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Effect of Supplementation of *Tinospora cordifolia* (at Graded Levels) and Ascorbic Acid on Nutrient Metabolizability and Feed Passage Rate of Broiler Chickens

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ABSTRACT

A study was conducted September to October, 2016 to investigate the effect of *Tinospora cordifolia* (at graded levels) or ascorbic acid alone and in combinations on nutrient metabolizability and feed passage rate using 360 one day old broiler chicks. The experiment was done in 5×2 factorial experiment within a completely randomized design and divided into ten dietary treatments groups (T_1–T_{10}) in triplicate of 12 chicks/replicate. Following a 6-week feeding trial, five days metabolic trial was carried out at the Experimental Poultry Unit, located in poultry farm of College of Veterinary and Animal Science, Bikaner. The average temperature (31°C) during the study period was higher than the recommended normothermia zone *i.e.* 22–28°C established for poultry, which showed that birds were in chronic heat stress. The metabolizability (%) of all nutrients remained non-significant due to interaction (Giloy×Ascorbic acid) effect. Further, due to effect of Giloy only the metabolizability (%) of crude protein showed significant effect, while due to effect of ascorbic acid only metabolizability (%) of dry matter showed significant effect.Chronic heat stress in present study showed negative effect on nutrient metabolizability and feed passage rate in control group as compared to various treatment groups. Improvement in nutrient metabolizability and feed passage rate was observed due to supplementation of Giloy (*Tinospora cordifolia*) or ascorbic acid alone and their various combinations in broiler feeding. It was concluded that supplementation of *Tinospora cordifolia* (Giloy) at 0.25% level with ascorbic acid @ 0.025% may be as a beneficial management strategy in poultry feed during chronic heat stress.

KEYWORDS: *Tinospora cordifolia*, ascorbic acid, feed passage rate, metabolizability

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1. INTRODUCTION

Indian poultry industry has made a tremendous and remarkable progress and become highly organized and most sophisticated industry from a small scale backyard to the status of self-sufficient agro based industry (Shisodiyaet al., 2008). The environmental temperature during summer is recorded more than 34°C in arid zone of Rajasthan, which results into stress on the birds. Poorly developed poultry industry in the Western region of Rajasthan is mainly due to adverse climatic conditions. It is well known that in poultry the thermo neutral zone ranges from 13–24°C. Beyond that, temperature on either side of the critical zone (24–38°C and 0–13°C) is detrimental. At an average humidity, birds start panting, when the ambient temperature reaches 29.4°C (North andBell, 1990). The feed additives that hold great promise in the feeding of poultry comprises of anti-oxidants, antibiotics, coccidiostates, enzymes, probiotics, organic acid, mould inhibitor, hormones, herbals products etc. It was reported that the negative effects of environmental stress could be prevented by the use of feed additives such as vitamin C (Sahin and Kucuk, 2001) and phytogenic feed additives (Windisch et al., 2008). Natural feed additives of plant origin are generally believed to be safer, healthier and economic. Various herbs and plant extracts, and their essential oils have anti-microbial activities and antioxidant properties, which make them useful for quality safe meat production (Faixova andFaix, 2008). Phytogenic feed additives influence improvement of consumption and conversion of food, digestibility and gain of broiler chickens. However, in reference to the effect of plant- or plant extract-enriched diets on production traits of chickens, the results are inconsistent amongst different studies (Erener et al., 2011,Viveros et al., 2011, Farahat et al., 2016). The herb *Tinospora cordifolia* (Family- Menispermaceae) grows in the tropical and subtropical regions of India (Sengupta et al., 2011). *Tinospora cordifolia* (Menispermaceae) commonly known as 'Giloy', a Hindu mythological term refers to the heavenly elixir. Giloy stem and root is being used in the traditional medicinal system since ages.*Tinospora cordifolia* is a large, glabrous, deciduous climbing shrub, which exhibit various properties as described in ancient Ayurvedic texts like Jwarahara, Rasayana, Agnideepana, Vataraktanashaka, Sangrahi, Balya, Mehnashaka, Kasa-swasahara, Sarvakusthahara, Krimihara, Prameha, Arshnashaka, etc., and is remarkably validated by modern research evidences (Sharma and Murthi, 2019). Vitamin-C is a white to yellow-tinged crystalline powder. Vitamin-C occurs in two forms, namely dehydro-L-ascorbic acid (oxidized form) and L-ascorbic acid (reduced form) and both forms are biologically active. Vitamin-C is very susceptible to destruction through oxidation, which is accelerated by heat and light. Reversible oxidation-reduction of ascorbic acid with dehydroascorbic acid is the principal chemical property of vitamin-C and the basis for its well-known physiological activities and stabilities. Vitamin-C, an antioxidant, which is normally being synthesized by the chicken (Khan, 2011) and its biosynthesis mainly occurs in the kidneys. Although chickens are known to synthesize ascorbic acid, increased supplementation has proved to have beneficial effects in broilers reared under heat stress (Mahmoud et al., 2004). Though, it is not essential in birds but its role during various states is quite critical so total dependency in endogenous supply cannot be relied specially during heat stress. Therefore, the research study was planned to investigate the effect of *Tinospora cordifolia* (at graded levels) or ascorbic acid supplementation alone and in combinations on nutrient metabolizability and feed passage rate of broiler chickens during chronic heat stress.

2. MATERIALS AND METHODS

A 42-days feeding trial was carried out at the Experimental Poultry Unit, located in poultry farm of College of Veterinary and Animal Science, Bikaner during September to October, 2016. The 360 experimental day old broiler chicks were equally and randomly divided into ten dietary treatments groups (T_1–T_{10}) and each dietary group was replicated to three sub-groups (R_1–R_3) to make sure uniformly in various treatment groups. Diets included: T_1-basal diet with no supplementation;T_2-basal diet supplemented with 0.25% *Tinospora cordifolia*; T_3-basal diet supplemented with 0.50% *Tinospora cordifolia*; T_4-basal diet supplemented with 0.75% *Tinospora cordifolia*; T_5-basal diet supplemented with 1.0% *Tinospora cordifolia*; T_6-basal diet supplemented with 0.025% ascorbic acid; T_7-basal diet supplemented with 0.25% *Tinospora cordifolia* plus 0.025% ascorbic acid; T_8-basal diet supplemented with 0.50% *Tinospora cordifolia* plus 0.025% ascorbic acid; T_9-basal diet supplemented with 0.75% *Tinospora cordifolia* plus 0.025% ascorbic acid and T_{10}-basal diet supplemented with 1.0% *Tinospora cordifolia* plus 0.025% ascorbic acid. Broilers were maintained under standard managemental practices regarding brooding, feeding, watering and disease control throughout the trial period. The commercially available ascorbic acid (99.99% pure), was used. Good quality of Giloy (*Tinospora cordifolia*) stem was procured from reputed firm of Bikaner (Rajasthan). Thereafter, it was identified and authenticated by the Department of Botany, Govt. Dungar College, Bikaner (Rajasthan).The proximate analysis of experimental feed mixtures was carried out by the methods of Anonymous (2005). During the study period, digital thermohygrometer was used to record temperature and relative humidity thrice a day at 08:00, 14:00 and 22:00 hours, respectively. THI values were calculated from the recorded measurement as described below by (Kibler, 1964).

At the end of six weeks, metabolizability of different dietary principles were conducted using six chicks from each group for 5 days. During metabolizability study, the six birds from each treatment (two bird from each replicate) subjected to metabolizability study were transferred to metabolism cages. Polythene sheet of appropriate size was spread over the dropping trays for collection of mixed excreta in each group. The chicks were offered a weighed amount of experimental ration at a fixed morning hour (7:30 AM) every day during the trial period. The mixed droppings were collected at the end of every 24 hours and pooled to get the total excreta voided during the trial period. Daily feed intake was calculated after deducting the left over from the feed offered. Representative feed samples were drawn from the bulk, finally ground and stored in sample bottles for analysis. The group wise aliquots from droppings after thorough mixing with the help of spatula were drawn for dry matter and nitrogen estimation, separately. For nitrogen estimation, samples in duplicate were preserved in 5 percent sulphuric acid in wide-mouth glass stopperd bottles and kept in refrigerator. Dry matter determination of excreta was done in duplicate for each group by keeping the weighed excretal material in an oven at 85°C till constant weight was obtained. The treatment wise dry matter metabolizability (DMD %) of diet was determined using following formula:

Dry matter digestibility (%)=((A)-(B))/(A)×100………. (1)

Where: A=Weight of the dry matter consumed (g)

B=Dried weight of the excreta voided (g)

Similarly, the metabolizability of crude protein, ether extract, crude fiber, NDF and ADF was determined using following formula:

Digestibility cofficient of nutrient=((A)-(B))/(A)×100..(2)

Where: A=Unit nutrient intake (g)

B=Unit nutrient outgo (g)

Feed passage rate in chicks was assessed at the end of 6th week (43rd day) of age by using an inert dye Cr_2O_3, a known amount of which was mixed well in the respective diets. The six birds from each treatment (two birds from each replicate) were fasted overnight. Clean drinking water was available *ad lib.* On the day of trial, at 10 am, the respective feed and faecal trays were introduced into the cages of all replicates and the test diet-containing marker was offered to chicks for a period of half an hour only. Thereafter the respective feed (without marker) was offered. Droppings from the respective lots of chicks were watched for appearance of green color and the time of 1st appearance of droppings for each lot of chicks was recorded separately to observe the feed passage rate. The data obtained in the experiment were analysed statistically for main effect of *Tinospora cordifolia* or ascorbic acid alone as well as interaction (Giloy×Ascorbic acid) in factorial design (5×2) as per Snedecor and Cochran (2004) and significance of mean differences was tested by Duncan's New Multiple Range Test (DNMRT) as modified by Kramer (1957).

3. RESULTS AND DISCUSSION

The mean values of temperature (°C), relative humidity (%) and temperature humidity index (THI) recorded during different weeks have been presented in Table 1. The calculated THI for different weeks was obtained to be in range from 75.26 to 82.06; a value above the THI threshold of 70, established for poultry. The average temperature (31°C) during the study period was higher than the recommended normothermia zone *i.e.* 22–28°C (Donkoh, 1989) established for poultry in the tropical regions, which indicated that birds were in chronic heat stress.

Table 1 : Mean temperature (°C), relative humidity (%) and THI range observed during different weeks

Period (weeks)	Temperature (°C)				Relative humidity (%)				THI			
	8 am	2 pm	10 pm	Avg.	8 am	2 pm	10 pm	Avg.	8 am	2 pm	10 pm	Avg.
I	32.24	35.54	31.44	33.08	53.30	42.09	53.21	49.53	81.66	83.68	80.58	82.06
II	31.13	33.40	33.83	32.79	50.86	42.71	39.00	44.19	79.76	81.18	80.98	80.70
III	30.59	32.87	32.84	32.10	58.43	52.71	44.71	51.95	80.28	82.39	80.87	81.23
IV	28.94	31.14	29.87	29.99	66.71	59.00	60.14	61.95	79.22	81.15	79.56	80.01
V	27.16	30.46	29.53	29.05	46.43	43.29	42.00	43.90	74.00	77.66	76.32	76.01
VI	27.45	33.31	26.40	29.05	46.03	27.30	42.94	38.76	74.31	78.13	72.62	75.26
Mean	29.58	32.79	30.65	31.00	53.63	44.52	47.00	48.38	78.21	80.70	78.49	79.21

The chemical composition of experimental feed offered to the broilers has been presented in Table 2. The proximate composition of experimental feeds was almost similar for all the treatment groups and fed as per Anonymous (1992). The data of metabolizability coefficients (%) of nutrients in different treatment groups have been presented in Table 3. The statistical analysis of data of dry matter metabolizability (%) revealed non-significant effect on

Table 2: The chemical (% DM basis) composition of experimental feed offered to broiler chicks

Parameters	Treatment groups									
	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	T_9	T_{10}
DM	91.41	91.42	91.42	91.43	91.43	91.39	91.39	91.40	91.40	91.41
CP	20.32	20.27	20.23	20.18	20.13	20.32	20.27	20.22	20.17	20.13
EE	4.60	4.59	4.58	4.58	4.57	4.60	4.59	4.58	4.57	4.57
CF	3.75	3.78	3.80	3.83	3.85	3.75	3.78	3.80	3.83	3.85
TA	6.41	6.41	6.41	6.41	6.42	6.41	6.41	6.41	6.41	6.41
AIA	1.35	1.35	1.35	1.35	1.34	1.35	1.35	1.35	1.35	1.34
NDF	10.21	10.24	10.28	10.31	10.34	10.21	10.24	10.27	10.31	10.34
ADF	3.43	3.48	3.53	3.59	3.64	3.43	3.48	3.53	3.58	3.64

Table 3: Effect of supplementation of giloy and ascorbic acid on metabolizabilityof nutrients

Treatment groups	Metabolizability (%)					
	Dry matter	Crude protein	Ether extract	Crude fiber	NDF	ADF
Giloy×ascorbic acid						
T_1	72.04	74.00	75.01	20.60	17.94	9.18
T_2	74.06	76.61	78.22	23.37	18.63	9.21
T_3	73.70	76.55	77.99	23.12	18.34	9.57
T_4	73.59	76.37	77.63	23.33	18.56	9.53
T_5	73.53	76.28	77.36	23.22	18.63	9.47
T_6	73.66	74.93	77.38	23.19	18.28	9.33
T_7	75.25	77.32	78.43	23.51	18.39	9.58
T_8	75.15	77.20	78.19	23.46	18.54	9.40
T_9	75.07	77.15	78.17	23.26	18.13	9.22
T_{10}	75.01	77.19	78.19	23.24	18.67	9.59
SEm±	0.746	0.887	1.170	0.747	1.007	0.762
Effect of giloy						
0%	72.85	74.47a	76.19	21.89	18.11	9.26
0.25%	74.65	76.96b	78.33	23.44	18.51	9.40
0.50%	74.43	76.87b	78.09	23.29	18.44	9.49
0.75%	74.33	76.76b	77.90	23.29	18.35	9.37
1%	74.27	76.74b	77.77	23.23	18.65	9.53
SEm±	0.523	0.627	0.827	0.528	0.7123	0.539
Effect of ascorbic acid						
0 %	73.39^a	75.96	77.24	22.73	18.42	9.39
0.025%	74.83^b	76.76	78.07	23.33	18.40	9.42
SEm±	0.334	0.397	0.523	0.334	0.451	0.341

Giloy supplementation and interaction (Giloy×Ascorbic acid), while, highly significant (p<0.01) effect was observed due to ascorbic acid supplementation only. The statistical analysis of data of crude protein metabolizability (%) subjected to effect of ascorbic acid supplementation and interaction (Giloy×Ascorbic acid) revealed non-significant effect on crude protein metabolizability, while significant (p<0.05) effect was observed due to effect of Giloy supplementation. The statistical analysis of data of ether extract metabolizability revealed non-significant effect due to supplementation of Giloy at graded level and ascorbic acid as well as interaction. The statistical analysis of data of crude fiber metabolizability revealed non-significant effect due to graded levels of Giloy and ascorbic acid supplementation as well as interaction. The statistical analysis of data of NDF metabolizability revealed non-significant effect due to main effect of Giloy and ascorbic acid as well as interaction. The statistical analysis of data of ADF metabolizability revealed no significant effect due to main effect of Giloy and ascorbic acid as well as interaction. The feed passage rate obtained in different treatment groups have been presented in Table 4. The statistical analysis of data of feed passage rate revealed non-significant effect due to Giloy and ascorbic acid supplementation as well as interaction.

The results obtained in present study regarding non-significant effect of Giloy supplementation on dry matter metabolizability are in accordance with the observations of Singh (2014) who observed non-significant improvement in dry matter metabolizability on inclusion of graded levels of Giloy in broilers diets. The present findings of highly significant (p<0.01) effect with respect to effect of ascorbic acid supplementation are in line with Dhore et al. (2014) who reported highly significant (p<0.01) results of dry matter metabolizability @ 0.025% ascorbic acid supplementation in broiler diets. Crude protein metabolizability was found to be non-significant in ascorbic acid supplemented group, which are in contrastwith findings of Lohakare et al. (2005), who reported significant improvement in crude protein metabolizability on supplementation of ascorbic acid. With respect to effect of ascorbic acid supplementation, non-significant effect on ether extract metabolizability was in

Table 4: Effect of supplementation of giloy and ascorbic acid on feed passage rate

Treatment groups	Feed passage rate (minute)
Giloy×ascorbic acid	
T_1	251.33
T_2	243.17
T_3	245.50
T_4	244.67
T_5	249.33
T_6	247.00
T_7	246.17
T_8	248.33
T_9	243.50
T_{10}	245.67
SEm±	3.9637
Effect of giloy	
0%	249.17
0.25%	244.67
0.50%	246.92
0.75%	244.08
1%	247.50
SEm±	2.8028
Effect of ascorbic acid	
0 %	246.80
0.025%	246.13
SEm±	1.7726

contrast to findings of Lohakare et al. (2005), who reported significant improvement in ether extract metabolizability on supplementation of ascorbic acid @ 200 ppm in broilers. The nutrient metabolizability of different components of ration increased with the increase in body weight but no definite trend of significant improvement in metabolizability was observed with respect to significant improvement in body weight indicated that metabolizability might not always related with body weight of broilers. Further, the present findings gain support from the findings of Yang et al. (2013) who reported no correlation between body weight and metabolizability on different body weight of chickens. Metabolizability of crude protein was lower than carbohydrate are in line with the findings of (Noy and Sklan, 1995). The effect of supplementation of Giloy or ascorbic acid alone and their various combinations in diet of broiler chicks revealed a numerical improvement in metabolizability of different nutrient components of diet as compared to control diet, which could be explained because of negative effect of chronic heat stress in control diet. Further, the present findings are well supported with the findings of different researchers (Wallis and Balnave, 1984, Sahin and Kucuk, 2001), they reported decrease in metabolizability of different components of the ration due to heat stress as observed in control group. Better nutrient metabolizability on supplementation of Giloy may be due to improved digestive activity, increased digestive secretion (Puvaca et al., 2013), improved gut morphological characteristics (Jamroz et al., 2003) and enhancement of intestinal activities of trypsin, lipase, amylase and are the major mechanisms through, which phytobiotics exerts their beneficial effect on the nutrient metabolizability. Whereas, mode of action of ascorbic acid could be explained due to protective roles on pancreatic tissues to oxidative stress which may help the pancreas to function properly, thus improving the metabolizability of nutrients (Attia et al., 2009). Broilers reared under chronic heat stress impacted poor metabolizability coefficients (%) of various nutrients in control group as compared to various treatment groups. Improvement in metabolizability coefficients (%) of various nutrients was observed due to *Tinospora cordifolia* (at graded levels) or ascorbic acid supplementation alone or in combinations. From the research study, it was concluded that supplementation of *Tinospora cordifolia* (Giloy) at 0.25% level with ascorbic acid @ 0.025% may be as a beneficial management strategy in poultry feed during chronic heat stress.

The non-significant findings of faster feed passage rate in Giloy and ascorbic acid either alone or in combinations corroborated with the observation of Washburn, (1991) who reported a low correlation between better feed passage rate and FCR. Frankic et al. (2009) observed that phytobiotic extracts speed up the digestion and shorten the time of feed passage through the gastrointestinal tract. Numerous factors influence transit time of ingesta through the GIT *i.e.* excitement, the age of bird, temperature, genotype, amount of feed consumed, dietary composition of feed. Antimicrobial activity of Giloy leads to reduced microbial population, which causes thinning of the gut lining, and increases the nutrient absorption, which may be one of the causes of higher feed passage rate due to Giloy supplementation.

4. CONCLUSION

Supplementation of *Tinospora cordifolia* (Giloy) at 0.25% level with ascorbic acid @ 0.025% might be beneficial management strategy in poultry feed during chronic heat stress as reflected by improved nutrient metabolizibility and feed passage rate in Giloy supplemented broilers in study in text.

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