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# PERFORMANCE OF BROILERS SUPPLEMENTED WITH A MICROBIAL PREPARATIONS

**Fahmy, M.O.**

**Faculty of Agric. and Veterinary Med., Dep. of Animal Prod. &Breeding, AL-Qasseem University, Buriedah P.O. Box 6622, Saudi Arabia.**

## ABSTRACT

An experiment was conducted to study the effects of effective microorganisms (EM) supplementation on broilers performance, immunity and intestinal gross anatomy. Two hundred 1-d-old broiler chicks were randomly assigned to equal five treatment groups A, B, C, D and E. Groups B and C were fed on starter and finisher diets supplemented with 0.4 and 0.6 % EM (Pokashi), while groups D and E were fed starter and finisher diets with the addition of 0.5 ml and 1 ml of Pokashi per liter of drinking water, respectively. Group A was used as control. The EM supplementation improved live body weight and gains during the starter period than those of the control. Feed conversion ratio was not affected during the starter period, but from 3 to 4 weeks of age, 0.4 and 0.6 % Pokashi supplementation to the diets improved ($P<0.05$) feed conversion ratio by 13 % and 15.69 % than the control group. After 42 days of EM supplementation, the absolute or relative values of carcass, gizzard, duodenum, pancreas, liver, small intestine, caecum, heart, head+legs and gastrointestinal tract were not significantly affected. All the nutritional groups reduced significantly ($P<0.05$) spleen weight index than the control group. Chicks of groups B,C and E had higher antibody titers ($P<0.05$) against $SRBC_S$ injection after the primary and secondary response than chicks of group A. Chicks in group B showed significant ($P<0.05$) increase in anti-NDV antibody level than the control. In conclusion, supplementing broiler's diet or drinking water with EM showed some beneficial effects on live body weight ,weight gain and immunity (especially in young growing birds).

**Keywords**: Broilers, performance, effective microorganisms

## INTRODUCTION

Due to their several negative effects, antibiotics gradually been replaced by natural alternatives in poultry production (Fuller, 1992). Using of these alternatives instead of antibiotics in poultry production prevent damage to normal intestinal micro flora, drug-resistance as well as residues in carcass. Majority of these natural products are based mainly on *Lactobacillus acidophilus*, although other organisms have been studied such as *Streptococcus faecium*, *Bacillus subtilis* and yeast (Cheeke, 1991). Effects of yeast on poultry production have been recently reported by Stanley *et al* ., (2004); and Al Homidan and Fahmy (2007). Some studies have confirmed the effects of yeast culture in increasing concentrations of commensal microbes or suppressing pathogenic bacteria (Stanley *et al*., 2004a).

Recently, effective microorganisms (EM) have been used to improve poultry performance in intensive poultry production through, reducing odor (Yongzhen and Weijiong 1994); helps balance of micro flora within the animal's digestive tract; increases the coefficient of nitrogen utilized by the bird (Yongzhen and Weijion 1994); improve bird's health and immunity (Anjum *et al* 1996). Pokashi is the commercial name of a multi-strain effective microorganisms used in poultry production. It contains naturally occurring

nine different species of beneficial microflora which are generally regarded as safe by the American Food and Drug Administration (Fuller, 1989). However, information on the use of this probiotic, its levels in broiler diets and its effect on broiler production are not yet understood.

This study was conducted to investigate the effect of an EM on live body weight, weight gain, feed intake, feed conversion, gastrointestinal tract gross anatomy and the immune system of a commercial meat type chicken reared under local conditions.

## MATERIALS AND METHODS

**Birds and Management**

Two hundred day-old commercial broiler chicks of mixed sexes were reared for 42 days during November and December 2007 at the Poultry Farm of the Agricultural Experimental Station, Faculty of Agriculture and Veterinary Medicine, AI-Qassim University, Saudi Arabia. Chicks were weighed individually, and then randomly assigned to equal five treatment groups A, B, C, D and E (each of four replicates)each replicate of 10 chicks. The chicks in each replicate were kept in separate pen measuring 1.5 m long, 1.5 m wide and 3.5 m heights. A layer of 8 cm. shaving woods was used as litter in each pen. Pens were disinfected and then fumigated before the start of the experiment. Litter in each pen was stirred regularly during the experiment to keep it dry. All the recommended practices for broiler rearing were followed throughout the experimental period. Experimental diets (mash) and water were offered *ad libitum*.

**Diets Preparation and Feeding**

Pokashi is the commercial name of effective microorganisms (EM) contains a multi-strain probiotic in either liquid or dry brown powder form (biofeed) containing 3 main genera: phototrophic bacteria, lactic acid bacteria and yeast, the two forms of Pokashi were used as feed supplementation in this experiment. Pokashi (EM) was supplied through EM Research Organization 2-9-2 Ganeko, Ginowan-Shi.Okinawa, 901-2214.Japan. Commercial starter and finisher diets were obtained from a commercial feed mill Co., at Qassim. The two forms of Pokashi (EM) were supplemented to the nutritional groups in the starter and finisher diets (groups B , C) or in the drinking water (Groups D ,E) as follows:

Group B: Addition of 0.4 % Pokashi (biofeed) to the starter and finisher diets.
Group C: Addition of 0.6 % Pokashe (biofeed) to the starter and finisher diets,
Group D: addition of 0.5 ml liqued form of Pokashi per liter drinking water and
Group E addition of 1.0 ml liquid form of Pokashi per liter drinking water.
Group A was used as control (no addition of Pokashi in the diet or in the water (0 EM). The chemical analysis for the starter and finisher diets and the composition of the nutritional diets are presented in Tables (1,2).

**Live body weight, Weight gain, Feed intake and Feed conversion ratio**

All the birds were weighed individually at weekly intervals until 6 weeks of age. Weekly body weight gain and feed consumption of each experimental unit was recorded on per pen basis and feed conversion ratio (FCR) (Feed intake/body weight gain) and feed intake per bird per day (feed

intake ÷ (bird no×7)) were calculated. Therefore, feed consumption and FCR calculations were made from the average values of each pen.

**Table (1): Chemical analysis of starter and finisher diets**

| Items | Diet | |
|---|---|---|
| | Starter | Finisher |
| **Corn yellow %** | 44.08 | 55.00 |
| **Soybean meal 44 %** | 43.40 | 35.00 |
| **Vegetable oil %** | 8.53 | 6.52 |
| **Bone meal %** | 2.50 | 2.0 |
| **Limestone %** | 0.50 | 0.8 |
| **NaCl %** | 0.30 | 0.3 |
| **Premix (vita.+mine.)%** | 0.30 | 0.3 |
| **Methionine %** | 0.39 | 0.08 |
| **Total** | **100** | **100** |
| **Calculated nutritive values** | | |
| **ME ( K.cal./Kg )** | 3200 | 3200 |
| **Crude protein %** | 21.70 | 20.40 |
| **Methionine + Cystine %** | 1.02 | 0.73 |
| **Lysine %** | 1.3 | 1.10 |
| **Calcium%** | 1.1 | 1.05 |
| **Available Phosphorus %** | 0.46 | 0.38 |

**Table (2): Composition of the experimental diets: Group A (starter & finisher),Group B (starter & finisher + 4 % EM biofeed) and Group C (starter & finisher + 6 % biofeed)**

| Items | Nutritional groups | | |
|---|---|---|---|
| | A | B | C |
| ***Starter Phase*** | | | |
| **% Dry matter** | 89.75 ± 6.5 | 88.65 ± 5.2 | 88.09 ± 6.30 |
| **% Crude protein** | 21.70 ± 4.1 | 20.87 ± 3.3 | 20.45 ± 4.31 |
| **% Crude fat** | 4.92 ± 1.2 | 4.73 ± 1.4 | 4.50 ± 2.1 |
| **% Crude fiber** | 3.34 ± 0.5 | 4.49 ± 0.5 | 4.74 ± 1.0 |
| **% Ash** | 5.03 ± 0.2 | 5.55 ± 0.2 | 6.11 ± 0.8 |
| **% Nitrogen free extract** | 65.01 ± 2.0 | 64.36 ± 1.2 | 64.20 ± 1.9 |
| **ME Kcal/Kg** | 3156 ± 141 | 3120 ± 163 | 3091 ± 135 |
| ***Finisher Phase*** | | | |
| **% Dry matter** | 89.67 ± 5.4 | 88.67 ± 4.3 | 88.47 ± 3.7 |
| **% crude protein** | 19.95 ± 3.7 | 19.90 ± 4.2 | 19.13 ± 3.9 |
| **% Crude fat** | 8.13 ± 1.8 | 8.01 ± 2.3 | 8.12 ± 2.8 |
| **% Crude fiber** | 3.55 ± 0.7 | 4.66 ± 0.9 | 4.76 ± 1.1 |
| **% Ash** | 6.09 ± 0.9 | 6.50 ± 1.1 | 6.87 ± 1.2 |
| **% Nitrogen free extract** | 62.28 ± 3.1 | 60.93 ± 4.2 | 61.12 ± 4.1 |
| **ME Kcal/Kg** | 3216 ± 120 | 3199 ± 140 | 3191 ± 125 |

**Slaughtering, Carcass and Gastrointestinal gross anatomy data**

At the end of the experiment, randomly twelve birds from each group (three per replicate) were starved over night, individually weighed then slaughtered by the Moslems method. Slaughtered birds were then

immediately eviscerated. Carcass weight and percentage as well as heart, liver, gizzard, pancreas, spleen and gastrointestinal gross anatomy were recorded as absolute amount and/or as percent live body weight. Gastrointestinal tract was used as it is without empting its content in the gross anatomy study. Carcass weight to live body weight percentages of chicks were calculated by the formula: Carcass weight / live body weight x 100. Carcass weight was recorded after removing of head, neck, feathers, lungs, heart, liver, spleen, tarsometatarsus, toes with feet and gastro-intestinal tract.

**Immune response**

Two immunization tests were carried out to evaluate the effects of Pokashi (EM) supplementation to the diets or to the drinking water on the immune system of the experimental chicks. Testing of chickens for NDV antibodies was done by the method of Allan and Gough (1974). To measure the immunity of the birds against Sheep Red Blood Cells (SRBCs), 10 chicks per group were injected intramuscularly with freshly prepared $SRBC_S$ (40% suspension in PBS, 0.5 ml/bird) at four weeks of age, following by a booster injection of SRBCs suspension at 6 wks of age. Seven days after each injection, blood samples were collected. Plasma from each sample was harvested and stored at -20$^o$C until tested. Titers were expressed as the log 2 of the reciprocal of the highest dilution given complete agglutination (Wegman and Smithes, 1966).

**Statistical Analyses:**

Data were analyzed using GLM procedure of SAS program (SAS, 1999) followed by testing of significant differences among treatments using LSR method (Duncan), The following model was applied for all studied traits:

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

Where:

$Y_{ij}$= Observation.

$\mu$ = Overall mean.

$T_i$ = Effect of $i^{th}$ treatment (out of five; control and 4 nutritional groups).

$E_{ij}$ = Random error.

## RESULTS AND DISCUSSION

**Live body weight and Mortality**

Live body weight and number of dead birds of the experimental broilers are given in Table 3. The data showed that only four birds died throughout the experimental period, tow birds died in group E at 3 and 5 weeks of age and the other two birds died at 6 weeks of age (one in group B and the other in group D). No mortality was recorded in groups A and C. Examination of the data in this table clearly showed that after one week of treatment, live body weight was significantly ($P<0.05$) greater in broilers given 1 ml of EM solution in the drinking water than the control and the other treatment groups. From the 2nd week post treatment onward to the 4th week of age, live body weight was significantly greater in broilers given 1ml EM solution or 0.4 % biofeed simultaneously than the control broilers ($P<0.05$). At the 5th week of age, broilers given 0.4 or 0.6% biofeed EM showed significant ($P<0.05$) live body weight than the control and the other treatment

groups. At the end of 42 days of treatment, live body weight was 4.23 % greater in broilers given 0.4% EM biofeed than (1949g compared with 1870 g) control broilers. The above mentioned growth stimulating effect of the probiotic Pokashi is in agreement with the results reported by several researchers on different probiotics. Kermanshahi and Rostami (2006), Thitaram *et al*. (2005) , Nayebpor *et al*., (2007) and Ashayerizadeh *et al*., (2009), reported that probiotics can improve the weight of birds. Wenk (2000) reported that probiotics supplementation had more pronounced effect in young growing animals which substantiates the findings of our study. In contrast, Midilli *et al*., (2008) reported that probiotic did not significantly affect broiler body weight. The stimulating effects of Pokashi on live body weight confirm growth promoting activity of EM as reported by Hussein and El-Ashry, 1991; and Hussain *et al*., (1996). The better weight gain in EM-treated broilers could be related to better digestibility of crude protein and crude fiber (Hussain *et al*., 1996). The Pokashi consumption enters a large amount of lactic acid bacteria in the broiler's gastrointestinal tract. These microorganisms by producing acids (such as acetic and lactic) and other compounds might cause an inhibition to the pathogenic bacteria growth and help the adhesion or colonization to the intestinal mucosa and rapid proliferation of beneficial bacteria in animal's intestine (Fuller, 1989). So that, probiotics by improving the microflora intestinal microbial balance has beneficial effects on growth performance. In addition, the probiotics increased available energy by increasing the digestibility of carbohydrates, improved the organic matters digestibility, increased the amylase enzyme activity (Jin *et al*., 2000). However, probiotics with making better microbial environment in intestine and by activating the enzymes in the feed and in the bird's gut help the digestion and absorption of nutrients, efficiency of utilization of feed and at last resulting in the improvement of the growth performance of birds.

**Table (3): Effect of supplementing effective microorganism's preparation on broilers body weight and mortality during the period from 1 to 6 weeks of age**.

| Age (Wks) | Parameter | Treatment | | | | | SE |
|---|---|---|---|---|---|---|---|
| | | Control (0 EM) | Effective microorganism added in | | | | |
| | | | Feed | | Water | | |
| | | | 0.4% | 0.6% | 0.5 ml | 1ml | |
| 1 | Body weight, g | 139.02$^{b}$ | 141.64$^{ab}$ | 148.14$^{ab}$ | 138.68$^{b}$ | 150.96$^{a}$ | 3.1 |
| | No of dead birds | 0 | 0 | 0 | 0 | 0 | |
| 2 | Body weight, g | 307.11$^{b}$ | 346.24$^{a}$ | 325.44$^{ab}$ | 323.08$^{ab}$ | 341.81$^{a}$ | 7.1 |
| | No of dead birds | 0 | 0 | 0 | 0 | 0 | |
| 3 | Body weight, g | 578.39$^{b}$ | 638.23$^{a}$ | 617.24$^{ab}$ | 603.08$^{ab}$ | 637.16$^{a}$ | 12.8 |
| | No of dead birds | 0 | 0 | 0 | 0 | 1 | |
| 4 | Body weight, g | 940.14$^{c}$ | 1052.44$^{a}$ | 1051.68$^{a}$ | 982.56 $^{b}$ | 1037.08$^{ab}$ | 20.2 |
| | No of dead birds | 0 | 0 | 0 | 0 | 0 | |
| 5 | Body weight, g | 1359.58$^{b}$ | 1489.60$^{a}$ | 1463.37$^{a}$ | 1356.41$^{b}$ | 1438.39$^{ab}$ | 29.1 |
| | No of dead birds | 0 | 0 | 0 | 0 | 1 | |
| 6 | Body weight, g | 1870.18$^{a}$ | 1949.29$^{a}$ | 1902.68$^{a}$ | 1826.47$^{a}$ | 1894.04 $^{a}$ | 38.3 |
| | No of dead birds | 0 | 1 | 0 | 1 | 0 | |

**Means in the same row with different superscripts are differ significantly at P ≤ 0.05**

**Feed intake, Weight gain and Feed conversion ratio**

Data in Table (4) clearly show that, there was no significant effect of Pokashi supplementation neither to broiler's diets nor to drinking water on feed intake or on feed intake/bird/day. The present results agree with (Watkins and Kratzer, 1983, 1984; Estrada *et al*., 2001; O'Dea *et al*., 2006; Midilli *et al*., 2008) and In contrast, with (Zulkifli *et al*., 2000; Kalavathy *et al*., 2003; Khaksefidi and Ghoorchi, 2006; Panda *et al*., 2006).

**Table (4): Effect of effective microorganisms supplementation on feed intake, weight gain, feed conversion and feed intake/bird/day**

| Period / week | Parameter | Treatment | | | | |
|---|---|---|---|---|---|---|
| | | Control 0 EM | Effective microorganism added in | | | |
| | | | Feed | | Water | |
| | | | 0.4% | 0.6% | 0.5 ml | 1ml |
| 1 – 2 | Feed intake(g) | 3159.50 $^{a}$ | 3477.50 $^{a}$ | 3305.25 $^{a}$ | 3463.75 $^{a}$ | 3443.00 $^{a}$ |
| | Weight gain(g) | 168.10 $^{c}$ | 204.61 $^{a}$ | 177.31 $^{bc}$ | 190.86 $^{ab}$ | 184.41 $^{bc}$ |
| | Feed conversion | 1.90a $^{a}$ | 1.70 $^{a}$ | 1.87 $^{a}$ | 1.82 $^{a}$ | 1.89 $^{a}$ |
| | Feed intake/bird/d(g) | 45.14 $^{a}$ | 49.68 $^{a}$ | 47.22 $^{a}$ | 49.48 $^{a}$ | 49.19 $^{a}$ |
| 2 - 3 | Feed intake(g) | 5604.00 $^{a}$ | 5595.25 $^{a}$ | 5990.25 $^{a}$ | 6018.50 $^{a}$ | 5449.75 $^{a}$ |
| | Weight gain(g) | 271.28 $^{a}$ | 291.98 $^{a}$ | 291.79 $^{a}$ | 295.35 $^{a}$ | 278.93 $^{a}$ |
| | Feed conversion | 2.07 $^{a}$ | 1.91 $^{a}$ | 2.05 $^{a}$ | 2.06 $^{a}$ | 2.01 $^{a}$ |
| | Feed intake/bird/d(g) | 80.06 $^{a}$ | 79.93 $^{a}$ | 85.57 $^{a}$ | 85.98 $^{a}$ | 77.85 $^{a}$ |
| 3 - 4 | Feed intake(g) | 8059 $^{a}$ | 7992.50 $^{a}$ | 8151.00 $^{a}$ | 7959.75 $^{a}$ | 7884.75 $^{a}$ |
| | Weight gain(g) | 361.75 $^{c}$ | 414.21 $^{ab}$ | 434.44 $^{a}$ | 399.92 $^{abc}$ | 379.48 $^{bc}$ |
| | Feed conversion | 2.23 $^{a}$ | 1.94 b $^{c}$ | 1.88 $^{c}$ | 2.00 $^{abc}$ | 2.15 $^{ab}$ |
| | Feed intake/bird/d(g) | 115.13 $^{a}$ | 114.18 $^{a}$ | 116.44 $^{a}$ | 113.71 $^{a}$ | 112.64 $^{a}$ |
| 4 - 5 | Feed intake(g) | 8594.00 $^{a}$ | 8954.00 $^{a}$ | 8890.75 $^{a}$ | 8830.25 $^{a}$ | 8690.75 $^{a}$ |
| | Weight gain(g) | 419.44 $^{ab}$ | 432.80 $^{a}$ | 411.70 $^{ab}$ | 403.60 $^{ab}$ | 373.84 $^{b}$ |
| | Feed conversion | 1.89 $^{b}$ | 2.12 $^{ab}$ | 2.20 $^{ab}$ | 2.24 $^{ab}$ | 2.40 $^{a}$ |
| | Feed intake/bird/d(g) | 122.77 $^{a}$ | 127.91 $^{a}$ | 127.01 $^{a}$ | 126.15 $^{a}$ | 124.15 $^{a}$ |
| 5 - 6 | Feed intake(g) | 12436.50 $^{a}$ | 11346.00 $^{a}$ | 11607.75 $^{a}$ | 11744.75 $^{a}$ | 11626.00 $^{a}$ |
| | Weight gain(g) | 510.60 $^{a}$ | 459.69 $^{a}$ | 439.31 $^{a}$ | 455.66 $^{a}$ | 470.06 $^{a}$ |
| | Feed conversion | 2.45 $^{a}$ | 2.65 $^{a}$ | 2.69 $^{a}$ | 2.66 $^{a}$ | 2.60 $^{a}$ |
| | Feed intake/bird/d(g) | 177.66 $^{a}$ | 162.09 $^{a}$ | 165.83 $^{a}$ | 167.78 $^{a}$ | 166.09 $^{a}$ |

**Means in the same row with different superscripts are differ significantly at P ≤ 0.05**

The results of this experiment demonstrated that birds given feeds or water containing EM (Pokashi) as an additive substance had a higher weight gain during the periods(1-2, 2-3, 3-4 weeks) than those of the control. In the meanwhile, this higher weight gain was significant (P<0.05) between groups B&C and D than the control group A during the period from 1to 2 weeks of age, and between groups B and C only than the control (A) during the period from 3 to 4 weeks of age. No significant differences were observed among treatments on weight gain during the period from 5 to 6 weeks of age.

Feed conversion ratio (amount of feed consumed to gain per unit body weight) was not affected by Pokashi supplementation to the diets or to the drinking water during the starter period (from 1 to 3 weeks of age). From 3 to 4 weeks of age, 0.4 and 0.6 % Pokashi supplementation to the diets improved significantly (P<0.05) feed conversion ratio, the more pronounced

improvement in feed conversion ratio was noticed in group C which was supplemented with 0.6 % EM piofeed in the diet (15.69 % lower than the control group). However, Pokashi supplementation to the drinking water has no significant effect on feed conversion ratio during this period. This finding is in agreement with Watkins and Kratzer, 1984 and Timmerman *et al* .2006 who reported that, administration of probiotics in the drinking water generally resulted in a lower increase of average daily gain when compared with studies with probiotic administration via the feed (Zulkifli *et al* ., 2000; Kalavathy *et al* ., 2003). However, during the period from 4 to 5 weeks of age, no significant difference was observed in the feed conversion ratio between the control group (A) and groups B,C and D, while group (E) which was supplemented with 1 ml of EM solution in the drinking water, feed conversion ratio increased significantly($P<0.05$) than the control group (+ 27 %). Thereafter, there was no significant difference between the control group and the treatment groups in the feed conversion ratio. The present results also agree with Zulkifli *et al*., (2000) who observed that broilers fed a diet containing LC consumed less feed and had better feed efficiency ratio during the growing period (1-21 days), and this superiority in food efficiency did not extend to the finishing period (22-42 days).

The results presented in Table (4) demonstrate that during the starter period, birds given feeds or water containing EM (Pokashi) as an additive substance had a higher performance than those of the control. Wenk (2000) reported that probiotics supplementation had more pronounced effect on young growing animals which substantiates the findings of our study during starter phase. The EM consumption by the birds during this period enters a large amount of lactic acid bacteria in the bird's gastrointestinal tract. These microorganisms by producing acids (i.e acetic and lactic) and other compounds cause to inhibit the pathogenic bacteria growth and help the adhesion or colonization to the intestinal mucosa and rapid proliferation of beneficial bacteria in animal's intestine (Fuller, 1989). So that, EM by improving the microflora intestinal microbial balance has beneficially effects on growth performance. Moreover, the EM increased available energy by increasing the digestibility of carbohydrates, improved the organic matters digestibility, increased the amylase enzyme activity (Jin *et al*., 2000). However, ME with making a better microbial environment in intestine and by activating the internal and external enzymes of animals help to digestion and absorption of nutrients, efficiency of utilization of feed and at last causing an improvement to the growth performance of birds (Nayebpor *et al* ., 2007). In older ages (finisher period), broiler's GIT of the control group is colonized with a diverse and dynamic microbial population. Immediately following hatching the GIT is sterile but becomes rapidly colonized by competing microbial populations and a stable population is only established until after day 49(Van Eys, 2006). Edens *et al*., (1997) showed that in ovo and ex ovo administration of *Lactobacillus reuteri* resulted in an increase villus height, indicating that probiotics are potentially able to enhance nutrient absorption and thereby improving growth performance and feed efficiency.

**Carcass and Gastrointestinal tract gross anatomy**

After 42 days of EM supplementation to broiler's diets or drinking water the carcass weight, gizzard length, duodenum length, duodenum weight, pancreas length, liver weight, small intestine length, small intestine weight, caecum length, caecum weight, rectum weight, heart weight, head+legs weight and gastrointestinal tract weight had values similar to these of control group.

In our experiment, EM supplementation reduced significantly ($P<0.05$) proventriculus length and spleen weight than the control group. However, 0.4 % EM supplementation to the diet and 0.5 ml EM supplementation to drinking water increased proventriculus weight than the control group. Moreover, supplementation of 0.4 % EM to the diet increased gizzard weight, pancreas weight and rectum weight significantly ($P<0.05$) than the control group.

The data on the effects of EM supplementation to the broiler's diets and drinking water on carcass and gastrointestinal tract organs as proportion to live body weight is given in Table 5. Examination of the data in this table clearly showed that most EM supplementation in this experiment had no significant effect on the weight index of the following parameters: gizzard, liver, small intestine, heart, head+legs, caecum and gastrointestinal tract.

**Table (5): Effect of effective microorganisms supplementation on live body Weight, carcass weight, head and legs weight and gastrointestinal tract gross anatomy**

| Trait | Treatment groups | | | | |
|---|---|---|---|---|---|
| | Control A (0 EM) | Effective microorganism added in | | | |
| | | Feed | | Water | |
| | | B (0.4%) | C (0.6%) | D (0.5 ml) | E(1ml) |
| **Live body weight, g** | 1962.00 [a] | 2052.50 [a] | 1977.50 [a] | 1904.50 [a] | 1929.00 [a] |
| **Carcass Weight, g** | 1553.10 [a] | 1630.60 [a] | 1565.00 [a] | 1536.10 [a] | 1419.50 [a] |
| **Proventriculus Length, cm** | 5.70 [a] | 4.80 [b] | 4.80 [b] | 5.60 a [b] | 5.00a [b] |
| **Proventriculus Weight, g** | 9.30 a [b] | 10.10 [a] | 7.80 [b] | 10.40 [a] | 7.80 [b] |
| **Gizzard Length, Cm** | 6.50 [a] | 6.80 [a] | 6.40 [a] | 6.80 [a] | 6.60 [a] |
| **Gizzard Weight, g** | 55.00 [b] | 64.60 [a] | 60.40 [a b] | 53.60 [b] | 53.70 [b] |
| **Duodenum Length, Cm** | 38.10 [a] | 37.20 [a] | 33.70 [a] | 38.50 [a] | 38.50 [a] |
| **Duodenum Weight, g** | 16.60 [a] | 17.50 [a] | 16.70 [a] | 14.60 [a] | 12.30 [a] |
| **Pancreas Length, Cm** | 13.80 [a] | 13.60 [a] | 14.50 [a] | 15.20 [a] | 14.20 [a] |
| **Pancreas Weight, g** | 4.65 a [b] | 5.92 [a] | 5.20 a [b] | 4.47 [b] | 4.52 [b] |
| **Liver Weight, g** | 61.00 [a] | 59.90 [a] | 60.10 [a] | 61.60 [a] | 56.30 [a] |
| **Small Intestine Length, Cm** | 206.50 [a] | 221.50 [a] | 219.90 [a] | 225.80 [a] | 203.20 [a] |
| **Small Intestine Weight, g** | 112.60 [a] | 112.20 [a] | 105.20 [a] | 111.10 [a] | 103.60 [a] |
| **Spleen Weight, g** | 3.60 [a] | 2.70 [b] | 2.70 [b] | 2.40 [b] | 2.40 [b] |
| **Caecum Length, Cm** | 25.50 [a] | 24.10 [a] | 25.70 [a] | 24.20 [a] | 23.70 [a] |
| **Caecum Weight, g (pair)** | 19.95 a | 20.57 [a] | 18.56 [a] | 18.30 [a] | 22.44 [a] |
| **Rectum Length, Cm)** | 12.17 [a b] | 10.41 [b] | 10.68 [b] | 13.05 [a] | 11.25 [a b] |
| **Rectum Weight, g** | 5.17 [a] | 5.02 [a] | 3.69 [a] | 9.44 [a] | 3.72 [a] |
| **Heart Weight, g** | 10.13 [a] | 10.35 [a] | 9.82 [a] | 10.43 [a] | 10.03 [a] |
| **Head +Legs, Weight, g** | 120.74 [a] | 126.97 [a] | 123.12 [a] | 1450.02 [a] | 118.29 [a] |
| **Gastro-Intestinal tract Weight, g** | 292.12 [a] | 300.62 [a] | 281.56 [a] | 269.98 [a] | 266.94 [a] |

**Means in the same row with different superscripts are differ significantly at P ≤ 0.05**

Meanwhile, EM supplementation to the drinking water (1 ml EM/liter) reduced significantly (P<0.05) carcass and proventriculus index as compared with the control group. All the nutritional groups that supplemented with EM reduced significantly (P<0.05) spleen weight index than the control group. Anjum *et al*., (2005) who found that liver index of EM treated broiler's was significantly lesser (P<0.05) than the control and the actual weight of proventriculus was significantly greater in EM treated broilers compared with the control (P<0.05) but its index did not differ significantly between EM treated broilers and the control.

**Table (6): Effect of effective microorganisms supplementation on carcass and gastrointestinal tract organs weights as proportional to live body weight**

| Trait | Treatment | | | | |
|---|---|---|---|---|---|
| | Control 0 EM | Effective microorganism (EM) | | | |
| | | Feed | | Water | |
| | | 0.4% | 0.6% | 0.5 ml | 1ml |
| **Carcass (C%LBW)** | 78.80 $^{a}$ | 79.30 $^{a}$ | 78.90 $^{a}$ | 80.50 $^{a}$ | 73.70 $^{b}$ |
| **Proventriculus (Pro%LBW)** | 0.48$^{ab}$ | 0.49$^{a\,b}$ | 0.40 $^{b}$ | 0.55 $^{a}$ | 0.40 $^{b}$ |
| **Gizzard (G%LBW)** | 2.80 $^{a}$ | 3.10 $^{a}$ | 3.00 $^{a}$ | 2.80 $^{a}$ | 2.70 $^{a}$ |
| **Liver (L%LBW)** | 3.10 $^{a}$ | 2.90 $^{a}$ | 3.00 $^{a}$ | 3.20 $^{a}$ | 2.90 $^{a}$ |
| **Small Intestine (SI%LBW)** | 5.70 $^{a}$ | 5.50 $^{a}$ | 5.30 $^{a}$ | 5.80 $^{a}$ | 5.40 $^{a}$ |
| **Spleen (S%LBW)** | 0.18 $^{a}$ | 0.13 $^{b}$ | 0.13 $^{b}$ | 0.13 $^{b}$ | 0.12 $^{b}$ |
| **Heart (H%LBW)** | 0.58 $^{a}$ | 0.50 $^{a}$ | 0.50 $^{a}$ | 0.54 $^{a}$ | 0.51 $^{a}$ |
| **Head +Legs (H+L%LBW)** | 6.10 $^{a}$ | 6.10 $^{a}$ | 6.20 $^{a}$ | 6.53 $^{a}$ | 6.10 $^{a}$ |
| **Caecum (C%LBW)** | 1.00 $^{a}$ | 1.00 $^{a}$ | 0.92 $^{a}$ | 1.20 $^{a}$ | 0.93 $^{a}$ |
| **Gastrointestinal Tract (GIT%LBW)** | 14.90 $^{a}$ | 14.70 $^{a}$ | 14.20 $^{a}$ | 14.20 $^{a}$ | 13.90 $^{a}$ |

**Means in the same row with different superscripts are differ significantly at P ≤ 0.05**

**Antibody titers**

The hemagglutination inhibition (HI) test and the antibody response to sheep red blood cells (SRBCs) is still the most widely used conventional serological method for measuring antibody levels in poultry sera (Xu, *et al* ., 1997). The immune responses against Newcastle virus and SRBCs of the experimental birds are presented in Table (7). Chicks of groups B, C and E had higher antibody titers (P<0.05) after the primary and secondary response than chicks of group A (control). Values during primary response were 3.9, 2.7, 3.5, and 3.3 in groups B, C, D and E respectively compared with 2.5 in the control group A. The counterpart antibody titers during the secondary response were 3.8, 3.1, 3.3, 3.6 and 2.8 in the previous corresponding groups. Chicks in group B which were supplemented with 0.4 % EM in the diet is the only group which showed significant (P<0.05) increase in anti-NDV antibody level than the control group. These results indicated that supplementing chicks with 0.4 % EM in the diet induced enhancement to the immune responses to the chicks in group B.

The present study showed that EM potentiated immune response in the experimental broilers. Previous studies demonstrated that Lactic acid bacteria administered orally or intraperitoneally enhanced activity of the mononuclear phagocytic system (Kato *et al*., 1983) and increased the production of circulating antibodies for certain antigens in mouse (Saito *et al*., 1983). However, further investigation is required for the elucidation of the mechanism through which EM produced systemic increase in the immune response.

EM supplementation to broiler diet showed some beneficial effects on it's performance. It increased immunity, live body weight and weight gain than the untreated group. Moreover, EM appears to have some growth stimulating effects especially in young growing birds. From the commercial point of view, this observation is very important to poultry breeders. However, further research is required to study, in details, the effect of Pokashi supplementation to broiler's diet on the composition of the microflora in the small intestine and to determine the most effective dose of Pokashi to be supplemented to the broiler's diet.

**Table: (7): Effect of effective microorganisms supplementation on immune response against inactive NDV vaccine and $CRBC_S$**

| Immune response against | Weeks after inject. | Treatment | | | | |
|---|---|---|---|---|---|---|
| | | Control 0 EM | Effective microorganism added in | | | |
| | | | Feed | | Water | |
| | | | 0.4% | 0.6% | 0.5 ml | 1ml |
| NDV | 1 | $4.2\pm0.040^{b}$ | $5.3\pm0.40^{a}$ | $4.1\pm0.23^{b}$ | $4.7\pm0.37^{ab}$ | $4.2\pm0.27^{b}$ |
| $SRBC_S$ | 1 | $2.5\pm0.34^{b}$ | $3.9\pm0.23^{a}$ | $2.7\pm0.23^{b}$ | $3.5\pm0.31^{a}$ | $3.3\pm0.33^{ab}$ |
| $SRBC_S$ | 2 | $2.8\pm0.17^{c}$ | $3.8\pm0.24^{a}$ | $3.1\pm0.19^{bc}$ | $3.3\pm0.22^{ac}$ | $3.6\pm0.22^{ab}$ |

**Means in the same row with different superscripts are differ significantly at $P \leq 0.05$**

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## كفاءة أداء الدجاج اللاحم المغذي علي تحضيرات ميكروبية

**محمد أسامه فهمي**

**كلية الزراعة والطب البيطري- قسم انتاج وتربية الحيوان - جامعة القصيم – – المملكة العربية السعودية – ص . ب.٦٦٢٢**

تم دراسة تأثير إضافة الميكروبات النافعة لعلائق الدجاج اللاحم علي الكفاءة الإنتاجية – المناعة وأوزان و أطوال أجزاء القناة الهضمية. تم تقسيم٢٠٠ كتكوت من الدجاج اللاحم عمر يوم عشوائيا إلي خمس مجاميع متساوية ( أ , ب , ج , د , هـ) - المجموعتين ب , ج تم تغذيتهم علي ألعليقه البادئة و الناهية مضاف إليهم ٠,٤ , ٠,٦ % علي التوالي من المنتج بوكاشي الذي يحتوي علي الميكروبات النافعة أما المجموعتان د , هـ فتم تغذيتهم علي ألعليقه البادئة و الناهية مع أضافه ٠,٥ مل , ١مل / لكل لتر ماء شرب علي التوالي من المنتج بوكاشي و استمرت التجربة لمدة ٤٢ يوم وقد استخدمت المجموعة أ كمجموعه مقارنه لم يتم إعطاءها المنتج بوكاشي في ألعليقه أو في ماء الشرب . أظهرت النتائج أن أضافه الميكروبات النافعة للعليقه أو في ماء الشرب قد حسنت معنويا ($P<0.05$) وزن الجسم الحي خلال المرحلة البادئة من عمر الطيور وأحدثت زيادة معنوية لوزن الجسم المكتسب خلال الفترات العمرية من ١-٢, ٢-٣, ٣-٤ أسبوع عن المجموعة المقارنة و لم يتأثر معدل التحويل الغذائي معنويا خلال المرحلة البادئة من عمر الطيور. خلال الفترة العمرية من ٣ ألي ٤ أسابيع المجاميع ب , ج المضاف لهم ٠,٤ , ٠,٦% بوكاشي في ألعليقه أظهر تحسنا معنويا ( $P<0.5$) في معدل التحويل الغذائي بمقدار ١٣%, ١٥,٦٩% عن المجموعة المقارنة. عقب ٤٢ يوما من التغذية علي الميكروبات النافعة - القيم المطلقة و النسبية من وزن الجسم الحي لكل من الزبيحه , القانصه , ألاثني عشر , البنكرياس , الكبد , الأمعاء الدقيقة , الأعورين , القلب الرأس+ الأرجل و القناة الهضمية كاملة لم تظهر أي فروق معنوية عن مثيلاتها في المجموعة المقارنة . حدث انخفاض معنوي ($P<0.05$) لوزن الكبد كنسبه مئوية من وزن الجسم في كل المجاميع الغذائية مقارنه بالمجموعة المقارنة. المجاميع الغذائية ب , ج ,هـ أظهرت زيادة معنوية ($P<0.05$) في تترالأجسام المضادة للاستجابة لحقن معلقSRBCs خلال المرحلة الأولي و الثانية للاستجابة للحقن ب SRBCs- المجوعة (ب) هي الوحيدة التي أظهرت فرق معنوي ($P<0.05$)في الأجسام المضادة ضد النيوكاسل عن المجموعة المقارنة. أضافه الميكروبات النافعة لعلائق و ماء شرب الدجاج اللاحم أظهرت بعض الفوائد خاصة بالنسبة لوزن الجسم – وزن الجسم المكتسب و المناعة وفيما يبدو أن الميكروبات النافعة تحدث تنشيط لنمو الكتاكيت الصغيرة أثناء مرحله التغذية علي ألعليقه البادءه و تنشط الجهاز المناعي للطيور. و من الناحية الاقتصادية هذه الملاحظات في غاية الاهميه لمربي الدجاج اللاحم لذا يجب أجراء دراسات أخري مستفيضة لنحدد بالتفصيل تأثير أضافه البوكاشي لعلائق و ماء شرب الدجاج اللاحم علي كفاءته الانتاجيه و دراسة مجموعات الكائنات الحية الدقيقة في الأمعاء و تحديد النسبة الفعالة من البوكاشي التي تضاف للعلائق أو لماء الشرب التي تحقق أفضل النتائج.