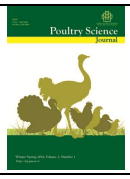

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Effect of Different Levels of Mushroom Waste (*Agaricus bisporus*) with or without Probiotic on Growth Performance, Carcass Characteristics, and Breast Meat Quality in Broiler Chickens

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Abstract

The aim of this study was to investigate the effect of mushroom waste and probiotic levels on growth performance, carcass characteristics, and meat quality in broiler chickens. A 2 × 3 factorial arrangement with two levels of probiotic supplementation (0 and recommended rate) and three levels of mushroom waste inclusion (0%, 3%, and 6%) was used in a completely randomized design using male broiler chickens (Ross 308 strain). Different levels of mushroom waste and probiotic had no significant effect on body weight gain and feed conversion ratio in broiler chickens. Neither different levels of mushroom waste nor probiotic independently had any significant effect on carcass characteristics. However, the use of mushroom waste and probiotics significantly reduced the malondialdehyde content in chicken breast meat 30 days after storage ($P<0.05$). Breast meat pH value was significantly reduced by supplementation with probiotic 1 and 30 days after storage ($P<0.05$). In conclusion, under the conditions of this experiment, inclusion of mushroom waste, particularly at 6% level, numerically improved growth performance and was effective in preventing meat oxidation.

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Introduction

In mushroom production units, large amounts of damaged, tiny, and deformed mushrooms as waste are obtained. Because this waste has a nutritional value, after drying, it can be used in the diet of broiler chickens. Mushroom (*Agaricus bisporus*) belongs to the kingdom of Fungi, which are considered as an important source of bioactive compounds possessing a medicinal value (Breen, 1990). The use of mushroom in poultry diet enhances growth performance and reduces gastrointestinal weight in poultry (Guo, 2003). Daneshmand *et al*. (2011) reported that adding mushroom to broiler diet improves feed conversion ratio but not body weight gain. Mushroom also contains considerable amounts of oligosaccharides, which have beneficial effects on the growth performance of broiler chickens (Falaki *et al*., 2011). It seems that the mechanism of action of fungi is similar to that of probiotic because fungi possess medicinal properties that can improve gastrointestinal function, which is due to the presence of polysaccharide compounds in the fungi (Cummings and Macfarlane, 2002).

It must be mentioned that different concentrations of methanol extract derived from mushrooms can eliminate free radicals. This antioxidant property of mushrooms is because of the presence of phenolic compounds (Yang *et al.,* 2002), which also possess antioxidant properties due to their renewal capacity as well as their chemical structure that enables them to neutralize free radicals (Rodriguez-Carpena *et al.,* 2011).

The use of probiotics, which are live microbial compounds, directly in poultry feed has quite desirable impacts on performance and health. The probiotics have allocated a special status to themselves, as their use does not decrease the durability of poultry carcass and also has beneficial effects on the productive properties of poultry (Cavasoni *et al*., 1998). There are controversial reports on the application of probiotics in poultry nutrition. In this regard, the research conducted by Silva *et al.* (2000) showed that the use of probiotic supplements in the diet of broilers led to an improvement in feed conversion ratio. A number of researchers also reported that probiotics have growth-stimulatory effects (Lan *et al.,* 2000; Mohan *et al.,* 1996). However, other studies showed that adding probiotic supplements had no effect on weight gain of broilers (Awad *et al*., 2009). The effectiveness of probiotics is further justified by the evidence that they impact the process of fat oxidation because broiler chickens receiving probiotics in their diet showed reduction in the amount of stored fat (Kot *et al.,*1995).

The aim of this study was to evaluate the growth performance, carcass characteristics, and meat quality parameters of broiler chickens fed on different levels of mushroom waste and probiotic.

Materials and Methods

Birds and housing

In this experiment, 108 male broiler chickens of the commercial Ross 308 strain were investigated in a completely randomized design with a 2 × 3 factorial arrangement consisting of two levels of probiotic (0 and recommended rate; 900 mg/Kg in the diet in starter period, 454 mg/Kg in grower period, and 225 mg/Kg in finisher period) and three levels of mushroom waste (0%, 3%, and 6%).

The experiment was performed using three replicates of six birds allocated to each of the dietary treatments. The birds were reared on deep litter floor pens for 35 days. They had free access to feed and water during the experiment. The temperature was maintained at 32°C during the first week and was gradually decreased by 3°C weekly until it decreased to 22°C, which was maintained constant up to the end of the experiment.

Dietary treatments

Before starting the experiment, mushroom waste was collected from local mushroom cultivation farms, washed, and ground in a mill. The chemical composition of mushroom wastes was measured based on AOAC (2009) procedures and contained 3.37% moisture, 3.29% crude protein, 6.13% crude fat, 21.19% crude fiber, 16.3% ash and 49.72% nitrogen free extract.

The experimental diets were formulated based on the requirements of Ross 308 strain (Avigen, 2009) for starter (0–10 days), grower (11–24 days), and finisher (25–35 days) periods. The composition of the experimental diets and the nutrient contents for starter, grower and finisher periods are shown in Table 1, 2 and 3, respectively. The probiotic was a trademark of Primalac, which included useful and viable microorganisms such as the bacterial strains, *Lactobacillus acidophilus* (2.5×10^7 cfu/g), *Lactobacillus casei* (2.5×10^7 cfu/g), *Bifidobacterium thermophilum* (2.5×10^7 cfu/g), and *Enterococcus faecium* (2.5×10^7 cfu/g).

Measuring growth performance and meat quality

Growth performance of broiler chickens was evaluated by recording the body weight, feed intake, and feed conversion ratio. Feed weight and chicken weight were determined in the beginning and at the end of each rearing period. At the end of the experiment (35 days), 36 birds with the body weight close to the related group mean body weight were selected (2 chickens per replicate), weighed, and sacrificed. After evisceration, hot carcasses were weighed immediately to determine the hot carcass yield. Weights of the cookable carcass, breast, thigh, and abdominal fat were recorded individually and were expressed as a percentage of preslaughter live weight of the birds.

The bird breasts were collected and assessed for meat quality after 1 and 30 days of storage in the freezer at −20°C. The pH was determined after homogenizing 10 gr of the sample in 50 mL double distilled water with a

standardized combination electrode attached to a digital pH meter (Thermo Orion, Model 420+, USA) (Naveena *et al.*, 2006). TBA was measured in duplicate according to the procedure described by Tarladgis *et al.* (1960). Water-holding capacity (WHC) was estimated by centrifuging 1 g of the muscles placed on a tissue paper inside a tube for 4 min at 1500×g. The water remained after centrifugation was quantified by drying the samples at 70°C overnight. WHC was calculated as [(weight after centrifugation - weight after drying)/initial weight] × 100 (Castellini *et al.*, 2002). The percentage of moisture was determined in duplicate according to the AOAC (2009) procedure.

Table 1. Ingredients and nutrient composition of experimental diets in starter period (0-10 days)

Ingredients (%)	Control	3%MW[1]	6%MW[1]	P[2]	3%MW+P	6%MW+P
Corn (7.29% CP)	47.36	42.53	37.71	47.18	42.36	37.53
Soybean meal (40.64% CP)	44.84	45.47	46.09	44.88	45.49	46.12
Soy oil	3.29	4.48	5.67	3.35	4.54	5.73
Mushroom waste (3.29%CP)	-	3.00	6.00	-	3.00	6.00
Dicalcium Phosphate	1.82	1.84	1.85	1.82	1.84	1.85
$Caco_3$	1.34	1.33	1.32	1.34	1.33	1.32
NaCl	0.37	0.37	0.38	0.37	0.37	0.38
Vitamin premix[3]	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix[4]	0.25	0.25	0.25	0.25	0.25	0.25
L-Lys	0.14	0.13	0.12	0.13	0.13	0.12
DL-Met	0.34	0.35	0.36	0.34	0.35	0.33
Primalac	-	-	-	0.09	0.09	0.09
Calculated composition						
ME (Kcal/Kg)	2900	2900	2900	2900	2900	2900
CP (%)	22	22	22	22	22	22
Ca (%)	1.05	1.05	1.05	1.05	1.05	1.05
P_a (%)	0.5	0.5	0.5	0.5	0.5	0.5
Na (%)	0.16	0.16	0.16	0.16	0.16	0.16
Lys (%)	1.43	1.43	1.43	1.43	1.43	1.43
Met (%)	0.7	0.7	0.7	0.7	0.7	0.7
Met + Cys (%)	0.08	0.08	0.08	0.08	0.08	0.08

[1]Mushroom waste; [2]Probiotic.

[3]The vitamin premix (11 Bro Basic, DSM, Delft, the Netherlands) provided the following per Kg of diet: 400 mg of Choline chloride, 12,000 IU of Vitamin A, 4,000 IU of Vitamin D3, 80 mg of Vitamin E, 9 mg of Vitamin K3 (Menadione), 3 mg of Thiamine, 7 mg of Riboflavin, 6 mg of Pyridoxine, 25 µg of Cyanocobalamin, 50 mg of Nicotinic acid, 15 mg of Pantothenic acid, 1.5 mg of Folic acid, and 150 µg of Biotin.

[4]The mineral premix (Rovimix Bro M, Roche, DSM) provided the following per Kg of diet:, 250 µg of Co, 1.5 mg of I, 300 µg of Se, 50 mg of Fe, 130 mg of Mn, 20 mg of Cu, and 100 mg of Zn.

Table 2. Ingredients and nutrient composition of experimental diets in grower period (11-24 days)

Ingredients (%)	Control	3%MW[1]	6%MW[1]	P[2]	3%MW+P	6%MW+P
Corn (7.29% CP)	50.54	44.56	40.93	50.45	45.66	40.84
Soybean meal (40.64% CP)	42.28	42.88	43.49	42.30	42.90	43.51
Soy oil	3.40	5.77	5.77	3.43	4.61	5.80
Mushroom waste (3.29% CP)	-	3.00	6.00	-	3.00	6.00
Dicalsium Phosphate	1.58	1.59	1.60	1.58	1.59	1.60
$Caco_3$	1.10	1.09	1.08	1.10	1.09	1.08
NaCl	0.37	0.37	0.30	0.37	0.37	0.38
Vitamin premix[3]	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix[4]	0.25	0.25	0.25	0.25	0.25	0.25
DL-Met	0.23	0.24	0.25	0.23	0.24	0.25
Primalac	-	-	-	0.045	0.045	0.045
Calculated composition						
ME (Kcal/Kg)	2950	2950	2950	2950	2950	2950
CP (%)	21	21	21	21	21	21
Ca (%)	0.9	0.9	0.9	0.9	0.9	0.9
P_a (%)	0.45	0.45	0.45	0.45	0.45	0.45
Na (%)	0.16	0.16	0.16	0.16	0.16	0.16
Lys (%)	1.27	1.27	1.27	1.27	1.27	1.27
Met (%)	0.58	0.58	0.58	0.58	0.58	0.58
Met + Cys (%)	0.95	0.95	0.95	0.95	0.95	0.95

[1]Mushroom waste; [2]Probiotic.

[3]The vitamin premix (11 Bro Basic, DSM, Delft, the Netherlands) provided the following per Kg of diet: 400 mg of Choline chloride, 12,000 IU of Vitamin A, 4,000 IU of Vitamin D3, 80 mg of Vitamin E, 9 mg of Vitamin K3 (Menadione), 3 mg of Thiamine, 7 mg of Riboflavin, 6 mg of Pyridoxine, 25 µg of Cyanocobalamin, 50 mg of Nicotinic acid, 15 mg of Pantothenic acid, 1.5 mg of Folic acid, and 150 µg of Biotin.

[4]The mineral premix (Rovimix Bro M, Roche, DSM) provided the following per Kg of diet:, 250 µg of Co, 1.5 mg of I, 300 µg of Se, 50 mg of Fe, 130 mg of Mn, 20 mg of Cu, and 100 mg of Zn**.**

Statistical analysis

All data were analyzed using the one-way ANOVA procedure of SAS (2002) for the analysis of variance. A 2 × 3 factorial arrangement with two levels of probiotic supplementation (0 and recommended rate) and three levels of mushroom waste inclusion (0%, 3%, and 6%) was used in a completely randomized design using male broiler chickens (Ross 308 strain). Significant differences among treatments were identified at 5% level by Duncan multiple range test.

Table 3. Ingredients and nutrient composition of experimental diets in finisher period (25-35 days)

Ingredients	Control	3%MW[1]	6%MW[1]	P[2]	3%MW+P	6%MW+P
Corn (7.29% CP)	57.17	52.36	47.57	57.02	52.32	47.52
Soybean meal (40.64% CP)	36.22	36.83	37.43	36.23	36.84	37.44
Soy oil	3.00	4.19	5.37	3.02	4.20	5.39
Mushroom waste (3.29% CP)	-	3.00	6.00	-	3.00	6.00
Dicalsium Phosphate	1.48	1.49	1.50	1.58	1.49	1.50
$Caco_3$	1.07	1.06	1.05	1.07	1.06	1.05
NaCl	0.37	0.37	0.37	0.37	0.37	0.37
Vitamin premix[3]	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix[4]	0.25	0.25	0.25	0.25	0.25	0.25
DL-Met	0.19	0.25	0.21	0.19	0.20	0.21
Primalac	-	-	-	0.022	0.022	0.022
Calculated composition						
ME (Kcal/Kg)	3000	3000	3000	3000	3000	3000
CP (%)	19	19	19	19	19	19
Ca (%)	0.85	0.85	0.85	0.85	0.85	0.85
P_a (%)	0.42	0.42	0.42	0.42	0.42	0.42
Na (%)	0.16	0.16	0.16	0.16	0.16	0.16
Lys (%)	1.12	1.13	1.13	1.12	1.13	1.13
Met (%)	0.52	0.52	0.52	0.52	0.52	0.52
Met + Cys (%)	0.86	0.86	0.86	0.86	0.86	0.86

[1]Mushroom waste; [2]Probiotic.

[3]The vitamin premix (11 Bro Basic, DSM, Delft, the Netherlands) provided the following per Kg of diet: 400 mg of Choline chloride, 12,000 IU of Vitamin A, 4,000 IU of Vitamin D3, 80 mg of Vitamin E, 9 mg of Vitamin K3 (Menadione), 3 mg of Thiamine, 7 mg of Riboflavin, 6 mg of Pyridoxine, 25 µg of Cyanocobalamin, 50 mg of Nicotinic acid, 15 mg of Pantothenic acid, 1.5 mg of Folic acid, and 150 µg of Biotin.

[4]The mineral premix (Rovimix Bro M, Roche, DSM) provided the following per Kg of diet:, 250 µg of Co, 1.5 mg of I, 300 µg of Se, 50 mg of Fe, 130 mg of Mn, 20 mg of Cu, and 100 mg of Zn**.**

Results and Discussion

Growth performance

Effects of mushroom waste, probiotic, and their interactions on growth performance of broiler chickens are shown in Table 4. Neither mushroom waste level nor probiotic independently had a significant effect on body weight gain. However, inclusion of mushroom waste and probiotic in the diet numerically increased body weight gain. Probiotic had no significant effect on feed intake. The effects of probiotic on weight gain were similar to the reports of Murry *et al.* (2006) and Awad *et al*. (2009) who reported that probiotic supplementation had no significant effect on the broilers weights. In contrast, Falaki *et al*. (2011) and Midilli and Tuncer, (2001) reported that probiotic supplementation increased body weight in broiler chickens.

It was difficult to directly assess different studies using probiotics because the efficacy of a probiotic application depends on many factors such as species composition and viability, administration level, application method, frequency of

application, overall diet, bird age, overall farm hygiene, and environmental stress factors (Patterson & Burkholder, 2003). In agreement with the results of our study, Pelicano *et al.* (2004) reported that probiotic supplementation had no statistically significant effect on the weights of broiler chickens. Ideal growing conditions and a non-stressful environment are effective in the biological response of the birds that consumed the probiotics (Mosenthin & Bauer, 2000).

The effect of different levels of probiotic and mushroom waste on feed intake of broiler chickens is shown in Table 4. Inclusion of mushroom waste in broiler diets resulted in an increase of feed intake. Broiler chickens that received mushroom waste at 6% level had a significantly higher feed intake than those birds fed on a control diet ($P<0.05$). Supplementation of probiotic to broiler diet had no significant effect on feed intake. No interactional effect was found between the levels of mushroom waste and probiotic on feed intake. Similarly, Hosseini *et al.* (2013) and Rahman *et al.* (2007) reported that probiotic had no significant effect on feed intake. It was hypothesized that probiotic not only enhanced the digestive rate but also increased the nutrients retention and decreased their passage rate (Rahman *et al.*, 2009).

The high feed intake in broiler chickens that consumed mushroom waste may be the result of the changing status of the digestive system. There is a substantial evidence that dietary mannan oligosaccharide (MOS) modifies the digestive enzyme activities, and amino acid transport in the digestive system, and can therefore increase feed intake (Iji *et al.*, 2001). In agreement with the results of this study, Kavyani *et al.* (2012) reported that the use of mushroom (*Agaricus bisporus*) had a significant effect on feed consumption. In contrast, there are some reports showing that the use of mushroom and mannan oligosaccharides has no effect on the feed consumption (Yalcinkaya *et al.*, 2008; Willis *et al.*, 2007). The discrepancy in these reports could be related to the differences in management and environmental conditions that exist in various experiments. It is suggested that under the beneficial management and/or environmental conditions, the effect of such feed additives may be worthless (Falaki *et al*., 2011).

Results on the effect of mushroom waste and probiotic on feed conversion ratio of the broiler chickens are shown in Table 4. Neither mushroom waste nor probiotic independently had a significant effect on feed conversion ratio. However, the use of mushroom waste or probiotic in the diet numerically decreased feed conversion ratio. No interactional effect was found between different mushroom waste levels and probiotic on feed conversion ratio.

Goodling *et al.* (1987) and Mutus *et al.* (2006) reported no significant improvement in feed conversion ratio with regard to adding probiotic. Timms, (1968) and Savage *et al.* (1968) reported that *Lactobacillus* is more effective under non-ideal research conditions such as intestinal damage due to coccidiosis *and mycosis.* Pelicano *et al.* (2004) reported an improvement in feed efficiency when MOS (1.1 g/Kg) was supplemented to the diet of broiler chickens from 1 to 21 days; however, this early improvement was not carried through 42 days of age.

These results combined with the earlier published studies show that the effects of oligosaccharides on growth performance of poultry are inconsistent under the research conditions (Biggs and Parsons, 2007).

Table 4. Effects of mushroom waste and probiotic on growth performance in broiler chickens during 1 to 35 d

Treatments	Body weight gain (g)	Feed intake (g)	Feed conversion ratio
Mushroom waste:			
0%	1802.30	3407.30[b]	1.90
3%	1960.20	3669.30[ab]	1.88
6%	2193.30	3847.80[a]	1.80
P-value	0.119	0.011	0.631
SEM[1]	196.680	221.570	0.051
Probiotic:			
0%	1968.00	3685.81	1.88
recommended rate	2002.50	3597.10	1.83
P-value	0.812	0.390	0.583
SEM[1]	24.363	62.730	0.036
Interaction:			
Control	1797.37	3535.53	2.08
3% Mushroom waste	2078.43	3619.62	1.74
6% Mushroom waste	2128.27	3902.28	1.83
Probiotic	1907.28	3279.00	1.71
3% Mushroom waste × probiotic	1841.88	3719.00	2.01
6% Mushroom waste × probiotic	2258.29	3793.28	1.76
P-value	0.420	0.371	0.050
SEM[1]	168.342	126.441	0.224

[1]Standard error of means.

[a,b] means with different superscripts differ significantly at $P<0.05$.

Carcass composition

The results of carcass composition including cookable carcass, thigh, breast, and abdominal fat relative weight are reported in Table 5. The results show that different levels of probiotics and mushroom waste had no significant effect on carcass composition. Moreover, no significant interactional effect was observed among the different levels of probiotics and mushroom waste on the body composition.

The present findings on carcass composition are in agreement with the findings of Kavyani *et al*. (2012) and Willis *et al*. (2007). Falaki *et al*. (2011) also reported no significant effect by supplementing different levels of Fermacto and Primalac to the broiler diet on the relative weight of thigh, carcass yield, and abdominal fat. Bitterncourt *et al.* (2011) mentioned that the efficacy or inefficacy of a probiotic product may be related to its microbial composition and viability, administration method and frequency, bird age, hygiene of the facilities, feed composition (cereals and their synergism or antagonism relative to the microbes in the product), as well as environmental stress factors.

Table 5. Effects of probiotic and mushroom waste on carcass characteristics of broiler chickens at 35 d (as % live body weight)

Treatments	Cookable carcass	Breast	Thigh	Abdominal fat
Mushroom waste:				
0%	64.38	23.25	20.11	2.43
3%	65.06	23.97	20.09	2.25
6%	66.22	24.40	20.10	2.43
P-value	0.358	0.584	0.999	0.478
SEM[1]	1.312	1.432	0.011	0.147
Probiotic:				
0%	64.16	22.20	19.83	2.25
recommended rate	66.28	21.74	20.38	2.49
P-value	0.050	0.477	0.215	0.209
SEM[1]	2.122	2.160	0.549	0.119
Interaction:				
Control	64.16	21.09	20.10	2.17
3% Mushroom waste	62.95	21.11	19.46	2.13
6% Mushroom waste	65.36	23.95	19.92	2.25
Probiotic	64.60	23.42	20.12	2.51
3% Mushroom waste × probiotic	67.16	24.84	20.72	2.18
6% Mushroom waste × probiotic	67.09	24.40	20.28	2.50
P-value	0.341	0.100	0.493	0.768
SEM[1]	1.903	1.642	0.633	0.196

[1]Standard error of means.
No significant difference was observed between treatments in each item ($P>0.05$).

Breast meat quality

The effects of mushroom waste, probiotics, and storage time 1 and 30 days after storage on the meat quality are shown in Table 6. Malondialdehyde content of the breast meat was not affected by different levels of mushroom waste and probiotic on day 1 after storage. In addition, no significant interactional effect was found in the malondialdehyde content of breast on day 1 after the slaughter. The malondialdehyde content of breast meat on day 30 after the slaughter was significantly affected by the levels of mushroom waste and probiotic ($P<0.05$). Broiler chickens fed on mushroom waste at 6% level significantly had the lowest malondialdehyde content after 30 days of storage among the other birds ($P<0.05$). Moreover, breast meat of broiler chickens that received probiotic also had a significantly lower malondialdehyde content than the birds not fed on probiotic ($P<0.05$). The interaction between mushroom waste and probiotic for malondialdehyde content in breast meat after 30 days of storage was statistically significant ($P<0.05$). This indicated that mushroom waste is more effective in reducing malondialdehyde content of breast meat in broiler chickens fed on a diet without probiotic supplementation.

Results related to the pH of the breast showed that after 1 and 30 days of storage, the pH was significantly lower in broiler chickens fed on probiotic diet than those birds not fed on probiotic ($P<0.05$). Neither mushroom waste nor the interactional effect between mushroom waste and probiotic had a significant effect on the pH value of the breast meat.

Results regarding the effect of dietary treatments on WHC showed that mushroom waste and its interaction with probiotic were significant at day 1 of storage ($P<0.05$). Mushroom waste at 6% level led to a significant increase of WHC ($P<0.05$). Different levels of mushroom waste and probiotic had no significant effect on the breast meat moisture content after 1 and 30 days of storage. A significant interactional effect was found between mushroom waste and probiotic at 1 and 30 days of storage for the meat moisture percentage.

Lin and Yen (1999) studied peroxidation of lipids by probiotics (*L. acidophilus* and *Bifidio bactriums*) and reported that probiotics have a protective role against lipid oxidation because of their ability to inhibit malondialdehyde. Moreover, the antioxidant effect of lactic acid bacteria has also been reported (Ahotupa *et al.,* 1996). Ivanovic *et al.* (2012) analyzed the limiting effect of probiotics on lipid oxidation and concluded that all probiotics have the ability to inhibit lipid peroxidation and reduce malondialdehyde content. The presence of heavy metals such as iron induces the process of fat oxidation. It seems that one of the mechanisms by which probiotics have antioxidant properties is by providing the conditions that lead to the release of iron from the tissues thus reducing the oxidation process. The other evidence justifying the effectiveness of probiotics on the process of fat oxidation is the reduction in the amount of fat stored in broilers receiving probiotics (Kot *et al.,* 1995).

Table 6. Effects of probiotic and mushroom waste on the meat quality of breast in broiler chickens

Treatments	Malondialdehyde (mg/Kg)		pH		WHC[1] (%)		Moisture (%)	
	1 d	30 d	1 d	30 d	1 d	30 d	1 d	30 d
Mushroom waste:								
0%	0.49	1.74^{a}	5.95	5.86	64.38^{b}	59.82	74.27	75.79
3%	0.48	1.71^{a}	5.90	5.82	65.74^{b}	60.26	76.06	76.14
6%	0.42	1.29^{b}	5.96	5.77	69.00^{a}	61.08	77.15	76.60
P-value	0.100	0.013	0.543	0.416	0.0003	0.310	0.094	0.141
SEM[2]	0.056	0.353	0.042	0.061	3.350	0.899	2.050	0.575
Probiotic:								
0%	0.47	1.74^{a}	6.01^{a}	5.90^{a}	65.83	60.30	75.54	75.77
recommended rate	0.45	1.43^{b}	5.86^{b}	5.69^{b}	66.91	60.48	76.11	76.58
P-value	0.448	0.022	0.002	0.0001	0.211	0.791	0.593	0.19
SEM[2]	0.021	0.308	0.151	0.266	1.07	0.177	0.567	0.802
Interaction:								
Control	0.55	2.44^{a}	6.00	5.98	57.93^{d}	58.62	70.02^{b}	74.44^{b}
3% Mushroom waste	0.45	1.71^{bc}	5.93	5.92	67.99^{ab}	61.04	77.82^{a}	76.01^{ab}
6% Mushroom waste	0.42	1.06^{bc}	6.11	5.95	71.58^{a}	61.24	78.80^{a}	76.88^{a}
Probiotic:	0.43	1.03^{c}	5.91	5.74	70.82^{ab}	61.03	78.52^{a}	77.14^{a}
3%Mushroom waste× probiotic	0.51	1.72^{b}	5.87	5.72	63.49^{c}	59.48	74.30^{ab}	76.28^{a}
6%Mushroom waste × probiotic	0.41	1.53^{bc}	5.80	5.59	66.42^{bc}	60.92	75.51^{ab}	76.31^{a}
P-value	0.058	0.0001	0.066	0.436	0.0001	0.059	0.0001	0.0008
SEM[2]	0.089	0.974	0.137	0.085	10.230	2.03	6.870	1.69

[1]Water holding capacity; [2]Standard error of means.

[a-c] mean values within a column with different superscripts differ significantly at $P<0.05$.

In this study, pH of the breast meat in broiler chickens fed on probiotic was significantly lower 1 and 30 days after storage, compared to that of the chickens not fed on probiotic. After sacrificing the birds, the blood flow is stopped in the body due to which the metabolic processes also start to cease slowly; but some of these processes continue for few moments at a faster rate after sacrificing by which glycogen is metabolized unaerobically resulting in the production of lactic acids. Lactic acid storage in the tissue thus lowers the pH (Asghar *et al.*, 2009).

Aksu *et al.* (2005) reported that the use of probiotics in the diet increases the WHC of breast meat. WHC and loss of moisture of meat after the slaughter depend on shortening of myofibrils, decrease of pH, denaturation of myosin, and formation of actomyosin. By protecting membrane phospholipids against oxidation, the available antioxidants in the diet can reduce the loss of moisture in the meat (Jensen *et al.,* 1998).

Few investigators reported that meat oxidation reduces sensitivity to hydrolysis and oxidation, lowers water storage between myofibrils, and finally lessens the meat moisture. Oxidation of lipids and proteins and all the factors that affect myofibrils have an impact on the loss of moisture in the meat. The presence of antioxidants reduces oxidation and its other secondary effects after the slaughter (Huff-Lonergan and Lonergan, 2005). Sazedul *et al*. (2010) reported that the use of probiotics increases the amount of moisture in the bristles.

Conclusion

The results of the present study showed that use of mushroom waste and also supplementation of probiotic to broiler diets improve growth performance and decrease meat oxidative indices, particularly after 30 days of meat storage.

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