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RESEARCH ARTICLE

ALTERNATE METHIONINE SUPPLEMENTATION OPTION IN BROILERS: A FIELD STUDY

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ABSTRACT

Objective of the experimental trial was to study efficacy of herbal methionine supplement Methiorep for improving growth, performance & carcass traits in broilers. Trial was conducted on 200 day-old unsexed broiler chicks (Vencobb), randomly allotted to two treatment groups, T_0 & T_1 comprising 100 chicks each. Group T_0 supplemented with commercial feed only (no over and above Methionine- DLM supplementation was done) and in group T_1 commercial feed supplemented with Methiorep (*M/S Ayurvet Limited, India*) @1.5 kg/tonne of feed. Statistical analysis of results showed that in group T_1 a significant ($P<0.05$) increase in mean final body weight (g) and LWG (g) was observed in comparison to unsupplemented control group T_0. FCR, dressing %, bursa weight (g) and Spleen weight (g) parameters were significantly ($P<0.05$) improved in Methiorep supplemented T_1 group in comparison to unsupplemented T_0 group suggesting efficacy of Methiorep in improving poultry health and carcass traits. Colour of fresh meat was significantly better and desirable in Methiorep supplemented group as compared to control group. Cooked meat colour, flavour, juiciness, texture and overall acceptability of T_1 group was observed to be significantly better. Non-significantly ($P<0.05$) different but numerically lower mortality occurred in Methiorep supplemented T_1 group as compared to untreated control T_0 group. The trial investigation revealed that over and above supplementation of Methiorep (*M/S Ayurvet Limited, India*) @1.5kg/tonne of feed alongwith commercial ration in broilers helped in improving growth, performance, feed efficiency, carcass traits. The product was found to be safe for usage.

INTRODUCTION

Feed additives are products used in animal nutrition to better the quality of feed and the quality of food from animal origin, or to improve the animals' performance and health (EC., 2003). Essential amino acids like methionine cannot be synthesized by the bird and must therefore be fed in order to supply the building blocks needed in the synthesis of body proteins thereby supporting growth (Pesti., 2009). Synthetic methionine role in increasing body weight, carcass yield, over all health of poultry and in foetal development has been reported previously (Mack *et al*., 2004; Meirelles *et al*., 2003; Wu *et al*., 1998). But synthetic methionine is listed among the prohibited synthetic substances and its usage has been questioned in organic farming practices (Anonymous 1999). Synthetic ("pure") amino acids are produced either synthetically or from genetically engineered sources and involve the use of highly toxic and hazardous chemicals such as hydrogen cyanide, ammonia, and mercaptaldehyde. Synthesis of DL-methionine, and DL-methionine hydroxy analogs also result in significant pollution of the environment (Methionine 2001). More synthetic methionine in the diet is metabolized into highly toxic compounds like methyl propionate, thereby, adversely altering the performance of birds (Bender., 1975). Herbal preparations composed of single or multiple plant ingredients traditionally have been used for the health management in the poultry i.e tone up the liver, improves appetite, and increases disease resistance (Mwale *et al*., 2005; Okitoi *et al*., 2007; Prabakaran 2003). So the concept of herbal supplementation of methionine came in to existence. Herbal methionine as a source of active methionine has been claimed to be effective on performance, cost benefit ratio, meat and feather quality of broiler chicken (Halder *et al*., 2007; Kumari *et al*., 2012). The aim of the study is to evaluate efficacy of herbal alternate methionine supplement (Methiorep, *M/S Ayurvet Limited, Baddi, India*) in improving overall growth, productivity and performance in poultry birds.

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MATERIALS AND METHODS

The research was carried out at a poultry house in Kamthana, Bidar (KA), India. 200 day-old unsexed broiler chicks (Vencobb) were randomly divided in to two treatment

groups, T_0 and T_1 comprising 100 chicks each. Group T_0 supplemented with commercial feed only (Table1) (no over and above Methionine- DLM supplementation) and in group T_1 commercial feed supplemented with Methiorep (*M/S Ayurvet Limited, India*) @ 1.5 kg/tonne of feed. The commercial feed is claimed to comprise Methionine 300g/100kg feed in starter ration and 500g/100kg feed in finisher ration as per NRC recommendations.

Table 1. Feed ration for broilers

Ingredients	Starter phase (%)	Finisher phase (%)	Methionine (content)
Maize	49.00	50.00	0.98g
Soybean meal	18.50	15.00	1.32g
Groundnut cake	15.00	11.00	0.6g
Fishmeal	2.00	2.00	0.36g
Wheat offal	12.45	19.05	0.25g
Bone meal	2.00	2.00	0.13g
Salt	0.25	0.25	-
Premix	0.25	0.25	-
Methionine	0.30	0.5	-
Lysine	0.25	0.20	-
	100	100	

Table 2A. Effect of Methiorep™ supplementation via feed for 42 days on LBW of broiler chicks

Body weights (g)	T_0 (DLM)	T_1 (DLM+MRP)
Day 0	36.36 ± 0.11^a	38.25 ± 0.22^b
Day 7	104.6 ± 0.10^a	107.21 ± 0.26^a
Day 14	297.9 ± 0.08^b	293.16 ± 0.11^a
Day 21	630.11 ± 0.21^a	681.96 ± 0.16^a
Day 28	919.13 ± 0.16^a	1116.96 ± 0.12^a
Day 35	1512.13 ± 0.13^a	1610.96 ± 0.17^a
Day 42	2008.13 ± 0.12^a	2170.96 ± 0.10^a

Different superscripts a, b in the same row indicate significant differences ($P<0.05$) between the treatment groups. Results are expressed as mean ± standard deviation.

Table. 2B Effect of Methiorep ™ supplementation via feed for 42 days on BWG of broiler chicks

BWG (g)	T_0 (DLM)	T_1 (DLM+MRP)
BWG0-7	68.27 ± 0.20^b	68.96 ± 0.21^a
BWG 7-14	193.3 ± 0.09^b	185.95 ± 0.16^a
BWG14-21	332.2 ± 0.10^b	388.8 ± 0.10^a
BWG21-28	289 ± 0.11^a	435 ± 0.13^a
BWG28-35	593 ± 0.12^a	494 ± 0.18^b
BWG35-42	496 ± 0.15^a	560 ± 0.12^b
cBWG0-42	1971.77 ± 0.16^a	2132.71 ± 0.12^a

BWG: body weight gain; BWGi-i+1: body weight gain calculated weekly; cBWG0-42: cumulated body weight gain calculated for a period of 42 days. Results are expressed as mean ± standard deviation. Different superscripts a, b in the same row indicate significant differences ($P<0.05$) between the treatment groups.

During the first three weeks of trial, the chicks were fed *ad libitum* with standard starter crumble feed and thereafter with finisher Crumble. Birds of all the groups were vaccinated with new castle disease (ND) vaccine on 7th and 21st and 31st day and with infectious bursal disease (IBD) vaccine on 14th day. *Body Weight, feed consumption, feed conversion ratio, physical evaluation and mortality record* The performance parameters were recorded at weekly intervals. The chickens were weighed individually on day 1, 7, 14, 21, 28, 35 and 42. Feed intake was recorded weekly and Feed Conversion Ratios (FCR) were calculated (Tanweer *et al*., 2014). According to the recommendations for the euthanasia of experimental animals (Close *et al*., 1997; Farrell., 2006), the animals were sacrificed without stress via cervical dislocation. To minimize the stress to the broiler chickens, those that were expected to be slaughtered were selected 2 hr prior to sacrifice and moved to an isolated place out of the sight of the other broiler chickens. Immediately after cervical dislocation, the carotid arteries were cut and the chickens were bled for 100 seconds, after which they were placed in hot water (60ºC) for about 4 min. The feathers were then removed by passing through a rotary drum mechanical picker for 30 sec. After the blood, feathers, feet, heads (cut at the first cervical vertebra) and shanks were removed and evisceration was conducted, the whole carcasses were air-chilled (air flow of 6 m^3s^{-1}) and stored in a chilling room at 4ºC until the next day. The carcasses were weighed and deboned 24 hr after slaughter, and the breast muscle, thigh meat with skin, liver, gizzard, abdominal fat, spleen, bursa of fabricius and thymus were then removed and weighed. The weight of abdominal fat was determined by collecting all fat spreading to the ischium, around the cloaca, and into the neighboring abdominal muscles. The dressing percentage was calculated as the ratio of the carcass weight to the live weight. The weight of the breast muscle and thigh meat were expressed as their rates to the carcass weight and the relative weight of liver, gizzard, abdominal fat, spleen, bursa of fabricius, and thymus and are reported as the percentage of live weight. Mortality was recorded daily in each group (Oh *et al*., 2013).

Meat Colour Evaluation

The breast muscle was cut and exposed to air for 15 min at room temperature prior to color measurement. The meat color was then measured three times using white ceramic tile (L*=92.30; a*=0.32 and b*=.33 as standard). The meat colour was expressed as Hunter Values (L*=Lightness; a*=redness; b*=yellowness) (Hunter and Harold., 1987; Jiang *et al*., 2007; Kruk *et al*., 2014).

Statistical analysis

The data from the study were pooled and subjected to suitable statistical analysis. The data were represented as Mean±S.D. at $P<0.05$ level of significance as suggested by Steel (Steel *et al.*, 1997)

RESULTS AND DISCUSSION

Body weight and weight gain

The weekly body weight and weight gain of broilers have been presented in table 2A and in table 2B. Statistical analysis of results showed that in group T_1 a significant ($P<0.05$) increase in mean final body weight (g) (2170.96 ± 0.10^a) was evident in comparison to unsupplemented control group T_0 (2008.13 ± 0.12^a) (table 2A). LWG in Methiorep supplemented T_1 group (2132.71 ± 0.12^a g) was observed to be significantly ($P<0.05$) higher by 160.94 g/bird in comparison to untreated control T_0 group (1971.77 ± 0.16^a g) as shown in table 2B. The results of the present trial are in accordance with the findings

of Kalbande, Narayanswamy and PN Onu who reported that the chicks in herbal methionine group showed a significant (p<0.01) gain in body weight as compared to the chicks in control group and numerically higher weight gain as compared to the chicks in synthetic methionine group (Kalbande *et al.*, 2009; Narayanswamy and Bhagwat., 2010; Onu *et al*., 2010). These studies showed that the herbal source of methionine can replace DL-methionine in the diet of commercial broiler chicks.

Table 3. Effect of Methiorep ™ supplementation via feed for 42 days on FCR of broiler chicks

FCR (g/g)	T_0 (DLM)	T_1 (DLM +MRP)
FCR 0-7	1.69± 0.02^{b}	1.19± 0.12^{a}
FCR 7-14	2.16± 0.22^{b}	1.92± 0.22 ^{a}
FCR14-21	1.91± 0.20^{ab}	1.62± 0.16^{a}
FCR21-28	2.68± 0.12^{ab}	1.62± 0.20^{a}
FCR28-35	1.64± 0.05^{b}	1.78± 0.12^{b}
FCR35-42	2.13± 0.07 ^{ab}	1.71± 0.08 ^{b}
cFCR0-42	2.01± 0.16^{b}	1.79± 0.20^{a}

FCR: Food conversion ratio; FCRi-i+1:Food conversion ratio measured weekly; cFCR0-i: cumulated Food conversion ratio determined for a period of i days. Different superscripts a, b in the same row indicate significant differences (P<0.05) between the treatment groups. Results are expressed as mean ±standard deviation.

Table 4. Effect of Methiorep ™ supplementation via feed for 42 days on the carcass traits and Immune organ weights in broiler chickens

Parameters	T_0 (DLM)	T_1 (DLM +MRP)
Slaughter weight (g)	2008.13±0.12 ^{a}	2170.96±0.10 ^{a}
Hot carcass weight (g)	1324.4 ± 14.2 ^{ab}	1545.1 ± 28.6 ^{a}
Carcass yield (%)	65.95 ± 0.8^{b}	71.71 ± 0.8 ^{b}
Spleen weight (g)	2.41±0.17^{a}	3.72±0.84^{b}
Spleen yield (%)	0.12± 0.01 ^{a}	0.17 ± 0.03 ^{b}
Bursa weight (g)	3.63 ± 0.43 ^{a}	5.42 ± 0.84 ^{b}
Bursa yield (%)	0.18 ± 0.01 ^{a}	0.24 ± 0.03 ^{b}

Organ yield was calculated by the following formula: 100 x organ weight / carcass weight. Different superscripts a, b in the same row indicate significant differences (P<0.05) between the treatment groups. Results are expressed as mean ±standard deviation.

Feed conversion ratio

Cumulative FCR of group T_1 (1.79± 0.20^{a}) was found to be significantly (P<0.05) better from the control group T_0 (2.01± 0.16^{b}) as shown in table 3. The results are in accordance with the findings of Garcia Neto M, Garlich JD and Chattopadhyay who claimed an increase in FCR by methionine supplementation (Garcia *et al*., 2000; Garlich., 1985; Chattopadhyay *et al.*, 2006).

Carcass traits and Immune organ weights

The effect of supplementation of Methiorep on carcass traits and visceral organs weight expressed as percent of live weight is presented in table 4. Carcass yield % was found to be significantly (P<0.05) higher in T_1 (71.71±0.8^{b}) as compared to T_0 group (65.95±0.8^{b}). Significantly (P<0.05) improved bursa weight (g) was observed in T_1 group (5.42±0.84^{b}) in comparison to control T_0 group (3.63±0.43^{a}) suggesting efficacy of Methiorep in improving immunity. Spleen weight (g) was observed significantly (P<0.05) higher (3.72±0.84^{b}) in Methiorep supplemented T_1 group in comparison to control T_0 group (2.41±0.17^{a}). The results are in accordance with Ojano-Dirain and Waldroup, who observed that a significant improvement (P<0.05) in dressing percentage and breast meat yield between the broilers fed NRC methionine level and those fed higher levels. Study showed that on Methionine supplementation there is an improvement in the carcass traits (Ojano-Dirain and Waldroup., 2002). Fresh meat colour, sensory evaluation of cooked chicken and mortality Color and odour of cooked poultry meat is important because consumers associate it with the product's freshness, and they decide whether or not to buy the product based on their opinion of its attractiveness. Colour of fresh meat was significantly better and desirable in Methiorep supplemented group as compared to control group as shown in Table 5A. Flavour, juiciness of meat and other important organoleptic characteristic have been presented in table 5B.

Table 5A. Effect of Methiorep ™ supplementation via feed for 42 days on fresh Meat colour

Parameters	T_0 (DLM)	T_1 (DLM+MRP)
Lightness	56.32^{b}	61.23^{a}
Redness	3.16^{a}	3.82^{ab}
Yellowness	14.13^{a}	17.27^{b}

Different superscripts a, b in the same row indicate significant differences (P<0.05) between the treatment groups.

Table 5B. Effect of Methiorep ™ supplementation via feed for 42 days on the Sensory attributes of cooked Chicken using 8-point hedonic scale

Parameters	T0 (DLM)	T1 (DLM+MRP)
Color/ Appearance	6.97 ^{a}	7.17 ^{b}
Flavour	6.84 ^{a}	7.17 ^{b}
Juiciness	6.76 ^{a}	6.98 ^{b}
Texture	7.11 ^{a}	7.07 ^{a}
Saltiness	7.02 ^{a}	7.07 ^{a}
Mouth Coating	6.47 ^{a}	6.86 ^{a}
Overall Acceptability	6.72 ^{a}	7.03 ^{b}

Different superscripts a, b in the same row indicate significant differences (P<0.05) between the treatment groups.

Table 6. Effect of Methiorep ™ supplementation via feed for 42 days on the mortality rates in broiler chickens

Parameters	T_0 (DLM)	T_1 (DLM+MRP)
Total No. of Birds in the group	100	100
No. of birds Died	7	6
Mortality percentage	7.00	6.00

Sensory attributes of cooked chicken of T_1 group was observed to be significantly (P<0.05) better and acceptable (7.03^{b} vs 6.72^{a}) as per the 8-point hedonic scale method given by Keeton (Keeton 2006). It can be inferred that supplementation of Methiorep might have lead to improvement in collagen and myofibrillar solubility, in turn improving the tenderness of edible muscles. Mortality in both groups was noted during the entire trial. The percentage mortality recorded in both groups is shown in table 6. In Methiorep supplemented T_1 group (6%) non-significantly (P<0.05) different but numerically lower mortality was recorded as compared to untreated control T_0 group (7%). The results are in accordance with Halder and Roy (Halder and Roy 2007) who observed

relatively lower mortality in herbo methionine supplemented group in comparison to synthetic DL-methionine supplemented group.

Conclusion

On the bases of present investigation it can be concluded that herbal feed premix Methiorep can successfully replace synthetic DL methionine in feed as it has been proven to be effective in improving commercial broiler performance (growth, FCR, and livability parameters).

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