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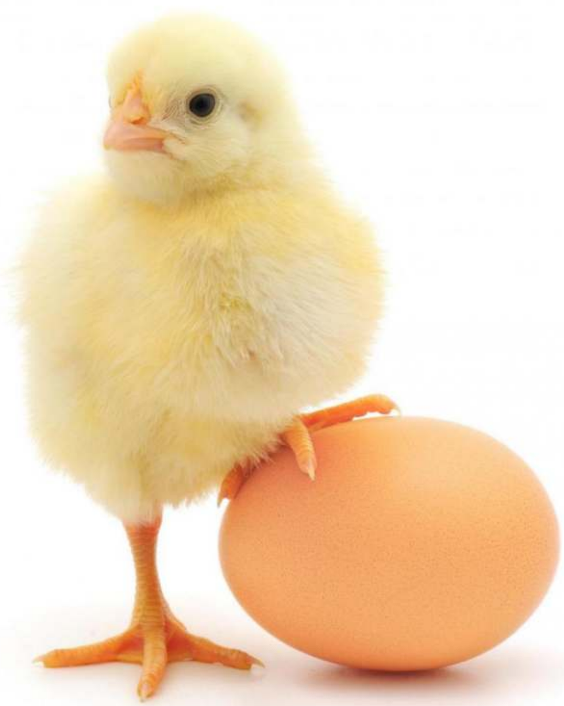

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# The Influence of Biological Feed Additives on Broiler Performance

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**Abstract:** An experiment was designed to evaluate the effect of some natural feed additives as alternatives of antibiotic growth promoters for broiler chicks. A total number of 1188 unsexed one-day old Ross 308 chicks were used. Two trials were conducted simultaneously using the same control treatments, a negative control (NC) with no feed additive and a positive control (PC) supplemented with Enramycine (0.025%) as antibiotic growth promoter. In the fist trial, natural mineral clay commercial products of Diatomaceous earth[1], at 0.2% (DE0. 2%) and 0.5% (DE0. 5%) and Farmagulator[2], at 0.125% (Fg. 125%) and, 0.25% (Fg 0.25%) were compared with the control treatments. In the 2[nd] trial, pre/pro-biotics commercial products of Diamond VXPC[3], 0.075% (DV 0.075%) and 0.10% (DV 0.10%); Tomoko[4], 0.05% (Tom 0.05%) and 0.1% (Tom 0.1%); and Primalac[5], 0.15% (PL 0.15) were compared with the two control treatments. Diets were formulated to meet nutrient requirements of chicks recommended for Ross 308. Results indicated that performance and immune response of the chicks fed diets containing pro/pre-biotic products were higher than those fed diets containing natural mineral clay products. The positive control or the treated group significantly recorded higher body weight than the negative control, with an exception of Diatomaceous earth treatment. Diatomaceous earth treatments (0.2% or 0.5%) significantly recorded the worst performance and the lowest economical efficiency compared to the negative control or the other treatments. From commercial point of view, biological feed additives of pre/pro-biotics or natural mineral clay as alternative of antibiotic growth promoter are beneficial with exception of Diatomaceous earth. Diets supplemented with biological feed additives enhanced the digestibility coefficients of most nutrients better than the controls, however, DE lowered the digestibility coefficients of most nutrients. It is concluded that performance and immune response of chicks fed on biological feed additives were equivalent or even superior to that of antibiotic growth promoter except for Diatomaceous earth. The tested materials were superior to the negative control when added to the diet in the term of performance.

**Key words:** Probiotic, prebiotic, diatomaceous, humic acid, broiler performance

## Introduction

During the decades, antibiotics have widely been used in poultry production as a growth promoter to enhance the performance. However, in, 2006, EU and many countries have banned using antibiotics as growth promoter in animal nutrition. This action encourages many investigators to search for alternatives to enhance performance. Patterson and Burkholder (2003) referred to an alternative approach to sub-therapeutic antibiotics in livestock is the use of probiotic micro-organisms, prebiotic substrates that enrich certain bacterial populations, or synbiotic combinations of prebiotics and probiotics. Probiotic (direct-fed microbials) is a generic term and products can contain bacterial cultures that stimulate micro-organisms capable of modifying the gastrointestinal environment to favor health status and improve feed efficiency (Dierck, 1989). Several studies have shown that addition of probiotics to the diet of broiler and turkey leads to improve the performance (Vicente *et al.*, 2007). Prebiotics have been defined as non digestible feed ingredients, which are growth substrates, specifically directed towards potentially beneficial bacteria already existing in caecum and colon. Several studies have shown that addition of prebiotics to the diet of broiler leads to improved performance through improving gut microflora (Xu *et al.*, 2003). The application of probiotics and prebiotics significantly improved the weight gain of broiler chickens (Mateova *et al.*, 2008). Most of the plant feed ingredients contain non-digested part (Cellulose, xylose arabinose, galactonic

[1]Composed of microscopic skeletal of unicellular alga-like plant called diatoms.
[2]Humates are formed from decayed plant matter, the composition of humates includes humus, humic acid, fulvic acid, ulmic acid, and trace minerals.
[3]Fermentation metabolities of yeast cells.
[4]Feed additive produced by fermentation using Koji (Aspergillus awamori), containing enzymes and active Aspergillus awamori mycelium.
[5]Lactobacillus acidophilus fermentation product, Minimum guarantee of 1.0 x 109 C.F.U./g Lactobacillus acidophilus.

acid) and some anti-nutritive factors which inhibit feed utilization and birds performance. These adverse effects can be overcome by supplementation of exogenous carbohydrase (Xylanase) enzymes which have been shown to lower viscosity of intestinal contents and to improve digestibility of starch, protein, fat and AME in broiler fed on diets containing wheat (Bedford, 1995). Exogenous enzyme may be used in broiler diet by proper selection of enzyme to get best result (Alam *et al.*, 2003). A number of research studies with broilers and ducks showed some improvements in feed conversion, body weight, uniformity and nutrient digestibility (Burrows *et al.,* 2002). Tomoko is a natural product of Koji, a Japanese fermentation process, used as broiler feed additive, is prepared by a fermentation technique, its composition is enzymes (Acidic Protease, a-amylase, Phytase, Glucoamylase, Cellulase) and Aspergillus Awamori cells. Tomoko is claimed to improve the performance of broiler when fed corn soybean meal diet and this effectiveness was not negatively affected by the heat treatment (Saleh *et al.,* 2006). Humates, a part of fertilizers are derived from plant matter decomposed by bacteria and contain humus, humic acid, fulvic acid, ulmic acid and some micro-elements (Stevenson, 1994). Body weights and feed conversions of broilers were significantly improved by the dietary humate treatments (Islam *et al.,* 2005). Diatomaceous earth (DE) is an inert dust formed by milling of fossilized remains of phytoplankton (diatoms), composed of silicon dioxide, commonly used for the control of insects infesting stored products (Alves *et al.,* 2006). Nutritionists indicate that DE improves the performance of livestock. Gao *et al.* (2002) concluded that adding suitable levels of diatomaceous earth in swine diets may effectively decrease volatile sulfide-associated odor and pollution to the environment. Fanatico (2006) reported that Producers sometimes give diatomaceous earth (DE) to the birds in the belief that the sharp edges of the fossilized diatoms will damage the parasites and reduce coccidiosis; however, there is no scientific data to support its use. The present work was carried out to study the effect of some natural feed additives as alternatives of antibiotic growth promoters on the broiler performance.

## Materials and Methods

A total number of 1188 unsexed one-day old Ross 308 chicks were used, the birds were weighed individually at the commencement of the experiment and those with extreme body weight were eliminated. Chicks were wing-banded and distributed randomly into 11 treatments of 108 chicks each in three equal replicates. All birds were housed in floor pens that located in semi-closed house with climate control condition. They kept under similar condition of management throughout the experimental period. Artificial lighting was used to provide chicks with 24 hours lighting daily during the whole experimental period. Initial brooding temperature was 33°C in the first week of age and reduced gradually 2°C per week up to 24°C. Diets and water were provided *ad libitum* all over the experimental period, which lasted for 35 days of age. The experiment divided into two trials (Trial 1 involved supplemental commercial natural mineral clay products and Trial 2 involved commercial pre/pro-biotic products) with negative control (with no additions) and positive control (received antibiotic growth promoter, Enramycine, 0.025%), the experimental treatments were illustrated in (Table 1). The compositions of the basal diets are illustrated in (Table 2). Diets were formulated using linear programme of least cost to meet nutrient requirements of chicks recommended to Ross 308 management guide (2006). At the end of the experimental period, a slaughter test was performed on 66 birds (6 birds from each treatment). The digestibility coefficients of nutrients of the experimental diets were examined at the end of the experimental period (5 weeks of age). Faecal nitrogen was determined according to the method outlined by Jakobsen *et al.* (1960), while the urinary organic matter fraction was calculated according to Abou-Raya and Galal (1971). The proximate analysis of feed and dried excreta were carried out according to the official methods (AOAC, 1990). Antibody production against SRBC was measured using microtitration-hemmagglutination technique with microtiter plate U-shape of 96 wells (8 row x 12 column) according to Bachman and Mashaly (1986). Individual blood samples were taken from jugular vein of 3 male birds within each treatment at 5 weeks of age. Serum was separated for determination of total lipids, cholesterol, transaminases (GOT and GPT), alkaline phosphates, creatinine, total protein, albumin and globulin, which were colorimetrically determined using commercial kits purchased from Bio-Merieux (France), following the same steps as described by manufactures. Data were subjected to the analysis of variance using General Linear Model (GLM) of SAS programme SAS ® software (1996). One way analysis of variance was carried out using the following model:

$$Y_{ij} = \mu + T_i + E_{ij}$$

Where $Y_{ij}$ = individual observation, μ = overall mean, $T_i$ = the effect of treatments, $E_{ij}$ = the experimental random error. Differences among treatments means were separated by Duncan's multiple rang test (Duncan, 1955).

## Results and Discussion

**Trail 1: Natural mineral clay commercial products as feed additives for broilers:** The performances of chicks fed experimental trail 1 diets are summarized in (Table 3). The initial live body weights of experimental chicks

Table 1: The experimental treatments (design)

| No | Abbr. | Treatment | Description | Active Form and mode of action | Inclusion rate |
|---|---|---|---|---|---|
| 1 | NC | No Additives | Negative control | | 0 |
| 2 | PC | Enramycine[1] | Antibiotic growth promotor | Enramycin produced by streptomyces fungicidus, Polypeptides antibiotic. Active on gram + bacteria, Clostridium, Streptoccocus, necrotic enteritis. Antibacterial activity inhibits bacteria wall. The major constituent is Enramycin A ($C_{107}$ $H_{138}$ $N_{26}$ $O_{31}$ $C_{l2}$), Enramycin B ($C_{106}$ $H_{140}$ $N_{26}$ $O_{31}$ $C_{l2}$) | 250 gm/Ton |
| Natural mineral clay commercial products | | | | | |
| 3 | DE 0.2% | Diatomaceous earth[2] | Composed of microscopic skeletal of unicellular alga-like plant called diatoms. | Chemical Name: Diatomaceous silica<br>Chemical Formulas: $SiO_2$*$nH_2O$ | 0.2% |
| 4 | DE 0.5% | | | | 0.5% |
| 5 | Fg 0.125 | Farmagulator[3] | Humates are formed from decayed plant matter. The composition of humates includes humus, humic acid fulvic acid, ulmic acid and trace minerals. | Humic acid have medium molecular size and their molecular weight is around 5,000 to 100,000 Dalton. Oxygen represents 33-36%, while nitrogen represents 4% in this substance. | 1250 gm/Ton |
| 6 | Fg 0.250 | | | | 2500 gm /Ton |
| Pre/pro-biotics commercial products | | | | | |
| 7 | DV0.075 | Diamondv XPC[4] | Fermentation metabolites of yeast cells, Saccharomyces cerevisiae yeast grown on a media of processed grain by-products, roughage products, cane molasses, malt and corn syrup. | Direct-fed microbials (DFM) are defined as microorganisms or their products that contribute to intestinal microbial balance. Modes of action of DFM include inhibition of pathogen growth in the gastrointestinal tract competitive exclusion increased mucosal villus heights and deeper crypts in the ileum, increases in digestive enzyme activity and increased antibody (IgA and IgG). | 750 gm/Ton |
| 8 | DV 0.10 | | | | 1000 gm/Ton |
| 9 | Tom 0.05 | Tomoko[5] | Dried Aspergillus Awamori fermentation product | | 500 gm/Ton |
| 10 | Tom 0.10 | | | | 1000gm/Ton |
| 11 | PL0.15 | Primalac[6] | Lactobacillus acidophilus fermentation product dehydrated. Minimum guarantee of 1.0 x $10^9$ CFU/g Lactobacillus acidophilus | | 1500 gm/Ton |

[1]Takeda Chemical Industries, LTD. Osaka, Japan, [2]Diatomate, Moputo, Mocambique, [3]Farmavet. International Inc, Kocaeli 41 400, Turkey., [4]Diamond V®, Cedar Rapids, Iowa, USA. [5]Biogenkoji Research Institute, 876-15, Mizobe, Kagoshima, Japan, [6]Star-Labs/Forage Research, Inc.

were almost alike with a little bit difference indicating the well randomization way for distributing chicks within the experimental treatments. Chicks fed diets containing Farmagulator 0.25% (Fg 0.25%), Farmagulator 0.125% (Fg 0.125%) and Enramycine, 0.025% as a positive control (PC) were significantly recorded the highest live body weight, being 1678, 1646 and 1630 gm, respectively. However, chicks fed diets containing Diatomaceous earth 0.5% (DE 0.5%) were significantly recorded the lowest values of live body weight (1449 gm). No significant differences between chicks fed diets containing Diatomaceous 0.2% (DE 0.2%) and the negative control were observed. Feed intake (FI) of chicks fed Diatomaceous 0.2% and 0.5% significantly higher than that of the control treatment. Feed conversion (FC) from chicks fed on diets containing Fg 0.25% and Fg 0.125% was significantly the best feed conversion (1.78 and 1.87, respectively). However, diets containing DE 0.5% or DE 0.2% were recorded significantly the worst feed conversion (2.28 and 2.19, respectively). These results agreed with Islam *et al.* (2005) who indicated that replacing antibiotic with humic acid (HA) as growth promoter in animal feed does not cause any loss in the performance of animals. He reported there is no doubt that HA has many beneficial effects like anti-bacterial, anti-viral and anti-inflammatory in animals that improves immune system, stress management and reduce odour in faeces. Humic acid has positive effect on liver functioning. Ultimately reduces mortality and increases growth in poultry. Although there is not enough evidence to hypothesize how humates promote the growth in broilers, it is assumed that humates might increase the uptake of nitrogen, phosphorus and other nutrients due to their chelating properties. This assumption needs to be further validated in poultry (Kocabagli *et al.,* 2002). The results reported herein were in accordance with the reports of Miles *et al.* (2006) who reported that birds given antibiotic were significantly heavier than those fed control diet. However, Sun *et al.* (2005) indicated that feeding birds on diets containing no antibiotic growth promoters resulted in higher mortality and decreased growth performance than did feeding a diet with an antibiotic. Mortality rate was within the normal range for

Table 2: The Composition and Chemical of the basal Diets

| Ingredients | Pre-starter 0-7 days | Starter % 8-18 days | Grower % 19-35 days |
|---|---|---|---|
| Yellow corn | 53.40 | 57.93 | 59.625 |
| Soybean meal (48%) | 40.00 | 35.00 | 32.45 |
| Corn oil | 2.50 | 3.0 | 4.00 |
| Di-calcium phoshate | 1.70 | 1.7 | 1.50 |
| Salt (Na Cl) | 0.35 | 0.35 | 0.35 |
| Premix[1] | 0.20 | 0.20 | 0.20 |
| Limestone | 1.30 | 1.30 | 1.40 |
| DL-Met. 99% | 0.25 | 0.22 | 0.175 |
| L- Lys-HCL. 78.4% | 0.20 | 0.20 | 0.20 |
| Choline chloride 50% | 0.10 | 0.10 | 0.10 |
| Total | 100 | 100 | 100 |
| Calculated: | | | |
| Crude protein % | 24.00 | 22.16 | 21.06 |
| Metabolizable energy (ME Kcal/Kg diet) | 3001 | 3075 | 3155 |
| Metabolizble energy (Kilojoules/Kg diet) | 12.55 | 12.86 | 13.20 |
| Crude Fiber% | 2.91 | 2.77 | 2.69 |
| Crude Fat% | 4.42 | 4.89 | 5.69 |
| Total P% | 0.78 | 0.78 | 0.73 |
| Available P% | 0.47 | 0.46 | 0.42 |
| Calcium % | 1.05 | 1.04 | 1.02 |
| Lysine % | 1.46 | 1.33 | 1.25 |
| Methionine % | 0.6 | 0.54 | 0.48 |
| Methionine + Cystine % | 0.97 | 0.89 | 0.82 |
| Threonine% | 0.92 | 0.84 | 0.80 |
| Tryptophan% | 0.29 | 0.26 | 0.25 |
| Arginine% | 1.64 | 1.47 | 1.39 |
| Isoleucine% | 1.08 | 0.98 | 0.93 |
| Na% | 0.16 | 0.16 | 0.18 |
| Price /kg (LE)[2] | 1.400 | 1.390 | 1.370 |

Each 2 kg of Vit. and Min. Mixture contains: Vit. A 12000, 000 IU, Vit. $D_3$ 2200, 000 IU, Vit. E 10, 000 mg, Vit. $K_3$ 2000 mg, Vit. $B_1$ 1000 mg, Vit. $B_2$ 5000 mg, Vit. $B_6$ 1500 mg, Vit. $B_{12}$ 10 mg, Pantothenic acid 10,000 mg, Niacin 30, 000 mg, Folic acid 1000 mg, Biotin 50 mg, Manganese 100, 000 mg, Zinc 80, 000 mg, Copper 10, 000 mg, Iron 50, 000, Iodine 1000 mg, Selenium 150 mg, Cobalt 100 mg, Ca $CO_3$ to 2, 000 gm. [2]LE Egyption pound, 1 $ = 5.40 LE

all experimental treatments except for chicks fed DE recorded the highest mortality rate, this could be as a result of contamination of clay by heavy metals (Stanley *et al.,* 2004). Percentages of dressing and abdominal fat for chicks fed different treatments are presented in (Table 3). Feeding diets with Fg 0.25%, PC or Fg 0.125% were significantly recorded the highest value for dressing percentages. No significant differences between chicks fed diet containing NC, DE 0.2% or DE 0.5% in the term of dressing weight were noticed. Negative control and PC significantly recorded the height values of abdominal fat, being 2.44 and 2.33% respectively, without significant differences between the other treatments. The results were supported by Karaoglu *et al.* (2004) who reported that humate supplementation to diets of broilers had no effect on performance, slaughter and carcass characteristics, a slightly improvement was observed in FCR for group fed with diet containing 0.1% humate. The effect of supplementing different biological additives on the immune response are illustrated in (Table 3). Female chicks fed diet containing Fg 0.250%, PC or Fg 0.125% were significantly recorded the highest antibody titer against SRBC, namely 8.66, 8.50 or 8.36, respectively. While Females fed diets with DE 0.5%, DE 0.2% or NC were significantly recorded the lowest antibody titer against SRBC, being 7.16, 7.33 or 7.85, respectively. Males fed diet containing Fg 0.125%, PC or Fg 0.25% were significantly recorded the highest antibody titer against SRBC, being 8.33, 8.07 and 7.83, respectively. Males fed diets supplemented with DE 0.5% or DE 0.2%, were significantly recorded the lowest antibody titer against SRBC, being 6.70 and 6.90, respectively. Generally, the female chicks revealed higher antibody titer against SRBC than the males. Increasing the levels of additives had no significant effect on the antibodies titer (DE 0.2% and DE 0.5% or Fag 0.125 and Fg 0.250). The results agreed with Belay *et al.* (1992) who reported no effect of dietary virginiamycin on the total, IgG, nor IgM antibody titers in 7 week old broilers. The influences of different experimental dietary treatments on economic efficiency (EE) measured by feed cost/kg dressed weight relative to control of broiler chicks are summarized in (Table 3). Chicks fed on diets containing Fg 0.25% or Fg 0.125% achieved the best economic efficiency 87% and 92%, respectively, while the worst economic efficiency achieved by DE 0.5% (112%) followed by DE 0.2% (108%). The economic efficiency of chicks fed on

Table 3: Performance, Immune response, Carcass characteristics and relative economic efficiency of broiler chicks at 35 days of age fed different natural feed additive

| Treatment | Initial body weight, gm | Average body weight[1], gm | Body weight gain, gm | Feed intake gm | Feed conversion, g/g | Mortality%[2] |
|---|---|---|---|---|---|---|
| Trail 1 | Natural mineral clay commercial products | | | | | |
| NC | 39.3 | 1570±11$^{b}$ | 1531±11$^{b}$ | 3086±21$^{bc}$ | 2.01±0.01$^{b}$ | 3.70 |
| PC | 39.2 | 1630±21$^{a}$ | 1591±21$^{a}$ | 3142±39$^{b}$ | 1.97 | 3.70 |
| DE 0.2% | 39.3 | 1562±19$^{b}$ | 1523±19$^{b}$ | 3331±49$^{a}$ | 2.19$^{b}$ | 4.62 |
| DE 0.5% | 39.2 | 1449±33$^{c}$ | 1410±33$^{c}$ | 3216±63$^{ab}$ | 2.28±0.01$^{a}$ | 4.62 |
| FG 0,125% | 39.2 | 1646±9.2$^{a}$ | 1607±9.2$^{a}$ | 3006±35$^{cd}$ | 1.87 | 0.00 |
| FG 0.25% | 39.3 | 1678±8.3$^{a}$ | 1639±8.3$^{a}$ | 2917±17$^{d}$ | 1.78±0.01$^{d}$ | 0.93 |
| Overall Means | 39.3 | 1589 | 1550* | 3116* | 2.02 | 2.54 |
| Probability | ND | 0.0001 | 0.0001 | 0.0001 | 0.0001 | ND |
| Trail 2 | Pre/Pro biotics commercial Products | | | | | |
| NC | 39.3 | 1570±11$^{c}$ | 1531±11$^{c}$ | 3086±21$^{a}$ | 2.01±0.01$^{a}$ | 3.70 |
| PC | 39.2 | 1630±21$^{b}$ | 1591±21$^{b}$ | 3142±39$^{a}$ | 1.97±0.01$^{a}$ | 3.70 |
| DV 0.075% | 39.4 | 1661±18$^{b}$ | 1622±18$^{b}$ | 2891±41$^{b}$ | 1.78±0.01$^{b}$ | 2.77 |
| DV 0.1% | 39.3 | 1705±14$^{a}$ | 1666±14$^{a}$ | 2879±28$^{b}$ | 1.73±0.01$^{c}$ | 1.85 |
| Tom 0.05% | 39.2 | 1642±04$^{b}$ | 1602±04$^{b}$ | 2937±62$^{b}$ | 1.83±0.04$^{b}$ | 1.85 |
| Tom 0.01% | 39.3 | 1652±15$^{b}$ | 1613±06$^{b}$ | 2907±49$^{b}$ | 1.80±0.01$^{b}$ | 2.77 |
| PL 0.15% | 39.2 | 1643±06$^{b}$ | 1603±06$^{b}$ | 2877±36$^{b}$ | 1.79±0.01$^{b}$ | 2.77 |
| Overall means | 39.3 | 1643 | 1604 | 3003 | 1.85 | 2.40 |
| Probability | ND | 0.0001 | 0.0001 | 0.0015 | 0.0001 | ND |

Table 3 Cont.

| Treatment | Immune response: Female | Immune response: Male | Carcass characteristics: Dressing % | Carcass characteristics: Abdominal fat % | Relative economic efficiency (EE)[4] |
|---|---|---|---|---|---|
| Trail 1 | Natural mineral clay commercial products | | | | |
| NC | 7.85±0.13$^{ab}$ | 7.40±0.21$^{bc}$ | 66.20±0.16$^{b}$ | 2.44±0.05$^{a}$ | 100 |
| PC | 8.50±0.17$^{a}$ | 8.07±0.09$^{a}$ | 67.03±0.09$^{a}$ | 2.33±0.03$^{ab}$ | 97 |
| DE 0.2% | 7.33±0.21$^{b}$ | 6.90±0.03$^{cd}$ | 66.09±0.12$^{b}$ | 2.30±0.06$^{b}$ | 108 |
| DE 0.5% | 7.16±0.18$^{b}$ | 6.70±0.23$^{d}$ | 66.35±0.20$^{b}$ | 2.26±0.02$^{b}$ | 112 |
| FG 0,125% | 8.36±0.43$^{a}$ | 8.33±0.29$^{a}$ | 66.94±0.18$^{a}$ | 2.30±0.04$^{b}$ | 92 |
| FG 0.25% | 8.66 ±0.23$^{a}$ | 7.83±0.20$^{ab}$ | 67.01±0.21$^{a}$ | 2.22±0.03$^{b}$ | 87 |
| Overall means | 7.98 | 7.55 | 66.61 | 2.31 | 97.6 |
| Probability | 0.0040 | 0.0004 | 0.0003 | 0.0116 | ND |
| Trail 2 | Pre/pro-biotics commercial products | | | | |
| NC | 7.85±0.13$^{c}$ | 7.40±0.21$^{b}$ | 66.20±0.16$^{b}$ | 2.44±0.05$^{a}$ | 100 |
| PC | 8.50±0.17$^{b}$ | 8.07±0.09$^{a}$ | 67.03±0.09$^{a}$ | 2.33±0.03$^{ab}$ | 97 |
| DV 0.075% | 9.00±0.11$^{ab}$ | 8.13±0.20$^{a}$ | 67.25±0.17$^{a}$ | 2.25±0.06$^{b}$ | 87 |
| DV 0.1% | 9.46 ±0.20$^{a}$ | 8.93±0.14$^{a}$ | 67.52±0.14$^{a}$ | 2.25±0.04$^{b}$ | 84 |
| Tom 0.05% | 8.66±0.29$^{b}$ | 8.23±0.17$^{a}$ | 67.20±0.12$^{a}$ | 2.25±0.04$^{b}$ | 90 |
| Tom 0.1% | 8.93±0.21$^{ab}$ | 8.33±0.18$^{a}$ | 67.47±0.14$^{a}$ | 2.23±0.03$^{ab}$ | 88 |
| PL 0.15 | 8.46±0.23$^{bc}$ | 8.07±0.20$^{a}$ | 67.25±0.19$^{a}$ | 2.24±0.03$^{b}$ | 88 |
| Overall means | 8.69 | 8.09 | 67.09 | 2.30 | 90.6 |
| Probability | 0.0024 | 0.0229 | 0.0001 | 0.0405 | ND |

a, b, Means with different superscripts within the same column are significantly different ( P = 0.0 5 %). [1]Initial body weight at 0 days was 39 gm ± S.E, [2]Mortality rate of the whole experiment was 2.44% (29/1188). [3]ND, Not statistically determined. [4]Assuming that the relative EE of control equal 100, economic efficiency as measured by feed cost/kg dressed weight. Negative control (NC), positive control Enramycine (PC), Diatomaceous earth 0.2% (DE 0.2%), Diatomaceous earth 0.5% (DE 0.5%), Farmagulator 0.125% (Fg 0.125%), Farmagulator 0.25% (Fg 0.25%), Diamond VXPC 0.075% (DV 0.075%), Diamond VXPC 0.01% (DV 0.1%), Tomoko 0.05% (Tom 0.05%), Tomoko 0.1% (Tom 0.1%), Primalac 0.15% (PL 0.15).

Farmagulator at either the two levels was better than those fed antibiotic growth promoter. The digestibility nutrients coefficients of the experimental diets are shown in (Table 4). Diets supplemented with DE lowered the digestibility of most nutrients. Digestibility coefficients of CP for chicks fed Fg 0.25% recorded the highest value (94.13%), while the negative control recorded the lowest value (90.70%). The effect of feeding diets with different feed additives on blood constituents are shown in (Table 5). Diets supplemented with Fg 0.125 or Fg 0.250 were significantly decreased blood cholesterol. Kidney functions (as measured by creatinine levels) were significantly decreased by supplemented Fg at a rate of 0.125 or 0.25%. Stepchenko *et al.* (1991) observed positive changes in the values of total protein, mineral substances and total

Table 4: Digestion coefficients of the nutrients as affected by dietary feed additive supplementations

| Treatment | %DM | %OM | %EE | %CF | %CP | %NFE |
|---|---|---|---|---|---|---|
| Trail 1 | Naturalmineral clay commercial products | | | | | |
| NC | 79.03±0.91$^{bc}$ | 81.36±0.84$^{a}$ | 79.60±0.47$^{a}$ | 29.00±0.30$^{a}$ | 90.70±0.30$^{b}$ | 79.45±0.59 |
| PC (Enr) | 80.93±0.35$^{a}$ | 81.96±0.58$^{a}$ | 77.86±0.21$^{a}$ | 28.96±0.29$^{b}$ | 91.16±0.52$^{b}$ | 81.06±0.74 |
| DE0.2% | 77.60±0.32$^{cd}$ | 79.22±0.18$^{b}$ | 72.12±0.31$^{c}$ | 24.06±0.93$^{b}$ | 90.83±0.35$^{b}$ | 79.87±0.27 |
| DE 0.5% | 77.16±0.32$^{d}$ | 79.10±0.15$^{b}$ | 72.96±0.29$^{c}$ | 26.06±0.43$^{a}$ | 90.73±0.43$^{b}$ | 79.90±0.70 |
| Fg 0,125% | 80.50±0.10$^{ab}$ | 81.26±0.59$^{a}$ | 79.50±0.30$^{a}$ | 28.30±0.42$^{a}$ | 92.14±1.12$^{b}$ | 81.03±0.81 |
| Fg 0.25% | 80.03±0.40$^{ab}$ | 80.39±0.51$^{ab}$ | 75.18±1.422$^{b}$ | 28.40±0.37$^{a}$ | 94.13±0.29$^{a}$ | 80.63±0.50 |
| Overall mean | 79.21 | 80.55 | 79.21 | 27.56 | 91.62 | 80.33 |
| Probability | 0.0004 | 0.0111 | 0.0001 | 0.0002 | 0.0070 | 0.384 |
| Trail 2 | Pre/pro-biotics commercial products | | | | | |
| NC | 79.03±0.91 | 81.36±0.84$^{abc}$ | 79.60±0.47$^{a}$ | 29.00±0.30 | 90.70±0.30 | 79.45±0.59 |
| PC (Enr) | 80.93±0.35 | 81.96±0.58$^{ab}$ | 77.86±0.21$^{ab}$ | 28.96±0.29 | 91.16±0.52 | 81.06±0.74 |
| DV0.075% | 80.16±0.78 | 82.53±0.62$^{a}$ | 76.29±1.10$^{ab}$ | 29.18±1.03 | 92.44±1.15 | 80.65±0.85 |
| DV 0.1% | 80.96±0.78 | 82.20±0.70$^{ab}$ | 79.06±0.68$^{a}$ | 29.60±0.93 | 93.20±0.98 | 81.18±0.43 |
| Tom 0.05% | 79.66±0.99 | 80.40±0.57$^{bc}$ | 75.34±1.63$^{b}$ | 27.40±0.70 | 91.76±0.49 | 80.27±0.63 |
| Tom 0.1% | 79.86±1.34 | 79.73±0.56$^{c}$ | 76.63±1.44$^{ab}$ | 28.80±0.88 | 92.60±1.21 | 80.54±0.54 |
| PL0.15 | 80.56±0.46 | 79.54±0.46$^{c}$ | 77.96±0.82$^{ab}$ | 29.60±0.52 | 92.84±1.52 | 80.68±0.92 |
| Overall mean | 80.17 | 81.11 | 77.55 | 28.86 | 92.10 | 80.55 |
| Probability | 0.6705 | 0.0201 | 0.1093 | 0.5155 | 0.5320 | 0.6975 |

a, b, Means with different superscripts within the same column are significantly different ( P = 0.05%). Negative control (NC), positive control Enramycine (PC), Diatomaceous earth 0.2% (DE 0.2%), Diatomaceous earth 0.5% (DE 0.5%), Tomoko 0.05% (Tom 0.05%), Farmagulator 0.125% (Fg 0.125%), Diamond VXPC 0.075% (DV 0.075%), DiamondVXPC 0.1% (DV 0.1%), Tomoko 0.1% (Tom 0.1%), Farmagulator 0.25% (Fg 0.25%), Primalac 0.1% (PL 0.15).

lipids in blood sera, liver, breast and leg muscles of chicken broilers after long-term feeding of a peat preparation (humic substances constitute 60 to 80% of peat). Feeding DE were significantly increased blood cholesterol, these results disagreed with Wachter *et al.* (1998) who indicated that diatomaceous earth is capable of reducing blood cholesterol and positively influencing lipid metabolism in humans.

**Trail 2: Pre/pro-biotic commercial products as feed additives for broilers:** The performances of chicks fed experimental trail 1 diets are summarized in (Table 3). The initial live body weights of experimental chicks were almost alike with a little bit difference indicating the well randomization way for distributing chicks within the experimental treatments. Chicks fed diets containing Diamond VXPC, 0.1% (DV 0.1%) were significantly recorded the highest body weight followed by Diamond VXPC, 0.075% (DV 0.075%), (Tom 0.1%), (PL 0.150), (Tom 0.05%) and Enramycine (positive control) in a decreasing order with insignificant differences among treatments. However, chicks fed diets containing Negative control (NC) were significantly recorded the lowest values of live body weight, being 1570 gm. Feed conversion of chicks fed diets containing DV 0.1%, were significantly recorded the best value of feed conversion being, 1.73 followed by (DV 0.075%), (PL 0.150), (Tom 0.1%) and (Tom 0.05%), respectively. The negative and positive controls were significantly recorded the worst feed conversion. The results of Diamond VXPC were in a good agreement with those of Banerjee and Pradharn (2006) who reported that live yeast supplementation to broiler chicks significantly improved feed conversion ration. Saleh *et al.* (2006) concluded that Tomoko is effective to improve the performance of broiler. The performance was in a good agreement with Jalaludeen *et al.* (2005) who reported that the 0.025% probiotic supplemented birds had a significantly higher body weight and weight gain. On the contrary of Maiolino *et al.* (1992) reported no significant differences in weight gain of chicken given diet with or without Lactobacillus cultures. Variation in the efficacy of probiotic may be due to differences in microbial species or strains of micro-organism used or methods of preparing the supplement (Jin *et al.,* 1998). The significant improvement of body weight and feed conversion ratio could be attributed to the effect of probiotics which improve absorption of nutrients and depressed harmful bacteria that cause growth depression (EL-Nagmy *et al.,* 2007). Similar findings were reported by Mateova *et al.* (2008) who confirmed the favorable effect of probiotics, prebiotcs and potentiated probiotics on growth and state of health of broiler chickens. The reason of variable effect of biological additives may be confounded by variations in gut flora and environmental conditions (Mahdavi *et al.,* 2005). Mortality rate was within the normal range (Table 3). In the present study mortality of chicks fed diets containing probiotic was in good agreement with Vicente *et al.* (2007) who reported that probiotic culture were significantly reduced mortality compared to the control group. However, Manickam *et al.* (1994) observed that broiler livability was higher in control groups than probiotic fed group, these chicks had lower body weights and feed conversion compared with the other treatments. Percentages of dressing and abdominal fat were significantly affected by dietary treatments

Table 5: Blood characteristics of the nutrients as affected by dietary feed additive supplementations

| Treatment | Cholesterol (mg/dl) | Total lipids (mg/dl) | A/G ratio | Globulin (g/dl) | Albumin (g/dl) |
|---|---|---|---|---|---|
| Trial 1: Natural mineral clay commercial products | | | | | |
| NC | 138±1.5$^{a}$ | 551±4.9$^{a}$ | 1.2±0.02$^{b}$ | 1.6±0.02$^{a}$ | 1.9±0.01$^{bc}$ |
| PC (Enr) | 130±1.7$^{a}$ | 547±8.8$^{a}$ | 1.2±0.03$^{ab}$ | 1.6±0.03$^{a}$ | 1.9±0.02$^{b}$c |
| DE0.2% | 132±4.2$^{a}$ | 543±6$^{a}$ | 1.2±0.05$^{ab}$ | 1.6±0.05$^{a}$ | 1.9±0.03$^{bc}$ |
| DE 0.5% | 135±3.4$^{a}$ | 542±3.5$^{a}$ | 1.2±0.04$^{ab}$ | 1.6±0.04$^{a}$ | 1.9±0.03$^{bc}$ |
| Fg 0,125% | 117±4.4$^{b}$ | 497±4.4$^{b}$ | 1.3±0.05$^{a}$ | 1.6±0.05$^{a}$ | 2.1±0.05$^{a}$ |
| Fg 0.25% | 107±3.8$^{c}$ | 492±17$^{b}$ | 1.29±0.03$^{ab}$ | 1.6±0.23$^{a}$ | 2.06±0.09$^{ab}$ |
| Overall mean | 126.4 | 528.7 | 1.24 | 1.6 | 1.97 |
| Probability | 0.0002 | 0.0009 | 0.175 | 0.772 | 0.0445 |
| Trail 2: Pre/pro-biotics commercial products | | | | | |
| NC | 138±1.5 $^{a}$ | 551±4.9$^{a}$ | 1.2±0.02$^{c}$ | 1.6±0.02$^{a}$ | 1.9±0.01$^{c}$ |
| PC (Enr) | 130±1.7$^{b}$ | 547±8.8$^{a}$ | 1.2±0.03$^{bc}$ | 1.6±0.03$^{a}$ | 1.9±0.02$^{bc}$ |
| DV0.075% | 117±2.6 $^{cd}$ | 498±6.6$^{a}$ | 1.3±0.05$^{abc}$ | 1.6±0.02$^{a}$ | 2.1±0.07$^{abc}$ |
| DV 0.1% | 112±2.7$^{d}$ | 502±5.5$^{b}$ | 1.4±0.10$^{abc}$ | 1.6±0.08$^{a}$ | 2.2±0.08$^{a}$ |
| Tom 0.05% | 121±1.7$^{c}$ | 488±12$^{b}$ | 1.4±0.04$^{ab}$ | 1.6±0.03$^{a}$ | 2.1±0.06$^{a}$ |
| Tom 0.1% | 118±1.2$^{cd}$ | 497±8.2$^{b}$ | 1.4±0.07$^{abc}$ | 1.5±0.04$^{a}$ | 2.1±0.06$^{ab}$ |
| PL0.15 | 115±2.3$^{cd}$ | 498±7.5$^{b}$ | 1.4±0.04$^{a}$ | 1.6±0.05$^{a}$ | 2.2±0.06$^{a}$ |
| Overall mean | 122 | 512 | 1.3 | 1.57 | 2.07 |
| Probability | 0.0001 | 0.0001 | 0.904 | 0.9229 | 0.0031 |

Table 5 Cont.

| Treatment | Total protein (g/dl) | GPT (U/l) | GOT (U/l) | Alkaline phosphatase (IU/l) | Creatinine (mg/dl) |
|---|---|---|---|---|---|
| Trail 1: Natural mineral clay commercial products | | | | | |
| NC | 3.5±0.02$^{a}$ | 15±0.9$^{c}$ | 87±0.09$^{a}$ | 604±5$^{a}$ | 1.5±0.02$^{a}$ |
| PC (Enr) | 3.5±0.04$^{a}$ | 14±0.3$^{c}$ | 86±1.9$^{a}$ | 603±11$^{ab}$ | 1.5±0.02$^{a}$ |
| DE0.2% | 3.5±0.03$^{a}$ | 16±0.3$^{c}$ | 84±2.03$^{a}$ | 570±11$^{c}$ | 1.4±0.04$^{a}$ |
| DE 0.5% | 3.6±0.01$^{a}$ | 15±0.3$^{c}$ | 86±2.01$^{a}$ | 579±8$^{bc}$ | 1.5±0.08$^{a}$ |
| Fg 0,125% | 3.7±0.07$^{a}$ | 21±0.7$^{b}$ | 64±3.2$^{c}$ | 555±6$^{c}$ | 1.14±0.04$^{b}$ |
| Fg 0.25% | 3.7±0.11$^{a}$ | 24±1.2$^{a}$ | 75±2.8$^{b}$ | 566±5$^{c}$ | 1.11±0.01$^{b}$ |
| Overall mean | 3.56 | 17.61 | 80.27 | 580.4 | 1.36 |
| Probability | 0.1250 | 0.0001 | 0.0001 | 0.0029 | 0.0001 |
| Trail 2: Pre/pro-biotics commercial products | | | | | |
| NC | 3.5±0.02$^{b}$ | 15±0.9$^{d}$ | 87±0.1$^{a}$ | 604±5$^{a}$ | 1.5±0.02$^{a}$ |
| PC (Enr) | 3.5±0.04$^{b}$ | 14±0.3$^{d}$ | 86±1.9$^{a}$ | 603±11$^{b}$ | 1.5±0.02$^{a}$ |
| DV0.075% | 3.6±0.06$^{ab}$ | 19±0.6$^{c}$ | 64±1.2$^{d}$ | 566±10$^{b}$ | 1.3±0.07$^{b}$ |
| DV 0.1% | 3.8±0.08$^{a}$ | 23±0.1$^{d}$ | 64±1.2$^{d}$ | 597±6$^{a}$ | 1.2±0.05$^{b}$ |
| Tom 0.05% | 3.7±0.02$^{a}$ | 20±1.2$^{c}$ | 59±0.6$^{e}$ | 555±9$^{b}$ | 1.4±0.08$^{b}$ |
| Tom 0.1% | 3.6±0.02$^{ab}$ | 24±0.3$^{ab}$ | 70±1.2$^{c}$ | 563±8$^{b}$ | 1.3±0.05$^{b}$ |
| PL0.15 | 3.8±0.10$^{a}$ | 26±1.5$^{a}$ | 75±1.5$^{b}$ | 563±8$^{b}$ | 1.1±0.03$^{b}$ |
| Overall mean | 3.64 | 20.2 | 72.10 | 579 | 1.33 |
| Probability | 0.0098 | 0.0001 | 0.0001 | 0.0013 | 0.0003 |

a, b, Means with different superscripts within the same column are significantly different ( P = 0.0 5 %). Negative control (NC) , positive control Enramycine (PC), Diatomaceous earth 0.2% (DE 0.2%), Diatomaceous earth 0.5% (DE 0.5%), Tomoko 0.05% (Tom 0.05%), Farmagulator 0.125% (Fg 0.125%), Diamond VXPC 0.075% (DV 0.075%), DiamondVXPC 0.1% (DV 0.1%), Tomoko 0.1% (Tom 0.1%), Farmagulator 0.25% (Fg 0.25%), Primalac 0.1% (PL 0.15).

(Table 3). Feeding negative control were significantly recorded the lowest value of dressing without significant differences among the other treatments. These results were confirmed by Abdulrahim *et al.* (1999) who studied the effect of Lacto-bacillus acidophilus and zinc bacitracin as dietary additives for broiler chickens. They observed that the combination treatment increased abdominal fat deposition. Abdel-Azeem (2002) indicated that carcass traits and internal organs were not affected due to addition of yeast culture to broiler diet. (Table 3) illustrated the effect of supplementing different treatments on the immune response. Female chicks fed diet containing DV 0.1%, DV 0.075% or Tom 0.10% were significantly recorded the highest level of antibody titer against SRBC, being 9.46, 9.00 and 8.93, respectively. While, NC or PL 0.15 were significantly recorded the lowest value, being 7.85 and 8.46, respectively. Generally, the females revealed higher antibody titer against SRBC than the males. No significant differences among treatments received different levels of the same feed additive DV 0.01%, DV 0.075% and Tom 0.05%, Tom 0.10%) were noticed. These results were in good agreement with AL-Homidan and Fahmy (2007) who reported that antibody titer concentration to Newcastle disease was significantly greater in broiler chicks supplemented with yeast culture (Diamond VXP) than the control treatment. Panda *et al.* (2000) compared the efficacy of a gut acidifier (GA), a probiotic (PB) and an antibacterial (AB) feed additive in female commercial broiler diets, Feeding PB or GA improved antibody titers against Newcastle disease and infectious bursal disease virus. Similar results were found by Kabir *et al.* (2002) who found that the antibody production in response to SRBC was significantly higher in experimental birds compared to the controls. The influences of different experimental dietary treatments on economic efficiency (EE) measured by feed cost/kg dressed weight relative to the control of broiler chicks are summarized in (Table 3). Treatment received DV 0.1% performed the best economic efficiency (84%), followed by DV 0.075% (87%), Tom 0.1 or PL 0.1 (88%) in decreasing order. While, the worst economic efficiency achieved by positive control (97%). All treatments achieved economical efficiency better than the negative control (NC) or those received antibiotic growth promoter. Tamilvanan *et al.* (2003) observed that the feed cost kg/live weight gain was less in case of probiotics fed broilers. The digestibility nutrients coefficients of the experimental diets are shown in (Table 4). There were no significant differences among treatments in the term of DM, CP, CF and NFE. The results were in a good agreement with Soliman *et al.* (2003) who reported that digestibility coefficients of ether extract (EE), crude fiber (CF) and crude protein (CP) were not affected significantly by yeast supplementation. The digestibility coefficients were on the contrary to the performance figures, these may be attributed to the level of drip water in the carcass, resulted in confusion for interpretation the clear difference of the figures of either digestibilities or performances. Abdel-Azeem (2002) indicated that addition of probiotic to broiler diet was significantly increased the digestibility coefficients of DM, OM, EE, CF and NFE. The levels of the blood constituents of different experimental treatments are listed in (Table 5). Diets supplemented with pre/pro-biotics significantly reduced blood cholesterol, total lipids and kidney functions (creatinine levels). The results were in a good agreement with Abdel-Azeem (2002) who found a slight reduction in blood cholesterol due to yeast supplementation. Mahdavi *et al.* (2005) realized that using different levels of probiotic caused significant decrease in plasma cholesterol, plasma triglyceride and egg cholesterol. Soliman *et al.* (2003) reported a slight reduction in blood cholesterol due to yeast supplementation. It is concluded that the biological feed additives used herein gave a similar or a better performance than the antibiotic growth promoter except for DE 0.5% or DE 0.2%. Besides, the economic efficiency of all treatments was superior to the antibiotic growth promoter with the exception of chicks fed DE 0.2% or DE 0.5%. Feeding chicks on diets containing antibiotic growth promoter had no superiority over diets containing biological feed additives such as probiotics or prebiotics in terms of body weight, feed conversion, mortality rate, carcass characteristics or immune response. However, taking into consideration, the possible risk of antibiotic growth promoter on human health. The overall mean of the experiment indicated that the performance and the immune response of the chicks fed diets containing yeast products were better than that fed diets containing clay products. Briefly, biological feed additives consider an alternative of antibiotic growth promoter with the exception of Diatomaceous earth.

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