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## Impact of probiotic containing *Lactobacillus sporogenes* on performance, feed efficiency and meat quality of broiler chicken

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### ABSTRACT

**Problem statement:** The use of antibiotics as growth promoters was completely banned in 1999 by the European Union (EU) European commission (2001). This was due to increases in microbial resistance to antibiotics and residues in chicken meat products which might turn to be harmful to the human consumers. Currently, in many parts of the world, feed additives such as probiotics and prebiotics are being experimented to alleviate the problems associated with the withdrawal of antibiotics from feed. The aim of this study was to investigate whether *Lactobacillus sporogenes* could improve the growth performance, feed conversion ratio and meat quality in broiler chicken.

**Approach:** A total of 200 broiler chicks were randomly assigned to 4 treatment groups, consisting of 2 replicates of 25 birds each. Commercial mash pre-starter, starter and finisher diets were supplemented with 50, 100 and 150 ppm of *L. sporogenes*.

**Results:** Growth performance was better in all the experimental groups than the control. Feed conversion ratio and TBA value were lowered in birds supplemented with 100 ppm of *L. sporogenes*.

**Conclusion:** This study concludes that supplementation of *L. sporogenes* to the broiler diet could improve the growth performance, decrease feed conversion ratio and TBA value, it could be recommended that addition of *L. sporogenes* at 100 ppm (100 mg/kg) could improve the growth performance of broiler chicken.

**Keywords:** Probiotic, Chicken, FCR, Performance, *Lactobacillus sporogenes*, TBA value.

**Introduction:**

Lilley and Stillwell (1965) first introduced the term "Probiotic" which means, "growth promoting factors" produced by microorganisms. The word "Probiotic" is derived from the Greek word 'probios' means 'for life'. Parker (1974) used the term probiotics for microorganisms or substances that contribute to intestinal microbial balance.

Probiotics are live micro-organisms that, when administered through the digestive tract, have a positive impact on the host's health. Mountzouris *et al.*, (2007) demonstrated that probiotic species belonging to *Lactobacillus*, *Streptococcus*, *Bacillus*, *Bifidobacterium*, *Enterococcus*, *Aspergillus*, *Candida*, and *Saccharomyces* have a potential effect on modulation of intestinal micro flora and pathogen inhibition. Supplementation of diets with a single strain of *Lactobacillus* sp. (*L. casei*, *L. fermentum*, *L. bulgaricus*, *L. reuteri*) was shown to improve the body weight and feed efficiency in broilers (Nakphaichit *et al.*, 2011; Salim *et al.*, 2013). Similar results were shown when broilers were given multiple strains of *Lactobacillus* sp. (Kalavathy *et al.*, 2003; Mookiah *et al.*, 2014). probiotics could protect broilers against pathogens by colonization in the gastrointestinal tract and stimulation of systemic immune responses.

Dietary probiotics (*S.cerevisiae*) could also improve meat tenderness and the oxidative stability of broiler meats (Takahashi and Jensen, 1984). Some antioxidant factors are present in probiotics (*S.cerevisiae*), if an antioxidant(s) from *S.cerevisiae* contributed to the lowered oxidation, that meats from the chickens fed *S.cerevisiae* contain less oxidative fat (or fatty acids)(Meyer *et al.*, 1994).

Lipid oxidation is one of the primary mechanisms of quality deterioration in meat products through adverse changes in flavor, color, texture and nutritive value. Chicken meat enriched with polyunsaturated fatty acids contains long fatty acids with a high number of double bonds, which increase the susceptibility of meat to oxidation. One of such product is malondialdehyde (MDA), which has long methods proposed for assessing MDA, the 2-thiobarbituric acid (TBA) has been widely adapted as a sensitive assay method for lipid oxidation in animal tissue. Zhang *et al.*, (2005) confirmed that TBARS values of both breast and thigh meat samples from broiler chickens that were fed with diets supplements of probiotic (*S.cerevisiae*) were significantly lower than those of control chickens.

The Thio Barbituric Acid (TBA) reactive substance test is most widely used method for quantifying lipid oxidation development in meat and meat products. The TBA test determines the amount of malondialdehyde (MDA), a major secondary by product of lipid oxidation, in an oxidized lipid. The present results show that the TBA value significantly decreases in broiler diet containing 100ppm of *L.sporogenes*. The TBA value is the measure of oxidative rancidity and carbonyl residue resulting from lipid per oxidation. It produces oxidative rancidity in foods and can decrease their nutritional quality.

In the same way, Zhang *et al.*, (2005) confirmed that TBARS values of both breast and thigh meat samples from broiler chickens that were fed with diets supplements of probiotics (*Lactobacillus*) were significantly lower than those of control chickens.

## Materials and Methods

### Birds, diet and Experimental Period

Two hundred day-old male broiler chicks (Ross) assigned to 25 chicks of 2 treatment groups, randomly. The experimental design was completely random, consisting of three dietary levels (50ppm, 100ppm and 150ppm of each two forms (powdery and granular) of *L. sporogenes* and a control group (without *Lactobacillus*) were formulated (Table 1). Each treatment had two replicates of 25 birds. Chicks fed three basal of Maize-soybean diets during three periods of 0-10d birds fed with broiler Pre-Starter, 11-20 birds fed with broiler Starter I, 21–30d birds fed with broiler Starter II, 31-36 d birds fed with broiler Finisher. The diets supplemented with amino-acids, minerals, and vitamins to meet all the Ross requirements (Ross 1994). The *L. sporogenes* (containing  $1 \times 10^9$  CFU/g) was provided from Pucheng (China).

### Chemical composition of the control diet (as fed):-

Ingredient (g/kg)		
Maize	-	563
Soya 48%	-	285
Fish meal	-	100
MBM	-	050
Rice bran oil	-	0.59
Methionine	-	0.26
Lysine	-	0.12
Threonine	-	0.31
Sodium carbonate	-	0.2
Salt	-	0.19
Water	-	0.49

**TABLE NO-1: Different Levels of *L. sporogenes* on Broiler Diet**

Treatment code	Experimental Diets	<i>L. sporogenes</i> (ppm)
T1	Basal diet without <i>L. sporogenes</i>	0.0 (consider as control)
T2	Basal diet with <i>L. sporogenes</i>	50
T3	Basal diet with <i>L. sporogenes</i>	100
T4	Basal diet with <i>L. sporogenes</i>	150

### Body Weight and Feed Consumption

Chicks were weighed individually and the feed consumption was measured weekly during the 10 days once of experiment period. Cumulative Weight Gain and Feed Consumption were determined, whereas weekly and cumulative Feed Conversion Ratio was calculated.

**Thiobarbituric Acid (TBA) value:****TBA- Thiobarbituric Acid Reactive substance:**

TBA value of Muscle and liver was determine according to the method describes by Tarladgis et al (1960). Muscle and liver samples that had been stored at  $-20^{\circ}\text{C}$  were thawed at  $4^{\circ}\text{C}$  and homogenized. Four subsamples, weighing approximately 2.5 g, from each of the Muscle and liver samples were weighed into 50-mL screw-capped centrifuge tubes and then incubated at  $30^{\circ}\text{C}$  for 10 d. After incubation, each subsample was immediately subjected to a malondialdehyde acid (MDA) assay to measure the extent of lipid oxidation. MDA, a secondary oxidation product, was determined (Sushil and Meliss, 1997). The amounts of 2-TBA-reactive substances (TBARS) were expressed as milligrams of MDA per kilogram of sample. The measurement of oxidative stability in skin samples was the same as outlined for Liver and Muscle samples except for the homogenization step. Intact skin samples were incubated from 0 to 10 d. immediately after incubation; skin samples were homogenized with 6 mL of 20% trichloroacetic acid and further processed as described above to measure the TBARS values.

**RESULTS AND DISCUSSION****Growth performance:**

The growth performance of broilers with different levels supplement with *L. sporogenes* was carried out from 1<sup>st</sup> day to 36<sup>th</sup> day. The mean body weight, feed consumption and feed efficiency of broiler chicks for 36 days are presented in Table 2-4.

The analysis of data on mean body weight (g) reveals significant differences ( $P<0.01$ ) at the end of the day. Birds fed with 100 ppm of feed *L.sporages* ( $2162.14 \pm 11.27$ ) has significantly increased body weight compared to control ( $1847.32 \pm 94.94$ ) birds (Table No 2).

**TABLE NO-2: Effect of supplementation of different levels of *L. sporogenes* on body weight (gm) of broiler chicks from 0-36 day.**

	10 <sup>th</sup> day	20 <sup>th</sup> day	30 <sup>th</sup> day	36 <sup>th</sup> day
Control	271.44 $\pm$ 14.126a <sup>#</sup>	794.7 $\pm$ 41.12a	1468.62 $\pm$ 75.67b	1847.32 $\pm$ 94.94b
50ppm	307.04 $\pm$ 4.03a	870.5 $\pm$ 5.70a	1671.34 $\pm$ 3.97a	1921.82 $\pm$ 116.16b
100ppm	310.06 $\pm$ 2.85a	908.92 $\pm$ 5.26a	1715.34 $\pm$ 7.68a	2162.14 $\pm$ 11.27a
150ppm	393.70 $\pm$ 13.95a	804.5 $\pm$ 49.83a	1510.08 $\pm$ 91.01b	2065.86 $\pm$ 10.16a
Treatment			892.06**	
Days			10.59**	
Treatment X Days			1.33 ns	

\*\* and <sup>ns</sup>, Significant at  $P<0.01$  and not significant respectively. Mean in a column followed by a same letter (s) are not significantly ( $P<0.05$ ) different according to Duncan's Multiple Range Test. <sup>#</sup> MeanQ  $\pm$ S.E

The feed intake was decreased significantly ( $P<0.01$ ) in 100ppm *L. sporogenes* (3207  $\pm$ 20.6) when compared to control (3497.92  $\pm$ 24.05) birds (table no 3).

**TABLE NO-3: Effect of supplementation of different levels *L. sporogenes* on feed intake (gm) of broiler chicks from 0-36 day.**

	10 <sup>th</sup> day	20 <sup>th</sup> day	30 <sup>th</sup> day	36 <sup>th</sup> day
Control	343.28±21a	1238 ±6.9a	2682 ±10.47a	3497.92 ±24.05b
50ppm	319.24±13.14a	1154.60 ±6.57a	2530 ±15.9ab	3465 ±23.75a
100ppm	318 ±3.99a	1144.96 ±96a	2362.56 ±12.11b	3207 ±20.6ab
150ppm	324.28 ±4.63a	1184.649±17.25a	2595.84 ±26.01ab	3260.80±96.64ab
Treatment			985.28**	
Days			2.85**	
Treatment X Days			<1	

\*\* Significant at  $P < 0.01$ , Mean in a column followed by a same letter (s) are not significantly ( $P < 0.05$ ) different according to Duncan's Multiple Range Test. # Mean ±S.E

The mean feed conversion ratio was significantly influenced by *L. sporogenes* fed groups ( $P < 0.01$ ) on 36<sup>th</sup> day. In 100ppm feed of *L. sporogenes* (1.766 ±0.09) showed significantly lower ( $P < 0.01$ ) feed conversion ratio than the control group (1.894 ±0.01) (Table No 4).

**TABLE NO-4: Effect of supplementation of different levels of *L.sporogenes* on feed conversion ratio of broiler chicks from 0-36 day.**

	10 <sup>th</sup> day	20 <sup>th</sup> day	30 <sup>th</sup> day	36 <sup>th</sup> day
Control	1.157±0.05a	1.455±0.07a	1.604 ±0.01a	1.894 ±0.01a
0.5%	1.110± 0.01a	1.416 ±0.04a	1.582 ±0.05a	1.868 ±0.09a
1%	1.022 ±0.01a	1.256 ±96b	1.472 ±0.09a	1.766 ±0.09a
1.5%	1.085 ±0.06a	1.420 ±0.06a	1.575 ±0.09a	1.847±0.07ab
Treatment			70.32**	
Days			5.47**	
Treatment X Days			<1	

\*\* and Significant at  $P < 0.01$  , Mean in a column followed by a same letter (s) are not significantly ( $P < 0.05$ ) different according to Duncan's Multiple Range Test. # Mean ±S.E

### STORAGE STABILITY

The mean (±S.E) TBA value of the muscle and liver of birds were influenced by dietary supplementation of *L.sporogenes* up to four week of storage period. TBA value was significantly increased at 150ppm of *L.sporogenes* in liver and muscle shown in table 5. The results provide evidence that supplementation of *L. sporogenes* to a Maize-soybean meal base control diet could improve oxidative stability of broiler meat. It may indicate that there are some antioxidant factors present in probiotics (*L. sporogenes*) or that probiotics (*L. sporogenes*) supplementation may shift the oxidative fat (or fatty acids) profile in the meat. Some antioxidant factors in probiotics (*L. sporogenes*) have been reported, such as glucose tolerance factor fractions (acts as an antioxidant; Ampel *et al.*, 2000) and copper-zinc superoxide dismutase (acts as oxidation-retarding factor; Meyer *et al.*, 1994). The probiotics (*L. sporogenes*), which contains  $\alpha$ -glucan,

carboxymethylglucan, mannans, and some proteinous substances, has been reported to display relatively good antioxidative properties (Ferencsik, *et al.*, 1986)

The mean TBA value of the muscle revealed significant ( $P < 0.01$ ) difference between treatment groups of storage period (10 days). The lowest TBA value was recorded than the control ( $1.488 \pm 0.07$ ) birds (Table no 5) when fed with 100ppm feed of *L. sporogenes* ( $1.037 \pm 0.028$ ) fed group.

**TABLE NO-5: Effect of supplementation of different levels of *L. sporogenes* on Storage Stability – TBA value of muscle and liver (mg malonaldehyde/kg) of broiler chicks**

	Muscle	Liver
control	1.488±0.07a	0.333±0.015a
0.5%	1.12±.23b	0.368±0.0427a
1%	1.037±0.028b	0.175±0.0134b
1.5%	1.148±0.0679b	0.243±0.0271ab
Treatment	922.24**	
Days	20.28**	
Treatment X days	5.6**	

\*\* Significant at  $P < 0.01$ . Mean in a column followed by a same letter (s) are not significantly ( $P < 0.05$ ) different according to Duncan's Multiple Range Test. # Mean  $\pm$  S.E

#### DISCUSSION:

Results of the present study showed that the dietary supplementation of 100 ppm of *L. sporogenes* in broilers ration improved body weight gain, feed intake and feed efficiency. The obtained results confirmed the previous findings of several researchers The use of different probiotics and prebiotics has shown reduced lipid oxidation in chicken meat by displaying lower thiobarbituric acid reactive substances (TBARS) (Marek Bobko *et al.*, 2015; Zhang *et al.*, 2005; Capcarova *et al.*, 2010).

The findings of the present study indicate the storage stability in muscle was bare minimum with the usage of 100ppm of *L.sporogenes*. And the storage was minimal in the liver with the usage of 100ppm of *L.sporogenes*. In agreement with this study, Tsiapali *et al.*, (2001) reveal that Supplementation of probiotics to a corn-soybean meal base control diet could improve oxidative stability of broiler meat. It may indicate that there are some antioxidants factors present in *Lactobacillus* supplementation may shift the oxidative fat (or fatty acids) profile in the meat. Some antioxidant factors in *Lactobacillus* have been reported, such as glucose tolerance factor fractions (acts as an antioxidant).

Feeding of probiotics to broiler chickens could improve growth performance in a dose-dependent manner. Dietary *L.sporogenes* could also improve meat tenderness and the oxidative stability of broiler meats (Takahashi and Jensen, 1984 and Ampel *et al.*, 2000). Some antioxidant factors are present in probiotics if an antioxidant(s) from probiotics contributed to the lowered oxidation, that meats from the chickens fed probiotics contain less oxidative fat (or fatty acids) (Meyer *et al.*, 1994).

**CONCLUSION:**

Broiler chicks fed with 100ppm of *L.sporogenes* had the higher BWG, FI, compared with the control group or other dietary treatments. TBA value of muscle and liver was also decreased with the inclusion of 100ppm of probiotics (*L.sporogenes*)

**DECLARATION:**

The growth performance and TBA values were improved with supplementation of 100 ppm (100mg/kg) of *L.sporogenes* in broiler chicken.

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