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Effect of Effective Microorganisms (EM₄) on Health of Layers

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Abstract: Commercial laying hens (Babcock) 174-weeks-old, were given feed containing 1, 2 and 3% EM₄ for a period of 12 weeks. EM₄ did not influence live body weight. Egg production was greater in birds given feed containing 1 and 2% EM₄ but lower in birds given feed containing 3% EM₄ than the control birds. Blood glucose was significantly lesser in birds given feed containing 3% EM₄ than the control ($p < 0.05$). Serum phosphorus was significantly lesser in birds given feed containing 3% EM₄ than the control birds ($p < 0.01$). Serum total proteins, serum albumin, serum globulins, serum total lipids and serum cholesterol were not influenced significantly with the EM₄ treatment. The study suggests that EM₄ is a safe product for laying birds it increases egg production when mixed in feed.

Key words: Effective microorganisms, health layers

Introduction

Probiotics play a significant role as feed additives in poultry diet. These regulate the microbial environment of intestine, inhibit pathogenic intestinal micro-organisms and improve feed conversion efficiency (Barrow, 1992; Ohingra, 1993; Qiquo, 1994; Casas, 1995). Many of the existing probiotics are not stable in feed and have a shorter shelf life. Effective microorganism (EM₄) solution consists of wide variety of effective, beneficial and non-pathogenic micro-organisms of both aerobic and anaerobic types co-existing, having a longer shelf life. It is a newer technology invented by Professor Higa in Japan and introduced by Hussain *et al.* (1994) in Pakistan. EM₄ consists of lactic acid bacteria, yeasts, fermenting fungi, etc. The present project was designed to study the effect of EM₄ on health of poultry under experimental conditions.

Materials and Methods

Experimental design: The study was conducted on 72 White Leghorn (Babcock), 174-weeks-old, layers at the completion of 3rd moult. The hens were kept in cages randomly divided into 12 replicates, each having 6 hens. Three replicates were randomly assigned to one treatment. Total treatments were four including control, 1, 2 and 3% EM₄.

The birds were given a commercial layer mash having 16% crude protein and 2750 kcal/kg metabolizable energy. Each bird was served with 0.83 kg of feed weekly during the experimental period.

Effective microorganisms (EM₄): EM₄ solution was procured from Nature Farming Research and Development Foundation, University of Agriculture, Faisalabad, Pakistan. It was diluted as 1:100, 2:100 and 3:100. One liter of the resultant solution was thoroughly mixed in 50 kg feed for 12 weeks during the experiment. Data of Live body weight (kg) weekly. Number of eggs laid daily. Total lipids (g/dL) and cholesterol (g/dL) were determined photometrically (Khan *et al.*, 1994). Total serum proteins (g/dL) were estimated by Baush and Lomb Refractometer (Coles, 1974). Serum albumin (g/dL) was determined using Bromocresol Green (Asif *et al.*, 1995). Serum globulin concentration (g/dL) was obtained by subtracting the amount of albumin from that of total serum proteins. Serum glucose (mg/dL) was determined after enzymatic oxidation in the presence of glucose oxidase (Daly and Peterson, 1990). Phosphorus was determined following the method of Oser (1965).

Calcium was determined following the method of Richards (1981) and the data were analysed statistically.

Results

Live body weight: Live body weight was 1.62 kg in hens given feed containing 1% EM₄, 1.61 kg in hens given feed containing 2% EM₄ and 1.65 kg in hens given feed containing 3% EM₄ compared with 1.63 kg in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant.

Egg production: Average number of eggs per day were 14.16 in hens given feed containing 1% EM₄, 12.87 in hens given feed containing 2% EM₄ and 9.96 in hens given feed containing 3% EM₄ compared with 11.85 in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically significant ($p < 0.05$).

Total lipids: Total lipids were 2401 mg/dL in hens given feed containing 1% EM₄, 1660 mg/dL in hens given feed containing 2% EM₄ and 909 mg/dL in hens given feed containing 3% EM₄ compared with 1703 mg/dL in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant.

Cholesterol: Serum cholesterol was 113.3 mg/dL in hens given feed containing 1% EM₄, 137.3 mg/dL in hens given feed containing 2% EM₄ and 96 mg/dL in hens given feed containing 3% EM₄ compared with 123.7 mg/dL in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant.

Serum total Proteins: Serum total protein concentration was 5.1 g/dL in hens given feed containing 1% EM₄, 5.2 g/dL in hens given feed containing 2% EM₄ and 5.1 g/dL in hens given feed containing 3% EM₄ compared with 5.2 g/dL in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant.

Serum Albumin: Serum albumin concentration was 1.9 g/dL in hens given feed containing 1% EM₄, 2.1 g/dL in hens given feed containing 2% EM₄ and 2.0 g/dL in hens given feed containing 3% EM₄ compared with 2.3 g/dL in the control hens

Naqvi *et al.*: Effect of Effective Microorganisms (EM₄) on Health of Layers

Table 1: Body weight, egg number and some biochemical parameters of layers treated with different levels of EM₄

Parameter	Levels of EM ₄ (%)			
	Control	1	2	3
Body weight (kg)	1.63 ± 0.02	1.62 ± 0.01	1.61 ± 0.07	1.65 ± 0.03
Eggs per day	11.90 ± 0.72	14.20 ± 0.34*	12.90 ± 0.21	9.90 ± 0.36*
Total Lipid (mg/dL)	1703.00 ± 785	2401.00 ± 604	1660.00 ± 136	909.00 ± 464
Cholesterol (mg/dL)	124.00 ± 11.9	113.00 ± 55.2	137.00 ± 23.5	96.00 ± 41.5
Total proteins (g/dL)	5.20 ± 0.7	5.10 ± 0.7	5.20 ± 0.3	5.10 ± 0.8
Albumin (g/dL)	2.30 ± 0.05	1.90 ± 0.1	2.10 ± 0.1	2.00 ± 0.2
Globulin (g/dL)	2.90 ± 0.6	3.10 ± 0.6	3.20 ± 0.2	3.00 ± 0.7
Glucose (mg/dL)	208.00 ± 16	234.00 ± 10	220.00 ± 24	180.00 ± 13*
Phosphorus (mg/dL)	7.00 ± 1.0	5.60 ± 0.7	6.20 ± 0.8	4.20 ± 0.2*
Calcium (mg/dL)	14.00 ± 0.05	13.60 ± 0.6	10.20 ± 4.3	10.60 ± 0.8

Each figure represents mean (± standard deviation of the mean) of 18 birds. The data were subjected to analysis of variance.

*p < 0.05 compared with the control

(Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant).

Serum globulins: Serum globulin concentration was 3.1 g/dl in hens given feed containing 1% EM₄, 3.2 g/dl in hens given feed containing 2% EM₄ and 3.0 g/dl in hens given feed containing 3% EM₄ compared with 2.9 g/dl in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant.

Blood glucose: Blood glucose concentration was 234.3 mg/dl in hens given feed containing 1% EM₄, 220.0 mg/dl in hens given feed containing 2% EM₄ and 179.7 mg/dl in hens given feed containing 3% EM₄ compared with 208.0 mg/dl in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically significant (p < 0.05).

Serum Phosphorus: Serum phosphorus concentration was 5.6 mg/dl in hens given feed containing 1% EM₄, 6.2 mg/dl in hens given feed containing 2% EM₄ and 4.2 mg/dl in hens given feed containing 3% EM₄ compared with 7.0 mg/dl in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically significant (p < 0.01).

Serum Calcium: Serum calcium concentration was 13.6 mg/dl in hens given feed containing 1% EM₄, 10.2 mg/dl in hens given feed containing 2% EM₄ and 10.6 mg/dl in hens given feed containing 3% EM₄ compared with 14.0 mg/dl in the control hens (Table 1). The difference between hens given EM₄-treated feed and the control hens was statistically non-significant.

Discussion

The presence of beneficial micro-organisms in EM₄ has attracted its use in poultry. The present study was conducted on 72 commercial 174-weeks-old Babcock hens after the completion of 3rd moult for a period of 12 weeks. In this study, EM₄ did not show any adverse effect on many health parameters including clinical picture, live body weight and some of the biochemical parameters thus proving its safety for poultry. Qiquo (1994) compared the effect of antibiotics and EM₄ as feed additives in chicken diet. The results showed, more weight gain in the birds fed diet containing EM₄ than the birds fed diet with or without antibiotics. Moreover, there was marked reduction in smell from the dropping of birds using diet supplemented with EM₄. Average mortality was also lower in the birds having EM₄. Kalbande *et al.* (1992) also did not find

any positive effect of probiotic on the live weight of growing commercial pullets. Egg number was significantly greater in hens given feed containing 1 and 2% EM₄ (Table 1) than the control hens (p < 0.05). The data suggests egg enhancing activity of EM₄. This finding is in agreement with Anjum (1997) who observed 3% increase in egg production during a 6-week field trial when EM was administered at the rate of 1:1000 in drinking water to 58-weeks old layers. Chotisaitron *et al.* (1996) reported a significant increase in total egg production, feed conversion efficiency per dozen eggs and specific gravity of eggs with addition of EM in feed. However, supplementing EM in feed had no significant effect on feed intake, live weight gain, mortality and quality characteristics of eggs determined in terms of egg mass, weight, albumin weight, shell weight, colour and Haugh units. Ibanez (1990) supplemented *Lactobacillus acidophilus* to 28 to 52 weeks old commercial hens in feed. He recorded significantly higher egg production in treated hens than in the control hens. Shell strength, feed consumption, yolk pigmentation and mortality were not affected in the treated hens. Not much information is available on the blood chemistry of laying hens that were supplemented with any kind of probiotics in feed or in water. Hence, some of the biochemical profile was studied in the chicken using EM₄. Blood glucose concentration has been used as an indication of nutritional status of the birds. In this study blood glucose concentration was relatively higher in hens given feed containing 1 and 2% EM₄ than the control hens (Table 1). Interestingly, EM₄ significantly decreased blood glucose in hens given feed containing 3% EM₄. However, no other study is available for comparison. Various workers have tried a variety of useful micro-organisms to enhance productivity of poultry (Ibanez, 1990; Ohingra, 1993; Qiquo, 1994; Casas, 1995). The improvement in production is claimed to be due to restoring a stable microflora of the gut and to counteracting bacterial challenge by colonizing in the gut thus preventing attachment and proliferation of pathogenic bacteria and by enhancing digestibility of various feed ingredients. In animal husbandry, EM has been used with noticeable decrease in malodours, in the appearance of sickness and insect infestations, noticeable increase in fertility from artificial insemination and increase of the quality of meat, dairy and eggs (Hussain *et al.*, 1994). Gippert and Bodrogi (1992) reported the effect of Lacto-sacc in the feeding of broiler ducks. Penkin ducks were given diet supplemented with probiotic Lacto-sacc (*Streptococcus faecium*, *Lactobacillus acidophilus*, proteinase, cellulase, amylase and yeast culture) at 1 kg/ton for 45 days. Probiotic had no effect on mortality but increased live weight and feed intake. Phosphorus and calcium are essential constituents of bone and egg shell. Blood

Naqvi *et al.*: Effect of Effective Microorganisms (EM₄) on Health of Layers

concentration of phosphorus and calcium were relatively lesser in hens given 1 and 2% EM₄ compared with control (Table 1). Since shells thickness did not differ between EM treated and control hens, the decrease in Ca and phosphorus concentration in blood seems to have no clinical implication. No other studies are available for comparison. In conclusion, EM₄ when given in feed at the rate of 1 and 2%, proved beneficial compared with 3% which depressed many performance parameters. This suggests that EM is a safer product. It increased egg production in layers.

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