Probiotic supplementation in alleviating stress in broiler chicken

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Abstract: A study was conducted to evaluate the effect of probiotic supplementation in alleviating overcrowding stress and improving production performance in broiler chicken. The study was conducted in two phases, Phase I – in summer season, Phase II – in rainy season. In each phase 36 birds of one month of age was used for the study. Results of the study revealed that, in both phases of study overcrowding reduced the weekly body weight. Average weekly body weight gain was not affected by overcrowding. The weekly feed intake of birds reduced without much effect on body weight gain which thereby improved the feed efficiency. Probiotic supplementation further improved the feed efficiency. Overcrowding (348 cm²/bird) increased the H/L ratio of birds which was indicative of stress. The H/L ratio of 0.81 noticed in eight week old birds in summer indicated severe stress in them. In eight week old birds reared in summer the stress was more than those reared in rainy season. Probiotic supplementation was very effective in reducing stress and improving production during summer season.

Keywords: Broiler, stress, probiotic, production

I. Introduction

Broiler production is a highly flourishing industry in all parts of India and now the country is ranked as the sixth largest producer of poultry meat [1]. Genetic selection had improved the growth rate of birds and profit to farmers. However, it has increased sensitivity of these birds to stressors. Stress produces severe internal damages leading to a negative impact on economic traits. Broilers are subjected to frequent stress factors and therefore, it is important to have an effective management programme to minimize their effects on the performance and health of the birds [2]. One of the often-overlooked stressors that affects production efficiency and physiological responses of birds is social stress caused by overcrowding. Nutritional manipulations are easy and effective means to reduce stress. Now probiotics are widely used in commercial broiler production to reduce stress. Hence the present study was undertaken to scientifically evaluate the effect of probiotic supplementation in alleviating stress and improving production of broiler chicken subjected to overcrowding stress.

II. Materials and Methods

The study was carried out using four week old Vencobb strain of broiler chicken of either sex for a period of four weeks. The stress was induced by overcrowding (reducing the floor space by 50% so as to have a floor space of 348cm2/bird as against 696cm2/bird for the control group). The birds were selected randomly and separated into three groups with 12 birds in each group (G-I - Control, G-II - Stressed group of birds not supplemented with probiotic and G-III Stressed birds supplemented with Probiotic @ 0.025 per cent in diet). The probiotic used was Protexin one gram of which contains Lactobacillus plantarum-1.26x108cfu, Lactobacillus bulgaricus-2.06x108cfu, Lactobacillus acidophilus–2.06x108cfu, Lactobacillus casei-2.06x108cfu, Streptococcus thermophilus-4.1x108cfu, Streptococcus faecium-5.4x108cfu, Bifidobacteriumbifidum- 2x108cfu, Torulopsis species- 5.32x107cfu Aspergillusoryzae- 5.32x107cfu.

The birds were fed with standard broiler rations (starter and finisher) formulated as per [3] and proximate principles of the ingredients were estimated as per [4]

The experiment was carried out in two phases.

Phase I – In summer season (March to May)

Phase II – In rainy season (June to August)

Ad libitum feed and water with identical photoperiod schedule were provided to all the birds throughout the experiment. Daily total feed intake of each group of birds was recorded and at the end of each week the average daily feed consumption per bird was calculated. The body weight of individual bird was recorded at weekly intervals from fourth to eighth week of age.

Cumulative feed conversion efficiency (CFE) was calculated based on the data of body weight gain and feed intake.

Blood smears were prepared on a clean grease- free glass slide using fresh blood. Air-dried smears were stained with Leishman-Giemsa stain solution. Different leucocytes were counted and

Heterophil:Lymphocyte (H: L) ratio were calculated as per [5]. The data obtained were statistically analyzed using SPSS version 17.0. Student't' test was employed when only two groups were compared. One way analysis of variance followed by least significant difference test was adopted for comparison of more than one group.

III. Results & Discussion

The effect of probiotic supplementation on different production parameters (weekly feed intake, weekly body weight, weekly gain in weight, cumulative feed efficiency) of broiler chicken under overcrowding stress were analyzed. The level of stress was assessed by H/L ratio. The results of the study revealed that overcrowding (348cm^2 /bird) produced a significant (P ≤ 0.05) decrease in the body weight of 7 and 8 weeks old broiler chicks (table 1). The probiotic used could produce a significant improvement in the body weight of 8 week old stressed birds in summer. During rainy season in G-II birds, the reduction in body weight was observed from fifth week onwards and continued up to eighth week. Though the probiotic used was effective in summer it did not improve the body weight of overcrowded birds in rainy season. The body weight of four week old broilers in summer was found significantly (P ≤ 0.05) less than those in rainy season. This shows that heat stress reduced the growth of young birds than the old birds, may be by producing a more potential effect through thyroid hormones.

The weekly body weight gain of the broiler chicks are given in table 2. Body weight gain of broilers in control and experimental groups revealed that the mean total body weight gain of broilers was not significantly affected by overcrowding stress. However, in both summer and rainy seasons the gain in body weight at fifth week of age (1 week after application of stress) was significantly low in stressed control G-II group of birds. On continued application of the same stress the birds might have developed an adaptive response so that overcrowding stress did not produce any significant reduction in the body weight gain from sixth week onwards. At eighth week of age the probiotic supplementation could improve the body weight gain in stressed birds in summer.

[7] Observed that, when broilers were provided with a floor space of 0.095, 0.071, 0.051 and 0.048m²/bird the growth rate was not affected by increasing the stocking density. However, after six weeks of age, the birds at lowest stocking densities had the highest average daily gain and those at highest stocking density had the lowest daily gain. Supporting the earlier reports, in the present experiment, the weekly body weight gain of five and eight week old birds at lowest stocking density (696cm2/bird) was higher (441.67g and 445.83g) when compared to that at highest (348cm2/bird) stocking density (358.33g and 391.67g).However from sixth week onwards no significant difference in weekly body weight gain observed between the different group of birds. In eight week old birds in rainy season the birds at lower stocking density had a better weight gain (291.67g) than those at higher stocking density (280.17g), which might be due to better feed availability at lower stocking densities. Probiotic supplementation was effective to improve the weight gain of the stressed birds at eighth week of age in summer. [8] and [9] could not observe a better body weight gain in unstressed birds. Results of the present study were in accordance with the earlier reports. However the supplementation improve the gain in body weight when the intensity of stress was very high (Eight week old birds in summer). The body weight gain was not affected by the seasons (summer and rainy).

The average feed intake was least in G-II group of birds (table-3). The cumulative feed efficiency (CFE) of five week old G-1 group of birds was better than the other two groups (table-4). However, overcrowding improved the CFE of broilers from sixth week of age in both summer as well as rainy seasons. In both summer and rainy seasons the probiotic used was effective in further improving the feed utilization. The CFE was more in the summer than in the rainy season. [11] noticed a linear decrease in the feed intake with densities over 20 birds/m² with significant improvement in feed efficiency in the birds housed at high stocking densities. In this experiment also, the feed consumption was reduced in overcrowded birds without much affect on the growth rate, which improved feed efficiency. [12] obtained a highly significant increase in body weight and feed conversion in broilers on dietary supplementation of probiotic at the rate of 0.30g/60 chicks. In the present study too, it was seen that probiotic supplementation improved the feed efficiency in overcrowded as well as overcrowded heat stressed birds, which might be by improving the intestinal function.

In both phases of the study, the H/L ratio was significantly (P \leq 0.05) high in G-II group of birds (table. 5). In summer the probiotic supplementation was found effective in reducing the H/L ratio. [7] reported that increased stocking density produced stress indicated by higher H/L ratio from fourth week onwards. [13] suggested that in birds H/L ratio was a good measure of long-term stress (hours or weeks). They observed that H/L ratio of about 0.2, 0.5, and 0.8 characterized low, optimum and high levels of stress respectively. The high value of H/L ratio (0.7 – 0.8) observed in the present study indicated high level of stress at a stocking density of 29 birds/m². Also the increased value of H/L ratio observed in the summer indicated that high temperature (32.68°C) and humidity (88.25%) during the summer season is highly stressful to caged broilers. Supporting the findings of the present study [10] and [14] reported increased H/L ratio in birds subjected to heat stress.

		S	easons. Mean ± 3	SE (n=12)						
Seesons	Parameter		Body weight (g)/bird/week							
Seasons	Weeks Groups	4	5	6	7	8				
S	G -I	837.50±39.95** ^a	1279.17±47.06 ^a	1708.91±45.00 ^a	2071.41±62.71 ^b	2346.41±64.46 ^b				
\mathbf{S}_1	G -II	833.33±43.66** ^a	1191.67±53.60* a	1610.84±41.04 ^a	1940.01±40.36 ^a	2219.18±62.60 ^a				
	G -III	829.17±35.06* ^a	1229.17±45.42 ^a	1654.17±48.64ª	2025.00±49.43 ^{ab}	2337.50±50.42 ^b				
	G -I	908.33±36.32** ^a	1354.17±43.72 ^ь	1750.00±51.86 ^b	2137.50±50.42 ^b	2429.17±47.06 ^b				
~	G -II	829.17±29.17** ^a	1220.83±28.51* ^a	1604.17±33.97 ^a	1987.50±48.90 ^a	2247.67±56.35 ^a				
S_2	G -III	833.33±27.06* ^a	1237.50±48.51ª	1612.50±42.25ª	1991.67±34.72 ^a	2291.67±41.67 ^a				

IV. Tables Table-1. Weekly body weight of broiler chicken from fourth to eighth week of age in summer and rainy seasons. Mean ± SE (n=12)

G –**I** Unstressed control (floor space / bird = 696cm^2), **G** -**II** Overcrowded control (floor space / bird = 348cm^2), **G** -**III** Overcrowded Protexin supplemented group, **S**₁ Summer season. **S**₂ Rainy season. **a**, **b** Means within a column with no common superscripts are significantly different at 5% level

* Between seasons significantly different at 5% level

** Between seasons significantly different at 1% level

NS No significant difference between seasons \mathbf{s}_1 and \mathbf{s}_2

Table-2. Weekly body weight gain of broiler chicken from fifth to eighth week of age in summ	er and
rainy seasons. Mean ± SE (n=12)	

	Parameter	Body weight gain (g)/bird/week						
Seasons	Weeks Groups	5	6	7	8	Mean ±SE		
	G-1	441.67±43.88ª _{NS}	429.74±27.06 ° _{NS}	362.50±43.57 ° _{NS}	275.00±39.65ª _{NS}	377.23±22.69ª _{NS}		
S 1	G-II	358.33±40.75 ^b _{NS}	419.17±27.84 ^a _{NS}	329.17±29.81 ª NS	279.17±33.97ª _{NS}	346.46±14.47ª _{NS}		
	G-III	400.00±26.11 ^{ab} _{NS}	425.00±25.00 ° _{NS}	370.83±27.15 ª _{NS}	312.50±41.80 ^b _{NS}	377.08±11.60ª _{NS}		
	G-I	445.83±17.90 ° _{NS}	395.83±28.51 ° _{NS}	387.50±26.92 ° _{NS}	291.67±28.76 ^a NS	380.21±16.61ª _{NS}		
S ₂	G-II	391.67±33.05 ^b NS	383.33±44.52 ° _{NS}	383.33±29.73 ° _{NS}	280.17±23.40ª _{NS}	359.63±13.69ª _{NS}		
	G-III	404.17±45.00 [∞] _№	375.00±31.68 ° NS	379.17±26.44 ª NS	300.00±33.14 ^b NS	364.59±11.18 ^a NS		

G-IUnstressed control (floor space / bird = 696cm²), **G**-II Overcrowded control (floor space / bird = 348cm²), **G**-III Overcrowded Protexin supplemented group, **S**₁ Summer season. **S**₂ Rainy season. **a**, **b** Means within a column with no common superscripts are significantly different at 5% level **NS** No significant difference between seasons **s**₁ and **s**₂

Table-3. Weekly feed intake of broiler chicken from fifth to eighth week of age in summer and rainy seasons. Mean ± SE (n=12)

Parameter		Feed intake (g)/bird/week						
Seasons	Weeks Grou ps	5	6	7	8	Mean ±SE		
	G -I	854.17	958.33	979.17	945.83	934.38± 27.61		
S1	G -II	741.67	908.33	850.00	858.33	839.58±34.44		
	G -III	791.70	858.30	904.20	995.80	887.50±42.85		
	G-I	875.00	1008.33	1029.17	1029.17	985.42±37.13		
S ₂	G -II	791.67	916.67	916.67	920.33	886.33±31.57		
	G -III	858.33	854.17	900.00	941.67	888.54±20.51		

G -I Unstressed control (floor space / bird = 696cm^2), **G** -II Overcrowded control (floor space / bird = 348cm^2), **G** -III Overcrowded protexin supplemented group, **S**₁ Summer season. **S**₂ Rainy season.

Parameter		Cumulative Feed Efficiency						
Seasons	Weeks Groups	5	6	7	8	Mean ±SE		
S1	G -1	1.93	2.11	2.29	2.50	2.21±0.12		
	G -II	2.07	2.10	2.24	2.42	2.20±0.08		
	G -III	1.98	2.00	2.14	2.35	2.12±0.09		
S ₂	G-I	1.96	2.24	2.37	2.59	2.29±0.13		
	G -II	2.02	2.20	2.27	2.51	2.25±0.10		
	G -III	2.12	2.20	2.26	2.44	2.26±0.07		

Table -4. Cumulative feed efficiency of broiler chicken from fifth to eighth week of age in summer and
rainy seasons. Mean ± SE (n=12)

G –**I**Unstressed control (floor space / bird = 696cm²), **G** -**II** Overcrowded control (floor space / bird = 348cm²), **G** -**III** Overcrowded Protexin supplemented group, S_1 Summer season. S_2 Rainy season

Table-5. Heterophil/ Lymphocyte (H/L) ratio in broiler chicken . Mean ± SE (n=12)

Seasons	Weeks Groups	4	6	8
	G-I	0.49±0.02 *	0.65±0.01 *•	0.72±0.02*++
<u>\$1</u>	G -II	0.48±0.03 *	0.77±0.03 b	0.81±0.03 ^b •
	G -III	0.46±0.03 *	0.69±0.03 *	0.75±0.02*
	G-I	0.49±0.03 *	0.57±0.03* •	0.57±0.03 * ++
S2	G -II	0.48±0.03 *	0.68±0.04 b	0.72±0.03 • •
	G -III	0.48±0.02 *	0.69±0.04 b	0.77±0.03 b

G –**I**Unstressed control (floor space / bird = 696cm²), **G** -**II** Overcrowded control (floor space / bird = 348cm²), **G** -**III** Overcrowded Protexin supplemented group, S_1 Summer season. S_2 Rainy season.

a, **b** Means within a column with no common superscripts are significantly different at 5% level

* Between seasons significantly different at 5% level

** Between seasons significantly different at 1% level

NSNo significant difference between seasons s_1 and s_2

Table-	6. Weekly r	neteorological data	during the	experimental	period of eig	nt weeks - Su	mmer seasor
r							

Week	Temperature (°C)		Relative Hu (R	Rainfall (mm)	
			E		
	Maximum	Minimum	Forenoon	Anemoon	
I	34.70±0.30	25.40±0.10	86.90±1.00	51.10±2.30	0
п	35.10±0.30	24.00±0.40	86.40±2.40	52.60±1.40	2.20±1.20
III	33.80±0.90	24.60±0.60	84.90±1.90	55.70±3.40	1.30±1.30
IV	35.10±0.30	26.10±0.30	81.90±1.40	54.00±1.40	0
V	35.00±0.60	25.20±0.50	86.90±1.00	52.90±2.30	4.90±3.90
VI	34.80±0.30	24.80±0.40	84.90±1.90	55.00±2.50	12.30±10.80
VII	29.30±0.90	23.50±0.20	91.60±1.70	81.90±3.90	30.00±10.70
VIII	31.60±0.40	24.00±0.40	89.60±1.78	68.40±4.67	6.80±3.22
¹ Mean ±SE	34.68 ±0.30	25.03±0.20	85.03±0.90	53.35 ±1.10	0.88 ±0.50
$^{2}Mean \pm SE$	32.68±1.17	24.38 ±0.46	88.25±1.71	64.55±5.62	13.50±5.19
³ Mean ±SE	33.68 ±0.75	24.70 ±0.30	86.64 ±1.05	58.95 ±3.79	7.19±3.57

1 Mean values of I to IV weeks

2 Mean values of V to VIII weeks

3 Mean values of I to VIII weeks

Week	Temperat	ure (°C)	Relative H (F	Rainfall (mm)	
	Maximum	Minimum	Forenoon	Afternoon	
Ι	29.30±0.20	23.30±0.20	92.40±0.20	80.60±0.20	25.10±3.90
п	30.80±0.20	23.40±0.20	92.60±0.20	66.30±0.20	2.50±1.40
III	29.70±0.40	22.90±0.20	92.40±0.40	73.40±0.20	19.50±6.30
IV	29.90±0.40	23.40±0.20	93.60±0.40	74.90±0.20	6.70±2.70
V	28.60±0.60	22.90±0.30	93.10±0.60	78.10±0.30	11.90±6.50
VI	29.50±0.40	22.80±1.00	94.40±0.40	74.70±1.00	8.00±3.90
VII	29.00±0.40	22.90±0.20	93.90±0.40	76.30±0.20	21.60±5.30
VIII	28.60±0.75	22.70±0.34	94.00±0.75	82.30±0.34	34.60±17.35
$^{1}Mean \pm SE$	29.93±0.20	23.25±0.10	92.75±0.20	73.80±0.10	13.45±2.60
$^{2}Mean \pm SE$	28.93±0.26	22.83±0.12	93.85±0.22	77.85±1.40	19.03±5.21
³ Mean ±SE	29.43 ±0.26	23.04 ±0.10	93.30±0.29	75.83±1.74	16.24±3.83

Table-	7. Weekly	meteorological	data during t	the experimental	period of	f eight weeks -	Rainy Season
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1 Mean value of I to IV weeks

2 Mean value of V to VIII weeks

3 Mean value of I to VIII weeks

V. Conclusion

The study revealed that stocking density of 29/ birds/m² in cages is stressful to birds especially in eight week old birds in summer. Even though overcrowding reduced the body weight, the feed efficiency was better at higher stocking densities which increased the profit. The probiotic protexin @ 0.025 per cent in diet was effective in improving the production of stressed broilers. However, the stress may affect the meat quality which was not taken into account in the present study.

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