

# COMPARATIVE EFFICACY OF ANTICOCCIDIALS ON CLINICAL, GROSS AND HISTOPATHOLOGICAL CHANGES IN INDUCED BROILER CAECAL COCCIDIOSIS

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## ABSTRACT

For this study, 300 one day old Cobb-400 strain of broilers chicks were used to evaluate the comparative efficacy of commonly used anticoccidials. Fifty chicks each of T1, T2, T3 and T4 group were given Diclazuril (0.1 %), Salinomycin (12 %), Diclazuril (0.1 %) + Salinomycin (12 %) in shuttle programme and Maduramicin (1 %) in feed as coccidiostat, respectively. Other two groups, each with 50 chicks were kept as infected control (T5) and uninfected control (T6) without coccidiostat in feed. All the groups except T6 were given a challenge dose of 50,000 oocysts of *Eimeria tenella* on 22<sup>nd</sup> day of age. The highest mean qualitative faecal scores for T1 and T3, T2, T4 and T5 were  $2.4 \pm 0.25$ ,  $1.4 \pm 0.3$ ,  $1.6 \pm 0.3$  and  $4 \pm 0$ , respectively. Mean lesion score value of coccidiostat treated groups was in range of 1.10 to 1.70 ( $p < 0.05$ ) as compared to 2.25 in T5. The mean oocyst per gram and mean oocyst index revealed best efficacy of T4 and T2 groups followed by T1 and T3 groups. Observation suggests, Maduramicin and Salinomycin may be the better curative remedy against the caecal coccidiosis.

## INTRODUCTION

India is one of the largest and fastest poultry growing industry currently holding the third position in the world in egg production and poultry meat production has reached about 2.2 million tons per year (Maharana and Tewari, 2011). The Gujarat state is no more exception to this as the poultry population has increased by 12.39% from year 2007 to 2012 (Livestock census, 2012). Poultry meat is considered to be an important source of animal protein both in rural and urban areas owing to their relatively low fat and cholesterol contents than other meat (Umayya, 2014; Senapati *et al.*, 2015; Singh *et al.*, 2015). In intensive poultry farming, birds are exposed to several stress factors which enhance their susceptibility to various diseases that are already existing or emerging. Amongst the diseases, coccidiosis is a widely known, greatly studied and yet incompletely understood protozoan diseases of poultry. These protozoa inhabits the caeca and adjacent intestinal tissues causing a severe disease characterized by bleeding, high morbidity and mortality, low weight gain, emaciation and other signs attributed to coccidiosis (Muthamilselvan *et al.*, 2016).

Without the administration of anticoccidials, economic broiler production is inconceivable. In spite of advances in immunological, biotechnological and genetical methods, control of coccidiosis chiefly depends upon prophylactic chemotherapy with anticoccidial drugs (Maharana and Tewari, 2011; Witcombe and Smith, 2014). The introduction of ionophores anticoccidials has changed the ability to control

coccidiosis (Maharana and Tewari, 2011; Chapman *et al.*, 2010). However, the emergence of drug resistance in coccidia is a great problem which, in due course, may limits their use (Abbas *et al.*, 2012). Middle Gujarat being the major belt of avian production and considering the problems of drug resistant, the objective of the present investigation was to assess the comparative efficacy of coccidiostats on clinical, gross and histopathological changes in broilers by giving experimental infection of *E. tenella*. The knowledge of comparative efficacy of anticoccidials might be helpful for the proper selection of drug for prevention of most pathogenic caecal coccidiosis and better improvement in broiler production.

## MATERIALS AND METHODS

### Experimental design

Total of three hundred Cobb-400 broiler chicks of either sex were used for the study obtained from Venky India Limited, Mogar, Anand District of Gujarat. They were reared under coccidia-free conditions. Fifty chicks of T1, T2, T3 and T4 group were given Diclazuril (0.1 %), Salinomycin (12 %), Diclazuril (0.1 %) + Salinomycin (12 %) in shuttle programme (Diclazuril for initial three week followed by Salinomycin for last three week) and Maduramicin (1 %) at a dose rate of 100 gm, 50 gm, 100 + 50 gm and 50 gm per 100 Kg broiler feed as coccidiostat, respectively. One group of 50 chicks will be kept as infected control (T5) and another group of 50 chicks

will be kept as uninfected control (T6) without coccidiostat in feed. They were vaccinated against Marek' Disease, Ranikhet Disease and Gumboro Disease as per the schedule on first, seventh and fourteenth day, respectively. The whole experimental work was approved by the Institutional Animal Ethics Committee (IAEC), Veterinary College, Anand having CPCSEA Registration number 486/01/A/CPCSEA during the second meeting of the year 2011 with approval letter No. AAU/GVC/CPCSEA-IAEC/72/2011 dated 08/12/2011.

#### Faecal score

Randomly about 5-10 g of fresh faecal sample was collected from birds of each group in polythene bags and was transported fresh to the laboratory for further processing daily. Faecal score was made upon the qualitative observation of the appearance of the dropping and assigned score no. 0, 1, 2, 3 and 4 for normal dropping, slightly abnormal dropping, loose motion, dropping mixed with blood and fresh bloody diarrhoea, respectively. Faecal score was observed regularly for 11 days started from day 2 to day 12 post infection (Shameem *et al.*, 2010).

#### Lesion score

Lesion score described by Johnson and Reid (1970) according to severity of the intestinal/ caecal changes at the time of necropsy of birds infected with *Eimeria tenella* was followed. The lesions score were studied on third, fifth, seventh and ninth day post inoculation in battery trials as follows. Lesion score numbered as (0) for no gross lesions, (+1) for very few scattered petechiae on the caecal walls, no thickening of caecal wall, normal caecal contents present, (+2) for lesions more numerous with noticeable blood in the caecal contents, caecal wall was somewhat thickened, normal caecal contents present and (+3) for large amount of blood or caecal cores present, caecal walls greatly thickened, little faecal contents in the caeca and caecal wall greatly distended with blood or large caseous cores, faecal debris lacking or included in cores. Dead birds scored as +4.

#### Oocyst per gram (OPG)

It was done to know the number of oocysts passed in the faeces of each group of caged birds to observe the ability of a

drug to suppress oocyst production (Williams, 2001). Oocyst per gram was calculated regularly up to 9 days started from day 4 to day 12 post infection. The procedure followed for OPG was McMaster chamber method as described by Long and Rowell (1958).

#### Oocyst index

Oocyst index was determined by microscopic examination of mucosal scrapings from the caeca on day 7 post infection as per the method of Hilbrich (1978) with some modification. The oocyst index was graded as 0, 1, 2, 3, 4, 5 for oocysts per field as <1, 1-10, 11-20, 21-50, 51-100 and >100 oocysts, respectively.

#### Collection of tissue and histopathological examinations

Tissue samples from broilers died of coccidiosis and from two random sacrificed broiler birds in each group at 2, 3, 4, 5 and 6 weeks of age were examined for faecal score, lesion score and oocyst study. Caeca were collected in 10 % neutral formalin for histopathological study at Department of Veterinary Pathology. Tissue pieces of caeca preserved in 10 % neutral buffered formalin were processed by paraffin wax embedding method. Sections were cut at 4-5 micron thickness with the help of microtome and stained with Ehrlich's Haematoxylin and Eosin (H and E) method for examinations as described by Luna (1960). Typical lesions were photographed at different magnifications.

## RESULTS

### Clinical picture

Clinical symptoms such as, anorexia, unthriftiness, greenish or reddish diarrhoea, huddling, ruffled feathers, paleness of comb and wattles, anaemia were less prominent in treatment groups compared to infected control group.

### Gross lesions

The gross lesions observed in the caecum of the birds infected with *E. tenella* in T1 to T5 groups shown slight enlargement, distention with partially clotted or unclotted blood with reddish brown contents and exudate containing tissue debris on five day of post infection (dpi) (Figure 1), it was gradually reduced

**Table 1: Faecal score value (Mean  $\pm$  S.E.) in different treatment group from 2 to 12 dpi (n=5)**

Days Post Infection (dpi)	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Period Mean
2	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 <sup>s</sup>
3	2.20 $\pm$ 0.20	1.40 $\pm$ 0.24	1.20 $\pm$ 0.20	1.40 $\pm$ 0.24	3.40 $\pm$ 0.24	1.92 <sup>b</sup>
4	2.40 $\pm$ 0.24	1.60 $\pm$ 0.24	2.40 $\pm$ 0.24	1.60 $\pm$ 0.24	4.00 $\pm$ 0.00	2.40 <sup>a</sup>
5	2.40 $\pm$ 0.24	1.40 $\pm$ 0.24	2.40 $\pm$ 0.24	1.20 $\pm$ 0.20	4.00 $\pm$ 0.00	2.28 <sup>a</sup>
6	2.20 $\pm$ 0.20	1.60 $\pm$ 0.24	1.80 $\pm$ 0.20	1.00 $\pm$ 0.00	3.40 $\pm$ 0.24	2.00 <sup>b</sup>
7	1.20 $\pm$ 0.20	0.80 $\pm$ 0.20	1.20 $\pm$ 0.20	0.40 $\pm$ 0.24	3.40 $\pm$ 0.24	1.40 <sup>c</sup>
8	0.40 $\pm$ 0.24	0.60 $\pm$ 0.24	0.60 $\pm$ 0.24	0.60 $\pm$ 0.24	3.20 $\pm$ 0.20	1.08 <sup>d</sup>
9	0.40 $\pm$ 0.24	0.40 $\pm$ 0.24	0.60 $\pm$ 0.24	0.80 $\pm$ 0.20	2.60 $\pm$ 0.24	0.96 <sup>de</sup>
10	0.40 $\pm$ 0.24	0.60 $\pm$ 0.24	0.40 $\pm$ 0.24	0.20 $\pm$ 0.20	2.40 $\pm$ 0.24	0.80 <sup>e</sup>
11	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	1.80 $\pm$ 0.20	0.36 <sup>f</sup>
12	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	0.00 $\pm$ 0.00	1.20 $\pm$ 0.20	0.24 <sup>fg</sup>
Treatment mean	1.06 <sup>b</sup>	0.77 <sup>c</sup>	0.96 <sup>b</sup>	0.66 <sup>c</sup>	2.67 <sup>a</sup>	
	T	C.D	P	C.D	T $\times$ P	C.D
	S Em	C.D	S Em	C.D	S Em	C.D
	0.06	NS	0.09	0.24	0.19	0.54

The means bearing different superscript within same row differ significantly from each other (P < 0.05)

**Table 2: Lesion score value (Mean  $\pm$  S.E.) in different treatment group from 3, 5, 7 and 9 dpi (n = 5)**

Days Post Infection (dpi)	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Period Mean
3	2.40 <sup>a</sup> $\pm$ 0.24	1.60 <sup>a</sup> $\pm$ 0.24	2.20 <sup>a</sup> $\pm$ 0.20	1.20 <sup>a</sup> $\pm$ 0.20	2.40 <sup>a</sup> $\pm$ 0.24	1.96 <sup>a</sup>
5	1.80 <sup>a</sup> $\pm$ 0.20	1.60 <sup>a</sup> $\pm$ 0.24	1.80 <sup>a</sup> $\pm$ 0.20	1.80 <sup>a</sup> $\pm$ 0.20	2.40 <sup>a</sup> $\pm$ 0.24	1.88 <sup>a</sup>
7	1.40 <sup>a</sup> $\pm$ 0.24	1.20 <sup>a</sup> $\pm$ 0.20	1.40 <sup>a</sup> $\pm$ 0.24	1.00 <sup>a</sup> $\pm$ 0.00	2.40 <sup>a</sup> $\pm$ 0.24	1.48 <sup>b</sup>
9	1.20 <sup>a</sup> $\pm$ 0.20	0.60 <sup>a</sup> $\pm$ 0.24	0.80 <sup>a</sup> $\pm$ 0.20	0.40 <sup>a</sup> $\pm$ 0.24	1.80 <sup>a</sup> $\pm$ 0.20	0.96 <sup>c</sup>
Treatment mean	1.70 <sup>b</sup>	1.25 <sup>cd</sup>	1.55 <sup>bc</sup>	1.10 <sup>d</sup>	2.25 <sup>a</sup>	
T			P		T $\times$ P	
S Em		C.D	S Em	C.D	S Em	C.D
	0.11	0.31	0.1	0.28	0.22	NS

The means bearing different superscript within same row differ significantly from each other ( $P < 0.05$ ).

**Table 3: Oocyst per gram value (Mean  $\pm$  S.E.) in different treatment group from 4 to 12 dpi (n = 10)**

Days Post Infection (dpi)	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	Period Mean
4	10690.50 <sup>c</sup> $\pm$ 312.06	8728.80 <sup>d</sup> $\pm$ 134.68	11069.00 <sup>b</sup> $\pm$ 211.78	8790.80 <sup>d</sup> $\pm$ 134.91	13565.68 <sup>a</sup> $\pm$ 265.23	10568.94 <sup>f</sup>
5	13509.40 <sup>b</sup> $\pm$ 245.35	10943.00 <sup>d</sup> $\pm$ 221.97	12848.00 <sup>c</sup> $\pm$ 164.26	10909.10 <sup>d</sup> $\pm$ 167.62	20318.40 <sup>a</sup> $\pm$ 97.42	13705.58 <sup>e</sup>
6	17014.10 <sup>c</sup> $\pm$ 371.93	13912.30 <sup>d</sup> $\pm$ 214.74	18103.60 <sup>b</sup> $\pm$ 220.57	12725.40 <sup>e</sup> $\pm$ 188.39	21970.90 <sup>a</sup> $\pm$ 249.91	16745.26 <sup>c</sup>
7	22227.70 <sup>b</sup> $\pm$ 204.65	19927.90 <sup>c</sup> $\pm$ 182.69	19056.50 <sup>d</sup> $\pm$ 605.72	10922.00 <sup>e</sup> $\pm$ 168.99	22539.50 <sup>a</sup> $\pm$ 171.69	18934.72 <sup>a</sup>
8	22227.90 <sup>b</sup> $\pm$ 259.77	15445.60 <sup>d</sup> $\pm$ 154.51	19788.90 <sup>c</sup> $\pm$ 137.71	10679.90 <sup>e</sup> $\pm$ 125.37	23247.40 <sup>a</sup> $\pm$ 212.16	18277.94 <sup>b</sup>
9	18951.40 <sup>a</sup> $\pm$ 546.74	14237.80 <sup>d</sup> $\pm$ 124.48	17425.40 <sup>c</sup> $\pm$ 244.15	8568.90 <sup>e</sup> $\pm$ 71.03	18595.70 <sup>b</sup> $\pm$ 34.14	15555.84 <sup>d</sup>
10	9768.80 <sup>c</sup> $\pm$ 50.04	8575.00 <sup>d</sup> $\pm$ 70.24	10541.40 <sup>b</sup> $\pm$ 124.21	6474.40 <sup>e</sup> $\pm$ 123.59	17595.10 <sup>a</sup> $\pm$ 91.54	10591.54 <sup>f</sup>
11	8599.70 <sup>c</sup> $\pm$ 56.92	7324.00 <sup>d</sup> $\pm$ 131.39	9415.30 <sup>a</sup> $\pm$ 177.94	5537.30 <sup>e</sup> $\pm$ 90.51	16861.80 <sup>a</sup> $\pm$ 256.62	9548.02 <sup>g</sup>
12	6393.30 <sup>b</sup> $\pm$ 78.38	5403.50 <sup>c</sup> $\pm$ 76.14	6441.00 <sup>b</sup> $\pm$ 121.38	4675.20 <sup>d</sup> $\pm$ 111.02	14358.60 <sup>a</sup> $\pm$ 81.69	7454.32 <sup>h</sup>
Treat mean	14375.87 <sup>b</sup>	11611.10 <sup>d</sup>	13854.35 <sup>c</sup>	8809.23 <sup>e</sup>	18784.00 <sup>a</sup>	
T		P	T $\times$ P			
S Em		C.D	S Em	C.D	S Em	C.D
	2.40	70.69	1.96	94.84	1.47	212.07

The means bearing different superscript within same row differ significantly from each other ( $P < 0.05$ ).



**Figure 1: Photograph showing blood filled and opened ceaca with haemorrhagic blood clots**

after seven dpi and was very mild on nine dpi onwards whereas in T<sub>5</sub>, caecal wall was greatly thickened because of edema and cellular infiltration with the formation of scar tissue.

#### Comparative efficacy of coccidiostats on faecal score

Qualitative faecal score was made upon the observation of the appearance of the dropping from normal values as given in Table 1. In T<sub>1</sub> group mean value ranged from 0 to 2.4 between two to twelve dpi. Highest mean value  $2.4 \pm 0.25$  was observed in T<sub>1</sub> and T<sub>3</sub> group on 4<sup>th</sup> and 5<sup>th</sup> dpi among coccidiostats treatment groups, while on 3<sup>rd</sup> dpi less faecal score ( $1.2 \pm 0.2$ ) was observed in T<sub>3</sub> group as compare to T<sub>1</sub> group ( $2.2 \pm 0.2$ ). In T<sub>2</sub> and T<sub>4</sub> group similar mean value 1.4

$\pm 0.3$  and  $1.6 \pm 0.3$  were observed on 3<sup>rd</sup> and 4<sup>th</sup> dpi, while non significant lower mean value were observed in T<sub>4</sub> group as compared to T<sub>2</sub> group on 5 to 7 dpi. In T<sub>5</sub>, mean faecal score ranged from 0 to 4.0 with highest mean value ( $4 \pm 0$ ) being observed on 4<sup>th</sup> and 5<sup>th</sup> dpi.

#### Comparative efficacy of coccidiostats on lesion score

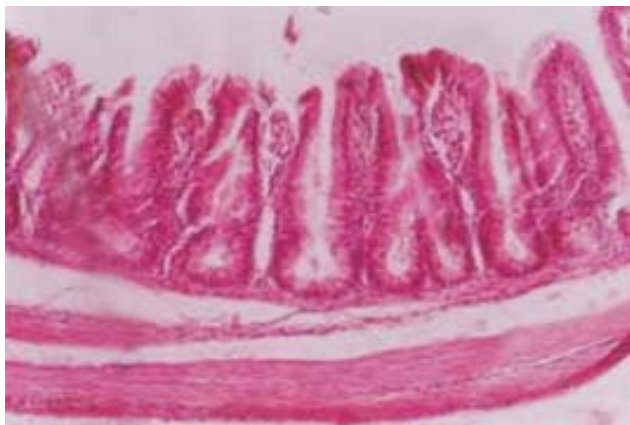
Observed lesion scores are presented in Table-2. In the present study the mean lesion score value of *E. tenella* infected and coccidiostat given group was in range of 1.10 to 1.70, which was significantly low ( $p < 0.05$ ) as compared to 2.25 in infected non medicated birds. Statistically all treatment groups were showing significant lower lesion score as compare to infected control.

#### Comparative efficacy of coccidiostats on oocysts per gram (OPG)

The efficacy of coccidiostats was evaluated based on their ability to suppress the oocyst production. The mean OPG of faeces excreted in different five groups experimentally infected with *E. tenella* on different days of infection are presented in Table 3. The overall treatment mean results indicates best efficacy of Maduramicin and Salinomycin in OPG reduction followed by Diclazuril and Diclazuril + Salinomycin. The overall trend of period mean showing increase in OPG count was observed up to seven dpi and there after decreasing trend was observed up to 12 dpi.

#### Comparative efficacy of coccidiostats on oocyst index

The Oocyst index was studied on seventh dpi. It was 0, 1, 2, 2, 3 in Maduramicin group, 0, 3, 3, 2, 1 in Salinomycin group, 2, 0, 3, 4, 4 in Diclazuril group, 2, 2, 3, 3, 4 in Shuttle group and



**Figure 2:** Normal caecal section showing mucosa makes up about two thirds of the total thickness of the wall containing well developed villi and lymphoid cells in the lamina propriety at 3 dpi in T6 group bird (100x).

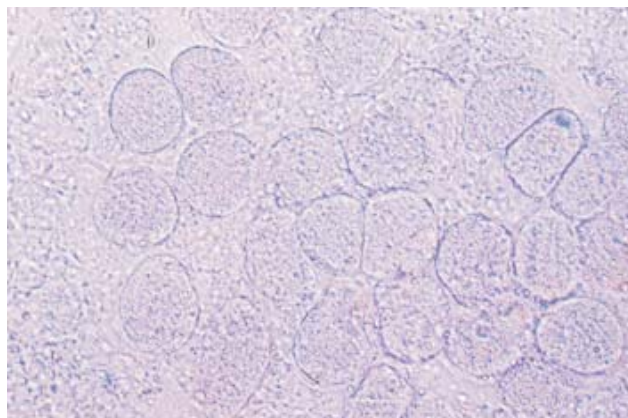


**Figure 3:** Caecal section showing cellular infiltration in lamina propria and desquamation of the epithelial lining of the vill containing schizonts at 3 dpi in T5 group bird (100x).



**Figure 4:** Caecal section showing desquamation of the epithelium with edema and cellular infiltration of lymphocytes in the mucous and submucous layers at 3 dpi in T4 group bird (100x)

3, 4, 4, 4, 5 in positive control group. Average oocyst index was highest in infected non-medicated control (4.0).



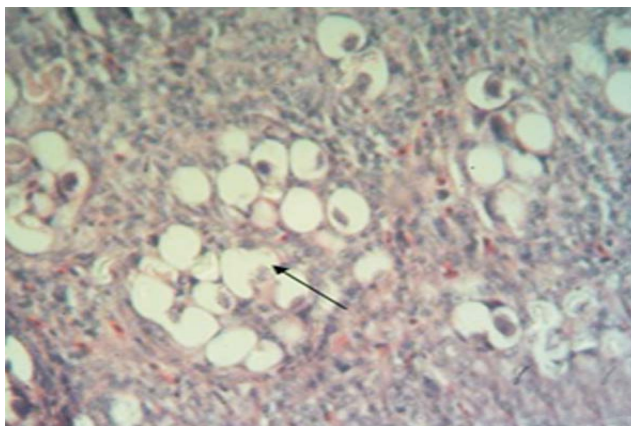
**Figure 5:** Caecal section showing development of large second stage schizont in haemorrhagic mucosal smear with presence of blood Schizont at 5 dpi in T2 group bird (100x)

Maduramicin and Salinomycin groups have lower oocyst index as compared to Diclazuril and Diclazuril + Salinomycin Shuttle group.

#### Microscopic lesions

The histological examination of normal caeca did not show any changes and had the normal histological structure (Figure 2). On 3<sup>rd</sup> dpi, caeca of group T5 showed cellular infiltration in lamina propria and desquamation of the epithelial lining of the villi. The edema of submucosa and muscular layers of caecum with more number of schizonts in the epithelial cells of villi were observed. Due to developing schizonts, the epithelial cells ruptured, resulted in congestion and hemorrhages, which were seen in the mucosa, submucosa and muscular layers (Figure 3). Similar histopathological lesions were observed in T1, T2 and T3. While in group T4, similar histopathological lesion with less intensity were noticed. (Figure 4).

On 5<sup>th</sup> dpi, caeca of group T1 and T5 showed number of second stage schizont with numerous inflammatory cells especially lymphocytes in the disrupted mucosal smear. In T2 group, there was development of large second stage schizont in haemorrhagic mucosal smear with presence of blood cells (Figure 5). In T3 group, there was cluster of large schizont with severe tissue damage with lymphocytic infiltration (Figure 6), while in T4 group less severe haemorrhage was observed in sub mucosal layer with mild destruction of caecal epithelium. At seven dpi, the caecum revealed a large number of second generation schizonts packed with merozoites and liberating of merozoites along with the presence of developing oocysts were observed in T1 and T5 groups. In addition, congestion of vessels, multifocal areas of haemorrhages and leukocyte infiltration predominately heterophils with, degeneration and desquamation of crypt epithelium were also noticed. The T2 group showed similar histopathological lesions with reduced intensity, along with few developing schizonts and merozoites in the crypt epithelium. However, hyperplastic changes in crypt epithelium characterized by regenerating epithelium with lymphoblast and numerous mitotic bodies were noticed on 7<sup>th</sup> dpi. In T3 group there was cluster of large developed oocyst in the caecal crypts with number of inflammatory cells.



**Figure 6:** Caecal section showing cluster of large schizont with severe tissue damage and lymphocytic infiltration at 5 dpi in T3 group bird (40x)



**Figure 7:** Caecal section showing regenerating changes of connective tissue proliferation in submucosa and muscular layer with cellular infiltration and hyperplasia of lymphoid follicles at 10 dpi in T1 group bird (400 x)

While group T4 showed infiltration of lymphocytes and fibroblasts, hyperplasia of mucosal glands. Numbers of glandular cells were observed in some places, with slight destruction of epithelial cell. On 10<sup>th</sup> dpi, caeca of group T5 showed development of numerous oocysts in the distended caecal crypts surrounded by leucocytic infiltration. T1 group showed connective tissue proliferation in submucosa and muscular layer and cellular infiltrations were also observed. Hyperplasia of lymphoid follicles and the commencement of tissue regeneration were observed (Figure 7). The group T2 showed regeneration of caecal mucosa, hyperplasia of lymphoid follicles with oocyst, while group T3 showed degeneration of gametocytes with destructed oocyst structures and cellular infiltration in the lamina propria, while in group T4 showed regeneration of caecal mucosa. No oocysts could be detected in the epithelial cells.

## DISCUSSION

Gross findings, Faecal score, lesion score, oocyst output and oocyst index values are helpful for knowing the intensity of infection and degree of pathological damage in the intestine.

Results of gross findings are in accordance with other studies on caecal coccidiosis. However, these lesions were mild in anticoccidial group T1 to T4 with variation in the intensity of gross pathological changes. Less intensity necropsy lesion were observed in T2 and T4 group (Misra and Goutham, 1970; Clarke, 1979; Mc Dougal and Reid, 1991; Soomro *et al.*, 2001).

All faecal score mean value of treatment group was significantly lower as compared to infected control group (T5) with lowest faecal score mean was observed in T4 group followed by T2 group indicating better efficacy of Maduramicin and Salinomycin. In the present study the mean lesion score value of *E. tenella* infected and coccidiostat given group was in range of 1.10 to 1.70, which was significantly lowered as compared to 2.25 in infected non medicated birds. Statistically all treatment groups showed significant lower lesion score as compared to infected control. The lowest treatment means values 1.10 was in Maduramicin group followed by Salinomycin group 1.25 indicating better efficacy of Maduramicin and Salinomycin. This observations are in accordance with the reports by the earlier workers (Majumdar *et al.*, 1993; Rana and Tikaram, 2002; Abbas *et al.*, 2008; Raju *et al.*, 2012; Shameem *et al.*, 2010; Anish *et al.*, 2007). Reduction in the faecal score values is due to the suppression of schizogony process by the coccidiostats resulting in less intestinal damage as compare to infected non treated control.

Similarly, Maduramicin and Salinomycin treated birds also showed lowered oocysts production which was more effective than Diclazuril and Diclazuril+Salinomycin groups. Our findings are in conformity with previous observations by various workers (Salisch and Shakshouk, 1990; Abbas *et al.*, 2008; Azizi *et al.*, 2010). The Oocyst index was studied on seventh day post infection which indicates better efficacy of Maduramicin and Salinomycin. Mzurkiewicz *et al.* (1987) studied the similar effect of ionophore coccidiostats against *E. tenella* and *E. acervulina* infections and noted best oocyst index for Maduramicin followed by Lasalocid, Narasin, Salinomycin and Monensin. Georgieva *et al.* (2010) reported that Maduramicin at 5 ppm dose rate found to reduce oocyst index and lesion score at 7 dpi given mixed 80000 *Eimeria spp.* oocysts. These results are in agreement with our study in which Maduramicin fed group shown lowest oocyst index, which might be due to beneficial effects of Maduramicin on lipid peroxidation, reducing oxidative stress as reported by Georgieva *et al.* (2010).

Histopathology is very much important to differentiate the abnormalities between normal and healthy structure at microscopic level. It is concerned with the demonstration of minute alterations in tissue structures in diseased conditions (Culling, 1963). Gross and microscopic pathology were specifically used to demonstrate the severity of the disease in chickens infected with *E. tenella*. The presence of high numbers of oocysts, schizonts and severe tissue damage in the caeca indicated the severity of *E. tenella* infection. The major histopathological changes observed in caeca of infected chicks in the group T5 are due to infection of *E. tenella* pathogen revealed marked degeneration, desquamation of superficial epithelium, leucocyte infiltration in submucosa, denudation of villi and hemorrhages. The tissue contained various stages of schizonts. These observations are in agreement with the

earlier reports by various workers (Mc Dougald and Reid, 1991; Jaipurkar *et al.*, 2004). But there was less severe histopathological lesion in infected but treated groups. In group T2 and T4, the regeneration of caecal mucosa is observed, while in group T1 and T3 most of the part of caecal mucosa is seen affected due to parasite. There were no oocysts of *E. tenella* in the epithelial cells in T4 and T2 groups and recovery of caecal tissue was observed during experimentation. This might be attributed to the fact that ionophores cause marked inhibition of asexual development by reducing viability and infectivity of sporozoites at 3rd dpi (Smith *et al.*, 1981; Long and Jeffers, 1982; Chapman, 2010; Kant *et al.*, 2013) and it was found active against sporozoites and early schizogenous stages (Conway *et al.*, 1999; Smith *et al.*, 1981). Chappel (1979) also reported the similar efficacy of Salinomycin. These finding however, correspondingly related to those recorded by Babu *et al.* (1976), Soomro *et al.* (2001), Lakkundi *et al.* (2002), Dauton Luiz Zulpo *et al.* (2007), Ogbe, *et al.* (2008), Siddiki, *et al.* (2008), and Chandrakesan, *et al.* (2009) in different drugs and infection. In the present study, early recovery was also indicated by the presence of regenerative changes on the 10<sup>th</sup> dpi in anticoccidial treated group compared to infection control group.

From this study it is concluded that, the Maduramicin and Salinomycin treated group showed very less mechanical damage to tissue hence these can be used as a curative remedy against caecal coccidiosis. However, further studies are requisite to ascertain the effectiveness and economical impact of the anticoccidial usage in the poultry operation.

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