Effect of supplementation of *lactobacillus* and *Saccharomyces boulardii* on the performance of broilers

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Abstract

A trial was conducted to evaluate the effect of *Lactobacillus* and *Saccharomyces Boulardii* on performance of broilers. A total of 300 day old broilers were randomized in 5 groups, each with 6 replicates with (5x6) 10 birds per replicate. Basal diet (D1) (control) was prepared as per BIS specification. In that of *Lactobacillus* was added @ 100 and 200g/t in D2 and D3 ; *Saccharomyces Boulardii(Sb)* was added @ 500 and 1000g/t of feed respectively up to 42 days of age. The diets were iso nitrogenous and iso caloric. The results revealed, a significant ($P<0.05$) improvement in body weight gain, better FCR with increased concentrations of *Lactobacillus* (D3) and *Saccharomyces Boulardii* (D5) in feed. The results suggest that supplementation of *Lactobacillus* and *Saccharomyces Boulardii* @ 200g/t and 1000g/t of feed respectively improved the performance of the broilers by increasing the absorption capacity of the gut.

Introduction

Pathogenic bacteria are always present in the gut but the balance between pathogenic and beneficial bacteria determines whether disease will occur or not (Ivanov 2003). Maintaining a healthy balance between all microflora with in the gut is known as Eubiosis (Jensen, 1980) and can be influenced by bacteria endemic to the micro flora. To reduce the pathogenic bacteria and to enhance growth, to minimize the disease prevalence using of antibiotic growth promoters in broiler ration is an age old practice. With increasing public concerns there is a worldwide attempt to reduce antibiotic use in Poultry production (Dibner and Richards, 2005). A convincing alternative of antibiotic has been the use of probiotics as a sub therapeutic and growth promoting agent (Yang et al., 2009).

This study was designed to study the effects of Lactic acid producing bacteria and *Saccharomyces Boulardii* i yeast on broiler performance and intestinal morphology.

Material and Methods

Commercial day old vencobb broiler chicks (n=300) were wing banded, weighed individually and randomly assigned to 5 treatments on the basis of initial body weight in a randomized complete block design. Each treatment had 60 broilers arranged in 6 replicates of 10 chicks each. The broilers were reared for a period of six weeks in battery brooders with provision of continuous lighting throughout the experimental period. The temperature was maintained at 34±1ÚC up to 7 days of the age and gradually reduced to 26±1ÚC by 21 days of age after which, chicks were maintained at room temperature. On first day chicks were offered only crushed maize and then given commercial diet from 2^{nd} day onwards along with ad-libitum drinking water. All the birds were kept under uniform management conditions throughout the experimental period. The birds were vaccinated against Mareks disease, New castle disease and Infectious bursal disease as per the routine vaccination schedule and Dose.

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Experimental design: basal diets (D1) were prepared for both starter (0-4 weeks) and finisher (5-6 weeks) satisfying the nutrient requirement (BIS, 1992). Lactic acid producing bacteria at 100g and 200g/ton in D2 and D3 , *Saccharomyces Boulardii* strain @ 6x 10^9 /gram at 500 and 1000g/ton in D4 and D5 were supplemented to the basal diet and test diets were prepared. Representative feed samples were ground well by passing 1 mm sieve and proximate principles, calcium and phosphorus were analyzed as per AOAC (2000).

Birds were weighed at weekly interval. Body weight and feed consumption were recorded at weekly interval up to 6th week. Feed conversion ratio (FCR) was calculated as the feed consumed per kg body weight gain.

Statistical analysis

The data was analyzed using general linear model procedure of statistical Package for social sciences (SPSS)15th version and comparison of means was done using Duncan's multiple range test (Duncan, 1955) and significance was considered at $P \leq 0.05$.

Results and Discussion

The impact of *Lactobacillus* and *Saccharomyces Boulardii* supplementation on body weight, feed intake, feed conversion ratio and gut morphology on broiler chicks are shown in Table 1, 2, 3 and 4 respectively.

Significantly higher body weights were noted in groups supplemented with higher levels of *lactobacillus* (D3) and *S.boulardii* (D5). In pre starter and starter phases no significant ($P \geq 0.05$) variation in body weights, where as significantly ($P \leq 0.05$) higher but were observed in finisher phase. These were on par with Kumar *et al.,* 2013 who reported higher body weight in birds fed with diet supplemented with *Lactiflora* alone (@ 0.05% or in combination with *Sacchromyces cervacae* (@ 0.05% supplemented groups than control. Similarly improvement in the growth performance and nutrient retention due to probiotics (*Lactobacillus*) supplementation reported by Panda *et al.*, 2006 and Talebi *et al.*,2008. Whereas improvement in growth performances and nutrient retention due to supplementation of *Saccharomyces* was confirmed by Kumpretchtova *et al.*, 2000 and Zhang *et al.,* 2005. However recently Khaksefidi and Rahimi 2005, Singh *et al.*, 2009, Chae *et al.*, 2012 reported that improvement in the performance in broiler chickens by supplementation of probiotics (*Lactobacillus, Saccaaromyces and Streptococcus*) in diets.

Feed intake was significantly ($P \leq 0.05$) less in *Lactobacillus* and *Saccaaromyces* supplemented groups up to 3 weeks of age. Later (4 to 6 weeks of age) there was increase in feed intake with increase in *Lactobacillus* and *Saccaaromyces* in diet. However numerically lower ($P \geq 0.05$) feed intake values was observed than control on cumulative basis.

Alkhalf *et al.*, 2010; Rajput *et al.*, 2013 reported significant increase in live body weight of broilers in Probiotics (*lactobacillus and S.boulardii*) supplemented groups than control without any significant variation in feed intake.

Feed conversion ratio was better ($P \geq 0.05$) at higher levels of *lactobacillus* and *S.boulardii* supplemented groups than control. This improvement in Body weight and FCR with supplementation of *lactobacillus* and *S.boulardii* might be due to maintenance of beneficial microbial population, improved feed utilization and digestion than altering bacterial metabolism.

Mechanism by which the probiotics improve growth performance include reinforcement of intestinal mucosal integrity by stimulating enzymatic activities, improving epithelial cell integrity, increasing immune response and better utilization of the diet. *lactobacillus* and *S.boulardii* has shown an improvement at the bird performance and decreased the mortality. This improvement may be related with the balanced microbial population in the gastrointestinal tract which has an important role in the health and performance of broilers.

The results of this research are similar to Paryad and Mahmoudi (2008) by incorporation of yeast @ 1.5%. Likely Zhang *et al.,* 2005 reported that yeast culture contains yeast cells as well as metabolites such as peptides, organic acids, oligosaccharides, amino acids, flavor and aroma substances, and possibly some unidentified growth factors, which have been proposed to produce beneficial performance response.

Gut morphology

observations are presented in Table 4. The current findings revealed significant ($p<0.05$) increase in intestinal villus height, width of villus, goblet cell number with linear increasing the level of Sb in the jejunum and ileum. These were on par with Rajput *et al.,* 2013. Maiorka (2000) and Loddi (2003) reported higher villi in the intestinal mucosa of birds fed diets with monoligosaccharides (MOS) at 7 and 21 days of age respectively.

In contrast to this Santos et al., 2004 reported that no difference in villus height between the control group and in birds receiving diets containing probiotics based on *Lactobacillus acidophilus* and *Casei, Streptococcus lactis* and *faecium, Bifidobacterium bifidum* and *Aspergillus oryzae* or prebiotics based on MOS.

Pelicano et al. 2005 reported greater cryptal depth (CD) (P<0.01) in birds which received Probiotics based on *Bacillus subtilis*, smaller in those diets without additives or with probiotics based in bacterial pool.

This could be attributed to Probiotics enhancing nutrient absorption by increasing the villus height in the small intestine (Zhang *et al.*, 2005; Panda *et al.*, 2006) and thus improve broiler performance. Probiotics compete with the harmful bacteria, change the pH in the gut, prevent infection and modifies mucin biosynthesis and /or degradation , which in turn influences gut function resulting in improved nutrient uptake (Smirnov et al ., 2005).

Increase in the villus height and cryptal depth than control suggests an increased surface area capable of greater absorption of available nutrients (Caspary, 1992). Likewise, greater villus height increases the activity of enzymes secreted from the tip of the villi resulting in improved digestibility (Hampson, 1986). Cell wall components of yeast may provide protective function to mucosa by preventing pathogens from binding to villi and allowing fewer antigens to be in contact with the villi. Taller villi indicate more mature epithelia and enhance absorptive function due to increased absorptive area of the villus.

Table 1: Effect of supplementation of *lactobacillus* and *saccharomyces boulardii* in diet on body weight (g/bird/week) in broilers

Levels	Day old	1stWeek	2nd week	3rd week	4th week	5th week	6th week
D1(Control)	47.10	117.8a	229.4	604.8	1039.1	1258.5d	2081d
D2(100g/T)*	47.75	108.5b	232.9	654.3	1067.0	1449.0c	2229c
D3(200g/T)*	47.95	113.5ab	214.5	681.6	1084.5	1516.5b	2390ab
D4(500g/T)#	46.90	110.3b	209.6	626.2	1109.8	1534.6b	2262bc
D5(1000g/T)#	47.05	107.9b	214.2	658.4	1096.4	1601.8a	2400a
Sem	0.37373	0.99394	5.55163	16.03841	27.11783	32.11637	23.54034
N	10	10	10	10	10	10	10
P value	0.874	0.007	0.114	0.187	0.244	0.026	0.067

**Lactobacillus*

Saccharomyces Boulardii

Table 2: Effect of supplementation of *lactobacillus* and *saccharomyces boulardii* through diet on feed intake (g/d) in broilers

Levels	1ST Week	2nd week	3rd week	4th week	5th week	6th week	Cumulative
D1(Control)	90.1a	162.7a	336.2a	477.4a	627.3b	897.0c	2590.7
D2(100g/T)*	85.4ab	157.6b	291.2b	450.2ab	636.0b	976.3bc	2596.6
D3(200g/T)*	88.0a	144.6c	267.4c	470.5a	533.1c	1021.4b	2525.0
D4(500g/T)#	82.1b	147.5bc	314.1a	464.2ab	680.5a	1082.8a	2771.2
D5(1000g/T)#	76.1c	154.3b	263.9c	414.3b	600.6bc	1114.2a	2623.4
SEM	0.867	1.9485	3.348	2.714	2.932	3.483	3.416
N	5	5	5	5	5	5	5
P value	0.006	0.018	0.004	0.041	0.036	0.006	0.067

*Lactobacillus # Saccharomyces Boulardii

Table 3: Effect of supplementation of *lactobacillus* and *saccharomyces boulardii* on feed conversion ratio in broilers

Levels	1ST Week	2nd week	3rd week	4th week	5th week	6th week	Cummulative
D1(Control)	1.307	1.410	2.037	2.386	2.195	2.320	1.942
D2(100g/T)*	1.271	1.478	2.247	2.370	2.203	2.283	1.975
D3(200g/T)*	1.290	1.483	2.362	2.304	2.461	2.340	2.041
D4(500g/T)#	1.343	1.421	2.312	2.389	1.978	2.089	1.922
D5(1000g/T)#	1.418	1.388	2.325	2.298	2.406	2.154	1.998
SEM	0.024	0.028	0.036	0.034	0.051	0.034	0.067
N	5	5	5	5	5	5	5
P VALUE	0.088	0.105	0.837	0.006	0.129	0.009	0.116

*Lactobacillus # Saccharomyces Boulardii

Table 4: Gut morphology of broilers supplemented with different levels of *lactobacillus* and *saccharomyces boulardii* through diet

Parameters (μm)	D1 (Control)	D2 (100g/T)*	D3 (200g/T)*	D4 (500g/T)#	D5 (1000g/T)#	SEM
			Jejunum			
Villus height	441.25	412.69	448.98	468.47	477.63	12.90
Cryptal depth	322.98	329.46	362.54	369.41	388.22	3.77
			Ileum			
Villus height	462.12	485.54	467 .36	502.41	517 .28	13.82
Cryptal depth	337 .65	341.54	358 .80	341.24	352 .71	9.15

**Lactobacillus*

Saccharomyces Boulardii

Conclusion

Beneficial effects were seen in production parameters as well as in histological indexes of the intestinal mucosa with the use of lactobacillus and *S.boulardii* in diets of birds in finisher phases.

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