



Effect of Herb *Urtica dioica* as Feed Additive on Growth Performance, Serum Biochemical, Immune Response and Oxidative Parameters of Broilers

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ABSTRACT

Background: Considering the increasingly high cost of poultry feed and the economic aspects of poultry rearing, search for organic feed supplements for optimal growth performance along with maintaining the health status of poultry has emerged as a challenge for animal nutritionists all over the world. Over the years, medicinal herbs have fascinated a lot of consideration for their important role as alternatives to antibiotic growth promoters in animals. These compounds display some important pharmacological and growth promoting effects in animals. With this background, exploratory research was designed to study the effects of a locally available medicinal herb, *Urtica dioica* at different levels on the performance, serum biochemical, immune response and oxidative parameters of broilers.

Methods: A total of 120-day old commercial broiler chicks were purchased; randomly distributed into four treatment groups. The groups were distributed as; group T₁-control offered basal diet, groups T₂, T₃, T₄ offered basal diet supplemented with *Urtica dioica* leaf powder @ 1, 1.5 and 2% respectively on dry matter basis. A growth trial of five weeks followed by a metabolic trial of seven days was conducted at the end of the experiment. The economics for broiler production was also calculated. In addition to this different biochemical, immune and oxidative parameter were also analyzed at the end of the experiment following standard procedures.

Result: Significant improvements in various production parameters viz. body weight, FCR, digestibility of DM, CP were observed in birds fed nettle as feed additive in comparison to control, however non-significant changes in the values of feed intake, digestibility of CF, EE and NFE among treatment groups was also recorded. On calculating the economics, a lower production cost per kg live weight gain and a higher profit/bird was calculated for treatment groups fed nettle as feed additive. Significant improvements in various biochemical, immune and oxidative parameters were also observed in birds of phytogetic supplemented groups in comparison to control.

Key words: Broiler, Immune response, Nettle, Oxidative status, Performance, Phytogetic, *Urtica dioica*.

INTRODUCTION

Poultry is one of the fastest growing animal industries and has a substantial contribution to food security and nutrition. The Indian poultry industry has also witnessed phenomenal growth during the last few decades primarily due to the exploitation of various modern growth promoting strategies, disease prevention measures coupled with the increasing preference of consumers towards consuming protein rich foods (Angelakis, 2013). The future growth of broiler industry is however challenged by several factors like increasing the production performance of birds without compromising on their health and immunity, managing oxidative stress in birds along with gaining the confidence of consumers about the safety and quality of products while keeping in view the economics of poultry rearing (Hafeez and Attia, 2020). All this has led to high production cost inputs due to incorporation of various growth promoting antibiotics/additives/synthetic anti-oxidants in the diet of broilers. The use of these synthetic growth promoters and anti-oxidants despite their effectiveness provokes occurrence of various chronic diseases among both birds

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and consumers, which significantly limits their use. This has led to a demand for additives of natural origin which are not only safe and cheap for both animals and humans

but are also able to prevent metabolic diseases in animals and oxidative reactions in animal products during their storage (Botsoglou *et al.*, 2002a). Development and utilization of antioxidants of natural origin are, therefore, in demand for making the industry more acceptable vis-à-vis human health. Exploration of locally available feed additives of plant origin has attracted considerable interest in the field of poultry science for healthy production of broiler meat. Amongst such medicinal herbs is a locally available herb known as *Urtica dioica* belonging to the Urticaceae family and commonly known as stinging nettle or common nettle. Due to its balanced protein composition and high content of minerals and vitamins, nettle has attracted great interest in human/animal nutrition (Joshi *et al.*, 2014). The World Health Organization, in its monographs on 'Selected medicinal plants', describes Urticaceae's valuable herbs for many medicinal uses (WHO, 2004). The leaves of nettle are rich in flavonoids, as well as phenolic compounds, organic acids, vitamins and minerals. The root contains lectins, polysaccharides, sterols and lignans. The most active flavonoid present in nettle is the quercetin. Nettle over the years has been used for its anti-oxidative (Toldy *et al.*, 2005) and growth-stimulating (Krukric, 2012) properties. Experimental studies over the years have also suggested that nettle as a feed additive increases the gut microflora which positively effects hosts nutrition, health and growth by better utilisation of nutrients (Hashemi and Davoodi, 2011). The aim of this study was to evaluate the effects of using different levels of dried nettle leaves on the performance, serum biochemical, immune and oxidative parameters of broilers.

MATERIALS AND METHODS

The study was carried out in the division of Animal Nutrition SKUAST-K Shuhama, Alusteng (J & K). Fresh nettle leaves were collected from the campus vicinity freed from dust and sand, spread on a clean polythene sheet and dried under shade. Leaves were separated from the stems and crushed to a fine powder and kept in clean, airtight containers after proper labelling. A total of 120-day old commercial broiler chicks were randomly divided into four treatment groups, having three replicates with ten birds each. The groups were distributed as; group T₁-control, offered basal diet without any feed additive whereas groups T₂, T₃, T₄ were offered basal diet supplemented with *Urtica dioica* leaf powder @ 1, 1.5 and 2% respectively on dry matter basis. Different rations *viz.*, pre-starter, starter and finisher were procured from a reputed supplier and analysed for proximate constituents, calcium and phosphorus content. The moisture, crude protein, crude fibre, ether extract, nitrogen-free extract and ash were estimated following the procedures laid down by AOAC (2005), while the calcium and phosphorus contents of the samples were estimated using the methods described by Talpatra *et al.* (1940) (Table 1). A growth trial of five weeks

was conducted in the months of December and January 2022. The body weight of all experimental birds was recorded weekly, whereas feed consumption was calculated by subtracting the weight of feed residue from the weight of total feed offered. At the end of the growth trial, a 7-day metabolic trial was conducted to assess the digestibility of nutrients in different treatment groups. Mortality if any was recorded daily. The economics was worked out by considering the purchase rate of chicks, the average feed consumption of birds, the cost of feed, the cost of feed supplements, mortality, total weight gain of birds and the managerial expenditures during the experimental period. To study the effect of nettle on the immune status of birds, parameters like cell mediated and humoral immunity were studied.

Humoral response was measured by using sheep red blood cells as antigen and the antibody titre against SRBC was determined by haem-agglutination technique. Cell mediated immunity was determined by using DNCB and the immune response was measured by determining the thickness of the skin on injecting DNCB. Various biochemical parameters were estimated by collecting blood samples in clot activator vials and estimated by using commercial diagnostic kits. The oxidative status of broilers (TOS and TAS) was estimated by the procedure given by Erel, (2004). The data was analysed by using one-way ANOVA. The research has been approved by the Animal Ethical Committee (IAEC) of SKUAST- Kashmir (regd.no: 1809/GO/ReBiS/ReL/15/CPCSEA).

RESULTS AND DISCUSSION

Production parameters

The results of various production parameters of our experiment are presented in Table 2, 3, 4, 5 and 6 respectively. It was observed that towards the end of experiment; a significant ($P \leq 0.05$) increase in body weights and gain was observed in birds fed nettle as feed additive groups with the highest body weights and gains in birds fed nettle @ 2%. The average and cumulative feed consumption during the entire experimental period revealed no significant differences between different treatment groups. The overall feed conversion ratio in birds fed nettle as additive was significantly ($P \leq 0.05$) lower/better when compared to the control with better FCR in birds fed nettle @ 2%. Similarly, a significantly ($P \leq 0.05$) higher dry matter digestibility and digestibility of crude protein was observed in treatment groups supplemented with nettle as feed additive with non-significant differences in digestibility of ether extract, crude fibre and nitrogen-free extract when compared to control. On calculating the economics of our experiment (Table 6) it was seen that the feed cost/kg liveweight gain worked out to be highest for birds of T₁ group (control) followed by T₂, T₃ and T₄ respectively. The highest profit was calculated in birds fed diets nettle @ 2%.

Serum biochemistry, immunity and oxidative parameters

Table 7 and 8 summarizes the impact of different levels of nettle on serum biochemical, immune and oxidative parameters in broilers. Significantly ($P<0.05$) lower serum glucose, cholesterol, triglycerides, LDL and ALT levels and significantly ($P<0.05$) higher HDL levels were observed in birds fed nettle as feed additive in comparison to control. The serum AST and creatinine levels were found to be within the normal range but did not differ significantly between the treatment groups. The total anti- SRBC antibody titre and DNCB values were significantly ($p<0.05$) higher in the treatment groups fed diets supplemented

with herb. Significantly ($P\leq 0.05$) lower mean TOS values and higher TAS values were observed in birds fed herb as feed additive when compared to control.

Incorporation of herbs and their products in the diets of poultry are known to stimulate and promote the effective use of feed nutrients which subsequently results in rapid body weight gain, higher production rates and improved performance (Ajaykumar *et al.*, 2024 and Vargas *et al.*, 2021). The positive effects of herbs on the average live body weight and body weight gain in our study can be attributed to the effects of bioactive compounds present in these herbs, which can control and limit the growth and

Table 1: Proximate composition of experimental diets and *Urtica dioica*.

Ingredients	Diet			
	Pre-starter	Starter	Finisher	Nettle
Proximate composition (%)				
Dry matter (DM)	91.00	92.30	90.32	88.6
Crude protein (CP)	23.20	20.16	18.00	24.9
Crude fibre (CF)	2.33	3.22	4.28	10.4
Ether extract (EE)	9.06	10.80	11.99	17.1
Ash	3.87	4.04	4.16	23.3
Calcium (Ca)	1.10	1.00	0.89	0.7
Phosphorus (P)	0.49	0.44	0.40	0.03
ME Kcal/kg (Calculated)	3,000	3,100	3,191	-

Table 2: Average weekly bodyweights (g) of broilers fed herb *Urtica dioica* as feed additive.

Experimental period (Weeks)	Treatment groups			
	T ₁	T ₂	T ₃	T ₄
1	170.16±1.01	171.82±1.02	172.06±1.06	172.78±1.23
2	357.32±1.70	359.23±1.74	358.68±2.90	360.66±2.22
3	625.40±7.54	628.23±4.94	629.86±6.85	634.95±5.73
4	983.43 ^a ±7.41	992.01 ^b ±2.73	998.26 ^b ±4.69	1031.66 ^c ±1.66
5	1498.36 ^a ±4.28	1527.33 ^b ±3.34	1565.50 ^c ±5.70	1620.50 ^d ±2.11

Table 3: Average weekly and cumulative feed consumption (g) of broilers fed herb *Urtica dioica* as feed additive.

Experimental period (weeks)	Treatment groups			
	T ₁	T ₂	T ₃	T ₄
1-2	216.58±1.37	220.88±1.96	223.39±2.51	225.72±1.85
2-3	421.11±1.96	428.08±1.97	436.74±0.85	446.75±1.35
3-4	655.18±4.16	646.12±2.08	643.13±2.30	651.52±3.64
4-5	932.94±1.49	937.88±1.50	944.47±2.08	953.78±1.25
1-5	2225.81±0.71	2232.96±3.63	2247.73±1.19	2277.77±1.15

Table 4: Average weekly and cumulative feed conversion ratios of broilers fed herb *Urtica dioica* as feed additive.

Experimental period (Weeks)	Treatment groups			
	T ₁	T ₂	T ₃	T ₄
1-2	1.15±0.015	1.17±0.032	1.19±0.019	1.20±0.018
2-3	1.58±0.044	1.59±0.02	1.61±0.017	1.62±0.027
3-4	1.82±0.04	1.77±0.08	1.75±0.048	1.64±0.038
4-5	1.81 ^d ±0.081	1.75 ^{cd} ±0.022	1.66 ^b ±0.063	1.61 ^a ±0.052
1-5	1.67 ^c ±0.03	1.64 ^b ±0.014	1.61 ^b ±0.017	1.57 ^a ±0.020

colonization of numerous pathogenic and non-pathogenic species of bacteria in chicks' guts which in turn leads to greater efficiency in the utilization of food, resulting in enhanced growth (Bedford, 2000). The improvement in protein utilization, absorption and suppression of gram-negative bacteria by phytogenic feed additives might be the reason for the improvement in feed conversion ratios Abd El-Latif (2004). El-Gendy *et al.* (1996) also reported that the improvement in feed conversion ratio with feeding herbal extracts could be associated with improving the digestibility of dietary protein in the small intestine. The action of herbal extracts as antioxidants, anti-bacterial, anti-fungal and anti-protozoa also adds to the positive improvement in bird's performance (Jain *et al.*, 2008) Herbs

are also known to support the activity of digestive enzymes which in turn improves feed conversion ratios (Brenes and Roura, 2010). The results of the effects of nettle on various production parameters in broilers of our experiment are in accordance with Zamzami *et al.* (2014) who studied the effect of nettle leaf powder on performance of broilers and concluded that feeding different levels of nettle improved the body weight of broilers and nettle leaf powder supplementation @ 2% significantly increased ($P \leq 0.05$) production performance (body weight, body weight gain and FCR) in broilers. Similarly, Milosevic *et al.* (2021) in a study reported that the use of stinging nettle in broiler nutrition significantly ($P \leq 0.05$) improved the production performances of broilers like body weight, body weight gain

Table 5: Average nutrient digestibility (%) of broilers fed herb *Urtica dioica* as feed additive.

Parameters	Treatment groups			
	T ₁	T ₂	T ₃	T ₄
DM	63.59 ^a ±0.47	64.55 ^a ±0.48	65.90 ^b ±0.51	67.81 ^c ±0.52
CP	53.20 ^a ±0.32	54.94 ^{ab} ±0.35	56.20 ^{ab} ±0.41	57.78 ^b ±0.45
EE	60.21±0.43	61.56±0.48	62.72±0.51	63.50±0.55
CF	40.21±0.32	40.39±0.33	41.27±0.35	41.57±0.36
NFE	54.18±0.48	55.02±0.49	55.31±0.51	56.69±0.52

Table 6: Economics of broilers fed herb *Urtica dioica* as feed additive.

Economic parameters	T ₁	T ₂	T ₃	T ₄
Final body weight gain (g)	1328.2	1355.51	1393.44	1447.72
FCR (1-5week)	1.67	1.64	1.61	1.57
Total birds at end of study	30	30	30	30
Feed cost/kg live weight gain (Rs)	66.8	65.6	64.4	62.8
Feed cost/bird (Rs)	88.71	88.88	89.70	90.87
Input/bird (Rs) (Feed cost+Chick)	143.71	143.88	144.7	145.87
Cost+Misc.				
Input/Group (Rs)	4311.3	4316.4	4341	4376.1
Sale proceed/Bird (Rs)	146.08	149.05	153.23	159.17
Sale proceed/Group (Rs)	4382.4	4471.5	4596.9	4775.1
Profit/loss per group (Rs)	71.1	155.1	255.9	399
Profit/loss per bird (Rs)	2.37	5.17	8.53	13.3

Table 7: Bio chemical parameters of broilers fed *Urtica dioica* as feed additive.

Parameters	Treatment groups			
	T ₁	T ₂	T ₃	T ₄
Glucose (mg/dl)	233.33 ^d ±1.85	224.66 ^c ±2.02	225.33 ^c ±1.20	214.33 ^a ±2.33
Cholesterol (mg/dl)	206.33 ^a ±12.44	161.0 ^b ±1.73	169.33 ^b ±2.18	153.33 ^b ±5.23
AST	275.73±5.32	232.90±6.26	251.23±1.92	228.90±3.73
ALT	8.63 ^c ±0.38	6.66 ^b ±0.20	6.96 ^b ±0.18	5.83 ^a ±0.67
Triglycerides	77.01 ^c ±4.35	59.33 ^b ±3.48	54.66 ^{ab} ±2.40	51.33 ^a ±1.66
HDL	59.00 ^a ±3.51	71.66 ^b ±1.44	77.33 ^{bc} ±2.72	80.13 ^c ±1.73
LDL	106.66 ^d ±2.40	78.66 ^b ±2.90	76.33 ^b ±1.76	75.66 ^a ±2.02
Creatinine	0.34±0.029	0.35±0.029	0.31±0.0296	0.32±0.020

and FCR. Behboodi *et al.* (2021) in his study reported that supplementation of broiler diets with nettle extract at 0.25 ml/l significantly improved performance (body weight and FCR) of broiler chickens. Also, in agreement with our results Ahmadipour and Khajali (2019), reported that nettle powder at the level of 1.5% had positive effects on growth, weight gain and FCR in the broilers exposed to oxidative stress. In an experiment conducted by Hashemi *et al.* (2018) it was reported that nettle as safe additive in broiler diets had strong growth promoting ability and nettle leaf extract at 0.25% significantly improved ($P<0.05$) the body weight and body weight gain in broilers at the end of 42 days. Safamehr *et al.* (2012) in his study reported that supplementation of nettle at 1% significantly ($P<0.05$) increased the body weight of broilers at 42 days of age. In accordance with our study, Minh *et al.* (2010) reported that supplementation of herb like dried ginger to broiler diets led to an improved performance in broilers and reduced their cost of feed. Similarly, Oleforuh- Okoleh *et al.* (2014) recorded highest revenue and net return from birds fed herbal supplemented diets. Puvaca *et al.* (2016) also recorded lowest feed cost per kg body weight gain in herbal powder supplemented diets of broilers.

To ascertain the effects of nettle supplementation on health status of broilers, different biochemical, immune and oxidative parameters were analyzed at the end of experiment. All the parameters were within normal range indicating no deleterious effects of nettle on broilers. The blood glucose concentration of experimental birds revealed significant differences among various treatment groups, it was seen that birds of group control had the highest concentration of blood glucose in comparison to birds fed herb as an additive. A number of recent studies on nettle have revealed that it possesses anti-diabetic properties (Sharafetdinov *et al.*, 2017). The results of the lipid profile of birds fed different levels of nettle revealed significant differences. A significant reduction in the values of triglycerides, cholesterol and LDL and an increase in the values of HDL of birds fed nettle as feed additive was seen. The results of our study are in accordance with Mansoub (2011), who reported that supplementation of broilers with feed additives like *Urtica dioica* resulted in significant difference on the levels of total cholesterol and triglycerides in the serum however HDL was not significantly different among the treatment groups. In another study conducted by Safamehr *et al.* (2012) it was reported that supplementation of nettle@1% in broiler diets significantly

lowered the values of triglycerides and cholesterol. Also, in agreement with our results, Behdoodi *et al.* (2021), reported the effects of nettle supplementation on biochemical parameters in broilers and revealed that birds fed nettle had decreased serum concentration of blood urea nitrogen alkaline phosphatase, alanine aminotransferase, aspartate aminotransferase, triglycerides and cholesterol ($P<0.05$). In a study conducted by Majedi *et al.* (2021) it was reported that supplementation of nettle in broiler diets led to an improvement of most biochemical indices (AST, ALT, HDL, LDL, total protein and globulin. The cholesterol reduction activity of nettle could depend on its sterol and phenolic content (Kregiel *et al.*, 2018). Righi *et al.* (2021) in a study suggested that the positive effect of nettle on lipid profile may be related to its high content of antioxidants as polyphenols and Vitamins along with this nettle is also known to inhibit liver damage with its potent antioxidant activity (Yildizhan *et al.*, 2020).

A significantly ($P<0.05$) higher antibody response to SRBC in birds fed different levels of nettle was seen in comparison to control. In accordance with our study, Ngugi *et al.* (2015) in his study reported that nettle as an additive has a potential to improve the immune system of birds. Similarly, Sandru *et al.* (2016) in a study stated that alcoholic extract of *Urtica dioica* increased cell-mediated innate immune potential in chickens. In another study conducted by Poudel and Khanal (2011), it was reported that supplementation of nettle in laying hens improved the egg quality and the vitelline membrane integrity besides enhancing antibody titre against Newcastle disease. Presence of various phytochemicals like quercetin, kaempferol and isorhamnetin in the aerial parts of the nettle are known to contribute to its immuno-modulatory activity. The anti-oxidative activity of nettle was accessed by determining the total oxidative and total anti-oxidative status of broilers. A significant improvement in the oxidative status of birds fed nettle as feed additive was observed as they had reduced levels of TOS and increased levels of TAS in comparison to control. In a study by Loetcher *et al.* (2013b) it was reported that nettle contains alpha tocopherol, flavonoids, ferulic acid and carotenoids as natural antioxidants. Extracts of nettle have also been known to have a neutralizing role on reactive oxygen species (Gulcin *et al.*, 2004). In accordance with our results; Ahmadipour and Khajali(2019) revealed that nettle powder has potent antioxidant activity and its supplementation @1.5% had positive effects on broilers exposed to oxidative stress.

Table 8: Immune and oxidative parameters of broilers fed herb *Urtica dioica* as feed additive.

	Treatment groups			
	T ₁	T ₂	T ₃	T ₄
SRBC	1.56 ^a ±0.14	1.96 ^{ab} ± 0.120	2.16 ^b ±0.17	2.36 ^b ±0.176
DNCB	1.06 ^a ±0.07	1.37 ^b ± 0.082	1.49 ^b ± 0.090	1.97 ^c ±0.39
TOS (µmolTrolox Eq/L)	7.94 ^a ±0.48	7.11 ^b ±0.11	7.12 ^b ±0.88	6.93 ^c ±0.49
TAS (µmolH ₂ O ₂ Eq/L)	1.23 ^a ±0.14	1.27 ^b ±0.14	1.31 ^{bc} ±0.14	1.37 ^c ±0.145

Behboodi *et al.* (2021) in his experiment, studied various oxidative parameters in broilers fed nettle as feed additive and observed that nettle increased the activity of various anti-oxidative enzymes (T-SOD) and improved the anti-oxidative status (T-AOC) of birds.

CONCLUSION

The results of our study indicated that nettle supplementation at 1-2% had positive effects on performance, blood biochemical and various immune and oxidative parameters of broilers. It can be concluded that nettle can be safely used as a feed additive for sustainable broiler nutrition. The preferred level of inclusion according to our research is 2% on a D.M basis.

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Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal care and handling techniques were approved by the University of Animal Care Committee.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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