CLINICAL AND HAEMATO-BIOCHEMICAL STUDIES ON LAMB COCCIDIOSIS AND CHANGES FOLLOWING AMPROLIUM AND SULPHADIMTHOXINE THERAPY

Ghanem, M.M and Abd El-Raof, Y.M.

Department of Animal Medicine, Fac. Vet. Med. (Moshtohor), Benha University

ABSTRACT

This study was carried out on 18 lambs collected from different localities at Kalubia Governorate. Five lambs were clinically healthy and free from external and internal parasites and considered as control. The other 13 lambs had bloody diarrhea and their faeces had oocysts of *Eimeria* species. Haematological changes included a significant reduction (P<0.01) in RBCs count (8.16 ± 0.19 x 10^6/mm^3) and Hb content (8.4 ± 0.47 gm/dl). Also, there was a significant increase (P<0.05) of PCV% (39.68±1.55 %) and leucocytosis (18.22 ± 0.82 x10^3/mm^3) with neutrophilia (54.5 ± 3.2%), eosinophilia (5.5 ± 1.1%) and lymphocytopenia (37.8 ± 2.6%). Serum analysis showed a significant decrease of serum sodium (86.37 ± 2.71 Meq/L), iron (92.42±3.88 µg/dl), zinc (84.61 ± 3.81 µg/dl), and total protein (4.17 ± 0.42 gm/dl). In addition, there was a significant increase (P < 0.05) in serum aspartate aminotransferase (AST) (82.33 ± 3.57 U/L), alanine aminotransferase (ALT) (49.27 ± 2.41 U/L), alkaline phosphatase (ALP) (32.60 ± 0.46 IU/dl), gamma glutamyle transferase (GGT) (19.43 ± 0.46 IU/dl) and total bilirubin (1.30 ± 0.05 mg %) compared to control. On the other hand, there was a slight, albeit non-significant, decrease in the levels of serum calcium, copper and glucose and slight increase in serum potassium level. Treatment of lamb coccidiosis by oral sulfadimethoxine and amprolium was evaluated by determination of the haemato-biochemical parameters and comparing these values to those before treatment. Results showed that both amprolium and sulfadimethoxine were effective for treatment of eimeriosis in lambs via restoring the haemato-biochemical parameters to near the control values. However, sulfadimethoxine was little more effective than amprolium in controlling lamb coccidiosis.
INTRODUCTION

Ovine coccidiosis is one of the most common causes of enteritis in sheep. Its economic importance is attributed to the reduction of body weight, inefficient feed utilization and death of severely affected animals\textsuperscript{28}. The disease is caused by a protozoan parasite of genus \textit{Eimeria} which is characterized by sporulated oocysts and sporozoites\textsuperscript{11}. Coccidiosis in lambs was always caused by a mixed \textit{Eimeria} infestation and at least three species of \textit{Eimeria} were recorded from each diarrheic lamb. Generally, a minimum of eight species of \textit{Eimeria} were recognized in sheep including \textit{E. Parva}, \textit{E. Granulosa}, \textit{E. Ovinoidalis}, \textit{E. Ovina}, \textit{E. ahsata}, \textit{E. Pallida}, \textit{E. Crandallis} and \textit{E. Intricata}\textsuperscript{1,30}.

The different species of \textit{Eimeria} had a selective location within the intestinal tract\textsuperscript{3}. The general clinical findings of coccidiosis in sheep include depression, inappetance, grinding of teeth and abdominal pain. The faeces become watery, blood tinged, mucoid or containing blood clots. Affected animals sometimes suffered tenesmus. In addition, anemia, emaciation and dehydration were recorded\textsuperscript{22}.

Haematological changes recorded with coccidiosis included a reduction in erythrocytic count (RBCs) and hemoglobin concentration (Hb)\textsuperscript{17,29,35}. Moreover, the disease causing haemoconcentration indicated by an increase in packed cell volume\textsuperscript{17,27}. In addition, the disease is associated with leucocytosis\textsuperscript{9}, eosinophilia and neutrophilia\textsuperscript{9,29}.

The serum biochemical changes associated with sheep coccidiosis included a reduction in chloride and glucose\textsuperscript{35} and a reduction in serum sodium, calcium, phosphorus and copper\textsuperscript{1,4,18}. In addition, analysis of liver function in sheep coccidiosis produced an elevation in the levels of serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and bilirubin\textsuperscript{1}.

Regarding the medical intervention, symptomatic treatment of ovine coccidiosis included fluid therapy containing saline plus the use of hematinics as iron preparation and vitamin B complex, which is essential to overcome complications of dehydration and anemia\textsuperscript{28}. Specific treatment of coccidiosis by using anticoccidial drugs was
successfully attained by amprolium at a dose of 0.05 gm/kg B.wt for 10 days\textsuperscript{6,13}. Moreover, sulfonamides (sulfamerazine, sulfamethazine and sulfathiazole) have proved to be effective in treatment of coccidiosis after being given in drinking water for 3 days followed by oxytetracycline for 2 days\textsuperscript{24}.

This work aimed at recording the main clinical findings associated with coccidiosis among lambs and determination of the haematobiochemical changes associated with this problem. A further objective was to evaluate the effectiveness of two commonly used anticoccidial drugs in Egypt, namely and amprolium and sulfadimethoxine in controlling eimeriosis in lambs.

**MATERIALS AND METHODS**

**Animals and Experimental Design**

A total number of 18 native breed (Balady) lambs aging from 6 to 9 months, collected from different localities at Kalubia Governorate were used in this study. All lambs were subject to clinical examination\textsuperscript{21}. Fecal examination was performed according to Soulsby\textsuperscript{34}. Five lambs were clinically healthy and free from external and internal parasites and considered as control. The other 13 lambs had bloody diarrhea and fecal analysis indicated the presence of vegetative cells and oocysts of *Eimeria* species.

**Blood and Serum Analysis**

Two blood samples were drained from the jugular vein. The first sample was taken with anticoagulant (EDTA) for determination of blood picture (RBCs count, Hb content, PCV%, WBCs and differential leucocytic count) according to Jain\textsuperscript{19}. The second sample was collected without anticoagulant for the preparation of clear non-hemolysed serum.

Commercially available diagnostic kits were used for colorimetric determination of serum sodium\textsuperscript{15}, potassium\textsuperscript{15}, calcium\textsuperscript{14}, glucose\textsuperscript{23}, iron\textsuperscript{38}, total protein\textsuperscript{10}, total bilirubin\textsuperscript{20}, activities of ALT and AST\textsuperscript{31}, ALP\textsuperscript{5} and Gamma glutamyl transferase\textsuperscript{25}.

Serum copper and zinc levels were estimated by atomic absorption spectrophotometer (Perkin-Elmer Co., USA) according to a previously mentioned method\textsuperscript{39}.
**Therapeutic Trials**

Therapeutic trials were conducted by dividing diseased animals into two groups. The first group included 7 lambs that were treated orally by amprolium (El-Nasr Co. Therapeutic Chemicals, Abu-Zaabal, Kalubia) with a dose rate 67.5 mg/kg body weight for 14 days. Group 2 involved 6 lambs that were treated orally with sulfadimethoxine (Egyptian Company for Chemicals & Pharmaceuticals, ADWIA, 10th of Ramdan City, Egypt) 75 mg/kg B.wt twice daily for 5 days.

All diseased animals were injected with saline solution intravenously and drug containing ferrous sulphate was given orally at a dose rate of 2 mg/kg body 3 times daily for one week. In addition, Vit. B complex was injected intramuscularly. The comparative efficacy of both drugs (amprolium and sulfadimethoxine) was estimated by clinical examination of diseased lambs after treatment and by the haematobiochemical changes that occur three weeks post-treatment.

**Statistical Analysis**

All data were represented by the means ± standard error (SE). All pairwise comparison of infested lambs to control was analyzed by one-way analysis of variance (ANOVA) followed by Holm-Sidak test using SigmaStat 3.1, a statistical software for data analysis (SPSS Inc., Chicago, IL, USA). Unless otherwise indicated, all differences were considered statistically significant at P < 0.05.

**RESULTS AND DISCUSSION**

Coccidiosis in lambs is an intestinal infection characterized clinically by diarrhea and dehydration. A major cause of ovine coccidiosis is *Eimeria ovinoidalis*, but other species, such as *Eimeria crandallis*, may also be associated with this disease. In this study, clinical examination of diseased lambs revealed depression, reduced
appetite, pale mucous membrane (Figure 1) watery diarrhea with faeces frequently containing blood and mucus (Figure 2), tenesmus and dehydration. These signs were similar to those previously reported\(^6\). Faecal analysis demonstrated non-sporulated oocysts of \textit{E. Ovinoidalis} and \textit{E. Crandallis} (Figure 3), which are the most common species causing ovine coccidiosis\(^2\). The diarrhea observed in affected lambs might be attributed to the loss of surface epithelial cells and villus atrophy in the small intestine that are associated with first generation meronts of \textit{E. Crandallis} and the release of merozoites from them\(^12\).

Haematological changes of diseased lambs (Table, 1) included a significant decrease of total erythrocytic count and hemoglobin concentration. This result was in accordance with those recorded previously by several investigations\(^6,29,35\). The significant reduction of RBCs and Hb content might be attributed to the haemorrhagic enteritis associated with coccidiosis\(^7\). The PCV\(^\%\) was also significantly increased compared to control, which might be attributed to dehydration\(^26\). In this study, diseased lambs had leucocytosis with significant eosinophilia. This result was in agreement with a previous study\(^9\) that referred these changes to inflammation of intestine. The lymphocytopenia appeared in this study might be attributed to lymphocyte depletion and atrophy of the ileal Peyer’s patch follicles\(^2\).

Biochemical serum analysis for lambs with coccidiosis (Table 2 \& 3) demonstrated a significant reduction in the level of sodium (hyponatremia) and slight increase of the level of potassium. Changes of these electrolytes are usually related to diarrhea with loss of sodium ions. These results were comparable to those previously recorded\(^4,35\). Serum calcium, copper and glucose levels were slightly (non-significantly) decreased with coccidiosis, a result that was observed in other studies\(^1\). The decrease in these parameters might be attributed to suppression of appetite associated with coccidiosis. On the other hand, serum iron level was significantly decreased, which could be attributed to the bloody diarrhea and the inappetance occurring concurrently with eimeriosis\(^1\). Moreover, there was a significant decrease in serum zinc level that could be attributed to the secondary bacterial infection following coccidia infestation and the malabsorption syndrome occurring subsequently to damage of intestinal mucosa\(^9\) and loss of surface epithelial cells and villous
atrophy associated with first-generation meronts, crypt destruction and crypt hyperplasia. The significant decrease of serum total protein might be attributable to decreased absorption of nutrients from infection sites at intestinal mucosa due to damage and cell sloughing caused by coccidia.

Regarding liver function parameters, there was a significant increase in serum ALT, AST, ALP, GGT and total bilirubin in lamb with coccidiosis compared to control (Table 3). This result was nearly similar to those previously observed. Comparable results were demonstrated in calves during experimental and natural infections with *Eimeria Alabamenis*. The alterations of liver enzymes, total protein and bilirubin levels suggested that the liver might be adversely affected by coccidiosis.

The result of treatment of lamb coccidiosis with amprolium or sulfadimethoxine in conjunction with fluid therapy (saline) and hematinsics had a good response. Changed parameters returned to nearly normal values within three weeks post-treatment as there was no significant difference in these parameters between control and treated groups (Tables 1, 2 & 3). Despite of no significant changes in all parameters between amprolium-treated and sulfadimethoxine-treated lambs, values were more consistently close to control following sulfadimethoxine. This result implies that sulfadimethoxine is little more efficient in controlling coccidiosis than amprolium. Consistently with these findings, similar results were obtained in several other studies. However, other study showed that oral drenching of amprolium was highly efficacious in treatment and control of clinical coccidiosis in unweaned lambs.

In a conclusion, lamb coccidiosis was associated with bloody diarrhea, tenesmus, anemia, haemoconcentration, leucocytosis, neutrophilia, eosinophilia, hyponatremia and a reduction in serum iron, zinc and liver enzymatic activities. Treatment of coccidiosis in lambs is made possible with oral sulfadimethoxine or amprolium conjoined with fluid therapy and hematinsics. Sulfadimethoxine had little more efficiency in controlling lamb coccidian infestation than amprolium.
Table (1): Mean values of haematological parameters in control and lambs with coccidiosis.

<table>
<thead>
<tr>
<th>Haematological parameters</th>
<th>Negative control (n=5)</th>
<th>Lambs with coccidiosis</th>
<th>Lambs with coccidiosis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before treatment (n=13)</td>
<td>After treatment with amprolium (n=7)</td>
<td>After treatment with sulfadimethoxine (n=6)</td>
</tr>
<tr>
<td>RBCs (10^6/Cu mm)</td>
<td>11.92±0.24</td>
<td>8.16±0.19**</td>
<td>10.82±0.32</td>
</tr>
<tr>
<td>Hb (gm/dl)</td>
<td>12.20±0.27</td>
<td>8.4± 0.47**</td>
<td>11.72 ±2.92</td>
</tr>
<tr>
<td>PCV %</td>
<td>36.7 ± 1.49</td>
<td>39.68±1.55</td>
<td>37.43±1.37</td>
</tr>
<tr>
<td>WBCs (10^3/Cu mm)</td>
<td>9.14±0.19</td>
<td>18.22±0.82**</td>
<td>10.72±0.96</td>
</tr>
<tr>
<td>Lymphocytes %</td>
<td>55.50±2.08</td>
<td>37.8±2.6**</td>
<td>53.4±2.3</td>
</tr>
<tr>
<td>Monocytes %</td>
<td>2.50±0.25</td>
<td>2.2± 0.01</td>
<td>2.65±0.37</td>
</tr>
<tr>
<td>Neutrophils %</td>
<td>39.50±2.00</td>
<td>54.5±3.2</td>
<td>41.2±3.9</td>
</tr>
<tr>
<td>Eosinophils %</td>
<td>2.50±0.24</td>
<td>5.5±1.1**</td>
<td>2.75±0.09</td>
</tr>
<tr>
<td>Basophils %</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

* Significant different from control at P < 0.05

** Significant different from control at P < 0.01
Table (2): Mean values of some serum biochemical parameters in control and lambs with coccidiosis.

| Parameters | Negative control (n=5) | Lambs with coccidiosis | | | |
|------------|------------------------|------------------------|------------------------|------------------------|
|            | Before treatment (n=13) | After treatment with amprolium (n=7) | After treatment with sulfadimethoxine (n=6) | |
| Sodium (Meq/L) | 148.25±3.21 | 86.37±2.71 ** | 137.62±0.5 | 140.20 ± 0.72 |
| Potassium (Meq/L) | 5.42±0.12 | 6.93±0.81 | 5.82±0.11 | 5.66 ± 0.32 |
| Calcium (mg%) | 9.23±0.38 | 8.74±0.92 | 8.85±0.22 | 9.12 ± 0.25 |
| Iron (µg/dl) | 142.4±5.72 | 92.42±3.88 ** | 128.60±4.62 | 130.70 ± 5.01 |
| Copper (µg/dl) | 106.76±3.84 | 97.22±4.40 | 100.24±5.61 | 103.01 ± 3.21 |
| Zinc (µg/dl) | 105.41±2.80 | 84.61±3.81 ^ | 93.77±4.21 | 101.20 ± 3.92 |
| Glucose (mg%) | 52.12±3.15 | 47.35±4.21 | 49.72±3.66 | 51.54 ± 3.17 |

* Significant different from control at P < 0.05
** Significant different from control at P < 0.01
Table (3): Mean values of some serum liver function parameters in control and lambs with coccidiosis.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Negative control (n=5) Before treatment (n=13)</th>
<th>Lambs with coccidiosis After treatment with amprolium (n=7)</th>
<th>After treatment with sulfadimethoxine (n=6)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>before treatment (n=13)</td>
<td>After treatment with amprolium (n=7)</td>
<td>After treatment with sulfadimethoxine (n=6)</td>
</tr>
<tr>
<td>AST (U/L)</td>
<td>54.20±3.10</td>
<td>82.33±3.57 *</td>
<td>58.41±2.34</td>
</tr>
<tr>
<td>ALT (U/L)</td>
<td>28.60±1.71</td>
<td>49.27±2.41 *</td>
<td>30.21±1.54</td>
</tr>
<tr>
<td>ALP (IU/dl)</td>
<td>17.54±0.58</td>
<td>32.60±0.46 *</td>
<td>20.13±3.42</td>
</tr>
<tr>
<td>GGT (IU/dl)</td>
<td>16.14±0.72</td>
<td>19.43±0.46 *</td>
<td>18.33±0.29</td>
</tr>
<tr>
<td>Total bilirubin (mg%)</td>
<td>0.08±0.01</td>
<td>1.30±0.05 *</td>
<td>1.10±0.007</td>
</tr>
<tr>
<td>Total protein (gm/dl)</td>
<td>6.55 ± 0.82</td>
<td>4.17±0.42 *</td>
<td>5.97±0.37</td>
</tr>
</tbody>
</table>

* Significant different from control at P < 0.05

** Significant different from control at P < 0.01
Figure 1. A lamb with coccidiosis showing pale conjunctival mucous membrane.

Figure 2. A lamb with coccidiosis showing diarrhea and bloody feces.
Figure 3. Microphotograph of non-sporulated oocysts of *Eimeria Ovinoidalis* (A) and *Eimeria Crandallis* (B) in feces of affected lambs (Magnification 400 X).

REFERENCES


دراسات إكلينيكية ودموية وبيوكيميائية على الكوكسيديا في الحملان والتغيرات التي تحدث بعد العلاج بالأميروليم و السلفاداي ميتوكسین

محمد محمد غانم و يسین محمود عبد الرؤوف
قسم طب الحيوان – كلية الطب البيطري بمنحور – جامعة بنيها

المختصر العربي

أجريت هذه الدراسة على 18 حمالة تجميعها من مناطق متفرقة في محافظة القيروانية، حيث تم إجراء فحص إكلينيكي لهم وكذلك فحص طفيلي للبراز وقد تبين وجود خسارة حملان سليمة إكلينيكيا وخالية من أي طفيليات خارجية أو داخلية وقد اتخذت كمجموعة ضابطة أما الثلاثة عشر حيوانا الأخرى فكانت تعلاني من برازو دموي وثبت الفحص الطفيلي للبراز وجود حوصلات الكوكسيديا. ببينت صورة الدم نفس معنى ملحوظ في عدد كرات الدم الحمراء وكذلك نسبة الهيموجلوبين وكذلك زيادة في نسبة الخلايا المضغوطة. وقد لوحظ أيضا زيادة معنوية في عدد كرات الدم البيضاء مع زيادة في الخلايا المتدللة والحمضية وتناقص في نسبة الخلايا اللبماوية. وقد بين التحليل البيوكيميائي لعمل الدم نفس معنى في الصوديوم والحمض والزئبق والبيروتين الكلى وكذلك زيادة في نشاط إنزيم الألمنين أمينوتاراس فرينز وإنزيم الأسيرات أمينوتاراس فرينز والفيروسات القلوى وإنزيم الجاما جلوتاميك ترانس فرينز وكذلك زيادة في البيروتين. وكان هناك تناقص غير ملحوظ في قيم الكالسيوم والنحاس والسكر بينما زيادة بسيطة في مستوى عصر البروتامين. وقد أثبتت محاولات العلاج كفاءة كل من مركب السلفاداي ميتوكسین و الأميروليم ولكن مركب السلفاداي ميتوكسین كان أكثر فاعلية بقليل من العلاج بمركب الأميروليم.