



Research Article

Effect of Different Herbs on the Histomorphology and Performance of Coccidiosis Challenged Broiler Chickens

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Abstract | The present study was designed to evaluate the effect of herbs on the performance of broiler chickens challenged with coccidiosis. A total of 450 broiler chickens were allotted into 6 groups (n = 75/group). Group A controlled without supplementation, Group B, C and D were supplemented with *Aloe barbadensis* (5ml/L), *Ferula foetida* regal (500mg/L), *Tamarindus indica* (50mg/L), respectively. Group E and F were supplemented with a mixture of all 3 herbs/plants at 2ml/L; mixed with distilled water and another with citric acid. The dressing percentage was observed significantly ($P < 0.05$) higher in compound II supplementary group F and lowest in control group A. Whereas, the fat pad percentage recorded significantly ($P < 0.05$) lowest in the compound II supplementary group F and highest in control group A. The relative weight of organs; liver, pancreas, spleen, heart, duodenum, jejunum, ileum, thymus and ceaca was observed significant ($P < 0.05$) in the herbs supplementary groups as compared to control group. The compound-II supplementary group F significantly ($P < 0.05$) increased the percentage of crude protein and fat contents as compared to control and other groups. Histomorphological examination of liver of broiler chicken supplemented with Compound-I and Compound-II in the coccidiosis challenged broiler chickens showed few pathological changes as compared to control (coccidiosis induced and without supplementation) group. In conclusion, supplementation of herbs mixture (Group E and F) at the dose 2ml/L to the coccidiosis challenged broiler chickens improve the overall performance and liver function.

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Introduction

The antibiotic development promoters as feed additives have been forbidden in many countries due to cross-resistance adjacent to pathogens and residues in tissues of animal and poultry (Hajati *et al.*, 2014). There are several evidences have been reported by different scientists on influence of antimicrobial drugs on the unlike herbal extorts in various parts of world (Rajput *et al.*, 2012; Jarić *et al.*, 2015; Alabi

et al., 2017). Medicinal herbs and their extracts are among the alternative sources of allopathic growth enhancers are already being in use (Jarić *et al.*, 2015). The medicinal herbs could be an excellent alternative source of allopathic growth promoters, with lower expenditure of manufacturing, and abridged danger of toxicity, least hazards for health and friendly environment, since the physical condition of intestinal gut plays vital role in the continuance of better development with most important function of absorption

and amalgamation of nutrients in birds (Rajput *et al.*, 2013; Alabi *et al.*, 2017; Sobolewska *et al.*, 2017). Herbs supplementation decrease the mortality rates, the feeding cost, accumulation of lipids and increases dressing percentage weight of liver, spleen, whole giblet weight in poultry birds (Jahejo *et al.*, 2019). Furthermore, Aloe vera gel, Amla, curcumin, mushrooms produced anti-microbial properties against pathogenic microbes and positively influenced on growth of gut microflora in broiler chicken (Mbangi *et al.*, 2010). Darabighane *et al.* (2012) documented that Aloe vera have strongly therapeutic effect against pathogenic microbes and significantly influenced on the gut microflora and enhance immunity in broiler chickens.

Coccidiosis is one of the most economically significant diseases of poultry around the globe. Depending upon the prevailing market conditions, the total economic losses (treatment, prophylaxis, and supportive therapy) from coccidiosis were estimated to be US \$45,405.00. Different species of *Eimeria* cause an estimated loss of US\$ 800 million per year to the world's commercial poultry producers (Williams, 1998). Medicinal herbs each and every plant or their extracts and essential oils consist of many bioactive chemical compounds which can be used as a growth promoter, diuretic, anthelmintic, appetizer, alkaline phosphates stimulator, antibacterial and antifungal agent (Hajati *et al.*, 2014).

The dietary supplementation effect with natural carotenoids curcumin and lutein on the broiler pigmentation and immunity. In addition, we have also found the immunomodulatory effect of *Ocimum basilicum* seeds supplementation to the heat-stressed broiler chickens (Rajput *et al.*, 2013; Jahejo *et al.*, 2019). The most common herbs and plant extracts, which are being used as an alternative source of antimicrobials in poultry are aloe vera, giloy, tabasheer, vaghayani, anwara, gadamri, garlic, cumin, black cumin, wild mint, pumpkin, thyme, cinnamon, chestnut, clove, alfalfa, turmeric, sumac, mushroom, grape seed, goldthread, mulberry leaf and honeysuckle (Aroche *et al.*, 2018). Birds challenged with *Eimeria* resulted in structural and functional damages in the small intestine, digestive enzymes, nutrients absorbers and but can be improved with the supplementation of various therapeutic agents and medicinal plants (Rajput *et al.*, 2013; Kim *et al.*, 2017). Medicinal plants increase the productive performance of poultry and health generally by stabilizing the microorganisms of the intestine

inhibiting the growth of pathogenic microbes and improving the performance and production of digestive enzymes (Tekeli *et al.*, 2006).

Keeping the significance of herbal plants in view of the performance of broiler chickens, the present study has been designed to evaluate the role of herbs in the coccidiosis challenged broiler chickens.

Materials and Methods

Experimental design

A total of four hundred fifty (450 day-old-chicks) were purchased from a well reputed hatchery and equally allocated into 6 groups/5 replicates.

Group-A was kept as control (without medicinal herbs supplementation), while birds of group-B were supplemented with Aloe vera (*Aloe barbadensis*) group-C with Vaghayani (*Ferula foetida* regal), group-D with Imli; *Tamarindus indica*, group-E with compound-I (Equal proportion of Aloe vera, Vaghayani and Imli properly mixed and dissolved in potable water). The group-F was supplemented with compound-II (Equal proportion of Aloe vera, Vaghayani and Imli properly mixed and dissolved in citric acid). All the medicinal plants/herbs and their mixture compound were initially analyzed for their nutritive value *i.e.* protein, fiber, fat, carbohydrates and minerals, respectively. The chemical/proximate composition of used medicinal plants/herbs and two compounds were given in Table 1. The dose concentration of each medicinal herb is given in Table 2. The feed was offered twice a day and refusal was collected once at morning and as necessary water was provided. The iso-caloric and iso-nitrogenous basal diet was prepared as recommended by National Research Council (1994) for broiler chicken with complete nutrients profile (Table 3).

The birds were vaccinated against New Castle disease (ND), Infectious bronchitis (IB) and Infectious bursal disease (IBD) diseases and kept under environmentally controlled houses. The lighting program consisted of 23 hours of light during the whole experimental period. Temperature was maintained around 95 to 90 °F for first week, 90 to 85 °F second week, 85 to 80 °F third week, 80 to 75 °F fourth week, and 75 to 70 °F during 5th and 6th weeks. Around 45% to 65% of humidity level was controlled within the shed. The Institutional Committee for Animal use and Care,

Table 1: Chemical/proximate composition of medicinal plants/herbs used as growth promoters for the rearing of broiler chicken.

Medicinal plants/herbs	Chemical Composition (%)				
	Crude protein	Crude fiber	Crude Fat	Carbohydrate	Total Minerals
Aloe vera (<i>A. barbadensis</i>)	10.50	0.15	1.83	56.30	19.50
Vaghayani/Hing (<i>F. foetida regel</i>)	4.00	4.00	1.00	68.00	7.00
Gadamri/Imli (<i>T. indica</i>)	23.50	3.50	6.94	10.00	6.24
Compound-I	25.32	6.50	3.94	15.00	8.20
Compound-II	27.50	8.50	4.41	21.00	9.78

Group-A = Control (fed on free antibiotic and anticoccidial diet), Group-B = Aloe vera (*Aloe barbadensis*) herb supplementation, Group-C = Vaghayani/Hing (*Ferulafoetida regel*) herb supplementation, Group-D = Gadamri/Imli (*Tamarindus indica*) herb supplementation, Group-E = Compound-I and Group-F = Compound-II supplementation.

Table 2: Medicinal herbs and compounds with dose level used for research trial.

Groups	Local name	Scientific name	Part used	Dosage	Source
A	Control	N/A	Basal diet	N/A	N/A
B	Aloe vera	<i>Aloe barbadensis</i>	Leave juice	5ml/L	DW
C	Vaghayani/Hing	<i>Ferula foetida regel</i>	Gum resin	500mg/L	DW
D	Gadamri/Imli	<i>Tamarindus indica</i>	Fruit pulp	50mg/L	DW
E	Compound-I	Mixture of 6 herbs dissolved in water		2ml/L	DW
F	Compound-II	Mixture of 6 herbs dissolved in citric acid		2ml/L	DW

N/A = Not applicable, DK= Drinking water, Group-A = Control (fed on free antibiotic and anticoccidial diet), Group-B = Aloe vera (*Aloe barbadensis*) herb supplementation, Group-C = Vaghayani/Hing (*Ferulafoetida regel*) herb supplementation, Group-D = Gadamri/Imli (*Tamarindus indica*) herb supplementation, Group-E = Compound-I and Group-F = Compound-II supplementation.

Table 3: Feed ingredients and nutrient composition of basal diets for broiler chicken.

Ingredients (%)	Starter (1 to 21 d)	Finisher (22 to 38 d)
Rice	60.25	65.47
Soybean meal	32.5	28
Corn gluten meal	3	2.5
Limestone	1.2	1.2
Dicalcium phosphate	2	1.8
NaCl	0.3	0.3
Feed premix ¹	0.5	0.5
L- lysine	0.1	0.1
DL- methionine	0.15	0.13
Chemical composition of diet		
ME (kcal/kg)	2998	3023.9
CP %	21.19	19.20
Lysine %	1.21	1.09
Methionine %	0.496	0.440
Methionine+cystine %	0.80	0.72
Ca %	0.988	0.949
P %	0.531	0.485

Sindh Agriculture University Tandojam, approved all experimental procedures.

Coccidiosis infection

All the nine (6) groups, including group-A which was kept as control (fed on the free antibiotic and anti-coccidial diet) at the age of 21d, the birds were orally challenged with *Eimeria* populated oocysts (10'000 oocysts per broiler) diluted in 0.5 ml of distilled water as performed in our previous study [Rajput et al. \(2013\)](#).

Dressing percentage

After weighing of live bird, it was sacrificed, de-feathered and visceral organs have been removed, and carcass weight was recorded on analytical weighing balance, while dressing percentage of birds from each group was estimated with the help of following formula;

$$\text{Dressing (\%)} = \text{Carcass weight} \times 100$$

Relative weight of organs

Total Six (6) broiler chickens from each supplement-

ed group were indiscriminately chosen, off feed for 6 hours, separately weighed on digital weighing machine and slaughtered at the completion of research trial. Weight of gizzard, liver, pancreas, spleen, heart, duodenum, jejunum, ileum, thymus, ceaca and fat pad content weights were measured with the help of analytical balance.

Nutrients digestibility

The feces dropping of the broiler gathered from all allocated groups, and dried into a hot oven, weighed and later powdered to pass through a 1.0mm sieve for determination of crude protein and crude fat method as described by AOAC (2000). Further nutrient digestibility was estimated by method documented by (Rajput *et al.*, 2013). To get the percentage of crude protein, 1g fecal sample was initially weight and 5g digestion mixture containing (copper sulfate, ferrous sulfate and potassium sulfate) added in digestion flask along with 30ml commercial HCl. Well shake and mix sample in digestion flask then put on 1000watt heater for 3 hours subjected to digestion till light green color appeared, after digestion sample was cool down at normal room temperature for 30 minutes. Sample was diluted with distilled water. 10ml diluted sample and 10ml NaOH 40% was put in Kjeldhal apparatus for distillation and ammonia was captured in 10ml boric acid 2% in beaker from another end, beaker was filled up to 50ml. Finally, titration with H_2SO_4 to captured ammonia till yellowish color change into light pink color, its reading was noted down for crude protein calculation.

$$\text{Constant} = 10.937$$

Reading = Volume used \times Constant Formula
as given bellow used for estimation of apparent
nutrients retention

(Nutrients in feed - Nutrients in meat / Nutrients in feed \times 100) expressed as a percentage.

To get the percentage of crude content, it was analyzed through Soxhlet extraction apparatus. 5g dried sample was wrapped in to filter paper, the process with solvent diethyl ether for a time period of 5-6 hours at 55-60°C in Soxhlet extraction flask filled $\frac{3}{4}$ with diethyl ether and run water condenser for 2-3 hours. Sample was removed and all the diethyl ether-was evaporated by using hot air oven, flask was dried, and final weight of the flask was taken and result was computed through formula mentioned bellow:

$$\text{Fat (\%)} = \frac{W2 - W1}{\text{Weight of sample}} \times 100$$

W1: Empty weight of Soxhlet flask

W2: Final weight of Soxhlet flask

Histomorphological examination of liver

For the histomorphological analysis of liver, the tissue samples from liver was collected from the coccidiosis challenged slaughtered broiler birds at last day of research trial and fixed in 10 percent buffered formalin. Liver tissues dried by dipping in the alcohol with increasing from 70% to its purity, permeated by Xylene, fixed in paraffin. Casting of tissue blocks was done in L-molds which the assist of manipulation of size as per the need of the protocol. The paraffin sections were sliced with the help of tissue microtome. The paraffin fixed tissue blocks with the thickness of 5mm were properly trimmed and sectioned. Continuous ribbons (6-7inches) of the tissue sample were cut and kept into the water bath at constant temperature of 55°C. Tissue sections were divided with hot sharp blade after they extend entirely. Clean glass slides were used for mounting of tissue segments by using section adhesive made up of Mayer's egg albumin. Mounted tissue glides were dehydrated at 60°C in oven for 1h. Harris hematoxylin and eosin staining methods were used for the staining of tissue sections. Graded alcohol along with distilled water was used during de-paraffinizing the paraffin sections by Xylene before hydration. After dehydration clearing was performed by using Xylene, and distrain plasticizer Xylene mountant drop was positioned on a cover slip and section on the slide pushed on it, slide was up-turned and cover slip was pushed with a rod to take away air bubbles if any intent. Oculometer at a magnification of 10X was used for calculating the values under a light microscope fixed with stage micrometer.

Results and Discussion

Dressing and fat pad percentage

The results of dressing and fat pad percentage of broiler chickens supplemented with different medicinal plants/herbs and compounds are given in Figure 1. The dressing percentage was observed significantly ($P < 0.05$) higher in compound II supplementary group F and lowest in control group A. However, overall, the results of dressing percentage within the herb supplementary groups B, C, D, E and F recorded significant ($P < 0.05$) as compare to control group A. Whereas, the fat pad percentage recorded significantly

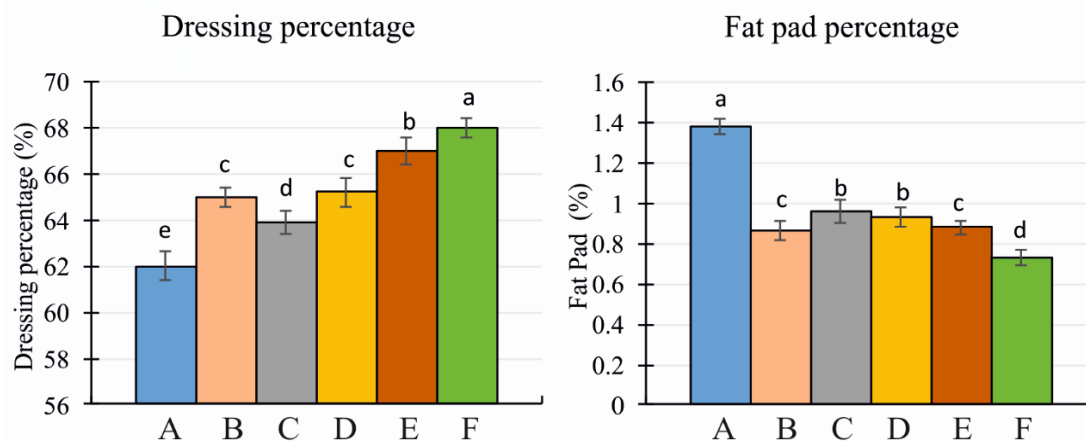


Figure 1: Influence of most effective medicinal plants/herbs and compounds on dressing and fat pad content percentage of coccidiosis challenged broiler.

^{a-f}Superscripts with different letters in same column varied statistically to each other.

Group-A = Control (fed on free antibiotic and anticoccidial diet), Group-B = Aloe vera (*Aloe barbadensis*) herb supplementation, Group-C = Vaghayani/Hing (*Ferulafoetida regel*) herb supplementation, Group-D = Gadamri/Imli (*Tamarindus indica*) herb supplementation, Group-E = Compound-I and Group-F = Compound-II supplementation.

Table 4: Influence of most effective medicinal plants/herbs and compounds on the relative weight of organs (g) of coccidiosis challenged broiler chickens.

Groups	Weight of internal edible organs (g)								
	Liver	Pancreas	Spleen	Heart	Duodenum	Jejunum	Ileum	Thymus	Ceaca
Group-A	2.32 ^{ab}	0.81 ^d	0.089 ^c	0.56 ^a	0.49 ^c	0.76 ^d	0.64 ^c	0.16 ^{bc}	0.38 ^a
Group-B	1.94 ^c	1.31 ^c	0.087 ^c	0.53 ^b	0.57 ^b	0.96 ^c	0.68 ^{cd}	0.21 ^b	0.34 ^b
Group-C	2.27 ^{ab}	1.64 ^{ab}	0.106 ^a	0.52 ^b	0.64 ^a	0.99 ^c	0.88 ^c	0.21 ^b	0.28 ^c
Group-D	2.03 ^c	0.69 ^d	0.103 ^{ab}	0.52 ^b	0.52 ^b	0.98 ^c	0.86 ^c	0.15 ^{bc}	0.32 ^c
Group-E	2.30 ^{ab}	1.68 ^a	0.083 ^c	0.50 ^b	0.61 ^{ab}	1.02 ^a	0.92 ^a	0.21 ^b	0.28 ^c
Group-F	2.38 ^a	0.86 ^d	0.084 ^c	0.50 ^b	0.52 ^b	1.00 ^{ab}	0.90 ^{ab}	0.33 ^a	0.37 ^a

^{a-f}Superscripts with different letters in same column varied statistically to each other.

Group-A = Control (fed on free antibiotic and anticoccidial diet), Group-B = Aloe vera (*Aloe barbadensis*) herb supplementation, Group-C = Vaghayani/Hing (*Ferulafoetida regel*) herb supplementation, Group-D = Gadamri/Imli (*Tamarindus indica*) herb supplementation, Group-E = Compound-I and Group-F = Compound-II supplementation.

($P < 0.05$) lowest in the compound II supplementary group F and highest in control group A. The fat pad percentage of groups C, D and B, E were insignificant with each other ($P > 0.05$).

Relative weight of organs

The influence of herbs on the relative weight of organs; liver, pancreas, spleen, heart, duodenum, jejunum, ileum, thymus and ceaca are given in Table 4. The results of relative weight were significant ($P < 0.05$) within the groups A, B, C, D, E and F. The relative weight of liver was significantly ($P < 0.05$) higher in group F as compare to all other groups but insignificant with group E.

The relative weight of pancreas was significantly ($P <$

0.05) higher in group E as compare to all other groups but insignificant with group C. However, the control group A and compound II supplementary group E were insignificant ($P > 0.05$) with each other.

The relative weight of spleen was significantly ($P < 0.05$) higher in group C and D as compare to all other groups but insignificant ($P > 0.05$) difference was observed within groups A, B, E and F.

The relative weight of spleen was significantly ($P < 0.05$) higher in group A as compare to all other groups but insignificant ($P > 0.05$) difference was observed within groups B, C, E, E and F.

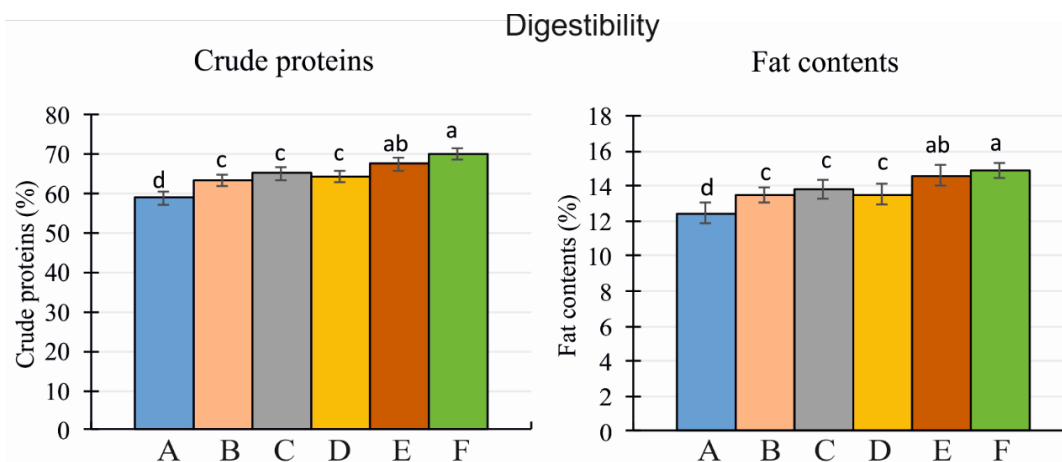


Figure 2: Influence of most effective medicinal plants and compounds on the crude protein and fat contents percentage of coccidiosis challenged broiler.

^{a-f}Superscripts with different letters in same column varied statistically to each other.

Group-A = Control (fed on free antibiotic and anticoccidial diet), Group-B = *Aloe vera* (*Aloe barbadensis*) herb supplementation, Group-C = *Vaghayani/Hing* (*Ferulafoetida regel*) herb supplementation, Group-D = *Gadamri/Imli* (*Tamarindus indica*) herb supplementation, Group-E = Compound-I and Group-F = Compound-II supplementation.

The relative weight of duodenum, jejunum and ilium was significantly ($P < 0.05$) higher in group E and F as compare to all other groups.

The relative weight of thymus was significantly ($P < 0.05$) higher in group F as compare to all other groups but insignificant ($P > 0.05$) difference was observed within groups B, C, D, E and F.

The relative weight of caeca was significantly ($P < 0.05$) higher in group A and F as compare to all other groups but insignificant ($P > 0.05$) difference was observed within groups C, D and E.

Nutrients digestibility of crude protein and crude fat percentage

The influence herbs and herb mixture compounds were observed on the nutrients digestibility of crude protein and fat percentage in coccidiosis challenged broiler chickens' feces and the results are presented in Figure 2.

The dietary supplementation of different medicinal plants and their associated compounds considerably ($P < 0.05$) increased utilization of crude protein and fat contents in the coccidiosis challenged broiler chickens as compared to the control group A chickens. The compound-II supplementary group F significantly ($P < 0.05$) increased the percentage of crude protein and fat contents. Whereas, the insignificant results were observed within the groups B, C and D.

Histomorphology of liver

The results of histomorphological examination of liver in coccidiosis challenged broiler chicken are presented in Figure 3. The results revealed that the extensive degeneration of hepatocytes with sporadic deformation of hepatic cords and hyperplasia of Kupffer cells was recorded in control group. In addition, the perivascular area of the liver of non-supplemented group of bird's numerous noticeable permeations of mononuclear cell was observed and irregular change was noticed in the size of liver parenchyma. However, results of histomorphological examination of liver of broiler chicken supplemented with *Aloe vera* (*Aloe barbadensis*) and Compound-II and Compound-I challenged with coccidiosis infection showed few pathological changes like the noticeable propagation of poly-morphonuclear and mononuclear cells and activation of immune response in shape of activation of liver macrophage system for the repairing of the Kupffer cells, the size and shape of the liver seems to be normal contrast to broiler allocated in control group infected with coccidiosis. Whereas, the histomorphology of the liver of broiler infected with coccidiosis treated with the *Gadamri/Imli* (*Tamarindus indica*), *Vaghayani/Hing* (*Ferulafoetida regel*), *Anwara* (*Emblica officinalis*), *Tabashir* (*Bambusa arundinacea*) and *Giloy/Satgullo* (*Tinaspora cordifolia*) medicinal plants/herbs illustrated rare and mild permeation of perivascular mononuclear cells, low degree of hepatocellular degeneration, minute enlargement of Kupffer cells and sporadic heterophile intensify in parenchyma, though few variations noticed, while severe noted

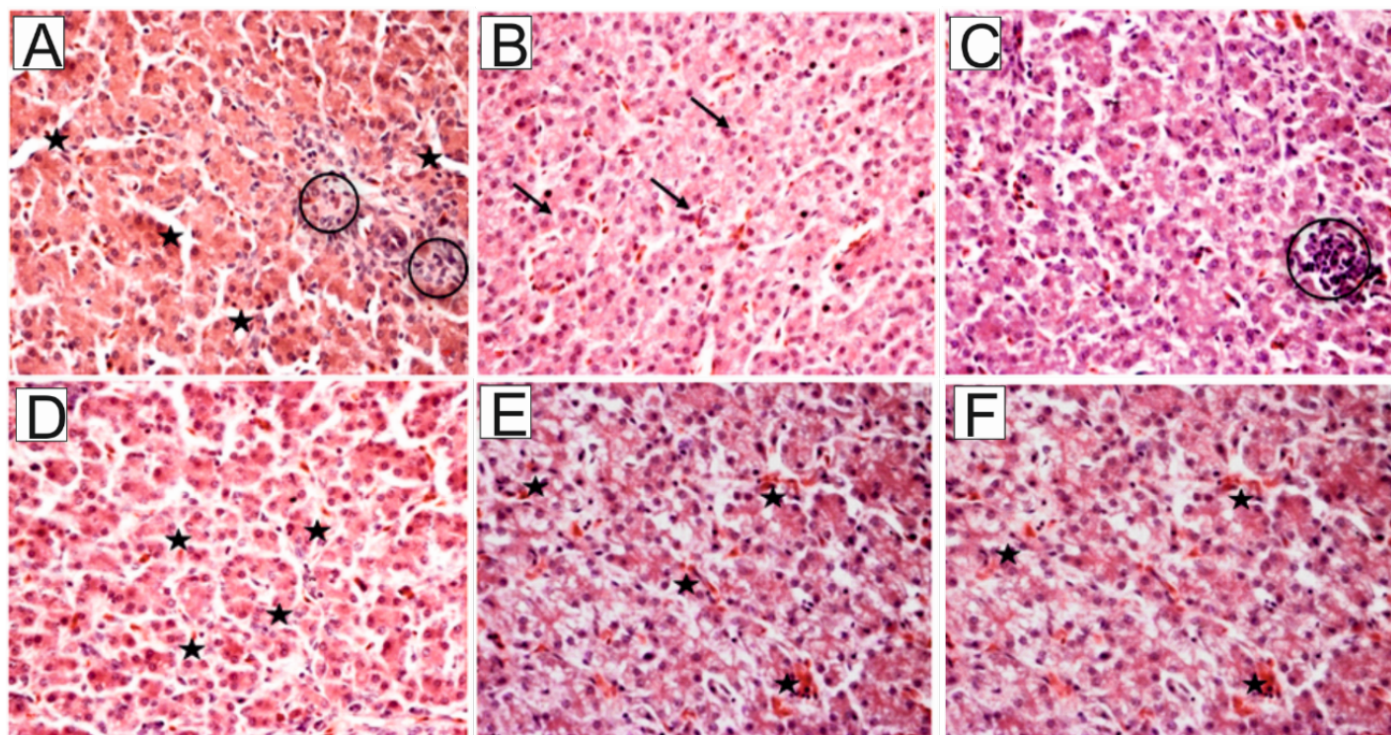


Figure 3: Influence of most effective medicinal plants and compounds on the histomorphological examination of liver in coccidiosis challenged broiler chickens.

A) Micrograph of Liver of coccidiosis challenged broiler without any supplementation (Control) showing degeneration with occasional hyperplasia of Kupffer cell (circle) and irregular change in Parenchyma of liver (star).

B) Micrograph of Liver of coccidiosis challenged broiler supplemented with hing herb showing low degree of perivascular mononuclear cell infiltration (circle).

C) Micrograph of Liver of coccidiosis challenged broiler supplemented with imli showing low degree of hepatocellular Kupffer cell degeneration (star).

D) Micrograph of Liver of coccidiosis challenged broiler supplemented with Alovera showing regeneration (arrows) of hepatocytes and Kupffer cell.

E) Micrograph of Liver of coccidiosis challenged broiler supplemented with mixture of herbs (Compound-I) showing low degree of hepatocellular Kupffer cell infiltration (star).

F) Micrograph of Liver of coccidiosis challenged broiler supplemented with mixture of herbs (Compound – II), showing less degeneration of hepatocytes & Kupffer cell (star).

in control group. Moreover, among all the herb supplemented groups of birds with few hepatocellular degenerative variations, inflammatory cell permeation was mellowed, and process of liver cell regeneration and normal architecture of liver was seen in broilers of supplemented groups contrast to birds in control group.

Herbs are being investigated on a large scale worldwide. Our laboratory has done several works on this and found beneficial herbs/plants which have therapeutic usage and also can help to increase production (Rajput *et al.*, 2012; 2013; Jahejo *et al.*, 2016a; 2016b; 2019). In this study, we have used different herbs/plants to investigate their effects on the performance of coccidiosis challenged broiler chickens. Furthermore, we have investigated the effect of those herbs on the histomorphology of liver.

In correlation with present findings Ashayerizadeh *et al.* (2009) reported that leaves, stem, seeds, roots and barks of medicinal plants contained active constituents had significant impact against various diseases and improved digestion need for increase in weight gain and carcass yield of the broiler. Mixed supplementation of capsicum and curcumin increased dressing percent, carcass weight and minimized lesion scores of gut in poultry contrast to infected birds fed on non-supplemented diet (Lee *et al.*, 2013; Rajput *et al.*, 2013). In accordance of present findings with several reports reported helpful consequences of curcumin herbs on weight gain and carcass yield of broiler (Kumari *et al.*, 2007).

In the current investigation in majority of case the relative weight of fat pad broiler supplemented with herbs was recorded high. In contrast, similar findings reported by Kumari *et al.* (2007), they reported that

the in non-LPS-treated birds had low weight of fat pad supplemented with curcumin was lower than that of control and lutein-supplemented birds, respectively. In addition, (Koutsos *et al.*, 2006) documented greater relative fat pad in broiler fed without lutein supplementation and those non-supplemented chicks suffered a severe systemic inflammatory response after LPS injection. In agreement with present findings Kim *et al.* (2013) stated that *Curcuma longa*, *Capsicum annuum* and *Lentinus edodes* herbs supplementation improved weight gain, weight of relative organs and serum antibody titers against proflin, reduced fecal oocysts shedding in coccidiosis challenged broilers contrast to control group of birds.

In the current investigation in majority of case the relative weight of organs; liver, pancreas, spleen, heart, duodenum, jejunum, ileum, thymus and ceaca of broiler supplemented with Compound-II, Compound-I, Vaghayani/Hing (*Ferulafoetida regel*), Gadami/Imli (*Tamarindus indica*), and Aloe vera (*Aloe barbadensis*) recorded markedly higher than control group. In agreement Lee *et al.* (2013) reported that cinnamaldehyde herbs increased spleen weight and formed lymphocyte propagation and triggered the activity of macrophages required for production of nitric oxide. Results of (Rajput *et al.*, 2013) correlated with the present results; they reported that relative weight of the thymus, bursa, duodenum, jejunum and ileum notably improved with the supplementation of curcumin in broiler contrast to control groups, though the weight of spleen was noted higher in control by authors. In contrast, Brenes *et al.* (2010) found no change in weight of spleen and in other organs of broiler supplemented with curcumin (phenolic compound). The restriction of herbal supplementation restricted in coccidiosis challenged group of birds will reduce the relative weight of organs. Nevertheless, turmeric modifies containing active ingredients may boostup the restoration process and/or revival of liver cells (Rahardja *et al.*, 2015).

In current investigation utilization of crude protein and fat was recorded noticeably at high level in coccidiosis challenged birds supplemented with Compound-II, Compound-I. In accordance Rajput *et al.* (2013) observed that the addition curcumin herb in basal diet of broiler noticeably improved the nutrient digestibility of crude protein, fat and Metabolizable energy when compared with non-supplemented control group of birds. In the current study positive

impact of herb supplementation on the utilization of nutrients in broiler chicken supported Giannenas *et al.* (2010) and (Viveros *et al.*, 2011) they reported that the supplementation of phenolic compounds present in various medicinal plants *i.e.* curcumin, Tabashir (*Bambusa arundinacea*), Amla (*Emblica officinalis*), Vaghayani/Hing (*Ferulafoetida regel*), Gadami/Imli (*Tamarindus indica*), Aloe vera (*Aloe barbadensis*) and Giloy/Satgullo (*Tinaspora cordifolia*) may reduce inflammation of gut, improved nutrients absorption and metabolic activity. These compounds may also have ability to alter the structure and function of gastrointestinal tract particularly at what time of growth-promoting antibiotics are banned in feed. Similarly, El Amin (2003) recorded high level of crude protein and crude fat digestibility, live body weight, improved FCR value and feed consumption with supplementation of garlic paste in diet of broiler.

According to Dragen *et al.* (2014), garlic plant extract administration increased the serum albumin, globulin, and total protein level due to garlic anti-inflammatory and immuno-modulatory action that refurbish the intestinal lesions produced due to Eimeria infection. However, in the present investigation, the low oocyst excretion rate, higher percentage of survival of infected birds, negligible signs of bloody diarrhea, minimum shedding of oocyst, and higher percent protection against caecal and intestinal lesions in medicinal plants supplemented groups of broiler chicken challenged with coccidiosis infection compared to control group of birds suggested the participation of some immune effectors present in the medicinal plants/herbs in shape of the tannins, phenols and phenolic acids, alkaloids, flavones, flavonoids, and flavonols is probably due to their ability to complex with extracellular and soluble proteins and to complex with parasite cell walls, more lipophilic flavonoids may also disrupt parasitic cell membranes that might inhibit the development of the parasites life cycle in the host (Kaleem *et al.*, 2014). Moreover, during coccidial infection, the cytokine metabolite environment, produced within the microenvironment of the bird's intestine, may lead to physiological alterations including vasodilation, which caused increased hemorrhagic lesions in severely infected negative control chickens (Habibi *et al.*, 2014).

The current investigation histomorphological examination of liver in coccidiosis challenged broiler chicken showed few pathological changes like the

noticeable propagation of poly-morphonuclear and mononuclear cells and activation of immune response in shape of activation of liver macrophage system for the repairing of the Kupffer cells, the size and shape of the liver seems to be normal compared to control group of broiler (Fed on free antibiotic and anticoccidial basal diet) showed extensive degeneration of hepatocytes with sporadic deformation of hepatic cords and hyperplasia of Kupffer cells. Similar trend of results for histomorphology of liver of herb supplemented groups of broilers challenged with coccidiosis infection against control infected group of birds was recorded by [Chand et al. \(2016\)](#). They reported beneficial effect of medicinal herbs on the liver of coccidiosis challenged birds; liver cells were regenerated, low degree of hepatocellular degeneration and diffuse infiltration of inflammatory cells and normal architecture of liver was observed compared to control infected group of birds. In accordance [Tabassum et al. \(2010\)](#) reported protective role of dandelion, curcumin, *Tamarindus indica* and *Ferulafoetida regel* medicinal herbs on the normal function, shape and health of liver in coccidiosis infected birds compared to non-supplemented infected broiler ([Rajput et al., 2013](#); [Gulfraz et al., 2014](#)). Likewise, in the current investigation, the better health and histomorphology of liver of medicinal plants supplemented groups of broiler challenged with coccidiosis infection contrast to control group might be due to the presence of bio-active compounds like A, C, thiamine and riboflavin vitamins, lactones, triterpenes, carotenoids, myristic fatty acids, apigenin and luteolin flavonoids in *Aloe vera* (*Aloe barbadensis*), *Gadamri/Imli* (*Tamarindus indica*), *Vaghayani/Hing* (*Ferulafoetida regel*) and in other medicinal plants herbs and their associated compounds, they enhanced the hepatic function and possessed antioxidant activity against coccidial agents ([A., 2011](#)).

Conclusions and Recommendations

In this study, it was evaluated that the performance and liver function (based on histopathology) of coccidiosis challenged broiler chickens improved with the supplementation of *Aloe barbadensis* (5ml/L), *Ferulafoetida regel* (500mg/L), *Tamarindusindica*(50mg/L). The best result of this experiment was obtained when supplemented the mixture of *Aloe barbadensis*, *Ferulafoetida regel*, and *Tamarindus indica* at the dose of 2ml/L. Future studies be conducted to find the effects of these herbs alone or in combination with *Ocimum*

basilicum on the growth performance of broiler chickens challenged by heat stress.

Novelty Statement

The first study which screened useful herbs and evaluated the effect of those herbs' supplementation on the performance and histomorphology of liver in coccidiosis challenged broiler chickens. Comparatively, the compound mixture of 3 herbs *Aloe barbadensis* and *Ferulafoetida regel*, *Tamarindusindica* increase the performance of coccidiosis challenged broiler chickens.

Author's Contribution

Nasir Rajput: Designed the experiment.

Ahmed Ali Moryani: Data collection, experimental work and wrote the article.

Muhammad Naeem, Atta Hussain Shah and Hidayatullah Soomro: Collected and analyzed the data. All authors read and approved the final manuscript.

Conflict of interest

All authors state that, there is no conflict of interest.

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