The hematological changes associated with ovine subclinical mastitis.

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Abstract

The aim of the present study was to investigate ovine subclinical mastitis and associated with the hematological changes were studied in awassi sheep flock consists of ninety ovine lactating (58 normal and 32 ewes subclinical mastitis) both groups aged 1.5-4 years in AL-Najaf province /Iraq. Milk samples were collected for California mastitis test (CMT) and blood samples were collected from jugular vein during the period February 2016 till April 2016.

The results showed that the means ± SE in normal lactating and ewes subclinical mastitis were as follows; Packed cell volume (PCV) 31.9 ± 0.4 % and 25 ± 0.3 %, hemoglobin (Hb) 10.3 ± 0.16 g/dl and 7.3 ± 0.12 g/dl, red blood cell count (RBC) 10.2 ± 0.13 ×10⁶/µL and 8.9 ± 0.28 ×10⁶/µL, mean cell volume (MCV) 31.3 ± 0.36 fl and 30.9 ± 0.62 fl, mean cell hemoglobin (MCH) 10.1 ± 0.12 pg and 9.7 ± 0.21 pg, mean cell hemoglobin concentration (MCHC) 32.2 ± 0.18 g/dl and 31.2 ± 0.36 g/dl, erythrocyte sedimentation rate (ESR) 9.9 ± 0.46 mm/24 hrs and 17.9 ± 2.08 mm/24 hrs, Platelets 283.8 ± 16.6×10³/µL and 184.5 ± 8.3×10³/µL, white blood cell count (WBC) 10784 ± 268.3×10³/µL and 13956 ± 578.4×10³/µL, lymphocytes 54.3 ± 1.36 and 50.9 ± 2.51/µL, neutrophils 40.7 ± 1.41 and 52.8 ± 1.84/µL, monocytes 1.83 ± 0.16 and 3.0 ± 0.27/µL, eosinophil’s 2.84 ± 0.31 and 5.81 ± 0.76/µL and basophils 0.28 ± 0.05 and 0.78 ± 0.07/µL respectively.

The PCV, Hb, RBC and platelets were significantly higher (P<0.05) in normal lactating compared to that of ovine subclinical mastitis. The MCV, MCH and MCHC showed no significant differences between normal and ewes with subclinical mastitis. While ESR and WBC count were significantly lower in normal lactating.

Key words: Hematological parameters, clinically lactating normal, subclinical mastitis awassi sheep.
Introduction:

Subclinical mastitis is a worldwide problem and its economic importance attributed mainly to its higher prevalence and effect on quality and quantity of the produced milk (1). Also, subclinical mastitis has adverse effects on the hygienic quality and physicochemical properties of milk (2).

On the other hand, subclinical mastitis is the most prevalent type of mastitis characterized by no detectable changes in the udder and no visual abnormalities in milk (3). However, (4) reporting the absence of clinical signs specific to subclinical mastitis in sheep.

Diagnosis of such cases mainly depends upon bacterial culture results and/or indirect methods as CMT and SCC. Somatic cell count (SCC) considered a very helpful diagnostic tool in diagnosis of subclinical mastitis in dairy ewes (5). The limits of somatic cell counts of sheep milk, have not yet been definitely established, although it has been suggested that a threshold level for subclinical mastitis in sheep should be close to 1,500,000 cell mL, much higher than that set for cows (<500,000 cell mL) (6).

Materials and Methods

The California Mastitis Test (CMT, Jorgensen laboratories, Inc. Loveland, Colorado) was applied to all samples collected using the method of (7). According to the reactions obtained, the results were classified as: (negative), (traces), 1, 2 and 3, recorded as(−), (±), (+), (++), and (+++), respectively.

On the other hand, blood samples were collected into EDTA tubes (EDTA-K3 Italy) from the jugular vein of 90 ovine lactating (58 normal and 32 ewes with subclinical mastitis) during the period from February 2016 till April 2016 both aged 1.5–4 years in Al-Najaf governorate.

The blood were used directly for hematological parameters investigations. PCV was measured by
using micro hematocrit centrifuge according to (8), the hemoglobin was determined by acid hematin method (9). Red blood cells and white blood cell counts were measured by using hemocytometer (10). The MCV, MCH and MCHC were calculated according to the following formulas; MCV = PCV/RBC × 10fl, MCH = Hb/RBC×10pg and MCHC = Hb/PCV × 100g/dl (9). ESR was measured using westergren tubes according to (11).

A general guideline for estimating platelet numbers on a blood smear is to determine the average number of platelets in 10 oil immersion fields using a 100 objective and multiply the average by 15×10³ to obtain the estimated number of platelets per microliter (12).

The differential leukocyte count was carried on 200 WBC in giemsa stained blood film according to the method of (9).

**Statistic**

Data were analyzed using SPSS version 20. The least significant differences test (LSD) was used to determine differences among groups. Data were subjected to analysis of variance statistically using one-way ANOVA and the Duncan range test (Statistic a).

**Results and Discussion**

In general, diagnosis of subclinical mastitis mainly depends upon bacterial culture results and/or indirect methods as CMT and SCC. (13) as they stated that the CMT is very useful technique in diagnosis of subclinical mastitis in lactating ewes. According to the results of CMT method.

Table (1) illustrated The hematological parameters for normal lactating and ovine subclinical mastitis. The ranges and means ± SE of hematological parameters in normal lactating and ovine subclinical mastitis were as follows: PCV 28 - 40% and 31.9 ± 0.4 %, 20 - 27 and 25 ± 0.3, Hb 8.6 - 13.5 and 10.3 ± 0.16 g/dl, 5.6 - 8.1 and 7.3 ± 0.12 g/dl, RBC 8.8 - 13.3 and 10.2 ± 0.13 ×10⁶/μL, 5.3 - 11.4 and 8.9 ± 0.28× 10⁶/μL, MCV 27.5 - 37.1 and 31.3 ± 0.36 fl, 23.4 - 37.4 and 30.9 ± 0.62 fl, MCH 8.6 - 12 and 10.1 ± 0.12 pg, 8.1 - 12.1 and 9.7 ± 0.21 pg, MCHC 30.5 - 35.7 and 32.2 ± 0.18 g/dl, 28.1 - 35.6 and 31.2 ± 0.36 g/dl, ESR 3 - 17 and 9.9 ± 0.46 mm/24 hrs, 8 - 63 and 17.9 ± 2.08 mm/24 hrs, Platelets 122 - 723 and 283.8 ± 16.6 ×10³/μL, 103 - 280 and 184.5 ± 8.3 ×10³/μL respectively.

The means of PCV, Hb, RBCs and platelets were significantly high (P<0.05), there was no significant differences in MCV, MCH and MCHC, while ESR showed a significant decrease (P<0.05) in normal lactating compared to that of ewes with subclinical mastitis (table1).

### Table (1). The hematological parameters for normal lactating and ovine subclinical mastitis; ranges and means± SE.

<table>
<thead>
<tr>
<th>parameters</th>
<th>groups</th>
<th>Normal lactating N=58</th>
<th>Subclinical mastitis N=32</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCV (%)</td>
<td></td>
<td>28-40</td>
<td>20-27</td>
</tr>
<tr>
<td>Parameter</td>
<td>Value</td>
<td>Value</td>
<td></td>
</tr>
<tr>
<td>---------------------</td>
<td>-------------</td>
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<td></td>
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<tr>
<td>Hb (g/dL)</td>
<td>8.6-13.5</td>
<td>5.6-8.1</td>
<td></td>
</tr>
<tr>
<td>RBC (x10^6/μL)</td>
<td>8.8-13.3</td>
<td>5.3-11.4</td>
<td></td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>27.5-37.1</td>
<td>23.4-37.4</td>
<td></td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>8.6-12</td>
<td>8.1-12.1</td>
<td></td>
</tr>
<tr>
<td>MCHC (g/dL)</td>
<td>30.5-35.7</td>
<td>28.1-35.6</td>
<td></td>
</tr>
<tr>
<td>ESR mm/24h</td>
<td>3-17</td>
<td>8-63</td>
<td></td>
</tr>
<tr>
<td>Platelets (x10^3/μL)</td>
<td>122-723</td>
<td>103-280</td>
<td></td>
</tr>
<tr>
<td>WBC (/μL)</td>
<td>3950-12950</td>
<td>8150-23550</td>
<td></td>
</tr>
<tr>
<td>L%</td>
<td>31-72.5</td>
<td>13.5-76.5</td>
<td></td>
</tr>
<tr>
<td>N%</td>
<td>22-67.5</td>
<td>33.5-73.0</td>
<td></td>
</tr>
<tr>
<td>M%</td>
<td>0.5-6.0</td>
<td>0.5-6.5</td>
<td></td>
</tr>
<tr>
<td>E%</td>
<td>0-9.5</td>
<td>0-20.0</td>
<td></td>
</tr>
<tr>
<td>B%</td>
<td>0-1.5</td>
<td>0-1.5</td>
<td></td>
</tr>
</tbody>
</table>

The differences in capital letters horizontally refer to the presence of significant value at (<0.05).

The normal reference range of PCV, Hb, RBC, WBC and platelets obtained in the present study agreed with the range of healthy sheep by (12, 14, 15), although some hematological parameters were different from the reference range of these studies, and this may be due to one or the following: nutritional, geographical, physiologic status and genetic factors (16). On the other hand, in normal lactating the hematological parameters; MCV, MCH, MCHC, platelets and ESR of this study were no significant difference with the values reported (17).
Table (1) Results showed the ranges and means ± SE of WBCs and differential leukocytes in normal lactating and ovine subclinical mastitis were as follows; WBC 3950 - 12950 and 10784 ± 268.3 ×10³/μL, 8150 - 23550 and 13956 ± 578.4 ×10³/μL, lymphocytes 31 - 72.5 and 54.3 ± 1.36, 13.5 - 76.5 and 50.9 ± 2.51 /μL, neutrophils 22 - 67.5 and 40.7 ± 1.41, 33.5 - 73.0 and 52.8 ± 1.84 /μL, monocytes 0.5 - 6.0 and 1.83 ± 0.16, 0.5 - 6.5 and 3.0 ± 0.27 /μL, eosinophil’s 0 - 9.5 and 2.84 ± 0.31, 0 - 20.0 and 5.81 ± 0.76 /μL and basophils 0 - 1.5 and 0.28 ± 0.05, 0 - 1.5 and 0.78 ± 0.07 /μL respectively.

There was a significant increase (p<0.05) in WBC count of ovine subclinical mastitis compared to that of normal lactating. However in differential leukocyte counts neutrophils, monocytes, eosinophil’s and basophils were significantly higher(p<0.05) in ewes with subclinical mastitis compared to normal lactating. On the other hand, the lymphocytes showed no significant difference in normal and ovine subclinical mastitis, the differences may be attributed to immune status of the host at time of infection and the pathogenicity of the causative bacteria (18).

The lymphocytes, neutrophil, monocyte, eosinophil and basophil reference range of (12, 14, 15) were as follows; Lymphocytes 2000-9000 /μL, neutrophils 700-6000 /μL, monocytes 0-750 /μL, eosinophils 0-1000 /μL and basophil 0 – 300 /μL. These references range were similar in monocytes and eosinophils counts, narrower range in lymphocytes and neutrophils counts and a wider range in basophils counts compared to that of present data. This may be attributed to type of feed or genetic.

The values of WBC, lymphocyte, neutrophil, monocyte and eosinophil showed a significant decreased, while, basophil revealed no significant difference with (19). The WBC, neutrophils, lymphocyte, monocyte, eosinophil and basophil counts in the present study was agreement compared to that of (17). There was a non-significant increase of lymphocytes counts in normal lactating ewes compared with subclinical mastitis ewes, this is agree with (20,21).

However, this result could be due to the fact that lactating ewes were more susceptible to systemic and local infection through the udder, also serum lactic dehydrogenase (LDH) levels increased in lactating ewes (22). Moreover, the lactic dehydrogenase (LDH) may produce leukocytosis (23). Also, infection by bacteria can cause bone marrow suppression, resulting in thrombocytopenia and anemia (24, 25).

Conclusions

The data presents reference range and mean ± SE of hematological parameters in normal lactating ewes. However, in normal lactating a significant increase(p<0.05) in values of PCV, Hb, RBC and platelets and a significant decrease(p<0.05) in ESR and WBC in comparison with ovine subclinical mastitis.

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