Effects of Ascorbic acid, Giloy (*Tinospora cordifolia*) along with various bedding materials on Japanese quail egg quality attributes

ABSTRACT

The study investigated the effects of giloy (*Tinospora cordifolia*) and ascorbic acid supplementation along with different bedding materials (sand B_1, saw dust B_2 and wheat straw B_3) on egg production status of Japanese quails. A total of 432 chicks (7 day-old) of Japanese quails were divided into three equal groups (144 each) for different bedding material used and each group was further subdivided into four groups (each of 36 chicks) on the basis of dietary treatment (control T_0, giloy T_1, ascorbic acid T_2 and combination of both T_3). The present experiment was conducted for 24 weeks starting from October 2020 to April 2021 at the Poultry unit, Livestock Farm Complex, College of Veterinary and Animal Science, Bikaner. Thus birds were randomly and uniformly distributed in total 12 treatment groups comprising of 36 birds in each group and each group was further divided into two replicates comprising 18 birds in each. Quails were fed a basal diet (control, T_0) and the basal diet supplemented with giloy 5 g/kg of diet, ascorbic acid 240 mg/kg diet and a combination of giloy 5 g/kg and ascorbic acid 240 mg/kg diet in dietary treatment groups T_1, T_2 and T_3, respectively. Highly significant ($p<0.01$) effect of incorporation of supplements and different bedding material was found on egg shell thickness and egg shell weight of Japanese quail. In present experiment, egg shell thickness and weight were higher in saw dust bedding material group. The interaction between dietary supplementation and different bedding materials was found non-significant on these traits.

Key words: Ascorbic acid, Giloy, Japanese quail, egg shell thickness, egg shell weight

INTRODUCTION

Poultry is one of the fastest growing components of the agricultural sector in India. Poultry plays an important role as animal protein source in human diet in terms of egg and meat. India ranks fourth in total production of poultry meat in the world (Basic Animal Husbandry Statistics, 2019). Japanese quail (*Coturnix coturnix japonica*) is one of the most efficient biological machines for converting feed into animal protein of high biological value (Das *et al*., 2012). They have less feeding requirement (about 20-25 g per day) compared to chicken (120-130 g per day) (Ani *et al*., 2009).

Various types of feed additives, such as antibiotics, enzymes, hormones, prebiotics, probiotics, herbal products *etc*., are being used as growth stimulants in poultry production to

improve efficiency and get maximum returns in shortest possible time. *Tinospora cordifolia*, which is known by the common names guduchi, giloy and heart-leaved moonseed, is a herbaceous vine of the family Menispermaceae indigenous to tropical regions of the Indian subcontinent (Sengupta *et al*., 2011). Giloy is a rich source of protein and micronutrients, such as iron, zinc, copper, calcium, phosphorus, and manganese (Saeed *et al*., 2020). The most clearly established functional role for vitamin C involves collagen biosynthesis. Beneficial effects result from ascorbic acid in the synthesis of "repair" collagen (Bera *et al*., 2010).

Wood sawdust is the most common used bedding material, however there are many alternative materials that may be used such as peanut hulls, rice and wheat straw, rice hull ash (Chamblee and Yeatman, 2003), and other dry, absorbent and low-cost organic materials. Moreover, sand is occasionally used as a bedding material (Shields *et al*., 2005). Birds spend their entire life in contact with the litter material. Therefore, its quality is considered a crucial factor of poultry welfare. Hence, the present experiment was planned to study the effect of bedding materials such as sand, wheat straw and saw-dust with dietary supplementation of giloy herb (*Tinospora cordifolia*) and ascorbic acid and their combination on egg quality traits such as egg shell thickness, egg shell weight in Japanese quail.

MATERIALS AND METHODS

The present study was conducted at Poultry unit, Livestock Farm Complex, College of Veterinary and Animal Science, Bikaner, Rajasthan University of Veterinary and Animal Sciences, Bikaner (India), 28.0325° N, 73.3295° E following approval of Institutional Animal Ethics Committee of the College.

Experimental Design and Material used

The study was undertaken on four hundred thirty two (432) seven-day old Japanese quail chicks which were purchased from central poultry development organization, Chandigarh. Out of 432 birds 72 birds were slaughtered at the age of 8 weeks for evaluating carcass characteristics and remaining 360 birds were further used for remaining traits (growth traits and egg production traits) for the entire experimental trail of 24 weeks.

The factorial design (3x4) was adopted for the present study. The chicks were equally and randomly divided into four dietary treatment groups (n=108) and one-third of each (n=36) were reared on each of three bedding materials (sand, saw-dust and wheat-straw) using two replicates each of 18 birds (R1-R2) to ensure the uniformity in various treatment

groups. The chicks were reared on sand, saw-dust and wheat straw as group B_1, B_2 and B_3, respectively (n=144 each), and each bedding group was subdivided equally in one control and three dietary treatment groups (giloy @ 5 g/kg diet, ascorbic acid @ 240 mg/kg diet, and a combination of both at the said rates) denoted by T_0, T_1, T_2 and T_3, respectively (n=36 each) to study the effect of bedding material, dietary treatment and their interaction on egg production of birds.

Commercially available readymade starter and finisher and layer rations were procured as basal diets and feed additives such as giloy (*Tinospora cordifolia*) and ascorbic acid were supplemented in them. During first seven days, newspapers were spread on litter material and from 8^{th} day onward till the completion of experiment chicks were reared on respective litter material of about 6 inches of depth.

Egg shell thickness (mm)

Egg shell was dried in the air, thereafter drying shell thickness was measured by using screw gauge and three measurements were taken for each egg shell one on large end, one on small end and the other one on the equator region. The mean of all three measurements was considered as thickness of egg shell.

Egg shell weight

The egg shell along with the membrane was sunk in 5 % EDTA solution for 30 minutes. After that shell membrane was removed carefully, egg shells were dried in the air and weighed using an electronic balance.

Statistical Analysis

The data generated was analyzed for two way ANOVA using factorial RBD and means were compared using Duncan's post-hoc test at $P<0.05$ on SPSS software (Snedecor and Cochran, 1994).

RESULTS AND DISCUSSION

Egg shell thickness (mm)

Effect of dietary supplementation and bedding material

The statistical analysis of variance of data revealed highly significant ($P<0.01$) effect of dietary supplementation and bedding materials on the overall mean egg shell thickness of Japanese quail during experiment(Table 4). However, during 8^{th}, 10^{th}, 12^{th}, $14^{th}$and 18^{th} week of study there was non-significant effect of bedding materials on egg shell thickness. The overall means of egg shell thickness (mm) for various dietary treatment groups were recorded

to be 0.19 in T_0, 0.19 in T_1, 0.20 in T_2 and 0.20 in group T_3, respectively (Table 1). So, the overall study indicated that there is beneficial effect of incorporation of giloy and ascorbic acid in the diet on egg shell thickness of the Japanese quails. Numerically highest egg shell thickness (mm) was found in sawdust and wheat straw (0.20) as compared to sand (0.19) (Table 2). So, the overall study indicated that there is beneficial effect of sawdust and wheat straw on egg shell thickness of the Japanese quails.

Table 1: Effect of dietary supplementation on Egg shell thickness (mm) at different weeks

Supplement effect	Age in weeks									
	8W	**10W**	**12W**	**14W**	**16W**	**18W**	**20W**	**22W**	**24W**	**8-24W**
T_0	0.17^{a}	0.18	0.18	0.18^{a}	0.18^{a}	0.19^{a}	0.20^{a}	0.20^{a}	0.21^{a}	0.19^{a}
T_1	0.17^{a}	0.18	0.18	0.18^{a}	0.19^{ab}	0.2^{ab}	0.21^{ab}	0.21^{b}	0.21^{a}	0.19^{b}
T_2	0.18^{b}	0.18	0.19	0.19^{ab}	0.19^{b}	0.20^{b}	0.21^{bc}	0.21^{b}	0.21^{a}	0.20^{b}
T_3	0.18^{b}	0.19	0.19	0.19^{b}	0.20^{c}	0.21^{c}	0.22^{c}	0.23^{c}	0.23^{b}	0.20^{c}
SEM	0.0018	0.0020	0.0022	0.0027	0.0025	0.0028	0.0032	0.0030	0.0034	0.0012

Means having different superscripts in a column differ significantly ($P \leq 0.05$)

The findings of the above experiment are in close agreement with Shit *et al.*(2012) conducted a trial in which total one hundred and twenty, sixty from each sex (15 week old) Japanese quail from same hatch were procured from the institutional experimental quail farm and supplemented with L- ascorbic acid. Among all the egg quality traits studied, only specific gravity, egg shell weight and thickness differed significantly ($P<0.05$) in the present study.

Similarly, Dhaliwal (2004) studied the effects of supplementation of vitamin C and E on growth and reproduction of Japanese quail (*Coturnix coturnix japonica*) kept in cages. Synthetic vitamin C and E or herbal CE supplementations were tested. Egg shell thickness increased significantly ($P<0.05$) on supplementation of vitamin E and herbal CE.

Results are also similar with findings of Karimi*et al*. (2015) who conducted a study to determine whether dietary chromium (1200 µg Cr/kg diet) and vitamin C (300 mg/kg) attenuated adverse effects of heat stress on external and internal egg quality traits in Japanese quails. They found increased ($P< 0.05$) egg shell thickness when supplemented with vitamin C and Cr-vitamin C groups.

Table 2: Effect of bedding materials on Egg shell thickness (mm) at different weeks

Supplement effect	Age in weeks									
	8W	**10W**	**12W**	**14W**	**16W**	**18W**	**20W**	**22W**	**24W**	**8-24W**
B_1	0.17	0.18	0.18	0.18	0.19^{a}	0.19	0.20^{a}	0.20^{a}	0.20^{a}	0.19^{a}

B_2	0.17	0.18	0.18	0.19	0.20^b	0.20	0.22^b	0.22^b	0.22^b	0.20^b
B_3	0.17	0.18	0.18	0.19	0.19^a	0.20	0.21^a	0.22^b	0.22^b	0.20^b
SEM	0.0015	0.0017	0.0019	0.0023	0.0022	0.0024	0.0028	0.0026	0.0029	0.0010

Means having different superscripts in a column differ significantly (P≤0.05)

Interaction effect of dietary supplements× bedding materials

The statistical analysis of variance of data revealed non-significant effect of interaction between dietary supplementation and different bedding materials on egg shell thickness of Japanese quail during entire period of study (Table 4). Further the comparison of means showed that highest egg shell thickness was recorded inT_{23} and T_{33} group (Table 3). Lowest egg shell thickness was found in $T_{10,}$ T_{11}, T_{12} and T_{30} groups (Table 3).

Table 3: Effect of dietary supplements × bedding materials Interaction on Egg shell thickness (mm) at different weeks

Interaction Effect	**Age in weeks**									
	8W	**10W**	**12W**	**14W**	**16W**	**18W**	**20W**	**22W**	**24W**	**8-24W**
T_{10}	0.17	0.18	0.19	0.18	0.19	0.19	0.20	0.19	0.20	0.19
T_{11}	0.17	0.19	0.19	0.19	0.19	0.20	0.20	0.20	0.21	0.19
T_{12}	0.18	0.18	0.19	0.19	0.19	0.20	0.21	0.21	0.21	0.19
T_{13}	0.18	0.19	0.19	0.20	0.20	0.21	0.22	0.22	0.22	0.20
T_{20}	0.18	0.19	0.19	0.19	0.19	0.20	0.21	0.21	0.22	0.20
T_{21}	0.17	0.19	0.19	0.19	0.20	0.20	0.22	0.22	0.22	0.20
T_{22}	0.18	0.19	0.19	0.20	0.21	0.21	0.22	0.22	0.23	0.20
T_{23}	0.18	0.19	0.19	0.21	0.22	0.23	0.24	0.24	0.23	0.21
T_{30}	0.17	0.18	0.18	0.19	0.19	0.19	0.20	0.21	0.22	0.19
T_{31}	0.18	0.19	0.19	0.19	0.20	0.20	0.22	0.22	0.22	0.20
T_{32}	0.18	0.19	0.19	0.19	0.20	0.21	0.21	0.22	0.22	0.20
T_{33}	0.19	0.19	0.19	0.20	0.20	0.21	0.21	0.23	0.24	0.21
SEM	0.0031	0.0035	0.0038	0.0047	0.0025	0.0049	0.0057	0.0053	0.0059	0.0021

So, the overall study indicated that there is no beneficial effect of bedding material with supplementation of both giloy and ascorbic acid on egg shell thickness of the Japanese quails.

Table 4 Mean sum of squares for Egg shell thickness at different weeks

Source of variation	DF	Age in weeks									
		MEAN SQUARES									
		8W	10W	12W	14W	16W	18W	20W	22W	24W	8-24W
Supplement	3	0.00042**	0.00014	0.00010	0.00037*	0.00056**	0.00010**	0.00096**	0.0014**	0.00094**	0.00056**
Bedding	2	0.000043	0.000081	0.000027	0.00020	0.00057**	0.00030	0.0012**	0.0011**	0.0013**	0.00038**
Interaction (TxB)	6	0.000093	0.000036	0.000032	0.000015	0.000043	0.000058	0.00020	0.000057	0.000065	0.000011
Error	12	0.000038	0.000050	0.000058	0.000089	0.000078	0.000097	0.00013	0.0001125	0.00014	0.000018

*= significant (P≤0.05), **= highly significant (P≤0.01)

Egg shell weight

Effect of dietary supplementation and bedding material

The overall means of egg shell weight for various dietary treatment groups were recorded to be 1.44 in T_0, 1.53 in T_1, 1.55 in T_2 and 1.76 in group T_3, respectively (Table 5). So, the overall study indicated that there is beneficial effect of incorporation of giloy and ascorbic acid in the diet on egg shell weight of the Japanese quails. The statistical analysis of variance of data revealed highly significant (P<0.01) effect of incorporation of supplements and bedding materials on mean egg shell weight of Japanese quail throughout the experimental period (Table 8). However, during 8^{th}, 10^{th}, 12^{th}, $14^{th}$and 16^{th} week of study there was non-significant effect of bedding materials on egg shell weight (Table 6).

Table 5: Effect of dietary supplementation on Egg shell weight (g) at different weeks

Supplement effect	Age in weeks									
	8W	10W	12W	14W	16W	18W	20W	22W	24W	8-24W
T_0	0.95^a	1.14^a	1.16^a	1.41^a	1.50^a	1.65^a	1.70^a	1.72^a	1.72^a	1.44^a
T_1	1.06^b	1.24^b	1.26^b	1.50^b	1.65^b	1.75^b	1.76^b	1.77^b	1.76^{ab}	1.53^b
T_2	1.09^c	1.28^c	1.31^c	1.52^b	1.62^b	1.74^b	1.79^b	1.80^c	1.80^b	1.55^c
T_3	1.28^d	1.48^d	1.51^d	1.70^c	1.88^c	1.98^c	2.01^c	2.02^d	2.02^c	1.76^d
SEM	0.0089	0.0093	0.014	0.010	0.019	0.0098	0.012	0.012	0.015	0.0066

Means having different superscripts in a column differ significantly (P≤0.05)

Numerically highest egg shell weight (1.59) was found in sawdust as compared to other bedding material (Table 8). So, the overall study indicated that there is beneficial effect of sawdust on egg shell weight of the Japanese quails. Similarly, Bardakcioglu *et al*. (2005) carried out an experiment to investigate the effects of dietary vitamin C supplementation on some egg production traits and egg shell quality in Japanese quails reared under high ambient temperature. There was significant effect of vitamin C supplementation on egg shell weight. The findings of the above experiment are in close agreement with Shit *et al*. (2012) and Karimi *et al*.(2015) in japanese quail.

Table 6: Effect of bedding materials on Egg shell weight at (g) different weeks

Supplement effect	Age in weeks									
	8W	**10W**	**12W**	**14W**	**16W**	**18W**	**20W**	**22W**	**24W**	**8-24W**
B_1	1.08	1.27	1.30	1.52	1.64	1.75^a	1.78^a	1.79^a	1.79^a	1.55^a
B_2	1.10	1.30	1.32	1.54	1.67	1.79^b	1.84^b	1.85^b	1.85^b	1.59^b
B_3	1.10	1.29	1.32	1.54	1.66	1.79^b	1.83^b	1.84^b	1.84^b	1.58^b
SEM	0.0077	0.0080	0.012	0.0086	0.016	0.0085	0.011	0.010	0.013	0.0057

Means having different superscripts in a column differ significantly (P≤0.05)

Interaction effect of dietary supplements× bedding materials

The statistical analysis of variance of data revealed non-significant effect of interaction between dietary supplementation and different bedding materials on egg shell weight of Japanese quail during entire period of study (Table 8).

Table 7: Effect of dietary supplements × bedding materials Interaction on Egg shell weight (g) at different weeks

Interaction Effect	Age in weeks									
	8W	**10W**	**12W**	**14W**	**16W**	**18W**	**20W**	**22W**	**24W**	**8-24W**
T_{10}	0.94	1.14	1.16	1.41	1.48	1.61	1.65	1.66	1.67	1.41
T_{11}	1.04	1.25	1.26	1.49	1.66	1.74	1.76	1.76	1.76	1.52
T_{12}	1.08	1.27	1.30	1.50	1.59	1.72	1.74	1.77	1.77	1.52
T_{13}	1.28	1.46	1.50	1.70	1.88	1.97	1.99	1.99	1.97	1.75
T_{20}	0.96	1.16	1.18	1.43	1.55	1.68	1.74	1.76	1.76	1.47
T_{21}	1.08	1.26	1.27	1.51	1.66	1.75	1.78	1.79	1.78	1.54
T_{22}	1.10	1.30	1.32	1.53	1.63	1.77	1.84	1.83	1.84	1.57

T_{23}	1.29	1.50	1.55	1.72	1.87	1.99	2.03	2.06	2.04	1.78
T_{30}	0.95	1.14	1.17	1.40	1.48	1.67	1.73	1.76	1.76	1.45
T_{31}	1.07	1.24	1.28	1.51	1.64	1.77	1.77	1.77	1.77	1.53
T_{32}	1.10	1.30	1.34	1.56	1.65	1.74	1.82	1.82	1.81	1.57
T_{33}	1.30	1.50	1.50	1.70	1.90	1.99	2.02	2.03	2.06	1.78
SEM	0.015	0.016	0.024	0.017	0.033	0.017	0.022	0.020	0.026	0.011

Further the comparison of means showed the highest egg shell weight inT_{23} and T_{33} group (Table 7). The lowest egg shell weight was found in T_{10}group. So, the overall study indicated that there is no beneficial effect of bedding material with supplementation of both giloy and ascorbic acid on egg shell weight of the Japanese quails.

Table 8: Mean sum of squares for Egg shell weight at different weeks

Source of variation	**DF**	**Age in weeks**									
		MEAN SQUARES									
		8W	**10W**	**12W**	**14W**	**16W**	**18W**	**20W**	**22W**	**24W**	**8-24W**
Supplement	3	0.23**	0.24**	0.25**	0.18**	0.29**	0.24**	0.21**	0.21**	0.20**	0.22**
Bedding	2	0.0025	0.0022	0.0028	0.0026	0.0032	0.0074**	0.017**	0.018**	0.017**	0.0068**
Interaction (TxB)	6	0.00028	0.00044	0.0013	0.0011	0.0033	0.0011	0.0020	0.0019	0.0021	0.00039
Error	12	0.00095	0.0010	0.0023	0.0012	0.0044	0.0011	0.0020	0.0017	0.0027	0.00053

**= highly significant (P≤0.01)

CONCLUSION

The finding of the study indicates that supplementation of giloy and ascorbic acid in the diet improves egg quality traits of the Japanese quails, and that there is beneficial effect of saw dust and wheat straw as bedding material. Bedding material used with supplementation of both giloy and ascorbic acid improves egg shell thickness and egg shell weight.

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