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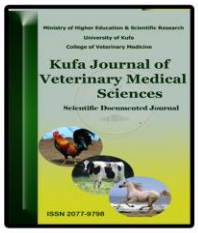

The hematological parameters in clinically normal lactating and ewes affected with mastitis

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Abstract

The aim of the present study was to the determine the ranges and means ± SE of hematological parameters in Naemi clinically healthy lactating and ewes with mastitis . The study was conducted on 50 normal and 50 ovine mastitis (19 clinical and 31 subclinical) infected with *staphylococcus aureus* , both groups aged 2-5 years in Salah al-ddin governorate / Iraq . The blood samples were collected from jugular vein during the period October 2012 till April 2013 .

Results showed that the means ± SEin normal lactating and ovine mastitis were as follows ; Packed cell volume (PCV) 28 ± 0.27 % and 21.76 ± 0.46 % , hemoglobin (Hb) 9.4 ± 0.12g /dl and 7.56 ± 0.14 g / dl , red blood cell count (RBC) 9.52 ± 0.11 $\times 10^6/\mu L$ and 7.67±0.19 $\times 10^6/\mu L$, mean cell volume (MCV) 29.39 ± 0.19 fl and 28.43 ± 0.32 fl , mean cell hemoglobin (MCH) 9.83 ± 0.08 pg and 9.88 ± 0.08 pg , mean cell hemoglobin concentration (MCHC) 33.51 ± 0.19 g/dl and 34.94 ± 0.33 g/dl , erythrocyte sedimentation rate (ESR) 6.28 ± 0.15 mm / 24 hrs and 27.08 ± 2.97 mm / 24 hrs , Platelets 247.22 ± 17.88 $\times 10^3/\mu L$ and 185.58 ± 14.55 $\times 10^3/\mu L$, white blood cell count (WBC) 8.18 ± 0.22 $x10^3/\mu L$ and 14.38±0.26 $\times 10^3/\mu L$, lymphocytes 4509.6 ± 127.6 and 5033.4 ± 412.7 / µL , neutrophils 2857.2 ± 129.2 and 7714.8 ± 415.5/µL , monocytes 371.3 ± 32.7 and 846.9 ± 38.8/µL , eosinophils 331.2 ± 18.8 and 294.7 ± 24.4/µL and basophils 100.6 ± 7.3 and 463 ± 33 / µL respectively . The PCV, Hb , RBC and platelets were significantly higher ($P < 0.05$) in normal lactating compared to that of ovine mastitis . The MCV, MCH and MCHC showed no significant differences between normal and ewes with matitis.

While ESR and WBC count were significantly lower in normal lactating. Moreover the results revealed significant differences ($P < 0.05$) in some hematological values of different subgroups .

Key words: Hematological parameters, clinically normal , mastitis , Iraqi , Naemi sheep.

القيم الدمية في النعاج الطبيعية سريريا والمصابة بألتهاب الضرع

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الخلاصة :

الهدف من هذه الدراسة كان لتحديد المديات والمعدلات للقيم الدمية في النعاج النعيمية الحلوبة الطبيعية والنعاج المصابة بالتهاب الضرع . اجريت الدراسة على 50 نعجة طبيعية و 50 نعجة مصابة بألتهاب الضرع (19 سريرية و 31 تحت سريرية) اصيبت بجرثومة المكورات العنقودية الذهبية ، تتراوح اعمارهم من 2 – 5 سنوات في محافظة صلاح الدين . جمعت عينات الدم من الوريد الوداجي خلال الفترة من تشرين الاول 2012 لغاية نيسان 2013 .

اظهرت النتائج للمعدلات ± الخطأ القياسي في النعاج الحلوبة الطبيعية والنعاج المصابة كما يلي : حجم الخلايا المضغوطة (% CV) 9.4 ± 0.12 غرام / 28 Hb (P± 0.27 % و 21.76 ± 0.46 % ، خضاب الدم ديسي لتر و 7.56 ± 0.14 غرام / ديسي لتر ، كريات الدم الحمر (10 × 0.11 ± 9.52 / ^{6}RBC) مايكرولتر و 7.67 ± 0.19 ×10^{6} / مايكرولتر ، الحجم الكاربي (0.19 ± 29.39 (MCV فيمتولتر و 38.43 ± 0.32 فيمتولتر ، خضاب الدم الكاربي (0.08 ± 9.8 (MCH بيكوغرام و 9.88 ± 0.08 بيكو غرام , تركيز خضاب الدم الكاربي (0.19 ± 33.51 (MCHC غرام / ديسي لتر و 34.94 ± 0.33 غرام / ديسي لتر ، نسبة تثقل كريات الدم الحمر (0.15 ± 6.28 (ESR ملم / 24 ساعة و 27.08 ± 2.97 ملم / 24 ساعة ، الصفائح الدموية (^{3}Platelet) 247.22 ± 17.88 × 10 / مايكرو لتر و 185.58 ± 14.55 × 10^{3} s ، كريات الدم البيضاء (10 × 0.22 ± 8.18 (^{3}WBC / مايكرو لتر و 14.38 ± 0.26 × 10^{3} / مايكرو لتر ، الخلايا اللمفية (127.6. ± 4509.6 (L / مايكرولتر و 5033.4 ± 412.7 / مايكرولتر ، العدلات (129.2. ± 2857.2 (N / مايكرولتر و 7714.8 ± 415.5 / مايكرولتر ، الخلايا احادية النواة (32.7. ± 371.3 (M / مايكرولتر و 846.9 ± 38.3 / مايكرولتر ، الحمضات (18.8. ± 331.2 (E / مايكرولتر و 294.7 ± 24.4 / مايكرولتر والقعدات (7.3. ± 100.6 (B / مايكرولتر و 463 ± 33 / مايكرولتر على التوالي واظهرت النتائج ان حجم الخلايا المرصوصة ، خضاب الدم ، كريات الدم الحمراء والصفائح الدموية كانت اعلى في النعاج الطبيعية ($P < 0.05$) مقارنة بالنعاج المصابة بألتهاب الضرع . لاتوجد فروق معنوية بين النعاج الطبيعية والمصابة بألتهاب الضرع في الحجم الكاربي ، خضاب الدم الكاربي و تركيز خضاب الدم الكاربي . بينما لوحظ انخفاظا معنويا في تثقل كريات الدم الحمراء واعداد كريات الدم البيضاء في النعاج الطبيعية . اضافة الى ذلك وجود اختلافات معنوية ($P < 0.05$) لبعض القيم الدمية في المجاميع الفرعية الاخرى.

Introduction:

The reference hematological value are useful tools for diagnosis and prognosis of many diseases , and the main laboratory hematological parameters were PCV, Hb , RBC, MCV, MCH and MCHC (1) . However, (2) studied hematological profile of twenty West African dwarf sheep fed on *Moringaoleifera* . Also (3, 4, 5) have documented the normal hematological values in sheep . In

Pakistan (6) they measured ESR at one hour in 400 sheep , 54 were infected with external and internal parasites and 346 clinically normal . While, (7) whom they studied hematological parameters in 60 normal and infected with internal parasite pregnant Akkaraman ewes in Turkey. Normal sheep WBC count were enumerated by (8) . Although, (9) they noted the influence of age and sex on the hematological values of goats and sheep in the arid zone of Borno state of Nigeria.

In Iraq , (10) were studied the hematological values in a total of 36 divided into 3 equal groups of clinically normal Awassi , Karadi and Arabi sheep both sexes aged 6 months - 5 years in Baghdad governorate .While , (11) who reported hematological values of 120 Awassi sheep in Baghdad governorate, these were selected for meat and twin production.The hematological parameters in clinically healthy Iraqi Awassi sheep were documented by (12) whom they studied hematological values in 40 normal lactating ewes . However the only study of hematological parameters ; PCV, Hb , RBC count, MCV, MCH , MCHC , ESR , WBC and differential leukocytes count in normal lactating and ovine mastitis have been reported by (13) .

Many of the above mentioned studies were conducted on smaller number or fewer hematological parameters; therefore, this investigation was carried out on a larger number of healthy lactating and ovine mastitis with as wide range of hematological parameters in Iraqi Naemi sheep.

Materials and Methods

Blood samples were collected into EDTA tubes from the jugular vein of 50 normal lactating and 50 ewes with mastitis (19 clinical and 31 subclinical) infected with *S. aureus* during the period from October 2012 till April 2013 both aged 2-5 years in Salah al-ddin governorate . The sheep affected with clinical mastitis were subdivided into acute and chronic subgroups . The blood were used directly for hematological parameters investigations . PCV was measured by using microhematocrit centrifuge according to (14) , the hemoglobin was determined by acid hematin method (15) . Red blood cells and white blood cell counts were measured by using hemocytometer (16) . The MCV, MCH and MCHC were calculated according to the following formulas ; MCV = PCV / RBC (m) ×10fl , MCH = Hb / RBC (m) x10pg and MCHC= Hb/PCV × 100g/dl (15) . ESR was measured using westergren tubes according to (17). Platelets count : stained blood film such as that prepared for routine hematological examination andthe number of platelets observed in fields scanned in 100 locating leukocytes , if WBC count is known, platelets number counted by the following formula : Platelets / µL= number of platelets × total WBC (15).While the differential leukocyte count was carried on 200 WBC in

giemsa stained blood film according to the method of (15) .

SAS program was used for statistical analysis . Data were subjected to Analysis of Variance (ANOVA) and significant means were compared by T- test at level ($P < 0.05$).

Results and Discussion

The ranges and means ± SE of hematological parameters in normal lactating and ovine mastitis were as follows : PCV 26 - 33 and 28 ± 0.27 % ,16 - 28 and 21.76 ± 0.46 % , Hb 8 - 12.5 and 9.4 ± 0.12 g / dl, 5 - 10.5 and 7.56 ± 0.14 g/dl , RBCs 8.4 - 12 and 9.52 ± 0.11× 10^6/ µL, 5 - 14 and 7.67 ± 0.19 ×10^6µL , MCV 25 - 32.6 and 29.39 ± 0.19fl , 24 - 32.5 and 28.43 ± 0.32 fl , MCH 8.3 - 11.4 and 9.83 ± 0.08 pg, 8.4 - 11.4 and 9.88 ± 0.08 pg , MCHC 30.76 - 36.76 and 33.51 ± 0.19 g / dl, 30.86 - 41.66 and 34.94 ± 0.33 g/dl , ESR 4.5 - 8 and 6.28 ± 0.15mm / 24 hrs , 9 - 93 and 27.08 ± 2.97mm/24 hrs , Platelets 108 - 613.6 and 247.22 ± 17.88 × 10^3µL , 35.4 - 409.2 and 185.58 ± 17.88 x10^3µL respectively . The PCV , Hb , RBCs and platelets means were significantly higher , there was no significant difference in MCV, MCH and MCHC , while ESR showed a significant decrease in normal lactating compared to that of ewes with mastitis. Also clinical mastitis revealed higher ESR compared to subclinical ones(table1).

Table (1). Hematological parameters in normal and ovine mastitis ; ranges and means ± SE

Parameters	Groups					
	Normal lactating ewes n=50	Ewes with Mastitis n=50	Subclinical mastitis n=31	Clinical mastitis n=19	Clinical mastitis	
					Acute n=11	Chronic n=8
PCV %	26-33 28±0.27 a	16-28 21.76±0.46 b	16-28 21.51±0.62 b	16-28 22.15±0.67 b	16-28 22.45±1.06 b	18-24 21.75±0.72 b
Hb g/dl	8-12.5 9.4±0.12 a	5-10.5 7.56±0.14 b	5-10.5 7.49±0.21 b	6.2-9.1 7.67±0.16 b	6.2-9.1 7.72±0.24 b	6.45-8.75 7.61±0.23 b
RBCs × 10^6/µL	8.4-12 9.52±0.11 a	5-14 7.67±0.19 b	5-14 7.59±0.3 b	6.2-9.2 7.81±0.16 b	6.5-9.2 7.93±0.21 b	6.2-8.8 7.65±0.28 b
MCV fl	25-32.6 29.39±0.19 a	24-32.5 28.43±0.32 a	24-32.3 28.64±0.93 a	24-32.5 28.08±0.55 a	24-32.5 27.9±0.74 a	25.7-32.2 28.33±0.87 a

MCH pg	8.3-11.4 9.83±0.08 a	8.4-11.4 9.88±0.08 a	8.4-11.4 9.97±0.11 a	8.8-10.7 9.73±0.1 a	8.8-10.1 9.58±0.12 a	9.4-10.7 9.95±0.16 a
MCHC g/dl	30.76-36.76 33.51±0.19 a	30.68-41.66 34.94±0.33 a	30.68-38.94 34.82±0.4 a	31.15-41.66 35.13±0.59 a	31.15-38.75 34.96±0.73 a	32.29-41.66 35.37±1.05 a
ESR mm/24 hrs	4.5-8 6.28±0.15 d	9-93 27.08±2.97 b	9-30 14.45±0.93 c	15-93 47.68±4.77 a	44-93 59.81±5.47 a	15-42 31±3.43 b
Platelets $\times 10^3\mu L$	108-613.6 247.22±17.88 a	35.4-409.2 185.58±14.55 b	52-409.2 206.17±20.16 b	38.4-296 151.97±17.61 bc	38.4-199.8 107.98±15.23 c	106.4-296 212.47±23.16 b

The differences in small letters horizontally refer to presence of significant value at (P < 0.05)

The lower limit in PCV, Hb , RBCs , MCV , MCHC and platelets ranges of this study were lower, while the MCH similar, moreover the upper limit in PCV , Hb , RBCs , MCV and platelets agreed , while the MCHC higher to that reported by (3,4). On the other hand the hemogram of this work including PCV, Hb , MCV, MCH , MCHC and platelets decreased , while the RBCs showed no significant difference compared to that of (18) .

In normal lactating the hematological parameters; PCV % , Hb , RBC, MCV and ESR of this study were decreased , no significant difference in MCH and platelets , while a significant increase in MCHC (12) .

Results showed the ranges and means ± SE of WBCs and differential leukocytes were as follows ; WBCs 6 - 11.8 and 8.18 ± 0.22 $\times 10^3$/ µL , 12.2 - 19.4 and 14.38 ± 0.26 × 10^3/ µL , lymphocytes 2472 - 8557 and 4509.6 ± 127.6/ µL , 769 - 15316 and 5033.4 ± 412.7/ µL and neutrophils 1064 - 5800 and 2857.2 ± 129.2/ µL, 1268 - 17122 and 7714.8 ± 415.5/ µL , monocytes 75 - 975 and 371.3 ± 32.7/ µL , 286 -1833 and 846.9 ± 38.8/ µL , eosinophils 0 - 931 and 331.2 ± 18.8/ µL , 0 - 764 and 294.7 ± 24.4/ µL and basophils 0 - 312 and 100.6 ± 7.3/ µL , 0 - 1274 and 463 ± 33/ µL respectively. There was a significant increase ($p<0.05$) in WBC count of ovine mastitis compared to that of

normal lactating , also a significant increase in clinical mastitis in comparison with subclinical mastitis .

However in differential leukocyte counts neutrophils , monocytes , basophils were significantly higher in ewes with mastitis compared to normal lactating .On the other hand the lymphocytes showed no significant difference in normal and ovine mastitis . However, eosinophils are significantly higher in normal lactating ewes. There were significant differences in ewes with mastitis subgroups (table 2) .

Table (2) The WBCs and differential leukocytes count in normal lactating and ovine mastitis ; ranges and means ± SE.

Parameters	Groups					
	Normal lactating ewes n=50	Ewes wth mastitis n=50	Subclinical mastitis n=31	Clinical mastitis n=19	Clinical mastitis	
					Acute n=11	Chronic n=8
WBCs $\times 10^3/\mu L$	6-11.8 8.18±0.22 d	12.2-19.4 14.38±0.26 b	12.2-14.4 13.18±0.11 c	14.4-19.4 16.33±0.36 a	16.2-19.4 17.34±0.4 a	14.4-15.4 14.95±0.9 b
Lymphocytes µL	2472-8557 4509.6±127.6 cd	769-15316 5033.4±412.7bc	3370-5400 4193.8±73.8 d	908-15182 6125.3±384 b	1022-3703 2313.1±265.3 e	3561-12158 11526.4±55.3 a
Neutrophils µL	1064-5800 2857.2±129.2 d	1268-17122 7714.8±415.5 bc	6259-8742 7379.4±64.5 c	1497-17122 8576.5±443 b	11485-17122 13384.7±305.1 a	1497-10100 1828.3±70.2 e
Monocytes µL	75-975 371.3±32.7 e	286-1833 846.9±38.8 b	286-1360 759.1±40.8 bc	437-1650 997.7±73.4 b	492-1385 456±64.1 de	938-1310 1194.6±41.8 a
Eosinophils µL	0-931 331.2±18.8 a	0-764 294.7±24.4 b	0-567 312.3±27.6 b	0-704 251.4±44 b	0-704 320.7±60.6 b	0-523 165.9±62.7 c
Basophils µL	0-312 100.6±7.3 d	0-1274 463±33 b	146-946 495.5±40.8 ab	0-894 383.7±45.7 bc	144-894 456±64.1 b	0-512 293±59.8 c

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

The WBC of the present study in normal lactating ewes were close, while the upper limit of monocytes and basophils were higher (3, 4, 5) .

The values of WBC , lymphocyte, neutrophil, monocyte and eosinophil showed a significant decreased ,while, basophil revealed no significant difference with (18) . The WBC, neutrophils and lymphocyte counts in the present study was significantly decrease, while monocyte , eosinophil and basophil showed a significant increase compared to that of (12) .

In ovine mastitis the increased numbers of WBC counts and neutrophils may be due to their role in defense mechanism against infections, or due to the increase serum LDH levels in ewes with mastitis (4 , 19) . There was a non significant increase of lymphocytes counts in total ewes with mastitis and a highly significant increase in chronic mastitis ,this is more or less agree with (13) who noted a significant increase in lymphocytes counts . Also this agree with (15) who recorded lymphocytosis in chronic diseases.

The differences in hematological parameters (tables 1 and 2) of this findings compared to other studies may be attributed to one or more of the followings : the different physiological status , type of feed , absence of scientific feeding program , season or genetic factors (12). While, the deficiency of proteins in ewes with mastitis of this study may be resulting in development of an anemia by interfering with hemoglobin production (15) . However the hematological values of ewes with mastitis in the present study showed a relation between blood parameters and infection in subclinical and clinical mastitis . Although , infection by bacteria can cause bone marrow suppression, resulting in thrombocytopenia and anemia (20,21) . Also , decreased blood parameters possibly due to the *staphylococcus aureus* hemolysins , this may cause lysis of red blood cells by damaging their cell membrane (22).

Conclusions

The data presents reference range and mean ± SE of hematological parameters in normal lactating ewes. However, in normal lactating a significant increase in values of PCV , Hb , RBC and platelets and a significant decrease in ESR and WBC in comparison with ovine mastitis. Moreover , there were significant differences in mastitis subgroups.

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