

## EVALUATION OF CLINICAL STATUS AND TREATMENT TRIALS IN SHEEP AND GOATS INFESTED WITH *EIMERIA* SPECIES

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### ABSTRACT

One hundred forty eight specimens were gathered from sheep and goats, to be inspected for the presence of *Eimeria* species by fecal analysis. The infection rate was 65.3%, 80%, 63.6% and 85.7 in sheep, goats, lambs and kids respectively. In histopathological examination *Eimeria* and oocysts were distinguished in the villi of epithelium, crypts and lieberkuhn glands. Eighteen animals were deliberately infested and classified into three groups according to the used anticoccidial drugs to estimate their efficacy. Hematological and biochemical analysis were done before and after treatment trials in all animals. In case of sheep, the sulphadimidine (33.3%) was more effective than amprolium and diclazuril while, the diclazuril has a better effect than amprolium and sulphadimidine (33.3%) in case of goats. All treated animals showed improved results in the hemogram and biochemical profile in comparison to untreated animals.

**Key words:** *Eimeria*, anticoccidial drugs.

### INTRODUCTION

Coccidiosis affect most domestic animals including ovine, caprine, canine, feline, equine, porcine, bovine, lagomorphs and avian, inducing a serious condition on general health status (Coetzer and Justin, 2004). The pervasiveness of *Eimeria* in sheep and goats is high that could be one hundred percentage in the flock. All *Eimeria* species share a similar monoxenous life cycle with an internal parasitic and external environmental phase that developed within cells at certain sites of the intestinal mucosa (Arslan *et al.*, 1999).

The best control methods are to avoid the overcrowdings of animals, the reduction of stressors, an adequate nutrition and the use of anticoccidial drugs (Radostits *et al.*, 2000). Anticoccidial drugs could be used to prevent or minimize the outcomes of the clinical cases (Foreyt, 1990; Taylor and Catchpole, 1994). Routine prophylactic medication in the feed and water supplies would usually control the

disease and allow the development of effective immunity especially in young lambs (Radostits *et al.*, 2000). Anticoccidial drugs that used in control and prevention of coccidiosis include sulphanilamide derivatives, monensin, lasalocid ionophorous antibiotics, amprolium, toltrazuril and diclarzuril (Platzer *et al.*, 2005 and Taylor *et al.*, 2003).

### MATERIALS AND METHODS

#### Clinical examination

One hundred forty eight samples were examined for presence of *Eimeria* species and specimens were gathered from all animals (95 sample from adult sheep, 11 samples from lambs, 35 samples from adult goats and 7 samples from young kids (all animals suffered from diarrhea), from different places in Qena province. Clinical examinations were recorded with especial reference to presences of diarrhea, body condition and mucous membrane examination using Famacha chart (Radostits *et al.*, 2000). Eighteen infected adult animals with *Eimeria* were selected from farm of faculty of veterinary medicine, South Valley University for exposure to anticoccidial drugs, these animals were divided into 3 groups, and each group contains 6 adult animals (3 sheep and 3 goats).

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### Fecal samples

Fecal samples were gathered either directly from anus or from barns from different places in Qena province (from small farms and private). Ninety five samples from adult sheep, 11 samples from lambs, 35 samples from adult goats and 7 samples from young kids.

### Blood samples

Thirty six samples were collected as a whole blood samples from 18 selected animals with interval 14 days pre and post treatment with anticoccidial drugs. For biochemical analyses 54 serum samples were gathered in two occasions in the third and fourteenth day post treatment.

### Histopathological examination

Specimens were settled in 10% formalin buffer, then the paraffin blocks were primed and five micrometer thickness section were made for histopathological examination, then stained using haematoxylin and eosin (Drury and Wallington, 1980).

### Statistical analysis

Sigma stat software SPSS was utilized to lead a two-path investigation of variance ANOVA and Student's t test to calculate the significance level between and among various treatment groups to test the effects of drug on the oocyst per gram of feces and on body weight. Results were communicated as mean  $\pm$  standard error at P = 0.05.

### Anticoccidial drugs

Three anticoccidial drugs used to treat the experimental animals; sulphadimidine 33.3% powder (100mg/ kg. B.W. given orally daily for 4 days - VETWIC. El Nasr pharmaceutical Co.), amprolium 20% powder (50mg /kg. B.W given orally daily for 4 days - ADWIA Co. S.A.E) and diclazuril 1% solution (1mg/kg. B.W given orally daily for 4 days -Diclosol. Pharma SWEDE- Egypt).

## RESULTS

### Rate of *Eimeria* infection

The rates of *Eimeria* infection by fecal examination for 148 animals revealed that lambs had a lower infection rate (63.6%) than adult sheep (65.3%), on the other side; kids had a higher infection rate (85.7%) than adult goats (80%) (Table 1).

### The efficacy of anticoccidial drugs

Using of sulphadimidine in treatment of coccidiosis, induced a gradual decrease in oocyst count with an efficacy 97.6% and 85.3% in sheep and goats respectively, while amprolium has an efficacy 95.6% and 87.6% in sheep and goats respectively but, there is a sharp decrease in oocyst count with an efficacy 95.2% and 100% in sheep and goat respectively in case of using of diclazuril.

### Hematological and biochemical parameters

Hematological analyses were evaluated for RBCs, WBCs and Hb. The previous parameters were detected two times, one time was done pretreatment with anticoccidial drugs and the second time was done post treatment by 14 days (Table 2). Haematological analyses showed reduction in RBCs and Hb in diseased animals more than treated, but the leukocytes number was significantly decreased in post-treated sheep and goat than pretreated. Biochemical parameters were done three times, one time was pretreatment and the second and third times were post treatment. The biochemical analysis revealed that an increment in the total protein, albumin, globulin and creatinine in the post treated animals more than diseased ones, there were some alterations in the level of serum ALT and AST (Table 3, 4).

### Clinical and post mortem examination

Clinical examination were done for 148 animals which suffered from a pale mucus membrane of eye, emaciation, lambs and kids suffered from pasted perineal region with watery diarrhea, dehydration and poor growth rate (Photo 1 A, B). Post mortem examination was done for one animal died and the lesions were thickness in the mucosa associated with scattered multifocal hemorrhagic areas in the mucosal surface of the intestine. The lesions observed mostly in the distal part of the jejunum, ileum and to lesser extent in the cecum and proximal colon (Photo 1 C).

### Histopathological examination

Histopathological examination revealed inflammatory reaction mainly infiltration of plasma cells, lymphocytes and eosinophils in the lamina propria. The formative phases of *Eimeria* were seen in the epithelium of influenced villi, crypts and Lieberkuhn glands. Oocysts were oval and had two layered walls which were seen in the mucosa and occasionally in necrotic-hemorrhagic exudates in the intestinal lumen (Photo 1 D).

**Table 1:** Infection rate of *Eimeria* species among all examined animals.

Animal species		No. of examined animals	No. of infected animals	%
Sheep	Female	75	50	66.6
	Male	20	12	60
<b>Total adult sheep</b>		95	62	<b>65.3</b>
Goats	Female	22	18	81.8
	Male	13	10	76.9
<b>Total adult goats</b>		35	28	<b>80</b>
Lambs	Female	3	0	0
	Male	8	7	87.5
<b>Total lambs</b>		11	7	<b>63.6</b>
Kids	Female	2	2	100
	Male	5	4	80
<b>Total kids</b>		7	6	<b>85.7</b>
<b>Total</b>		<b>148</b>	<b>103</b>	<b>69.6</b>

**Table 2:** Hematological parameters for pre and post treatment of selected sheep and goat.

Parameters	Group A				Group B				Group C			
	Sheep		Goat		Sheep		Goat		Sheep		Goat	
	Pre	Post										
RBCs $\times 10^6/\text{mm}^3$	9.84 $\pm 0.38$	10.95 $\pm 1.36^*$	13.52 $\pm 2.88$	15.49 $\pm 2.95^*$	11.16 $\pm 2.34$	12.57 $\pm 1.95^*$	12.23 $\pm 2.48$	16.43 $\pm 1.74^*$	10.05 $\pm 3.21$	12.25 $\pm 1.83^*$	18.50 $\pm 1.32$	18.80 $\pm 1.82$
WBCs $\times 10^3/\text{mm}^3$	10.58 $\pm 5.28$	8.45 $\pm 2.89^*$	6.56 $\pm 2.35$	8.58 $\pm 1.21^*$	7.41 $\pm 1.58$	10.46 $\pm 2.38^*$	8.45 $\pm 1.34$	7.98 $\pm 2.55$	12.26 $\pm 5.50$	9.05 $\pm 2.29^*$	9.60 $\pm 4.16$	6.95 $\pm 2.45^*$
Hb. g/dl	10.5 $\pm 1.35$	12.4 $\pm 1.60^*$	9.8 $\pm 0.45$	10.3 $\pm 0.95^*$	11.1 $\pm 1.21$	12.63 $\pm 2.10^*$	10.36 $\pm 0.32$	10.5 $\pm 0.1^*$	10.78 $\pm 2.56$	9.33 $\pm 0.40^*$	13.6 $\pm 0.98$	11.58 $\pm 0.92^*$

\* Significant different from mean value at  $P < 0.05$ 

RBCs: Red Blood Cell Count

WBCs: White Blood Cell Count

Hb: Hemoglobin Concentration

**Table 3:** Biochemical parameters in pre and post treatment of sheep

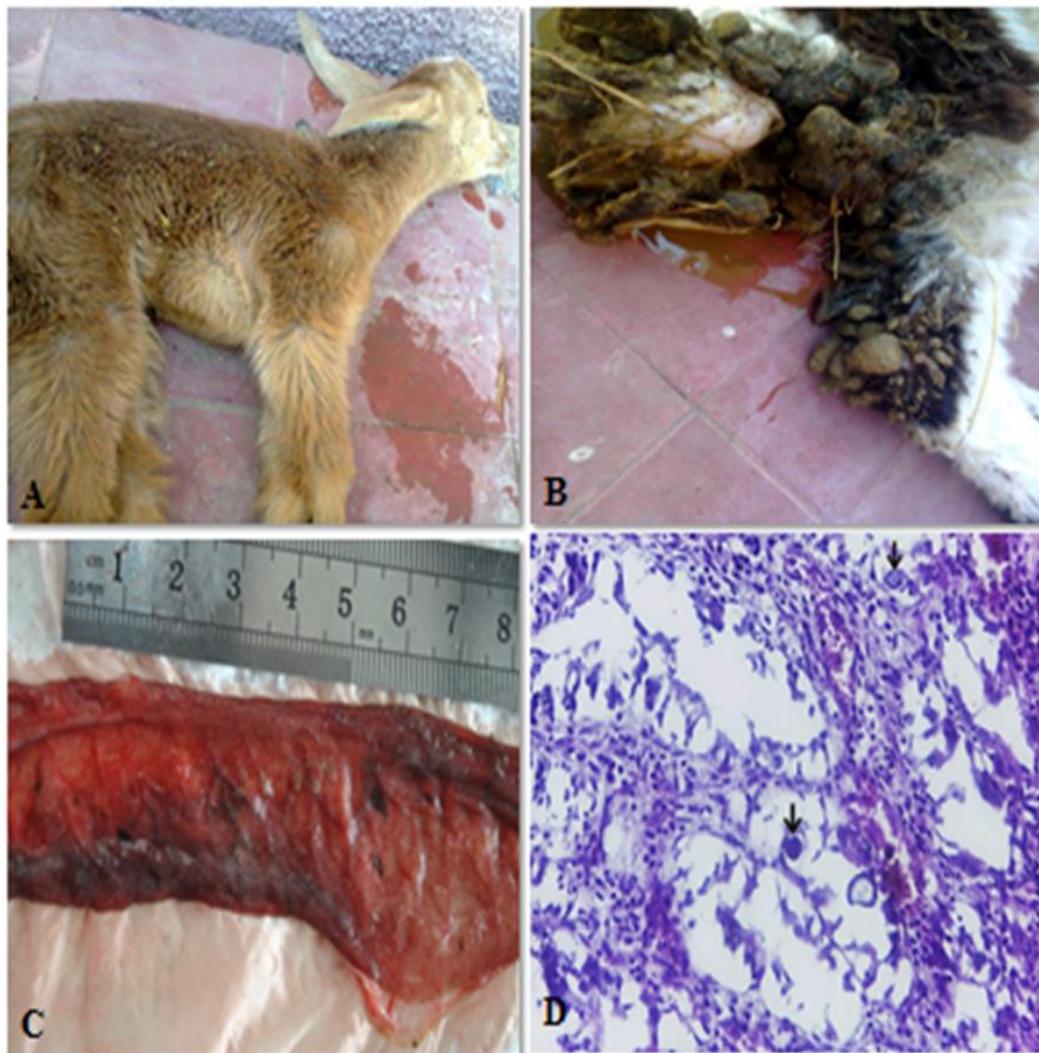
Animal	Group A			Group B			Group C		
	Pre	Sheep		Pre	Sheep		Pre	Sheep	
		3 <sup>rd</sup> day	14 <sup>th</sup> day		3 <sup>rd</sup> day	14 <sup>th</sup> day		3 <sup>rd</sup> day	14 <sup>th</sup> day
Total protein g/dl	5.9 $\pm 1.9$	5.5 $\pm 1.1$	5.5 $\pm 1.1$	5.5 $\pm 1.1$	5.2 $\pm 1.2$	5.8 $\pm 0.2^*$	5.2 $\pm 1.2$	5.2 $\pm 1.2^*$	5.1 $\pm 1.2^*$
Albumin g/dl	3.5 $\pm 0.3$	3.3 $\pm 0.2$	3.3 $\pm 0.2$	3.3 $\pm 0.2$	3.4 $\pm 0.3$	3.4 $\pm 0.3$	3.4 $\pm 0.3$	3.6 $\pm 0.4$	3.7 $\pm 0.0^*$
Globulin g/dl	2.4 $\pm 2.2$	2.2 $\pm 0.9$	2.2 $\pm 0.9$	2.2 $\pm 0.9$	1.8 $\pm 1.2$	2.3 $\pm 0.4^*$	1.8 $\pm 1.2$	1.6 $\pm 1^*$	1.5 $\pm 1.2^*$
Creatinine mg/dl	0.75 $\pm 0.3$	0.9 $\pm 0.2$	0.9 $\pm 0.2$	0.9 $\pm 0.2$	1.4 $\pm 0.4$	1.6 $\pm 0.3$	1.4 $\pm 0.4$	0.7 $\pm 0.2^*$	0.6 $\pm 0.2^*$
AST U/L	23.9 $\pm 9.6$	27.2 $\pm 21.9$	27.2 $\pm 21.9$	27.2 $\pm 21.9$	22.2 $\pm 16.3$	60.5 $\pm 11.7^*$	22.2 $\pm 16.3$	20.8 $\pm 18.9^*$	14.9 $\pm 18.9^*$
ALT U/L	11.1 $\pm 7$	6.6 $\pm 4.2$	6.6 $\pm 4.2$	6.6 $\pm 4.2$	8 $\pm 2.6$	4.9 $\pm 2.4^*$	8 $\pm 2.6$	8.3 $\pm 2.4$	10.9 $\pm 3.5^*$

\* Significant different from mean value at  $P < 0.05$

**Table 4:** Biochemical parameters in pre and post treatment of goat.

Animal	Group A			Group B			Group C		
	Goat			Goat			Goat		
	Pre	Post		Pre	Post		Pre	Post	
	3 <sup>rd</sup> day	14 <sup>th</sup> day		3 <sup>rd</sup> day	14 <sup>th</sup> day		3 <sup>rd</sup> day	14 <sup>th</sup> day	
Total protein g/dl	5.8 ±0.8	5.2 ±1.2*	5.1 ±1.2*	4.9 ±0.4	4.66 ±0.2*	4.7 ±0.2*	4.6 ±0.8	5.6 ±0.6*	5.6 ±0.1*
Albumin g/dl	3.5 ±0.1	3.6 ±0.4	3.7 ±0.0*	3.9 ±0.4	3.2 ±0.2	3.1 ±0.3*	3.9 ±0.3	3.7 ±0.2*	3.9 ±0.3
Globulin g/dl	2.3 ±0.8	1.6 ±1*	1.5 ±1.2*	1.5 ±0.7	1.4 ±0.1	1.6 ±0.3	0.6 ±1.0	1.8 ±0.55*	1.7 ±0.4*
Creatinine mg/dl	0.8 ±0.1	0.7 ±0.2*	0.6 ±0.2*	0.7 ±0.1	0.6 ±0.4	0.6 ±0.38	1.0 ±0.4	0.8 ±0.1*	0.6 ±0.4*
AST U/L	42.6 ±4.7	20.8 ±18.9*	14.9 ±18.9*	19.3 ±13.9	23.5 ±19	9.7 ±9.8*	9.9 ±6.4	3.5 ±0.0*	14.2 ±11.3
ALT U/L	6.6 ±4.6	8.3 ±2.4	10.9 ±3.5*	4.1 ±1.5	7.3 ±4.98*	3 ±2.5*	4.9 ±7.4	5.7 ±4.4	5.6 ±3.4

\* Significant different from mean value at P < 0.05



**Photos 1: A and B:** Diarrhea and dehydration in lamb and kid affected with coccidiosis. **C:** Multifocal necrohaemorrhagic areas were seen in the distal part of jejunum of lamb infected with coccidiosis. **D:** Different developmental stages of *Eimeria* and oocysts were seen in the epithelium of affected intestinal villi.

## DISCUSSION

Anticoccidial drugs had a significant variation in their effects in sheep treated with sulphadimidine, that having the highest percentage of efficacy 97.6% followed by amprolium 95.6% and diclazuril 95.2%; similar findings in buffalo calves (Ghanem *et al.*, 2008; Guo *et al.*, 2007). Sulphadimidine had a little efficiency in controlling goat coccidian infestation than amprolium (Abakar, Ghanem and Abd El-Raof, 2005), in contrary of diclazuril that has a higher efficacy against coccidiosis in goat (Ruiz *et al.*, 2012).

Haematological examination in ailing animals uncovered a diminishment in RBCs and Hb, which could be attributed to hemorrhagic enteritis associated with coccidiosis (Deghidy *et al.*, 1984). Treated animals with sulphadimidine and amprolium revealed a significant increase in total RBCs and Hb. The leukocytes number was significantly decreased in post-treated sheep and goat than pretreated; those changes could be as a result of recovery from disease (Amulya *et al.*, 2014, Ahmed *et al.*, 2006).

The biochemical analysis revealed an increase in the total protein, albumin and globulin in the pros-treated animals, because the gastro-intestinal parasitism attributed in plasma leakage through the gut which reflected on the protein level (Radostits *et al.*, 1994, Tanwar and Mishra 2001), in addition to the secondary bacterial infection and the malabsorption syndrome occurring subsequently to the damage of intestinal mucosa and the loss of epithelial surface which lead to decrease in the protein level (Catchpole and Gregory, 1985).

The level of creatinine in post treated group was increased that could be due to the toxic effect of drugs as nephropathy and hypersensitivity leading to increasing the level of urea and creatinine (Goodman and Gilman 1965). There were some alterations in the level of serum AST and ALT that could be attributed to the liver might be adversely affected by *Eimeria* and anticoccidial drugs (Ghanem and Abd-Elraof, 2005).

Histopathological examination showed a thickened mucosa with scattered small hemorrhagic patches, the developmental stages of *Eimeria* and oocysts were detected in the epithelium of villi, crypts and necrotic-hemorrhagic exudates in the intestinal lumen. These results in agreement with (Ghanem *et al.*, 2008). The intestinal mucosa of the untreated kids was characterized by hypertrophy of the intestinal villi that were severely infected with large number of the developmental stages of *Eimeria* species. The mucosal blood vessels were congested with blood and the lamina propria was infiltrated with leucocytes particularly lymphocytes and oesinophils. Large area

of the intestinal mucosa showed desquamation of the lining epithelia cells which seen in the intestinal lumen mixed with necrotic debris and inflammatory cells, similarly to (Gab-Allah, 1990).

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### تقييم الحالة الإكلينيكية في الأغنام والماعز المصابة بالكوكسيديا وطرق علاجها

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تم فحص مائة وثمانية وأربعين عينة من الأغنام والماعز عن طريق تحليل البراز لتحديد اصابتها بالكوكسيديا. وكان معدل الإصابة ٦٥.٣٪، ٨٠٪، ٦٣.٦٪ و ٨٥.٧٪ في الأغنام والماعز والحملان وصغار الماعز على التوالي. في الفحص النسيجي لخلايا الامعاء تم تمييز مراحل الاصابه بالكوكسيديا. قسّم عدد ١٨ حيوانامصابا إلى ثلاث مجموعات وفقا للأدوية المضادة لكوكسيديا وذلك لتقدير فعاليتها. تم إجراء بعض تحليل لصورة الدم والبيوكيميائية قبل وبعد التجارب العلاجية في جميع الحيوانات. في حالة الأغنام، كان السلفاديميدين (٣٣.٣٪) أكثر فعالية من الأمبروليوم وديكلازوريل في حين أن ديكلازوريل له تأثير أفضل من أمبروليوم والسلفاديميدين (٣٣.٣٪). في حالة الماعز أظهرت جميع الحيوانات المعالجه نتائج محسنة في صورة الدم والبيوكيميائية مقارنة بالحيوانات غير المعالجة.