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Influence of a *Piriformospora indica* plant-specific fungus and clove essence on broiler health and performance parameters during oxidative stress

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Abstract

The aim of this study was to evaluate the effects of dietary *Piriformospora indica* (PI) and clove essence (CE) and a combination of them (synbiotic) on performance and biochemical parameters after induction of stress using dexamethasone. The experiment was performed with 320 male, one-day-old chicks (Ross 308) using eight experimental groups and five replications. In each replication, eight chickens were fed a maize–soy-based diet. Experimental groups included group 1 (T1): control diet (based on maize and soybean without any feed additives); group 2 (T2): control diet with 0.2 cc dexamethasone injection; group 3 (T3): control diet with 11.6 cc CE; group 4 (T4): control diet with 10 mg PI; group 5 (T5): control diet with 11.6 cc of CE with 0.2 cc injection of dexamethasone; group 6 (T6): control diet with 10 mg PI and 0.2 cc of dexamethasone; group 7 (T7): control diet with synbiotic containing 10 mg of PI with 11.6 cc of CE; group 8 (T8): control diet with synbiotic containing 10 mg of PI and 11.6 cc of CE with 0.2 mg dexamethasone injection. PI and CE were poured into the drinking water during the rearing period. After 31, 33, 38, and 40 days, 2 mg/kg BW of dexamethasone was injected in the right breast muscle of experimental groups 2, 5, 6, and 8; after 35 and 42 days, samples were taken. The addition of CE, PI, and the synbiotic, caused an increase in body weight gain, feed intake, and reduced FCR compared to the control group in experimental groups 3, 4, and 7. The addition of 11.6 cc CE and 10 mg of PI caused an increase in body weight gain and feed intake and reduced FCR compared to the control group. Dexamethasone (0.2 mg) led to damage of the lymph organs, body weight loss, a reduction in humoral immunity, and side effects on performance parameters. Clove extract and the prebiotic reduced the adverse effects of dexamethasone.

Keywords: broiler, clove, microbial, performance, *Piriformospora indica*, oxidative stress
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Introduction

Medicinal herbs in poultry feed for the treatment of animal diseases are considered to be low-cost and non-hazardous additives, without side effects and complications for the environment (Sevim and Ayasan, 2020). Herbal supplements increase gastric secretions, stimulate blood circulation and gastrointestinal enzymes, and improve the digestibility and function of birds (Ashraf *et al*., 2013). Clove (*Syzygium aromaticum* L.) is one of the most important medicinal plants, and contains beta-caryophyllene, acetyl eugenol, iso-eugenol, eugenin, camphor, tannin, gallic acid, vitamin C, and minerals (boron, calcium, iron, manganese, potassium, and phosphorus) (Jirovetz *et al*., 2006). Clove essential oil contains more than 40% of phenol and flavonoids and also contains effective compounds such as eugenol acetate, while eugenol alone makes up more than 83% of the volume of essential oil (Jirovetz *et al*., 2006). Clove increases enzyme secretion and enhances the digestive activity of the gut. Eugenol, the active ingredient in clove essential oil, can have a marked effect on villi height and number of goblet cells, thereby increasing nutrient uptake (Jirovetz *et al*., 2006).

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Piriformospora indica is a root endophytic fungus rich in mannan oligosaccharides isolated from rhizosphere soil of plants growing in the extremely hot conditions of the Thar desert of Rajasthan, India (Varma *et al*., 1999). This fungus is similar to arbuscular mycorrhizal fungi; however, it can be axenically cultivated on a variety of synthetic media (Singh *et al*., 2011). It grows inter- and intracellularly and forms a symbiosis with the roots of a wide variety of plants and can promote the growth and enhance the activity of secondary metabolites in a range of economical and medicinally important plants (Ghabooli *et al*., 2013; Ghaffari *et al*., 2016). Besides stimulating growth, *P. indica* increases the uptake and metabolism of nitrogen and phosphate and allows plants to survive under extreme abiotic stress (Sun *et al*., 2010).

The adverse effects of a corticosteroid increase can be mimicked using dexamethasone in broilers and lead to an increase in the release of free radicals and oxidative stress (Hosseini and Farahvar, 2017). Glucocorticoids released in response to stress cause muscle damage and free radical release in the body, causing tissue damage. They also weaken the lymph by weakening the immune system and further reduce the consumption of food by affecting the thyroid (Hosseini & Farahvar, 2017). The use of high levels of tea polyphenols has been reported to reduce the negative impacts of corticosterone on weight loss, hyperlipidaemia, and hypertrophy of the liver in broilers (Azizipouri *et al*., 2021).

The purpose of this study was to evaluate the use of dietary CE and PI and a combination of PI with CE in feed on performance and biochemical traits in broilers after induction of dexamethasone stress.

Materials and Methods

Experiments were carried out based on procedures and guidelines approved by the Animal Care Committee of the Iranian Council of Animal Care 1995 (No. IACUC95). In this study, 320 day-old, male, Ross 308 broilers were allocated randomly to eight experimental groups with five replications. In each replicate, eight chickens were fed a diet based on maize and soybean for six weeks, adjusted using software (WUFFDA) and based on the Ross-308 broiler manual (2014) (Table1). Experimental groups included: group1 (T1), control diet, based on maize and soybean without any feed additives; group 2 (T2), control diet with injection of 0.2 cc dexamethasone; group 3 (T3), control diet with 11.6 cc clove essence (CE); group 4 (T4), control diet with 10 mg *P. indica* (PI); group 5 (T5), control diet with 11.6 cc clove essence (CE) with 0.2 cc injection of dexamethasone; group 6 (T6), control diet with 10 mg *P. indica* (PI) and 0.2 cc dexamethasone; group 7 (T7), control diet with symbiotic containing 10 mg of *P. indica* (PI) and 11.6 cc of clove essence (CE); group 8 (T8), control diet with symbiotic containing 10 mg of *P. indica* (PI) and 11.6 cc of clove essence (CE) with 0.2 mg of dexamethasone injection. After 31, 33, 38, and 40 days, 2 mg of dexamethasone per kilogram BW was injected in right breast muscle of experimental groups 2, 5, 6, and 8 in all replications, and after 35 and 42 days, samples were taken. The clove essence (CE) and *P. indica* (PI) were administered in the drinking water. In this study, Dexacoid® (Nasr Fariman Co., Iran) was used and contained 4 mg of sodium phosphate dexamethasone per millilitre.

Piriformospora indica was cultured in accordance with the procedure described by Ghabooli *et al*. (2013). Briefly, cultures were maintained on a complex agar medium supplemented with 15 g/L agar at 28 ± 1 °C. For liquid culture of the fungus, discs from the actively growing edges of the petri dish culture were punched out and inoculated in 250 mL Erlenmeyer flasks containing 100 mL complex medium without agar at 28 ± 1°C on a gyratory shaker at 150 rpm for 6–7 d. After completing the growth phase of the fungus, the culture medium was mixed with wheat bran (100, 200, 400, 600, and 800 mL of medium) and then added to the different diets to produce diets with different levels of *P. indica*. (Ghabooli *et al*. 2013; Babamir *et al*. 2016).

To prepare the clove essence, cloves were macerated with 70% ethanol (1:20; w/v) at room temperature for 2 d and filtered using Whatman no.42 filter paper. Other portions of the solvent were added to the pulp and the extraction was repeated until the last extract was colourless. The extracts were combined and concentrated under reduced pressure at 75 ºC, 30 rpm, for 110 min, using a rotary vacuum evaporator. The crude extract was then evaporated in a boiling water bath (HANSHIN, Scientific Co, South Korea) until a constant weight was obtained to produce the extract. It was poured into sterile containers and sealed in a package. After 24 h of refrigeration, it was transferred to Malayer University Research Farm. Clove essence was poured into the drinking water using an insulin syringe.

Analysis of the composition of the dried clove (including moisture, protein, fibre, fat, and ash) was obtained according to AOAC1990 methods. Carbohydrate content was calculated by difference. This experiment was repeated three times and the mean and standard deviation were calculated. Physical

and chemical characteristics of clove hydroalcoholic extract were obtained according to the AOAC1990 methods (including acid percentage, ester and free fatty acid content, and refractive index) (Tables 2 and 3).

Body weight gain (BWG), feed consumption (FC), and feed conversion ratio (FCR) were recorded weekly. To study serum biochemical parameters, in each pen, two chicks were selected, and individual blood samples were collected separately from the brachial vein into non-heparinized tubes at 5 and 6 weeks of age. The sera were analysed for biochemical parameters (triglycerides and cholesterol), as well as blood proteins (total protein, globulin, and albumin) using commercial kits (Boehringer Mannheim Hitachi 704 automatic analyser, Japan). Collected data were analysed using a statistical model in SAS software (version 9.2; 2009) (SAS, 2009). Comparison of treatments means were performed using Duncan's multiple range test (1995) and the significance was considered at P <0.05.

Table 1. Ingredients and chemical composition of the basal diet in the starter, grower, and finisher periods (g/kg)

Item	Starter diet (0–10 d)	Grower diet (11–24 d)	Finisher diet (25–42 d)
Maize	543.2	567.6	587
Soybean meal, 440 g/kg CP	394.3	365	319
Maize oil	21.6	30.5	59
Dicalcium phosphate	20.5	18	16.8
Oyster shell	9	8	7.9
Sodium chloride	3.7	3.5	3
Vitamin and mineral premix[1]	5	5	5
dl-Met	2	1.8	1.7
l-Lys HCL	0.7	0.6	0.5
Calculated composition (%, unless otherwise stated)			
ME (MJ/kg)	12.13	12.55	13.4
Crude protein	221.6	205.1	192
SID-Lys2	11.5	10.07	9.6
SID-Met	5	4.9	4.8
SID-Met + Cys	8.3	8	7.8
SID-Thr	7.9	7.4	7.1
Calcium	10	9	8.5
Phosphorus (available)	5	4.4	4.2

[1]Supplied per kg diet: Vitamin A 9000 IU, vitamin D_3 2000 IU, vitamin E 18 IU, vitamin K_3 2 mg, riboflavin 6.6 mg, pantothenic acid 10 mg, pyridoxine 3 mg, folic acid 1 mg, thiamin 1.8 mg, B_{12} 15 µg, biotin 0.1 mg, niacin 30 mg, choline 500 mg, selenium 0.2 mg, iodine 1 mg, copper 10 mg, iron 50 mg, zinc 85 mg, manganese 100 mg

Table2. The chemical composition of clove sprouts (%)

Sample/items	Protein	Fat	Fibre	Ash	Moisture	Carbohydrate
Clove sprouts	1.2	12.1	20	5.2	10	51.5

Table 3. Ingredients of clove essence (assay done at Malayer University)

Sample/items	Refractometer index (%)	Free fatty acid (%)	Ester (%)	Acid (%)
Clove sprouts	1.5310	1.92	38.22	3.843

Results and Discussion

The results of weekly BWG in the first to fourth weeks in the treatment groups was not substantially different from the control group. The addition of PI and CE and an effect of the synbiotic on BWG was evident from 28 to 42 d (P <0.05); the most BWG occurred in prebiotic group (T6; 2768 g) and the lowest weight gain was in the dexamethasone group (T2; 2072 g) (P <0.05) (Table 4). BWG

in chickens as result of the use of antibiotic, herbal, or probiotic growth stimulants can be attributed to increasing feed intake, dietary fat retention, apparent metabolizable energy; improved energy absorption efficiency; and an increase in dietary protein utilization. Recent research has also indicated that injecting corticosterone into broilers for seven days reduces insulin-like growth factor type 1 and impairs of the metabolism of thyroid hormones. Therefore, during corticosterone stress, it reduces the rate of growth and food consumption (Soccol *et al*., 2010). Glucocorticoids released in response to stress trigger proteolytic mechanisms in muscles, especially skeletal muscles, and cause muscle breakdown and a decrease in BW (Furukawa *et al*., 2016). Increased body weight gain has been shown in groups receiving PI spores (Babamir *et al*., 2016) and 0.4 g/kg PI in the diet (Soleimanpour *et al*., 2020). From 0–14d, the groups receiving PI and CE and synbiotic were similar in terms of FC (P <0.05). From 14–42 d, the groups receiving PI, CE, and the symbiotic, had a higher FC than the control (P <0.05). Form 0–42 d, T6 had a high FC (4415 g) and the dexamethasone injection group (T2) had a lower FC (4205 g) (Table 5).

The active ingredients in these plants, such as carvacrol, thymol, and eugenol, have a stimulating effect on the secretion of digestive leachate from organs such as the pancreas and liver, and adequate secretion of these leachates leads to better digestion, absorption, and metabolism of nutrients. It is assumed that the height of villi in the small intestine increases in treatments containing essential oils, which increases digestion. Increased uptake, secretion of enzymes from the villi, and nutrient delivery systems in the small intestine lead to weight gain and an improvement in FCR (Atassi and Servin, 2010). The addition of PI to broiler diets (0.4 g/kg) did not have a marked effect on feed consumption compared to the other groups in another study (Soleimanpour *et al*., 2020).

The FCR in weeks 1–3 was similar to the control group; there was a substantial difference in weeks 3–6 of the rearing period (P <0.05). The groups receiving PI, CE, and the synbiotic had a lower FCR than the control group (P <0.05). At 42 d, the FCR in the dexamethasone injection group increased to 2.02, and the lowest FCR was in T6 (1.59) (P <0.05) (Table 6). Additional PI spores in the diet had no effect on FCR (Babamir *et al*., 2016), but 0.4 g/kg in the diet decreased FCR (Soleimanpour *et al*., 2020).

Table 4. The effect of clove essence (CE) and *Piriformospora indica* (PI) on body weight gain (BWG) (g/bird) of broilers from 1 to 42 days of age

Treatment	0–7d	7–14d	14–21d	21–28d	28–35d	35–42d	1–42d
T1	147.4	200.6	260	738	753.3^{ab}	308.2 ^{ab}	2407.5^{ba}
T2	147.7	199.6	259.6	736.3	523.6^{bc}	205.7 ^{ac}	2072.5^{bc}
T3	144.7	203.6	268.6	794.3	825.3^{b}	380.3 ^{b}	2616.8^{c}
T4	143.4	203.6	271	958	754.6^{ab}	387.5 ^{b}	2718.1^{b}
T5	145.1	203.6	265.6	795	771.3^{c}	295.5 ^{ab}	2476.1^{ac}
T6	143.1	215	267.6	885.6	849.6^{a}	453.4 ^{a}	2768.8^{a}
T7	145.1	202	270	956	742.3^{ab}	258.6 ^{ac}	2619.5^{c}
T8	143.1	214.6	268.3	884.3	719^{ac}	315.5 ^{c}	2544.8^{ab}
SEM	1.2	1.4	1.3	2.1	2.9	2	2.3
***P*-value**	0.6942	0.2792	0.2473	0.7059	<0.0001	<0.0001	<0.0001

T1: (control diet, based on maize and soybeans without any feed additives); T2: control diet + 0.2 cc dexamethasone; T3: control diet + 11.6 cc CE; T4: control diet + 10 mg PI; T5: control diet + 11.6 cc CE + 0.2 cc dexamethasone; T6: control diet + 10 mg PI + 0.2 cc of dexamethasone; T7: control diet + symbiotic (10 mg PI + 11.6 cc CE); T8: control diet + symbiotic (10 mg PI + 11.6 cc CE) + 0.2 mg of dexamethasone.

Mean values within a row with different superscript letters (a, b and c) are significantly different (P <0.05); SEM, standard error of mean

Table 5. The effect of clove essence (CE) and *Piriformospora indica* (PI) on feed consumption (FC) (g/bird) of broilers from 1 to 42 days of age

Treatment	0–7d	7–14d	14–21d	21–28d	28–35d	35–42d	1–42d
T1	186.3	254.8	587.9^{b}	881.6^{c}	1446.7^{bc}	880.7^{bc}	4238^{ac}
T2	187.3	254	587.3^{b}	879.4^{c}	1413.4^{ac}	873^{c}	4205^{bc}
T3	179.6	252.2	612.5^{a}	895.3^{b}	1424^{ac}	884 ^{bc}	4237^{ab}
T4	179.9	250.7	613.4^{a}	901.6^{a}	1486.4^{c}	872^{c}	4304^{c}
T5	179.3	250.5	612.2^{a}	897.6^{b}	1407^{ab}	860.4^{c}	4207^{ac}
T6	179.3	250.7	614^{a}	908.3^{a}	1517.4^{a}	950^{a}	4415^{a}
T7	178.6	253.9	607.5^{a}	907.6^{a}	1461.3^{ab}	888.4^{ab}	4294^{c}
T8	177.9	252.7	610.4^{a}	904.6^{a}	1499^{b}	950.4^{b}	4395^{b}
SEM	0.8	1.2	1.2	1.4	2	1.7	1.31
***P*-value**	0.4645	0.0729	0.0247	0.0278	0.0302	0.0311	<0.001

T1: (control diet, based on maize and soybeans without any feed additives); T2: control diet + 0.2 cc dexamethasone; T3: control diet + 11.6 cc CE; T4: control diet + 10 mg PI; T5: control diet + 11.6 cc CE + 0.2 cc dexamethasone; T6: control diet + 10 mg PI + 0.2 cc of dexamethasone; T7: control diet + symbiotic (10 mg PI + 11.6 cc CE); T8: control diet + symbiotic (10 mg PI + 11.6 cc CE) + 0.2 mg of dexamethasone.
Mean values within a row with different superscript letters (a, b and c) are significantly different (*P* <0.05); SEM, standard error of mean

Table 6.The effect of clove essence (CE) and *Piriformospora indica* (PI) on feed conversion ratio (FCR) from 1 to 42 days of age

Treatment	0–7d	7–14d	14–21d	21–28d	28–35d	35–42d	1–42d
T1	1.26	1.27	2.26	1.19^{a}	1.92^{c}	2.85^{ac}	1.76^{b}
T2	1.26	1.27	2.26	1.19^{a}	2.71^{a}	4.24^{a}	2.02^{a}
T3	1.24	1.23	2.28	1.12^{b}	1.71^{bc}	2.32^{ba}	1.61^{ac}
T4	1.25	1.23	2.26	0.94^{ab}	1.96^{c}	2.25^{bc}	1.60^{ab}
T5	1.23	1.23	2.30	1.12^{b}	1.82^{ab}	2.91^{ab}	1.69^{ab}
T6	1.25	1.16	2.29	0.94^{ab}	1.71^{ac}	2.09^{ca}	1.59^{bc}
T7	1.23	1.25	2.25	1.01^{c}	2.04^{b}	3.43^{b}	1.63^{ac}
T8	1.24	1.17	2.27	1.02^{c}	2.08^{b}	3.01^{c}	1.72^{c}
SEM	0.06	0.05	0.01	0.008	0.007	0.001	0.001
***P*-value**	0.3581	0.6701	0.5504	0.0427	<0.0001	0.0063	<0.0001

T1: (control diet, based on maize and soybeans without any feed additives); T2: control diet + 0.2 cc dexamethasone; T3: control diet + 11.6 cc CE; T4: control diet + 10 mg PI; T5: control diet + 11.6 cc CE + 0.2 cc dexamethasone; T6: control diet + 10 mg PI + 0.2 cc of dexamethasone; T7: control diet + symbiotic (10 mg PI + 11.6 cc CE); T8: control diet + symbiotic (10 mg PI + 11.6 cc CE) + 0.2 mg of dexamethasone
Mean values within a row with different superscript letters (a, b and c) are significantly different (*P* <0.05); SEM, standard error of mean

The cholesterol and triglyceride results at 35 and 42d indicate that the addition of clove essence, *P. indica,* and the synbiotic substantially reduced cholesterol and triglyceride (*P* <0.05). The dexamethasone injection groups (especially T2), cholesterol and triglyceride were substantially increased, and in T3, TG and CHO were decreased (*P* <0.05). Treatment groups had no effect on low-density lipoprotein (LDL), high-density lipoprotein (HDL), and total protein (TP) at 35 d and 42 d. The addition of clove essence caused a substantial increase in GLO at 35 d. At 35 d and 42 d, Alb was increased in T2, in T1 at 35 d, and in T7 at 42 d (*P* <0.05) (Table 7 and 8).

In an experiment performed on broiler blood serum parameters, researchers found a marked phenotypic correlation between triglyceride, albumin, globulin, creatinine, LDL, very LDL, and abdominal cavity fat. Another study on broiler chickens showed that triglyceride, LDL, and VLDL concentrations had a significant phenotypic correlation with body fat and could therefore be used as an indirect method of reducing body fat (Ankari and Homeid, 1996). The concentration of different blood components varies under the influence of various factors such as age, sex, physiological status, nutrition, and genetics. In the current experiment, all factors except nutrition were fixed. Another study showed no differences between treatments in glucose, cholesterol, triglyceride, and LDL on day 14, but after 42 d, blood glucose was enhanced and cholesterol, triglyceride, and LDL were reduced in chicks fed diets containing *P. indica* (Soleimanpour *et al*., 2020).

Table 7. The effect of clove essence and *Piriformospora indica* (PI) on biochemical parameters in broilers at 35 days of age (mg/dL)

Treatment	CHO	TG	HDL	LDL	Alb	TP	Glo
T1	151.4^{ab}	90.4^{ab}	112.6	32.3	1.55^{b}	3.49	2.10^{ab}
T2	177^{a}	126.8^{a}	106.3	34.6	2.01^{a}	5.51	3.06^{b}
T3	126.7^{bc}	75.8^{ca}	126.3	29.3	1.60^{b}	5.37	3.52^{a}
T4	139.7^{ba}	77.8^{bc}	128	29.2	1.68^{b}	4.29	2.6^{b}
T5	149.7^{ac}	102.5^{c}	115.3	30.9	1.76^{b}	4.11	2.5^{b}
T6	157^{c}	108.8^{b}	117	32.3	1.86^{b}	4.46	2.67^{b}
T7	123.4^{ca}	82.4^{ba}	131.3	29	1.68^{b}	3.72	2.17^{c}
T8	160.4^{b}	85.8^{ac}	121.3	31	1.85^{b}	4.68	3.51^{a}
SEM	0.72	0.46	1.3	0.8	0.03	0.59	0.18
***P*-value**	<0.0001	<0.0001	0.0586	0.9692	0.0009	0.0507	<0.0001

T1: (control diet, based on maize and soybeans without any feed additives); T2: control diet + 0.2 cc dexamethasone; T3: control diet + 11.6 cc CE; T4: control diet + 10 mg PI; T5: control diet + 11.6 cc CE + 0.2 cc dexamethasone; T6: control diet + 10 mg PI + 0.2 cc of dexamethasone; T7: control diet + symbiotic (10 mg PI + 11.6 cc CE); T8: control diet + symbiotic (10 mg PI + 11.6 cc CE) + 0.2 mg of dexamethasone
Mean values within a row with different superscript letters (a, b and c) are significantly different (P <0.05); SEM, standard error of mean; CHO, cholesterol; TG, triglyceride; HDL, high-density lipoprotein; LDL, low-density lipoprotein; Alb, albumin; TP, total protein; Glo, globulin

Table 8. The effect of clove essence and *Piriformospora indica* (PI) on biochemical parameters in broilers at 42 d of age

Treatment	CHO	TG	HDL	LDL	Alb	TP	Glo
T1	135.3^{ac}	93^{b}	114	31.3	1.73^{c}	4.39	2.72
T2	174.7^{a}	110^{a}	107.6	34	2.63^{a}	4.81	2.67
T3	135.3^{ac}	59^{bc}	126.6	27.8	1.94^{b}	3.95	2.19
T4	133.7^{ac}	71^{ac}	128.3	28.4	1.76^{c}	3.63	2.09
T5	164.3^{b}	88^{c}	117.6	30.5	1.77^{c}	4.05	2.25
T6	159.3^{c}	80^{ab}	119.3	30.03	1.8^{c}	4.69	2.13
T7	129.3^{bc}	68^{ba}	131.6	29.8	1.67^{ab}	4.39	2.42
T8	151^{ab}	81^{ab}	120.8	32	1.92^{b}	4.52	2.37
SEM	0.7	0.98	1.1	0.8	0.07	0.39	0.31
***P*-value**	0.0010	0.0058	0.7108	0.6250	<0.0001	0.6948	0.7657

T1: (control diet, based on maize and soybeans without any feed additives); T2: control diet + 0.2 cc dexamethasone; T3: control diet + 11.6 cc CE; T4: control diet + 10 mg PI; T5: control diet + 11.6 cc CE + 0.2 cc dexamethasone; T6: control diet + 10 mg PI + 0.2 cc of dexamethasone; T7: control diet + symbiotic (10 mg PI + 11.6 cc CE); T8: control diet + symbiotic (10 mg PI + 11.6 cc CE) + 0.2 mg of dexamethasone
Mean values within a row with different superscript letters (a, b and c) are significantly different (P <0.05); SEM, standard error of mean; CHO, cholesterol; TG, triglyceride; HDL, high-density lipoprotein; LDL, low-density lipoprotein; Alb, albumin; TP, total protein; Glo, globulin

Conclusions

The addition of 10 mg PI caused a substantial increase in body weight gain and feed intake substantially reduced FCR compared to the control group. The use of 0.2 mg dexamethasone caused damage to lymphatic organs, body weight loss, a reduction in humoral immunity, and negative effects on performance parameters. The inclusion of 11.6 cc CE had a marked effect on some biochemical parameters. Therefore, it is recommended that *P. indica* and CE can be used to reduce stress in poultry and increase performance parameters.

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Authors Contributions

AF and MH established the experimental shed, arranged materials, and executed the methodology. AF and MH contributed to sample and data collection. MGH prepared *P. indica* for this study. MY analysed the data, and MH write the manuscript.

Conflict of Interest Declaration

Authors have no conflict of interests to declare.

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