J. Sher. Agric. Univ., 1(1): 99-107, July 2007

STUDY ON THE EFFECT OF FEEDS AT DIFFERENT LEVELS OF PROBIOTIC (PROTXIN) SUPPLEMENTATION ON PRODUCTION PERFORMANCE OF LAYING HEN

M.A.Wadud[1], M.J. Alam[2], M.A.H.Beg[3] & M.M. Hossain[4]

ABSTRACT

The study was conducted to determine the effects of dietary supplementation of a commercial probiotic (Protexin) on daily feed consumption, egg yield, egg weight, feed conversion, external and internal qualities of egg and livability in layer hens. To achieve the objectives a total of 96, 42 week-old layers arranged in four dietary treatment groups including control, containing 8 birds/ replication supplemented with 0, 0.5, 1.0 and 1.5% probiotic for a period of 16 weeks. The study reveals that the hen day egg production, feed conversion, egg weight and mass, egg shell weight, egg thickness and egg breaking strength increased linearly with the use of dietary protexin. However, livability and internal egg quality were not altered with the use of dietary protexin. Present study concluded that dietary supplementation of probiotic promote the laying performance and most of the external quality characteristics of eggs. In conclusion, it is recommended that dietary supplementation of probiotic at the rate of 1.5 gm per kg feed in the layer ration may be economically potential.

Keywords: Supplementation, probiotic, performance, layer

INTRODUCTION

Poultry feed influences the production cost of chicken and egg. Feed additives, like antibiotics, hormones and coccidiostats are used in the diet of poultry to improve the poultry feed efficiency and to reduce production cost. Recently, it is believed that Probiotics have beneficial effects to improve the productive performance of poultry. Probiotics are specific agents produced by micro-organism containing *Lactobacillus acidophillus, Lactobacillus casei, Bifidobactuium bifidum, Aspergillus oryzae* and *Torulopsis* (Mohan *et al.* 1995). Fuller (1988) defined probiotics as "A live microbial feed supplement, which beneficially affects the host birds by improving it intestinal microbial balance". The US National Food Ingredient Association defined probiotic (direct fed microbial) as a source of live naturally occurring microorganisms, and this includes bacteria, fungi and yeast (Miles and Bootwalla, 1991). Probiotic is a microorganism or a combination of microorganisms which selectively suppress the harmful bacteria (*Salmonella, E. coli,* etc) in the gut of living beings. Probiotics also contain other substances to improve the intestinal microbial balance. The adverse effect of antibiotic feeding encouraged a shift in favour of feeding probiotics to boost up productive performance of chickens (Fuller, 1988). Improved egg production, feed conversion ratio, egg weight, specific gravity was observed by adding probiotic in the diet of layer (Nahashon *et al.* 1992, 1993, 1994 and 1996; Mohan *et al.* 1995). During the laying phase, supply of 153g CP/kg diet with Lactobacillus produced significantly larger eggs ($P<0.05$) than those given a similar diet without *Lactobacillus* .But some authors found the reduced egg production and feed conversion efficiency in using probiotic in layer hen (Goodling *et al.* 1987).

The detailed objectives of the current investigation were:

1 To evaluate egg production, egg quality and livability of layers fed protexin as a probiotic
2 To find out the suitable dose of protexin for maximum production
3 To study the profitability of using protexin in layer diet.

[1] Prof., Dept. of Poultry Science, BAU, Mymensing ; [2&4]Asstt. Prof., [3]Assoc. Prof., Dept. of Animal Husbandry, Sher-e-Bangla Agricultural University , Dhaka-1207.

MATERIAL AND METHODS

The research was conducted for the period of 16 weeks at Bangladesh Agricultural University (BAU) Poultry Farm (2002), Mymensingh, to evaluate the efficiency of a probiotic (Protexin) for laying birds. An open sided house was used for rearing experimental birds and proper hygienic measures were taken in every step of management. Ninety six (96) commercial layers (Shavar 579) were selected randomly rearing in same farm at an age of 42 weeks and placed group wise in the experimental cages. The space given per bird was 450 cm^2. The details of layout of the experiment are shown in Table 1.

Table 1. Layout of the experiment

Treatment group	No of birds in each replication			Total
	R_1	R_2	R_3	
D_1	8	8	8	24
D_2	8	8	8	24
D_3	8	8	8	24
D_4	8	8	8	24
Total	**32**	**32**	**32**	**96**

D_1.control diet, i.e. without protexin in the feed; D_2- 0.5 g protexin/kg feed; D_3- 1 g protexin/kg feed; D_4- 1.5 g protexin/kg feed.

Collection of feed ingredients, probiotic and analysis of feed sample

Required feed ingredients (Table 2) for making diets for experimental birds were supplied by the poultry farm of BAU. Crushed maize, wheat, till oil cake and oyster shells were mixed by weight according as per requirement of each dietary treatment.

Table 2. Ingredients of experimental diets and their cost

Feed ingredients	Experimental diet (Amount in kg/100 kg)				Cost (Tk/kg)
	D_1	D_2	D_3	D_4	
Maize	50	50	50	50	9.00
Rice police	15	15	15	15	8.00
Til oil cake	6	6	6	6	8.50
Soybean meal	16	16	16	16	14.00
Protein concentrate	2.75	2.75	2.75	2.75	35.00
Meat & bone meal	3	3	3	3	19.00
Vegetable oil	1.25	1.25	1.25	1.25	40.00
Oyster shell	5.5	5.5	5.5	5.5	5.00
Common salt	0.5	0.5	0.5	0.5	7.00
Probiotic	0	0.5	1	1.5	75.00

D1- control diet i.e. without protexin in feed; D_2- 0.5 g. protexin/kg feed; D_3- 1g. protexin/kg feed; D_4- 1.5g. protexin/kg feed.

Table 3 indicates the requirements of ME, CP, critical and limiting amino acid, Ca and P content which were satisfied according to the recommendation of Shaver Breeding Company, Canada. All feed ingredients were mixed thoroughly in such way that uniform distribution of each ingredient can be made by manually in every week.

Probiotic (protexin) was purchased from the Ciba-Geigy company, Mymensingh. Probiotic was mixed properly as per requirements for different treatments. Analysis of feed ingredients was performed following AOAC (1990) and Krisna and Ranjhan (1980).

Table 3. Composition of the control diet (%DM Basis)

Nutrients	Calculated composition
ME (k cal/kg)	2792
CP (%)	17.9
CF (%)	3.7
Calcium (%)	3.5
Available P (%)	0.55
Lysine (%)	0.90
Methionine (%)	0.33
Tryptophan (%)	0.19

Nutrient composition close to breeder's Standard Shavar 579, commercial strain

Routine Management

The research birds were exposed to similar care and management throughout the experimental period. Strict hygienic measures were taken to protect pullets from unhygienic condition. The summer day length was less than 16 hours during the experimental period. So a provision was made by using 60-watt electric bulb to meet up 16 hours light per day for maximum laying performance according to the recommendation of Shavar 579 Breeding Company. Feeder and waterer were cleaned and disinfected regularly to avoid microbial contamination. The floor of the experimental house was kept clean. The experimental diets were prepared weekly and stored in tin containers. Feed and water were offered twice daily, once in the morning and again in the afternoon in such a way that feeders and waterers were not kept empty.

Data Recording

To achieve the objectives the following performance characteristics were recorded during the experimental period.

Body weight- Individual body weight for each replication was recorded at the beginning of the experiment and then at 4 weeks interval until the termination of experiment.

Egg production- Recorded daily; Egg weight- individually once a week.

Egg quality -Egg shape index, albumen index, yolk index, breaking strength, yolk colour score, Haugh unit, shell thickness etc. characteristics were recorded at 46, 50, 54, and 58 weeks of age.

Feed consumption- Weekly.

Mortality- Daily.

Determination of egg quality characteristics

Egg quality characteristics were measured for eggs laid by birds fed on different diets. One hundred forty four (144) eggs were collected randomly from different dietary groups taking 36 from each diet group and 12 eggs from each replication and 4 times at (46, 50, 54 and 58 weeks) of age. At first egg weight was taken by an egg scale and the length of egg was measured by a slide caliper. In every case, the mean of 3 records was considered for the length of a particular egg. The width of egg was measured using a slide caliper. To minimize the error, 3 measurements were taken for each egg; one from small end, one from large end and one from the middle of the egg. The mean of 3 measurements was considered as the width of a particular egg. The eggs were carefully broken on a glass plate (30 cm X 21 cm) to measure both internal and external quality. The following formulas were used to determine egg quality.

$$\text{Egg shape index} = \frac{\text{Average Width of egg}}{\text{Average length of egg}} \times 100$$ (Reddy *et al.* 1979)

$$\text{Albumen index} = \frac{\text{Average height of albumen}}{\text{Average diameter of albumen}} \times 100$$

$$\text{Yolk index} = \frac{\text{Average height of yolk}}{\text{Average width of yolk}}$$ (Wesley and Stadleman (1959)

HU (Haugh Unit) = 100 log (H + 7.57 - 1.7 $W^{0.37}$), Where HU=Haugh unit, H=Height of thick albumen, W= Egg weight (g) (Haugh (1937)

Yolk Color- Yolk color was determined by comparing with the Roche yolk color Fan (F. Hoffmann La Roche and Co. Ltd, Basle, Switzerland.)

Egg shell thickness -After oven drying, shell thickness was measured by means of an egg shell thickness meter (Ogawa sciki Co. Ltd.Tokyo, Japan) in mm. To minimize error, three measurements were taken for each egg shell. One from large end, one from the small end and one from the equator region of the egg shell. The mean of these measurements was considered as the shell thickness of a particular egg.

Egg Breaking strength (BS) = 50.86 X $EW^{0.915}$

Where, EW = Egg weight (g) and BS = Breaking strength (Arad and Madra (1982)

Statistical analysis

The collected data on laying performance and egg quality characteristics were analyzed using MSTAT computer package programme in a Completely Randomized Design (Steel and Torrie, 1980). Duncan's Multiple Range Test (DMRT) was used to compare of mean values for to identify significant differences.

RESULTS AND DISCUSSION

Effect of probiotic on egg production performance

Feed intake

Daily feed intake did not differ significantly (P>0.05) among the diets. However, the highest feed intake was found on diet D_1 followed by D_2, D_4 and D_3 respectively (Table-4 and Figure-1). The similar findings were observed by Nahashon *et al.* 1994 but this finding was different from those of other authors (Ahmed *et al.* 1994; Huthail Najib 1996 and Kaudela and Nyirenda. 1995) who reported that inclusion of probiotic in the diet increase feed utilization and thus reduced feed intake.

Table 4. Egg production performance of laying hen feed diets considering different level of protexin (43-58 weeks of age)

Variables	Diet				LSD value and level of significance	
	D_1	D_2	D_3	D_4		
Feed intake(g/pullet/day)	111.04	110.63	109.26	109.32		NS
Hen day egg prduction(%)	53.13^d	54.63^c	56.83^b	59.71^a	1.367	**
Egg weight(g/egg)	57.52^c	59.64^b	59.61^b	61.18^a	0.476	**
Egg mass(g/pullet/day)	30.63^d	32.53^c	33.7^b	36.36^a	0.903	**
Body weight gain (g)	113.66^c	221.66^a	223^a	198.33b	17.99	**
FCR (FI/egg mass)	3.7^a	3.47^b	3.30^c	3.08^d	0.119	**
Livability (per cent)	95.83	100	100	95.83		NS

NS (Non- Significant) P>0.05; **P<0.01;D1-Control died ;D2-Control +0.5% probiotic; D3-Control +1% Probiotic; D4- Control+1.5%Probiotic, Values in the same row bearing the different superscript differ significently (P<0.05)

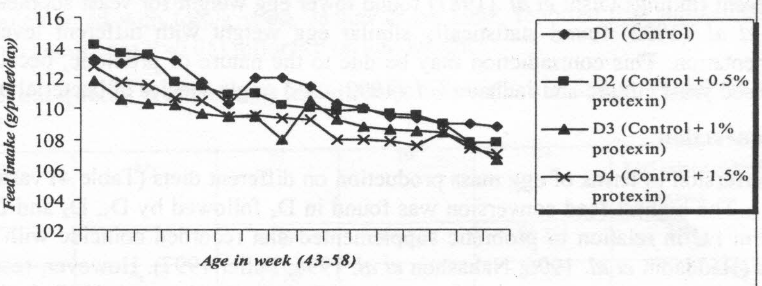

Fig.1. Weekly Feed intake pattern of layers fed on different levels of protexin

Body weight gain

Body weight gain significantly (P<0.01) differ among the diets. Body weight gain was highest on D_3 and D_2, intermediate on D_4 and lowest on D_1 diets (Table 4). It appears that probiotic supplementation made difference in body weight gain of egg laying pullets. This effect of probiotic supplementation on weight gain of pullets agreed with previous findings (Fuller, 1995; Nahashon *et al.* 1994) .The present findings contradicted with Kaudela *et al.* (1995). Jadhav *et al.* (1992) observed no significant(1>0.05) difference in body weight gains on diet containing dried *Lactobacillus sporogenes* (LSP) powder.

Egg production and mass production

Hen day egg production and egg mass production both were significantly (P<0.01) differ among the diets. Increasing protexin level increased both egg production and egg mass production (Table 4 and Figure 2). Bird that received the probiotic diets showed better performers than those on the control diet which was consistent with findings of Mohan *et al.* (1995) and Goodling *et al.* (1987) found the reduced egg production and egg mass production.

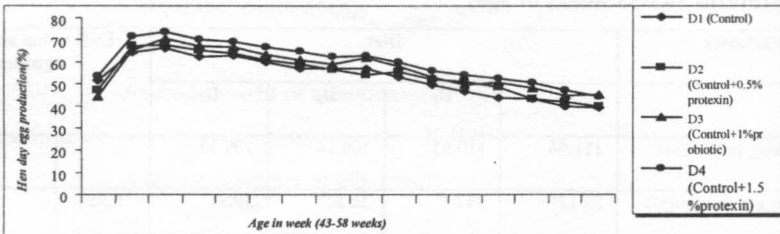

Fig.2. Weekly hen day egg production of pullets fed on different diet

Egg weight

Egg weight increased linearly with increasing concentration of dietary protexin (Table 4). The beneficial effect of dietary protexin on egg weight obtained in the present study, collaborated with findings of other workers (Gerendal *et al.* 1992; Nahashon *et al.* 1996; and Huthail Najib, 1996) found significantly ($P<0.01$) higher egg weight when diet contained probiotic. In contradictions with present findings Oishi *et al* .(1987) found lower egg weight for yeast supplemented diet but Jadhav *et al.* (1992) found statistically similar egg weight with different levels of probiotic supplementation. This contradiction may be due to the nature of probiotic, because Oishi *et al.* (1987) used yeast culture and Jadhav *et al.* (1992) used single species of bacterial product.

Feed Conversion

Feed conversion in terms of egg mass production on different diets (Table 4) varied significantly ($p<0.01$). The highest feed conversion was found in D_4 followed by D_3, D_2 and D_1 respectively. Change in FC in relation to probiotic supplemented diet recorded coincide with some previous findings (Haddadin *et al.* 1996; Nahashon *et al.* 1996; Fuller 1997). However, results obtained in this study do not agree with some other previous findings (Oishi *et al.* 1987; Jadhav *et al.* 1992; Nahashon *et al.* 1994) may be due to the nature of probiotic .

Overall performance

The overall performance of laying birds was not up to the level of breeder's performance standard. This was due to high environmental temperature during 43 to 51 weeks of the experimental period when temperature range from a minimum of 27 to a maximum of 31^0 C

Livability

In the present study, the supplementation of probiotic in the diet did not alter ($P>0.05$) survivability of birds (Table 4). Such a lack of effect of probiotic on survivability agreed with few previous findings. Goodling *et al.* (1987) reported no significant difference in livability when pullets fed viable *Lactobacillus* fermented product. The present result was contradicted by Gerendal *et al.* (1992); and Huthail Najib (1996). Kaudel aand Nyirenda (1995) who reported that the supplementation of probiotic to laying hen diet slightly increased mortality.

Feed cost

Cost of feed and cost for egg production (cost/ kg egg) were calculated and shown in Table 5. The feed cost was higher in D_4 and D_3 and than in D_2 and D_1. It reveals that feed cost for egg production was higher in D_1 and D_2 compared to D_3 and D_4. The cost of diet was increased as the level of protexin increased in the diet. Though cost of feed was increased by increasing of protexin .But the increased level of protexin decreased the cost of per kg egg production which was not significant (P> 0.05).

Table 5. Feed cost of different diets

Variables	Diet				Level of Significance
	D_1	D_2	D_3	D_4	
Feed cost Tk/kg mixed feed	10.79	11.16	11.54	11.91	-
Feed cost Tk/kg egg production	39.92	38.72	38.00	36.68	NS

NS (Non- Significant) P>0.05, D_1- Control diet i.e. without protexin in feed; D_2- 0.5g. protexin/kg feed; D_3- 1 g. protexin/kg feed; D_4- 1.5g. protexin/kg feed.

Effect of probiotic on the external qualities of egg

The external qualities of egg are shown in Table 6. The significant (P<0.05) differences were found among the diets for external quality traits of eggs in terms of egg weight, egg shell weight, shell thickness and breaking strength except shape index (P> 0.01). The effect of probiotic supplementation on external quality of eggs observed in the study agreed with some previous findings of (Huthali Najib,1995; Kaudela *and* Nyirenda 1995). The result of current findings contradicted by Oishi et al 1987, who reported that egg weight was lowered when basal diet supplemented with torula yeast. Jadhav *et al.* 1992, reported that supplementation of probiotic had no significant P>0.05) influence on egg weight.

Table 6. External quality traits of egg fed different diets (examined at 46, 50, 54 and 58th weeks of age)

Variables	D_1	D2	D3	D4	LDS value and level of significance
Egg sample	60.74^b	63.99^a	64.82^a	65.85^a	2.479* Weight(g)
Egg shape index	76.77	77.16	77.51	77.58	NS
Egg shell Weight(g)	5.54^c	5.90^b	6.07^b	6.53^a	0.168*
Egg shell thickness(mm)	0.34^c	0.37^b	0.37^b	0.40^a	0.018*
Breaking Strength(g)	2181.26^b	2286.06^a	2311.10^a	2338.84^a	78.26*

NS (Non Significant)P>0.01,*P<0.05; D_1- Control diet; D2- crontrol+ 0.5% protexin; D3- Control +1% protexin; D4- Control+1.5% protexin, Values in the same row bearing the different superscripts are significantly different (P<0.05)

Effect of probiotic on internal quality characteristics of egg

The internal characteristics of egg are shown in Table 7. Internal qualities of eggs in terms of albumen index, yolk index and Haugh unit did not differ significantly (P> 0.05) among the diets except yolk color score (p<0.05). This non-significant value agreed well with the results of Nahashon *et al.* 1996. It also observed that the yolk color score was maximum in D_2, intermediate in D_3 and D_4 least in D_1 diets respectively. Such an increase of yolk color due to the feedings of probiotic supplementation and got support from Nahashon *et al.* (1996).

Table 7. Internal quality traits of egg fed on different diets (examined at 46, 50, 54 and 58th weeks of age)

Variables	D_1	D_2	D_3	D_4	Level of significance
Albumen index	0.103	0.106	0.115	0.120	NS
Yolk index	0.417	0.423	0.417	0.415	NS
Yolk color score	4.333^c	6.167^a	5.667^b	5.833^b	*
Haugh unit	0.894	0.894	0.913	0.933	NS

NS (Non Significant) P>0.05.*P<0.05; D_1- control diet; D_2- control +0.5% protexin; D_3- Control+ 1%

protexin; d_4- control+1.5% protexin; Values in the same row bearing the different superscripts are significantly different (P<0.05).

On the basis of the results of the present study the following conclusions can be drawn.

1) The dietary supplementation of probiotic may promote laying performance and most of the external quality characteristics of eggs.

ii) Inclusion of probiotic at the level of 1.5 gm per kg of feed may be recommended to maximize layer performance and may economic

In Bangladesh very little work has been reported regarding the use of probiotic in poultry diet. But in abroad various significant works have been performed on meat and egg production. Therefore more research works in Bangladesh condition need to be conducted by using probiotic in poultry diet particularly when the climate becomes too hot and humid.

REFERENCES

Ahmed, H. F.; Sapeota, D. and Das, P. C. 1994. Effect of feeding probiotic on peak production in caged layers. *Indian journal of poultry Science*, 29(3): 269-271.

AOAC, 1990. Official methods of Analysis of the Association of Official Analytical Chemists, Washinhton D.C. edited by kenneth Helrich. 15:69-88.

Arad, Z. and Madra, J. 1982. Differences in egg quality among the Sinai Bedouig fowl, The commercial White Leghorn and their crossbreeds. British poultry Science, 23: 107-112.

Fuller, R. 1997. Nature of the determinant responsible adhesion of lactobacilli the chicken crop epithelial cells. *Journal of General Microbiology,* 87:245-250.

Fuller, R. 1988. Probiotics in man mad animal. *Journal of Applied Bacteriology*, 66:365-378.

Gerendal, D. and Gippest, T. 1992. *Effect* of Lacto-Sacc and yea-sacc products in diets of New Hampshire parent stock of fowls. Lacto sacc es yea-sacc keszitmenyek a New Hampdhire szulopa. *Baromfitenyestes es Feldolgozas*. 39(1); 11-15.

Goodling, A. C., Cernijglia,G. J. and Herbert, J. A. 1987. Production performance of White Leghorn layers fed Lactobacillus fermentation products. *Poultry Science*, 66(3): 480-486.

Haddadin, M. S. Y., Abdul Rahim, S. M., Haslamoun, E. A.R.and Robinson, R. K. 1996. The effect of *Lactobacillus acidophilus* on the production and chemical composition of hen's eggs. *Poultry Science,* 75(4): 491-494.

Haugh, R. R. 1937. The Haugh unit for measuring egg quality. U. S. Egg Poultry Magazine, 43:552-553 and 572-573 pp.

Huthail Najib. 1996. Effect of incorporating yeast culture *Saccharamyces cerevisiae* in to the Saudi Baladi and White Leghorn layers diet. *Journal of Applied Animal Research*, 10(2):181-186.

Jadhav, N. V., Siddqui, M. F. and Sakhare, P. G. 1992. Effect of probiotic feeding on the performance of pullets. *India Journal of poultry Science.* 27(3): 163-164.

Kaudela, K. and Nyirenda, C. C. S. 1995. Experimental per os application of probiotics Lactiferm in pullets and laying hens. Scientia Agriculture Bohemica, 26(2): 131-144.

Krishna , G. and Ranjhan, S. K. 1980. Laboratory manual for nutritional research. printed by Trypographers India at Rashtravavi printers. A 49/1 Mayapuri, phase 1, New Delhi.

Miles, R. D. and Bootwalla , S. M. 1991. Direct-fed microbials in animal production. A Review, National Food Ingredient association, West Des Moines, lowa, USA, 117-132 pp.

Mohan, B., Kadirve, R., Bhaskaran, M. and Natarajan, A. 1995. Effect of probiotic supplementation on serum/yolk cholesterol and on egg cell thickness in layers. *British Poultry Science*, 36(5): 799-803.

Nahashon, S.N., Naraue, H.S. and Mirosh, L.W. 1992. Effect of direct-fed microbials on nutrient retention and production parameters of laying pullets. *Poultry Science*, 71(sppli.1):111.

Nahashon, S. N., Naraue, H. S. and Mirosh, L. W. 1993. Effect of direct-fed microbials on nutrient retention and production parameters of Single Comb White leghorn pullets. *Poultry Science*, 72(2): 87.

Nahashon, S. N., Naraue, H. S. and Mirosh, L. W. 1994. Production variables and nutrient retention in Single Comb White Leghorn laying pullets fed supplemented with direct-fed microbials. *Poultry Science*, 73: 1699-1711.

Nahashon, S. N., Naraue, H. S .and Mirosh, L. W. 1996. Performance of Single Comb White Leghorn layers fed a diet with a live microbial during the growth and egg laying phases. *Animal Feed Science and Technology*, 57:25-38.

Oishi, T., Inusuka, S. and Aoki, S. 1987. Effect of low selenium feed with torula yeast on egg production rate and egg weight of laying hens. Memoirs of the Faculty of Agriculture, Kinki University. 20: 39-42.

Reddy, P. M., Reddy, V. R., Reddy, C. V. and Rao, P. S. P. 1979. Egg weight, Shape index and hatchability, in Khaki Campbell duck eggs. *Indian Journal of Poultry Science*, 14: 26-31.

Steel, R. G. D. and Torrie, J. H. 1980. Principles of producers of statistics: A Biometrical Approoch. McGraw Hill, New York.

Wesley, R. L and Stadelman. W. J. 1959. Measurement of interior egg quality. *Poultry Science*, 38: 479-481.