
| BW <sup>4</sup>        |                     |                      |                     |                      |       |
| 14d                    | 346.24a             | 325.80b              | 330.70ab            | 334.20ab             | 3.190 |
| 28 d                   | 1208.32a            | 1166.09b             | 1204.00a            | 1221.38a             | 7.148 |
| 42d                    | 2238.12             | 2227.14              | 2228.06             | 2251.05              | 5.440 |

Values in the same row not sharing a common superscript differ significantly ( $P \leq 0.05$ ). RC=Ross control; RM= Ross+ Myrtle essential oils; CM= Cobb + Myrtle essential oils CC=Cobb control, SEM = Standard error of mean

1. Daily Feed Intake (g per bird/day). 2. Daily Weight Gain (g/day) 3. Feed Conversation Ratio (g/g). 4. Body Weight (g) 5. Standard error of mean

Ref. [19] showed the reduction of *E. coli* and increasing of *Lactobacillus spp* population in gut by inclusion of myrtle essential oil in the diet. Also they reported that the dietary myrtle essential oil resulted in significantly longer villus height, lower epithelial thickness and lower goblet cell number of the small intestine at 42 day of age. It is thought that the activity of the intestinal microflora in the host is an important factor that may impact gut function. Inappropriate microflora population in the alimentary canal will result in poor nutrient absorption and the increasing energy requirements for maintenance [20]. This can be a possible mechanism for better FCR in broiler consuming MCE. The results of the present experiment are consistent with Mohamadi Saei *et al.*, [16] who by using 500mg/Kg MCE in diet observed positive effect on FCR. In contrast to this report, Ref. [18] observe negative effect on FCR in laying quails by using 5000 mg/Kg MCE in diet.

#### B. Immunity Parameters

As shown in Table III, the effect of MCE on immunity parameters was only significant ( $p \leq 0.05$ ) for relative weight of the bursa of Fabricius but mean differences between treatments for antibody titer against Newcastle and relative weight of the spleen were not significant. The relative weight of the bursa of Fabricius increased by using of MCE in diet in both strains. It increased by 12.5% and 10.3% for RM and CM groups respectively compared to control groups. This shows that in respect of this parameter the response of Ross strain to MCE was

slightly bigger than the Cobb strain. It is concluded that the active components of myrtle which have antibacterial, antiviral, antifungal, anti inflammatory and antioxidant activities induce positive effects on these organ. For the reason that the bursa is an important lymphoid organ in poultry, higher bursa weight can be an indicator of high immune activity. The increase in immune tissue weight may result in increase of immune cell proliferation and antibody production [21]. Ref. [22] reported that immunoglobulin G synthesis was higher in chicken with larger Bursa. In our previous research we had found the same results about the using of onion in diet [23]. Therefore It seems that the effect of medicinal plants such as Myrtle limit to the mucosal immune complement and not the systemic portion of the immune system. In agreement with our results, Ref. [24] reported that the use of a mixture include of myrtle oil had not significant effect on antibody titer level of Newcastle disease virus. Our findings about antibody titer are in contrast with Ref. [25] who reported that the supplementing the basal diet with MCE increased the antibody titers against Avian Influenza Virus and Newcastle disease Virus.

#### IV. CONCLUSION

The results showed that the dietary inclusion of 500 mg/kg Myrtle essential oils can improve FCR in broiler chickens. Also, the using myrtle essential oils in diet caused the larger bursa of Fabricius that is an indicator of high immune activity. There were not any differences between tow strains with respect to measured parameters.

TABLE III. EFFECT OF EXPERIMENTAL DIETS ON ANTIBODY TITERS AGAINST NEWCASTLE VIRUS AND THE RELATIVE WEIGHTS OF SPLEEN AND BURSA

| Variable          | treatments          |                    |                     |                    |       |
|-------------------|---------------------|--------------------|---------------------|--------------------|-------|
|                   | RC                  | RM                 | CM                  | CC                 | SEM   |
| New castle (log2) | 7.01                | 7.17               | 7.33                | 6.83               | 0.420 |
| Spleen            | 0.141               | 0.136              | 0.130               | 0.146              | 0.003 |
| Bursa             | 0.159b <sup>c</sup> | 0.181 <sup>a</sup> | 0.175 <sup>ab</sup> | 0.155 <sup>c</sup> | 0.004 |

Values in the same row not sharing a common superscript differ significantly ( $P \leq 0.05$ ). RC=Ross control; RM= Ross+ Myrtle essential oils; CM= Cobb + Myrtle essential oils CC=Cobb control, SEM = Standard error of mean

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