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Physico-chemical litter amendments and their impact on broiler chicks' performance

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ABSTRACT

The objective of the study was to compare the effects of alternative litter treatments on litter quality, growth, carcass traits and welfare of broiler chicks. Day old, commercial broiler chicks (180) having similar body weight range were randomly allocated to two different types of litter treatments, viz. treated litter at the rate of 120°C (T_1) and litter treated with sodium bisulfate 25 g/sq. ft. (T_2) along with the control group without any litter treatment (T_0). Birds of all the treated groups performed better in terms of growth, feed intake and FCR, etc. Cake formation was frequent, highest in T_0 followed by T_1 and T_2 groups which in turn affected the frequency of undesirably poor sanitary outlook of the birds. Significantly lower EPG (*E. coli* count per gram) count while a numerically lower microbial load of the faecal samples was noted in the T_2 than T_1 and control. This implied the change in the *p*H and acidification of litter materials prevented the growth of coccidia and microbial load in the litter, thus making the litter more suitable for bird welfare. It was concluded that chemical litter amendments had a beneficial effect on overall growth performance, carcass characteristics, health and welfare of broiler chicks.

Keywords: Broiler growth, Heat treated litter, Litter quality, Sodium bisulfate abatement

The majority of intensive commercial broiler chickens are raised indoors on a deep litter system of housing using different types of litter materials such as sawdust, rice husk, oat hulls, sugarcane bagasse, chopped straw, paper mill by-products, sand, wood shavings, corn cobs, and dried leaves, etc. Litter comprises the bedding material plus excrement, feed, feathers, and water. An ideal bedding material is one, i.e. dry and absorbent, able to dilute the accumulated moisture and excreta from the birds reared on top of it, thus making itself reusable. However, with continued reuse, the accumulation of moisture and manure leads to several challenges for broiler producers such as growth depression, disease susceptibility, and induce severe discomfort in the form of contact dermatitis in broiler chickens (Eichner *et al.* 2007). Microbial decomposition of the manure accumulated within litter leads to the volatilization of ammonia into the surrounding atmosphere, lower bodyweights at ammonia levels (>25 ppm) and huge mortality (Miles *et al.* 2004) at excessive levels of ammonia (>75 ppm). Enteric pathogens have been identified in broiler litter (Wei *et al.* 2013), which raises risks of horizontal transmission and pathogen carryover effects between successive batches reared on the reused litter. Therefore, litter material used in poultry houses must fulfill hygienic and sanitary standards and permissible ammonia level during the entire rearing period (Villagra *et al.* 2011). Sahoo *et al.* (2017) concluded that alum (ATL) and sodium bisulphate (SBTL) treatment had a substantial impact on litter quality and increased broiler chick performance without having any negative effects. Animal waste treatment with heat to transform litter that tests negative or at less than the detection limit for *Salmonella*, the temperature range of 65 to 80°C for 30 to 60 min is recommended (National Organic Program 2006). It is necessary to achieve a minimum temperature of 150°C for 60 min, a moisture content of less than 30%, and negative or lower than the detection limit for *Escherichia coli* O157:H7, *Salmonella*, and faecal coliforms (CLGMA 2010). Keeping these facts and figures in mind, the objective of the study was to compare study effects of alternative litter treatments on litter quality, growth, carcass traits and welfare of broiler chicks.

MATERIALS AND METHODS

Ethical approval: The proposed design of the study, which ensured the comfort and welfare of the birds, was accepted by the institution's animal ethical committee vide number (GADVASU/2021/IAEC/62/22).

Experimental design and management: The experimental trial was conducted upon 180 Vencobb-430 broiler straight run chicks, where they were randomly assigned to three equal groups. One being the CONTROL(T_0) group having fresh paddy husk, and the other two comprising of Heat

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Treated Litter (HTL/T1) group where paddy husk was heated by dry heat method @ 120°C for 2 h and Sodium Bisulfate Treated Litter group (SBTL/T2) @ 25 g/sq. ft.

Each treatment was equivalently distributed into four replicates, each replicate having 15 birds each, which were reared for 42 days interval. The experimental study was conducted in the winter months from November 2021 to January 2022. During 3-4 weeks of age, mean temperature and Relative Humidity (RH) was 19.0°C and 54.8%, respectively in the shed. The temperature decreased whereas the relative humidity increased during the trial period. Throughout the experimental trial the temperature dropped with the increase of RH due to progressing winter season. The observations of the shed average RH and temperature were used to compute the average THI (Temperature Humidity Index) which was found to be 69.2 for the pre-starter, 64.0 for the starter, and 63.2 for the finisher phase. The chicks were provided a pre-starter diet (3055.30 kcal ME/kg, 22.03% CP) for the first 2 weeks, a starter diet (3053.20 kcal ME/kg, 21.55% CP) for 3rd and the 4th week and the broiler finisher diet (3116 kcal ME/kg, 19.56% CP) for the following last 2 weeks (Table 1). *Ad lib.* supplies of feed and clean water were always made accessible. AOAC (Association of Official Agricultural Chemists) International's (2005) methodologies were used to analyze the feeds for various contents.

Growth performance: Weekly records of live weight and feed intake per pen were made in order to calculate weight gain, the feed conversion ratio (feed/gain in body weight), the energy efficiency ratio (energy intake/gain in live weight), and the protein efficiency ratio (gain in live weight/protein intake) for each week.

Table 1. Ingredient composition of broiler pre-starter, starter and finisher rations used in the experiment (ICAR-2013)

Ingredient (kg/100 kg)	Pre-Starter	Starter	Finisher
Corn yellow	53.8	55.4	61.0
Soybean meal	34.5	33.2	28.0
Rice polish	5.0	5.0	5.0
Oil	2.7	3.0	3.0
Dicalcium phosphate	2.5	3.0	3.0
Limestone powder	1.0	1.0	1.0
Common salt	1.0	1.0	1.2
Additives	+	+	+
Methionine	0.025	0.016	0.017
Calculated Chemical Composition			
CP%	22.03	21.55	19.56
ME, kcal/kg	3055.30	3053.20	3116.00
Lysine %	1.20	1.10	0.95
Methionine %	0.61	0.60	0.53
Calcium	1.13	1.03	1.01
Available phosphorous	0.67	0.56	0.47

Additives included (per 100 kg): Liver tonic (Superlive TM) 25 g, Vitamin C 20 g, Choline chloride 50 g, Trace mineral 50 g (Iron 4000 mg, Copper 0.5 g, Manganese 6000 mg, Zinc 4600 mg, Selenium 10 mg, Iodine 80 mg), Vitamin A 825000 IU, Vitamin D3 165000 IU, Vitamin E 500 mg, Vitamin B12 0.025 mg, Vitamin K 100 mg, Thiamine 80 mg, Riboflavin 6 mg, Vitamin B6 160 mg, Niacin 1200 mg, Biotin 0.2 mg, Folic acid 1.0 mg, TM200 25 g, Coccidiostat 25 g. CP%, Crude Protein %; ME, Metabolisable Energy.

Litter quality and hygiene Bacterial count: Litter samples were collected individually from six different locations of all the pens. The bacterial load was calculated using Spread Plate Technique (Thomas *et al.* 2012). To distinguish between lactose fermenters and non-lactose fermenters, Mac Conkey agar (HiMedia® Mumbai) was chosen, where pink colonies indicated lactose fermentation and yellow or white colonies showed non-lactose fermentation. The total bacterial count (White colonies) was determined by using Brain Heart Infusion agar media (HiMedia® Mumbai). The number of colony forming units (CFU) per gram from the original aliquot/sample can be calculated as CFU per gram = the average number of colonies for a dilution × dilution factor.

Parasitic load: Faecal samples were taken from each pen and routinely checked for *Eimeria* oocysts every two weeks. The samples were microscopically analyzed. Fresh faecal material that was taken straight away after defecation was subjected to a faecal examination. The simple flotation method was used to do the qualitative analysis of the faecal sample. The number of oocysts per field was counted as per the Mc Master method (Bhatia *et al.* 2010).

Litter pH and nitrogen content: At weekly intervals, 10 g of litter was collected in a 100 ml beaker from each individual pen, and 50 ml of distilled water was added and thoroughly mixed with a glass rod. The material was left at room temperature for 30 min. The *p*H was then measured using a portable *p*H meter (BOECO Germany PT-380, pocket *p*H tester), which was calibrated using 4 and 9 standard buffers at room temperature. The AOAC International standard (2005) technique was used to estimate the nitrogen content of the litter.

Bird's health and hygiene: In order to keep track of their overall health, broiler chicks in each treatment group were checked daily. Four birds selected randomly from each group were examined for general cleanliness at the time of weighing, at weekly intervals. The data were recorded in the form of foot pad score and breast blister score relative to the litter quality of each group (Garcia *et al.* 2018).

Statistical analysis: The collected data were subjected to statistical analysis using Software Package for Social Sciences (SPSS Version 16.0). The recorded data were subjected to a one-way analysis of variance (Snedecor and Cochran 1994) with a comparison among means done by Duncan's multiple range test (Duncan 1995) with a significance level of ($p \leq 0.05$).

RESULTS AND DISCUSSION

Growth performance: The average initial body weight of day-old broiler chicks at the time of procurement was 37.6 g. The growth data (Table 2) indicated that during the first phase of the growth, the average weight gain was 2.56%, and 1.89%, higher respectively, in SBTL and HTL

Table 2. Overall production indices of broiler chicks under different treatments

Parameter	Treatments (Mean±SE)			p-value
	Control	HTL	SBTL	
Initial average body weight, g/bird	37.6±0.00	37.6±0.00	37.6±0.00	0.105
Final average body weight, g/bird (at 42 days)	1673.58±27.83	1705.16±20.90	1740.60±23.36	0.281
Average weight gain, g/bird	1635.98^b±64.16	1666.57^b±19.26	1725.75^a±28.76	0.041
Average feed intake, g/bird	3567.24±26.06	3534.32±1.31	3532.23±13.64	0.311
Feed Conversion Ratio	2.19±0.10	2.12±0.02	2.05±0.02	0.122
Protein Efficiency Ratio	2.24±0.10	2.30±0.02	2.39±0.03	0.116
Energy Efficiency Ratio	6.78±0.33	6.55±0.07	6.33±0.08	0.121
Survivability, %	99.33±4.41	100.00±0.00	100.00±0.00	0.102

Mean values bearing different superscripts in a row differ significantly ($p<0.05$).

groups over the control group. Though, both the treatment groups (HTL & SBTL), only had numerical difference with respect to weight gain. The final body weight of the broiler chicks was 1740 g/bird in acidified litter group, followed by 1705 g/bird in the chicks reared on heat treated litter group and 1673 g/bird in the control group. The data for overall FCR indicated that the values for FCR varied from 1.675 to 2.382 in all the treatment groups. The efficiency of utilization of feed was numerically better in HTL and SBTL as compared to the control in the first phase of growth. The variation in the efficiency to utilize the feed by the broiler chicks under the treatment groups and the control had non-significant ($p>0.05$) difference. Treatment groups were associated with more weight gain thus improved FCR (Feed Efficiency Ratio) and PER (Protein Efficiency Ratio) values indicated better efficiency of utilization of feed and protein in litter amendment groups (HTL and SBTL) than the control groups, whereas the efficiency of utilization of energy was found to be highest in the control group.

Previous studies on litter amendment similarly revealed improved weight growth and feed conversion for broilers raised over litter treated with sodium bisulfate as opposed to untreated litter (Sahoo *et al.* 2017).

Moisture content and litter pH : The numerical values of moisture percentage (Table 3, Fig 1) were found to be significantly ($p<0.05$) different between SBTL group as compared to both HTL and control group. This finding revealed the efficiency of litter treatment to control the moisture of litter which might have decreased the volatilization of NH_3 from the litter and improved the ambient environment of the bird. The *p*H of the litter surface decreased significantly ($p<0.05$) as a result of the chemical compound utilized in litter treatment, which was relatively acidic in the presence of water (Table 3, Fig 2). In the treatment groups, lower levels of litter moisture and *p*H are positively correlated with decreased bacterial activity and higher performance in terms of growth, carcass, and survivability rate. The count of *E. coli*, *Salmonella*, total bacterial count and coccidia load reduced remarkably due to the highly acidic nature and less water content of litter. Moreover, due to heat treatment which perhaps gave a sterilization effect on the litter material, there was numerically less growth of bacterial load in the 2nd week of sampling in the HTL group, as compared to the control. Although the experiment came to an end with almost similar values of bacterial load. Similar findings of acidification with sodium bisulfate in poultry litter were conducted by Hunolt *et al.* (2015), Sahoo *et al.* (2017) and Prosch *et al.* (2019).

Nutritive value of litter: The per cent available nitrogen, which serves as a direct indicator of the crude protein in the litter content, was statistically higher ($p>0.05$) in the SBTL

Table 3. Litter quality evaluation of various treatments

Period (week)	Parameter	Treatments (Mean±SE)			p-value
		Control	HTL	SBTL	
End of 2nd week	*p*H	6.97^b±0.68	7.01^b±0.16	4.23^a±0.75	0.00
	Moisture, %	17.81^b±0.10	11.56^a±0.31	12.70^a±0.62	0.003
	Ash, %	17.18^b±0.04	16.91^b±0.11	19.53^a±0.32	0.012
	N, %	1.50^b±0.03	1.67^b±0.08	2.36^a±0.15	0.011
End of 4th week	*p*H	7.90^b±0.12	7.73^b±0.07	6.85^a±0.19	0.00
	Moisture, %	29.00^b±1.80	20.40^a±0.40	20.60^a±0.20	0.017
	Ash, %	17.18^b±0.04	17.46^b±0.09	21.42^a±0.77	0.005
	N, %	3.31^b±0.06	3.70^a±0.26	3.76^a±0.39	0.017
End of 6th week	*p*H	9.73^b±0.01	9.42^b±0.07	7.38^a±0.06	0.00
	Moisture, %	35.60^c±1.2	31.60^b±0.40	26.60^a±0.20	0.008
	Ash, %	22.60^b±0.16	22.22^b±0.10	25.54^a±0.38	0.004
	N, %	3.51±0.38	3.78±0.25	4.00±0.30	0.006

Mean value bearing different superscripts in a row differ significantly ($p<0.05$).

Table 4. Microbial load of litter of different treatments

Period (week)	Parameter	Treatments (Mean±SE)			p-value
		CONTROL	HTL	SBTL	
End of 2nd week	*E.coli*, CFU in log_{10}	5.57±0.37	4.85±0.37	4.41±0.69	0.335
	Salmonella and *Salmonella* like microbes, CFU in log_{10}	5.10±0.31	5.03±0.05	4.87±0.40	0.868
	TBC,CFU in log_{10}	7.59±0.11	7.24±0.40	7.16±0.48	0.717
	Parasitic Count, Oocysts/gm	2775.00^{b}±85.39	2650.00^{ab}±64.54	2475.00^{a}±47.87	0.035
End of 4th week	*E.coli*, CFU in log_{10}	6.42±0.21	6.23±0.18	5.92±0.08	0.257
	Salmonella and *Salmonella* like microbes, CFU in log_{10}	5.81^{b}±0.06	5.60^{ab}±0.03	5.51^{a}±0.04	0.049
	TBC,CFU in log_{10}	8.76±0.03	8.40±0.39	8.06±0.76	0.649
	Parasitic Count, Oocysts/gm	7025.00^{c}±125.00	6675.75^{b}±137.68	5450.00^{a}±119.02	0.000
End of 6th week	*E.coli*, CFU in log_{10}	6.75±0.05	6.61±0.15	6.52±0.11	0.433
	Salmonella and *Salmonella* like microbes, CFU in log_{10}	6.81±0.05	6.71±0.42	6.47±0.33	0.747
	TBC, CFU in log_{10}	10.74±0.01	10.69±0.10	10.64±0.08	0.719
	Parasitic Count, Oocysts/gm	9125.00^{b}±137.68	8775.00^{b}±131.49	7125.00^{a}±137.68	0.000

Mean value bearing different superscripts in a row differ significantly (p<0.05).

group than in the HTL and control groups when samples were subjected to proximate analysis at the conclusion of the second week of the trial (Table 3, Fig. 3). By the end of the fourth week of age, the nitrogen level in the HTL group had slightly but significantly (p>0.05) increased, but they had continued to follow the same pattern as the analyses of the first two weeks. The SBTL group consistently maintained a greater nitrogen level than the HTL group between the two litter-treated groups (p>0.05). The acidic character of the treated litter may have prevented the free ammonium ion from converting to ammonia, causing more nitrogen to be retained. This higher level of nitrogen content among the litter-treated groups may have contributed to the higher crude protein percentage. The total ash percentage of the litter samples was found to an increasing trend throughout the trial (Table 3, Fig.4). The SBTL group consistently maintained a higher value which also differ significantly (p<0.05) from HTL and control groups. Although the ash% content of the HTL group had a numerically higher value than control, but it didn't differ significantly (p<0.05). The study was found similar in findings to Hunolt *et al.* (2015), Sahoo *et al.* (2017), and Prosch *et al.* (2019).

Bacterial load: Since the chemical compound used in litter treatment was relatively acidic in the presence of water, it caused a large drop in litter surface *p*H. Lower values of litter moisture and *p*H correlate positively with reduced bacterial activity and better performance with respect to growth, carcass and survivability rate in the treatment groups. The count of *E. coli*, *Salmonella*, total bacterial count and coccidia load reduced remarkably due to the highly acidic nature and less water content of litter. Moreover, due to heat treatment which perhaps gave a sterilization effect on the litter material, there was numerically less growth of bacterial load in the 2nd week of sampling in the HTL group, as compared to the control. The various parameters with respect to litter microbiology and coccidia count have been depicted in the Table 4. Although the experiment came to an end with almost similar values of bacterial load. Similar findings were found by McWard and Taylor (2000) and Sahoo *et al.* (2017). Reduced pathogen levels in the litter and on bird carcasses are a result of lower litter *p*H, according to Lines (2002).

Coccidia load: Despite all treatments, the data on parasite count (Table 4) increased with the bird's age. The samples from the SBTL group showed superiority in lowering the parasite count at the conclusion of the second, fourth, and sixth weeks of sampling, followed by the HTL group, with a statistically significant difference (p>0.05). The count of SBTL oocysts was significantly lower than that of the HTL group between these two treatment groups. The numerical difference between all of the treatment groups was, however, statistically insignificant.

Carcass traits: The data for carcasses indicated that the eviscerated weight varied from 58.88 to 59.99% under different treatment groups. Both the treated groups had almost similar value for the eviscerated weight, the control had a slightly higher but non-significant (p>0.05) value than the treatment groups. The higher per cent yield of thigh (16.96%) and drumstick (17.18%) was found to be statistically significant (p<0.05) in the SBTL and HTL groups, respectively.

Health and sanitary outlook: Throughout the trial, no treatment-related significant health issues were noted. Only two instances of aberrant leg movement were seen in the control group, which signified that the foot pad scored 3 as opposed to 0 in all treatment groups. Both the treatment and control groups had breast blister scores of 0. When comparing the size and thickness of the cakes, the control groups' litter cakes formed more frequently and intensively than those in the SBTL groups.

The following conclusions were drawn from the study, i.e. heat treatment at 120°C for 2 h and chemical abatement with Sodium Bisulfate @ 25 g/sq.ft to the litter could be a suitable alternative to improve the condition of the litter and, consequently, the growth, carcass, health status, behavioral expression, and well being of broiler chicks. Litter treatments established a solid foundation for its continued use with appropriate protocols in successive batches without having any negative effects on the growth, welfare, carcass, health, or behavioral expression of broiler chicks.

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