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# Antibiotic Growth Promoters and Anise Seeds in Broiler Diets

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## ABSTRACT

The effect of replacing antibiotic growth promoter (Bacitracin Methylene Disalicylate: BMD) by anise seeds as natural growth promoter on broilers performance was studied. One-day old chicks were fed experimental rations for 42 days. The replacement of BMD by anise seeds significantly (P<0.05) increased live body weight, body weight gain and the antibody titer against Newcastle, inficious Bursal and infectious bronchitis diseases. The replacement of BMD by anise seeds also significantly (P<0.05) decreased feed conversion ratio and serum cholesterol and triglyceride concentrations. These findings suggest that anise seeds can replace BMD as growth promoter in broiler rations.

**Keywords**: Anise seeds, Antibiotics, Broiler, Growth promoters, Diets.

## INTRODUCTION

The use of antibiotic growth promoters at the same time with anticoccidials in broiler diets is a routine procedure used by poultry producers in Jordan as well as in many other developing countries. Combinations of growth promoters and anticoccidials showed general improvements in broilers performance, intestinal pathologies and intestinal microflora stability (Fairly et al., 1985; Waldroup et al., 1988; Truscott and Al-Sheikhly, 1997). However, the use of antibiotics in poultry rations has been banned due to the potential development of antibiotics-resistant human pathogenic bacteria and their residues in poultry products. Main advantage of using anise seeds over antibiotics is that they do not bear any risk regarding bacterial resistance or undesired residues in poultry products. Anise (*Pimpinella anisum* L.) is an aromatic annual herb widely grown in the Mediterranean region. The part used is the fruit, or the so-called seeds. Anise seeds have a number of active compounds, particularly volatile oil (1-4%), which consists of largely trans-anethol (70-90%) with estragole (methylchavicol), anisealdehyde, b-caryophlline, anise ketone (methyloxyphenylacetone) and the polymers of anethole (Ciftci et al., 2005). Moreover, as a medicinal plant, anise has been used as antibacterial (Singh et al., 2002; Tabanca et al., 2003), antifungal (Soliman and Badea, 2002), antiparasitic (Cabuk et al., 2003). Moreover, previous literature showed that essential oil of anise increased the digestion of protein, cellulose and fats (Jamroz and Kamel, 2002) and improved apparent whole tract and ileal digestibility of nutrients (Hamandez et al., 2004). Unfortunately, few studies have been conducted to evaluate the effect of replacing antibiotic growth promoters by natural growth promoters such as anise seeds. Therefore, the present experiment was designed to study the effect of replacing Bacitracin Methylene Disalicylate (BMD) by anise seeds on body weight, body weight gain, feed conversion ratio

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and immunity status of broiler chicks.

## MATERIALS AND METHODS

### Experimental Birds and Rearing Conditions

525 one-day old unsexed Lohman broiler chicks were purchased from a commercial hatchery with an average body weight of 43.0 g and randomly distributed in 15 pens each 1.15 m×2.10 m in an open-sided house (5 treatments × 3 replicates × 35 chicks). Feed and water were offered *ad libitum*. Chicks were vaccinated against Newcastle, infectious bronchitis diseases at 7 and 21days of age and against infectious Bursal disease at 13 days of age.

#### *Experimental Diets*

Chicks were fed starter diets from one to 21days of age, replaced by finisher diets from 22 to 42days of age (Table 1). All diets were formulated to cover the birds' requirements according to National Research Council (NRC, 1994). Randomized samples from each diet were collected for proximate analysis using the procedure of AOAC (1990). Five treatment groups were prepared; group one served as a control group (T1), group 2 (T2) was fed Lasalocid Na 0.06% and Bacitracin Methylene Disalicylate (BMD), group 3 (T3) was fed Lasalocid Na 0.06% and anise seeds 0.075%, group 4 (T4) was fed 3-nitro 0.025% and BMD 0.05% and group 5 (T5) was fed 3-nitro 0.025% and anise seeds 0.075% throughout the experimental period.

#### *Measured Parameters*

#### *Live Body Weight, Body Weight Gain, Feed Intake and Feed Conversion Ratio*

Chicks in each pen within each treatment were weighed individually every week throughout the experimental period and the average live Body Weight (BW) per chick was calculated. Body Weight Gain (BWG) was also calculated every week. At the beginning of each week, a known amount of the diet was weighed and offered for each replicate within each treatment. At the end of the week, the residues were weighed and subtracted from the offered amount to obtain the total feed intake during the week. The consumed feed was divided by the number of chicks to obtain the average amount of feed consumed per chick per week. Cumulative Feed Intake (CFI) was also calculated at the end of the experiment (42 days of age). Feed Conversion Ratio (FCR) was calculated using the following equation:

$$FCR = \frac{\text{Cumulative Feed Intake (kg/chick)}}{\text{average live BW (kg/chick)}}$$

#### *Mortality Rate (%)*

Mortality was daily recorded, then at the end of the experiment, Mortality Rate (MR) was calculated.

#### *Blood Constituents*

At the end of the experiment, blood samples were collected from the jugular vein of 5 chicks from each replicate within each treatment for the determination of blood constituents. Blood was collected in non-heparinized tubes and centrifuged at 3000 rpm for 3 minutes. Serum total cholesterol was assayed by JENWAY, 6105µ V/V spectrophotometer using commercial kits (Bis Mereuxas, France), while serum triglyceride was determined by commercial kits of triglycerides-GPO method (Biolabosa, France) after hydrolysis with lipase. High Density Lipoproteins (HDL) were determined by commercial kits (Labkit, Chemelex, Barcelona).

#### *Immunity Measurements*

At the end of the experiment (42days of age), antibody titers of IBD, IB and ND were quantified from 3 randomly selected chickens from each replicate within each treatment. Antibody titers were assayed using Enzyme Linked Immuno-Sorbent Assay (ELISA) for

IBD and Haemagglutination inhibition (HI) test for IB and ND as described by Thayer and Beard (1998).

## STATISTICAL ANALYSIS

Data were statistically analyzed by using the General Linear Model procedure of SAS (1990). The level of significance was set at P< 0.05 or less depending upon the F-values generated. Differences among treatments were found, means were separated by the Least Significant Difference (LSD) method following the procedure of SAS (1990).

## RESULTS

***Growth Performance***

Table (2) presents the means ± SE of live Body Weight (BW), Body Weight Gain (BWG) and mortality rate of broilers fed different dietary treatments at 42 days of age. Results showed that broiler chickens fed the basal ration containing anticoccidials (lasalocid Na or 3-nitro) supplemented with anise seeds had the highest (P<0.05) live Body Weight (BW), Body Weight Gain (BWG) and Feed Intake (FI) and the best (P<0.05) Feed Conversion Ratio (FCR) compared to those fed anticoccidial and BMD supplementad rations and the control group. The different dietary treatments failed to produce any significant improvement in mortality rate compared to the control group.

***Serum Constituents***

Table (3) shows that broiler chickens fed the basal ration containing anticoccidials and supplemented with anise seeds instead of BMD had the lowest (P<0.05) cholesterol and triglyceride and the highest (P<0.05) HDL concentrations compared with those fed the same anticoccidial and BMD and the control group.

***Immunity Status (Antibody Titer)***

Table (4) shows the means ± SE values of antibody titer against Newcastle Disease (ND), Infectious Bursal Disease (IBD) and Infectious Bronchitis (IB). Results demonstrated that broiler chickens fed the basal ration containing lasalocid Na or 3-nitro and supplemented with anise seeds had the highest (P<0.05) antibody titer against ND compared with other dietary treatments. With respect to IBD, the addition of anise seeds to basal ration containing lasalocid Na increased significantly (P<0.05) antibody titer compared with broiler chickens fed the basal ration alone or after the supplementation of 3-nitro and BMD. No significant differences were observed in antibody titer against IBD of broilers fed lasalocid Na and anise seeds or 3-nitro and anise seeds. Concerning IB, the present results showed that no significant effects were noticed in antibody titer among treatments, the only significant differences were observed between broiler chicks fed the basal ration alone and all other dietary treatments.

## DISCUSSION

The results of the present study clearly show that the addition of anise seeds instead of BMD to the basal ration containing either lasalocid Na or 3-nitro anticoccidials improves significantly (P<0.05) live BW, BWG,CFI and FCR (Table 2), suggesting that anise seeds can be used as a natural growth promoter. It has been reported that anise seeds stimulate digestion (Cabuk et al., 2003), particularly the digestion of protein, fats and cellulose (Jamroz and Kamel, 2002). In addition, several researchers reported that anise oil significantly improves daily live weight gain and FCR of broiler chickens (Ather, 2000; Williams and Losa, 2001; Giannenas et al., 2003; Ciftci et al., 2005). Moreover, it seems that the improved effects of using anise seeds in this study might be due to the improvement of apparent whole tract and ileal digestibility of the nutrients (Hemandez et al., 2004), increasing the effects of pancreatic lipase and amylase secretion (Ramakrishna et al., 2003). The present findings are in agreement with the previous

findings of Abu-Egla et al. (2001), El-Ghammary et al. (2002) and Hassan et al. (2004). Cabuk et al. (2003) postulated that the increase in live BW, BWG and the improvement in FCR may be due to the different active ingredients, particularly anothole and eugenol in anise which have digestive stimulating effects.

The reduction of cholesterol and triglyceride noticed in the blood serum in the present study may be attributed to the reduction of cholesterol synthesis or to the decreasing of its fractions by the small intestine (Brunton, 1999). Al-Harthi (2004) conducted an experiment to evaluate the efficiency of utilizing some herbs with or without antibiotic supplementation on broilers performance, and he reported that plasma cholesterol and triglyceride were significantly decreased by the addition of herbs or/and antibiotics to broiler chicken diets. Our present results were supported by McKenzine et al. (1989) and Saini et al. (2003) who reported that essential oils blend of medicinal plants shows the same effect of antibiotics. The noticeable improvement of antibody titer against ND, IBD and IB in the present study might be due to the effect of active components of anise seeds, particularly anothole, eugenol, methylchavicol, anisaldehyde and estragol (Ciftci et al., 2005). As a medicinal plant, anise had an antimicrobial effect (Tabanca et al., 2003) and an antifungal effect (Soliman and Badea, 2002). We assume that this could be considered another reason of improving the immune status of the chickens.

Finally, under the conditions of this study, it can be concluded that anise seeds can completely replace BMD in combination with anticoccidials. More studies are required to support and confirm the outcome of this study.

**Table 1. Composition of experimental rations.**

| | Ration | |
|---|---|---|
| **Ingredients (%)** | **Starter** | **Finisher** |
| Yellow corn | 50.0 | 54.0 |
| Soybean meal (48% CP) | 33.75 | 27.5 |
| Barley | 10 | 10 |
| Oil | 2.00 | 4.75 |
| Limestone | 1.70 | 1.50 |
| DCP | 1.30 | 1.00 |
| NaCl | 0.25 | 0.25 |
| * Vitamin: Mineral Premix | 1.00 | 1.00 |
| Total | 100 | 100 |
| **Calculated feeding values** | | |
| Crude protein | 21.72 | 19.03 |
| ME (kcal/kg) | 2939 | 3162 |
| EE (%) | 4.42 | 7.26 |
| Lysine (%) | 1.17 | 0.99 |
| Methionine (%) | 0.33 | 0.30 |
| Ca (%) | 1.05 | 0.89 |
| Total P | 0.51 | 0.45 |
| **Analyzed feeding values** | | |
| DM (%) | 91.2 | 90.2 |
| Crude protein (%) | 22.2 | 19.9 |
| EE (%) | 5.8 | 8.9 |
| Ash (%) | 8.9 | 6.9 |

* Vitamin: Mineral Premix: Provided the following (per kilogram of complete diets); 2,000,000 IU vitamin A; 400,000 IU Vitamin D3; 400 mg Vitamin E; 200mg Vitamin B1;800 mg Vitamin B2; 4,000 mg Nicotinic acid; 2,000 mg Pantothenic acid; 300 mg Vitamin K; 200 mg Folic acid; 300 mg Vitamin B6; 50 mg Co; 1,600 mg Cu; 6,421 mg Fe; 156 mg I; 12,800 mg Mn; 32 mg Se; 9,000 mg Zn; 100 mg Choline Chloride.

**Table 2. Effects of replacing BMD by anise seeds on growth performance of broiler chickens at 42 days of age.**

| | *Treatment* | Live body weight (kg) | Body weight gain (kg) | Cumulative feed intake (kg) | Feed conversion ratio | Mortality rate (%) |
|---|---|---|---|---|---|---|
| **1** | Lasalocid Na +BMD | 1.65$^{b}$ | 1.61$^{b}$ | 3.20$^{b}$ | 2.07$^{b}$ | 3.80 |
| **2** | Lasalocid Na +anise seeds | 2.10$^{a}$ | 2.06$^{a}$ | 3.85$^{a}$ | 1.87$^{c}$ | 3.20 |
| **3** | 3-Nitro+BMD | 1.67$^{b}$ | 1.63$^{b}$ | 3.42$^{ab}$ | 2.10$^{b}$ | 3.75 |
| **4** | 3-Nitro +anise seeds | 2.00$^{a}$ | 1.96$^{a}$ | 3.72$^{a}$ | 1.90$^{c}$ | 3.15 |
| **5** | Control (Basal diet) | 1.32$^{c}$ | 1.27$^{c}$ | 2.95$^{c}$ | 2.32$^{a}$ | 4.00 |
| | ± ***SE*** | 0.30 | 0.27 | 0.43 | 0.05 | 1.10 |

$^{a-c}$ Means with different superscripts in the same column are significantly different at $P<0.05$.

**Table 3. Effect of replacing BMD by anise seeds on some serum constituents at 42 days of age.**

| *Treatment* | | Constituents (mg/dl) at 42 days of age | | |
|---|---|---|---|---|
| | | **Cholesterol** | **Triglyceride** | **HDL** |
| **1** | Lasalocid Na +BMD | 133.02$^{a}$ | 48.04$^{a}$ | 42.59$^{b}$ |
| **2** | Lasalocid Na +anise seeds | 117.48$^{b}$ | 44.57$^{b}$ | 44.13$^{b}$ |
| **3** | 3-Nitro+BMD | 128.33$^{a}$ | 48.57$^{a}$ | 42.92$^{b}$ |
| **4** | 3-Nitro +anise seeds | 117.15$^{b}$ | 43.74$^{b}$ | 49.20$^{a}$ |
| **5** | Control (Basal diet) | 131.12$^{a}$ | 49.13$^{a}$ | 41.74$^{bc}$ |
| | ± ***SE*** | 7.176 | 3.292 | 2.922 |

$^{a-b}$ Means with different superscripts in the same column are significantly different at $P<0.05$.

**Table 4. Effect of replacing BMD by anise seeds on antibody titer of broiler chickens at 42 days of age.**

| *Treatment* | | Antibody titer at 42 days of age | | |
|---|---|---|---|---|
| | | **Newcastle disease** | **Infectious bursal disease** | **Infectious bronchitis** |
| **1** | Lasalocid Na+ BMD | $64.00^{bc}$ | $1788.53^{b}$ | $70.40^{ab}$ |
| **2** | Lasalocid Na+ anise seeds | $98.67^{a}$ | $1890.07^{a}$ | $84.27^{a}$ |
| **3** | 3-Nitro+BMD | $60.27^{bc}$ | $1748.60^{b}$ | $70.40^{ab}$ |
| **4** | 3-Nitro+anise seeds | $97.07^{a}$ | $1824.40^{ab}$ | $81.07^{a}$ |
| **5** | Control (Basal diet) | $53.33^{c}$ | $1708.33^{b}$ | $65.07^{c}$ |
| | ± ***SE*** | 16.519 | 73.549 | 13.213 |

[a-c] Means with different superscripts in the same column are significantly different at $P<0.05$.

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# تأثير الاستعاضة عن محفز النمو (باستيريسن مثيلين داي سلسليت) باليانسون في أداء دجاج اللحم

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## ملخـص

تمت دراسة تأثير الاستعاضة عن باستيريسن مثيلين داي سلسليت (محفز نمو من المضادات الحيوية) باليانسون كمحفز نمو طبيعي في أداء دجاج اللحم. تمت تغذية صيصان لحم بعمر يوم واحد على علائق التجربة لمدة 42 يوماً. أظهر استخدام اليانسون بدلاً من باستيريسن مثيلين داي سلسليت زيادة معنوية ($P<0.05$) في كل من وزن الجسم الحي والوزن المكتسب وكذلك مستوى المناعة لأمراض النيوكاسل والجمبورو والتهاب الجهاز التنفسي. كما أظهرت النتائج تحسناً معنوياً ($P<0.05$) في كفاءة التحويل الغذائي. وكان هناك انخفاض معنوي في تركيز الكوليستيرول والدهون الثلاثية في حين ارتفع تركيز الكوليستيرول الحميد (HDL). شهد مستوى المناعة تحسناً معنوياً ضد أمراض النيوكاسل والجمبورو نتيجة لتغذية الصيصان عليقة تحتوي على اليانسون في حين لم يشهد مستوى المناعة ضد مرض التهاب الشعب المعدي أي فرق بين اليانسون والباستيريسن مثيلين داي سلسليت، وشوهدت الفروق فقط مع مجموعة الشاهد. وعليه، فإنه يمكن استخدام اليانسون كمحفز نمو طبيعي بديلاً عن المضادات الحيوية.

**الكلمات الدالة**: دجاج لحم، محفزات نمو، يانسون، باستيريسن مثيلين داي سلسليت.

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