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Preliminary study regarding the effect of season on haematological parameters in Syrian Awassi rams

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ABSTRACT

Four apparently healthy Syrian Awassi rams were used for one year. Jugular blood samples were collected from the rams once a week to determine some essential blood components (13) using a Veterinary Haematology Analyzer. The overall means were: 8.92 x 10^3/mm^3 for White Blood Cells (WBC), 5.91% for Lymphocytes (LYM), 1.00 % for Monocytes (MON), 2.62% for Granulocytes (GRA), 7.94 x 10^3/mm^3 for Red Blood Cells (RBC), 10.24 g/dL for Haemoglobin (Hb), 30.93% for Haematocrit (HCT), 39.12fL for Mean Corpuscular Volume (MCV), 12.57 pg for Mean Cell Haemoglobin (MCH), 33.10g/dL for Mean Corpuscular Haemoglobin Concentration (MCHC), 16.34 % for Red Cell Distribution Width (RDW), 577.3 x10^3/mm^3 for Platelet Counts (PLT) and 3.99 fL for Mean Platelet Volume (MPV), with variations in the values among individuals. There were differences in the value means among the months of the year in some components, where the overall means of WBC, LYM, MON, GRA, HGB, MCH and MCHC were significantly ($P<0.05$) higher in winter as compared to summer months, whereas, MCV means were significantly ($P<0.05$) higher in summer as compared to winter months, whereas there were no significant ($P>0.05$) differences in the means of RBC, RDW, PLT or MPV among summer and winter months.

Keywords: WBC parameters, RBC parameters, platelets parameters, Awassi rams, season

INTRODUCTION

Measurement of haematological components can reflect the physiological and health status of animals, and is widely used in disease diagnosis and consequently the treatment in time. Values of haematological parameters are affected by several factors including breed, age, environment, reproductive status, husbandry and hormonal treatments (Arfuso et al. 2016; Habibu et al. 2017; Ahmed et al. 2018).

Many laboratories have no reference parameters and need to establish their own set for use. Reference values of haematological parameters for some species of animals were reported such as Italian Ariégeois, Bleu de Gascogne, Bracco italiano, Segugio italiano, and Briquet Griffon Vandeen hunting (Miglio et al. 2020) and Ghana locally raised dogs (Opoku-Agyemang et al. 2019), BALB/c mice in Brazil (Pessini et al. 2020), Chinese water deer (Nie et al. 2020), Brazilian geese (Benarrós et al. 2020), Egyptian buffaloes (Abd Ellah et al. 2019), Pakistani camels (Farooq et al. 2011), Italian Piemontese and Blonde d'Aquitaine cattle (Tarantola et al. 2020), Italian Heavy Draft horse (Miglio et al. 2019) and Polish half-bred and Wielkopolska horses (Burlikowska et al. 2015), and four Omani goat breeds (Al-Bulushi et al. 2017).

As for sheep, Al-Samarai and Al-Jbory (2017) studied the effect of sex, age, and region on the white blood cells (WBC), red blood cell (RBC), packed cell volume (PCV), haemoglobin (HGB), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) parameters in Iraqi Awassi sheep and reported that the effect of region and sex was significant, whereas no effect of age was reported. In other study in Iraq too, Hassan et al. (2019) reported that RBC number and MCH value did not change between the two seasons (summer and spring) in lactating Awassi ewes. In Poland, Wojtas et al. (2014) reported that heat stress led to serious changes in blood parameters (WBC, RBC, HGB, haematocrit: HCT, platelets: PLT, MCV, MCH and MCHC) in Merino rams, however, this effect could be minimised by air movement. Bezerra et al. (2017) compared erythrocyte and leukocyte parameters of Santa Inês and Morada Nova ewes in Brazil at different reproductive stages. In Kyrgyzstan, Kisadere et al. (2018) evaluated and compared 10 haematological parameters in Hissar and Edilbaev sheep breeds, and in Nigeria, Oluwatosin and Adebowale studied 6 haematological parameters, in an experiment lasted for 63 days, namely WBC, RBC, HGB, PCV, MCH and MCHC in the blood samples collected at the end of the feeding trial of West African Dwarf sheep to assess the effect of replacing *Pennisetum purpureum* with white *Morus alba* leaves on the above parameters.

The fat-tailed triple purpose (meat, milk and wool), (more than 14.5 million as compared to 1.8 million for goats and 0.8 million for cows in 2019: MAAR, 2021) Awassi is the local breed in Syria and many neighbouring countries. This breed is well-adapted to the harsh environment (Salhab et al. 2003).

In Syria, several studies were conducted on Syrian Awassi sheep such as productive and reproductive parameters in Awassi ewe lambs (Zarkawi and Al-Daker, 2018), body weight and reproductive parameters in Awassi ram lambs (Zarkawi and Al-Daker, 2016) and factors affecting serum testosterone levels in growing Syrian Awassi ram lambs (Zarkawi and Salhab, 2005). However, as for haematological parameters, very few studies and reports are available, and as there are no haematological reference values for Syrian Awassi rams so far, therefore, the aim of the current study was to define and establish the haematological reference values for the Syrian Awassi rams and to assess the effect of season.

MATERIALS AND METHODS

Location and experimental animals

This study was carried out at the Animal Production Division, in Deir Al-Hajar area, about 33 km south-east of Damascus. This area is considered a dry area with an annual rainfall of about 120 mm occurring mainly in December and January, and it is a part of the Syrian steppe region where the majority of sheep population is raised. Table 1 shows the average maximum and minimum temperature in the experiment site during the study period.

Table 1. Average maximum and minimum temperature throughout the year recorded in the study site.

Month	Average Maximum Temperature (^{o}C)	Average Minimum Temperature (^{o}C)
January	13.1	2.4
February	15.2	4.8
March	18.3	5.5
April	22	8.8
May	34	14.9
June	36	18.1
July	37	19.3
August	37.8	20.1
September	34.6	17.5
October	30.0	14.8
November	20.0	8.7
December	14.8	5.5
Overall	26.1	11.7

Four 2-4 year-old apparently healthy Syrian Awassi rams with no previous ill-health records were used for one year.

Ethical approval

This study was approved by the Local Scientific and Ethical Committee of the Atomic Energy Commission of Syria (AECS), Damascus, Syria (Permit Number 36/Z/M1-02/09/2019).

Blood sampling and analysis

Jugular blood samples (10 mL) were collected weekly at the same time (10 am) from the experimental Syrian Awassi rams using vacutainers (BD, Plymouth, UK) containing K_2E as an anticoagulant. Then, samples were immediately and directly transferred to the lab for determination of some blood components using a Veterinary Hematology Analyzer (Mythic™ Vet, Orphée, Geneva, Switzerland).

Parameters and statistical analysis

The following parameters were measured: WBC, LYM, MON, GRA, RBC, HGB, HCT, MCV, MCH, MCHC, RDW, PLT and MPV. Results were statistically analyzed and the differences among the means were calculated by ANOVA using Statview-IV programme on IBM system.

RESULTS AND DISCUSSION

This work gives, for the first time, values of 13 essential haematological parameters in Syrian Awassi rams throughout the year as reference intervals and is considered as a continuation to previous works to characterize this locally, regionally and internationally important Awassi sheep breed.

It is well known that blood tests and analyses are important diagnostic tools in animal diseases (Žaja et al. 2019). Blood contains 3 different types of cells namely red blood cells (RBC) or erythrocytes, white blood cells (WBC) or leukocytes or leucocytes and platelets or thrombocytes (Sahlol et al. 2020), and RBC form the majority of the blood cells (Aliyu et al. 2018).

Leukocyte parameters include WBC, LYM, MON and GRA; Erythrocyte parameters contain RBC, HGB, HCT, MCH, MCHC, RDW and MCV, whereas thrombocytes parameters consist of PLT and MPV.

Table 2 refers to the overall means of the haematological parameters for individual Awassi rams, whereas Table 3 shows the overall means of the studied components in the blood of the experimental rams during the different months of the year.

Leukocyte parameters

WBC counts. In the current study, the overall mean for WBC was 8.92 ± 5.05 $x10^3/mm^3$, ranging from 2.40 to 19.65 $x10^3/mm^3$ (Table 3), which is close to 9.8 $x10^3/mm^3$, reported in the blood of the Lika Pramenka ewes in Croatia (Vugrovečki et al. 2017). In addition, Rana et al. (2014) indicated that WBC counts in the blood of local Bangladeshi sheep subjected to 8 hours heat daily for 45 days were not affected ranging from 8.09 to 9.03 $x10^3/mm^3$ (mean value of 8.96 $x10^3/mm^3$) which is also close to our results, whereas in Poland, Wojtas et al. (2014) reported a decrease in WBC counts from 10.07 to 7.07 $x10^3/mm^3$ in the blood of Merino rams subjected to a heat exposure to about 30 ^oC and Temperature-Humidity Index (THI) of 78 for 7 days, as compared to 20 ^oC and THI of 70 for the control.

Results indicated that there was a significant ($P<0.05$) difference in the WBC counts between summer (5.04 and 4.16 $x10^3/mm^3$ for July and August) and winter (15.33 and 14.85 $x10^3/mm^3$ for January and February) months (Table 3).

LYM. The overall mean for LYM was 5.91 ± 3.92% ranging from 1.75 to 16.35% (Table 3), which was significantly ($P<0.05$) higher in winter (10.6 and 11.49% for January and February) as compared to 3.18 and 2.74% for July and August.

MON. The overall mean for MON was 1.00 ± 0.45%, with a significant ($P<0.05$) rise in winter (1.44 and 1.38% for January and February) as compared to 0.72 and 0.65% for July and August) (Table 3).

Table 2. Overall means (± SDs) for the haematological parameters in the Syrian Awassi rams used in the study

Ram No	Parameter	Overall Mean	Ram No	Parameter	Overall Mean	Ram No	Parameter	Overall Mean	Ram No	Parameter	Overall Mean
10	WBC ($x10^3/mm^3$)	11.86 + 4.63	20	WBC ($x10^3/mm^3$)	±5.33 8.10	30	WBC ($x10^3/mm^3$)	± 4.51 7.52	40	WBC ($x10^3/mm^3$)	± 4.61 8.21
	LYM (%)	8.41 ± 4.21		LYM (%)	5.4 ± 3.87		LYM (%)	4.25 ± 2.82		LYM (%)	4.92 ± 3.25
	MON (%)	1.35 ± 0.5		MON (%)	0.93 ± 0.45		MON (%)	0.79 ± 0.34		MON (%)	0.95 ± 0.27
	GRA (%)	3.78 ± 2.35		GRA (%)	1.92± 1.26		GRA (%)	2.33 ± 1.59		GRA (%)	2.53 ± 1.42
	RBC ($x10^3/mm^3$)	7.81 ± 0.63		RBC ($x10^3/mm^3$)	7.3 ± 0.99		RBC ($x10^3/mm^3$)	8.49 ± 0.68		RBC ($x10^3/mm^3$)	8.16 ± 0.51
	HGB (g/dL)	10.56 ±1.09		HGB (g/dL)	9.86 ± 1.41		HGB (g/dL)	10.65 ±1.02		HGB (g/dL)	9.93 ± 0.92
	HCT (%)	31.09 ±2.01		HCT (%)	31.0 ± 2.17		HCT (%)	31.78 ±2.29		HCT (%)	29.98 ±1.99
	MCV (fL)	40.15 ±2.07		MCV (fL)	41.3 ± 3.34		MCV (fL)	36.73 ± 1.69		MCV (fL)	36.08 ±1.99
	MCH (pg)	12.92 ±1.7		MCH (pg)	13.01 ±1.57		MCH (pg)	12.3 ± 1.27		MCH (pg)	12.04 ±1.18
	MCHC (%)	33.678 ±3.8		MCHC (%)	31.92 ±4.03		MCHC (%)	33.44 ±3.03		MCHC (%)	33.44 ±1.18
	RDW (%)	15.31 ±0.87		RDW (%)	16.16 ±1.53		RDW (%)	16.9 ± 0.93		RDW (%)	16.98 ±1.01
	PLT ($x10^3/mm^3$)	664.8 ± 132.1		PLT ($x10^3/mm^3$)	689.6 ±101.7		PLT ($x10^3/mm^3$)	598.7 ±135.1		PLT ($x10^3/mm^3$)	356.1 ± 119.4
	MPV (fL)	± 0.37 3.71		MPV (fL)	3.82 ± 0.33		MPV (fL)	3.96 ± 0.44		MPV (fL)	± 0.42 4.46

Table 3. Overall means (± SDs) and ranges for the haematological parameters in the Syrian Awassi rams during the months of the year

Parameter	MONTH												Overall Mean	Range
	January	February	March	April	May	June	July	August	September	October	November	December		
WBC ($x10^3/mm^3$)	15.33 ± 2.88	14.85 ± 2.41	13.31 ± 5.03	9.77 ± 3.87	7.70 ± 2.91	4.18 ± 1.88	5.04 ± 2.27	4.16 ± 1.53	4.77 ± 1.82	5.58 ± 1.84	11.878 ± 3.95	10.34 ± 4.41	8.92 ± 5.05	2.40-19.65
LYM (%)	10.6 ± 3.4	11.49 ± 2.74	8.48 ± 4.37	5.25 ± 2.42	4.25 ± 1.98	2.57 ± 0.96	3.18 ± 1.29	2.74 ± 0.84	2.95 ± 1.07	3.7 ± 1.36	8.16 ± 3.48	8.38± 2.58	5.91 ± 3.92	1.75-16.35
MON (%)	1.44 ± 0.52	1.38 ± 0.42	1.17 ± 0.38	0.95 ± 0.25	0.93 ± 0.25	0.72 ± 0.35	0.72 ± 0.21	0.65 ± 0.3	0.72 ± 0.19	0.9 ± 0.4	1.21 ± 0.5	1.26 ± 0.47	1.0 ± 0.45	0.3-2.65
GRA (%)	4.28 ± 0.98	4.29 ± 1.1	3.94 ± 1.17	3.09 ± 0.7	2.59± 0.84	0.98 ± 0.73	0.99 ± 0.6	0.88 ± 0.52	1.12 ± 0.79	1.5 ± 0.82	3.33± 1.51	3.53 ± 1.97	2.62± 1.8	0.2-8.7
RBC ($x10^3/mm^3$)	7.86 ± 0.67	7.8 ± 0.67	8.05 ± 0.78	8.06 ± 0.77	8.12 ± 0.84	7.75 ± 1.53	7.66 ± 0.99	7.67 ± 0.62	8.22 ± 0.35	8.1 ± 0.55	8.16 ± 0.95	7.9 ± 0.9	7.94 ± 0.85	5.15-9.96
HGB (g/dL)	11.27 ± 1.06	10.75 ± 0.82	11.06 ± 1.05	10.66 ± 0.78	9.95 ± 1.43	8.85 ± 1.23	9.42 ± 0.7	9.27 ± 0.46	9.96 ± 0.83	9.88 ± 0.68	11.09 ± 0.45	10.8 ± 0.9	10.24 ± 1.16	6.1-12.85
HCT (%)	30.24 ± 1.34	30.3 ± 1.83	30.84 ± 2.84	31.34 ± 1.92	31.18 ± 1.41	30.12 ± 1.71	31.18 ± 1.58	31.13 ± 2.1	31.56 ± 1.9	31.55 ± 3.79	31.03 ± 1.42	30.21 ± 2.39	30.93 ± 2.12	25.25-36.65
MCV (fL)	37.67 ±3.4	38.13 ± 2.9	38.43 ± 2.18	38.61 ±1.88	38.901 ±1.67	38.92 ±2.84	40.78 ±4.92	40.39 ±4.14	38.29 ± 0.58	37.97 ± 4.7	37.41 ± 2.43	37.3 ± 2.94	38.55 ±3.22	29.95-48.05
MCH (pg)	14.35 ±1.42	14.38 ±0.9	13.78 ± 0.89	11.96 ± 0.61	11.69 ± 0.51	11.3 ± 0.41	11.58 ± 0.79	11.34 ± 0.65	11.68± 1.81	11.9 ± 0.81	13.52 ± 0.81	13.44 ± 0.78	12.57 ± 1.49	7.45-15.9
MCHC (%)	38 ± 1.25	37.75 ± 0.92	35.27 ± 1.97	33.85 ± 1.16	32.08 ± 1.62	29.81 ± 1.88	30.13 ± 1.32	29.37 ± 1.14	29.3 ± 0.98	30.46 ± 2.54	35.63 ± 2.28	35.64 ± 1.79	33.1 ± 3.54	26.05-41.35
RDW (%)	16.66 ± 0.89	16.6 ± 0.94	16.14 ± 0.97	15.83± 0.54	15.83± 0.44	16.63 ± 2.07	16.28 ± 1.22	16.19 ± 1.05	16.03 ± 1.78	16.82± 2.47	15.88± 2.06	16.43± 0.62	16.34± 1.3	13.9-21.45
PLT ($x10^3/mm^3$)	619.1± 249.4	635.3 ± 244.4	553.9 ±192.8	569.3 ±190.5	540.7 ±176.4	525.3 ±182.3	583.9 ±189.4	519.8± 164.2	542.5± 122.6	529.3± 144.1	635.4± 108.0	645.5± 177.6	577.3± 179.8	227.5-978.5
MPV (fL)	3.8± 0.47	3.85 ± 0.62	3.9 ± 0.54	3.88 ± 0.43	4.1 ± 0.53	4.2 ± 0.38	3.94 ± 0.38	4.05 ± 0.27	3.98 ± 0.29	3.95 ± 0.65	4.0 ± 05	4.0 ± 0.56	3.99 ± 0.49	3.3-5.1

GRA. The overall mean for GRA was 2.62 ± 1.8%, ranging from 0.2 to 8.7%, with a significant (P<0.05) rise in winter (4.28 and 4.29% for January and February) as compared to 0.99 and 0.88% for July and August (Table 3).

Iyiola-Tunji et al. (2015) reported no effect of season on WBC parameters (WBC counts, LYM, Neutrophils, Eosinophils), except MON in lambs from the mixed flock of Balami, Uda, Yankasa sheep and their crosses in Nigeria, averaging 11.68 and 18.01 $x10^9$/L, for WBC and 55.79 and 53.33% for LYM during early dry and wet season, respectively.

Erythrocyte parameters

RBC Counts. The overall mean for RBC was 7.94 ± 0.85 $x10^3/mm^3$, ranging from 5.15 to 9.96 $x10^3/mm^3$ (Table 3), with no significant (P>0.05) differences among the means of the winter and summer months. Oluwatosin and Adebowale (2020) reported a mean value for RBC of 9.29 $x10^3/mm^3$ in the blood of West African Dwarf sheep given diet containing *Pennisetum purpureum* which is within our result.

HGB. The overall mean for HGB was 10.24 ± 1.16 g/dL, ranging from 6.1 to 12.85 g/dL, with a significant (P<0.05) rise in winter (11.27 and 10.75 g/dL for January and February) as compared to 9.42 and 9.27 g/dL for July and August (Table 3). Rana et al. (2014) reported a mean HGB value of 8.67 g/dL in the blood of local unstressed sheep in Bangladesh, which is within the range value of the results obtained in the current study.

HCT. Results indicated that the overall mean for the HCT was 30.93 ± 2.12%, ranging from 25.25 to 36.65% (Table 3), with no significant (P>0.05) difference in the value among the months of the year. Meneghini et al. (2016) reported that RBC and HTC value were higher in the blood of female Santa Ines lambs in Brazil as compared to the ewes, with a gradual decrease with advancing age.

MCV. The overall mean for MCV was 38.55 ± 3.22 fL, ranging from 29.95 to 48.05 fL, with a significant (P<0.05) decrease in winter (37.67 and 38.13 fL for January and February) as compared to 40.78 and 40.39 fL for July and August (Table 3).

MCH. The overall mean for this parameter was 12.57 ±1.49 pg, ranging from 7.45 to 15.9 pg with a significant (P<0.05) difference among summer and winter months (14.35 and 14.38 pg in January and February; and 11.58 and 11.34 pg in July and August) (Table 3).

MCHC. The overall mean for MCHC was 33.1± 3.54 g/dL, ranging from 26.05 to 41.35 g/dL with a significant (P<0.05) difference among summer and winter months (38 and 37.75 g/dL in January and February; and 30.13 and 29.37 g/dL in July and August) (Table 3).

RDW. The overall mean for RDW was 16.34 ± 1.3%, ranging from 13.9 to 21.45% (Table 3) with no significant (P>0.05) difference in the mean values among summer and winter months.

In summary, our results are in agreement with those reported by Vojta et al. (2011) in Croatia in female and male Dalmatian Pramenka sheep who reported 10 x 10^3/mm^3 for RBC, 10.2 g/dL for HGB, 1.73% for HCT, 31 fL for MCV, 10.1 pg for MCH, and 32.4 g/dL for MCHC; and with Ahmadi-Hamedani et al. (2015) in Iranian fat-tailed Sangsari sheep (HGB 5.4-12.52 g/dL, MCH 7.45-15.9 pg and MCHC 22.3-45.2 g/dL), close to the results reported by Vugrovečki et al. (2017) in the Lika Pramenka ewes in Croatia for the same parameters; and are within the results reported by Oluwatosin and Adebowale (2020) in West African Dwarf sheep in Nigeria fed *Pennisetum purpureum* diet (WBC: 7.26 x10^3/mm^3,RBC: 9.29 x10^3/mm^3, HGB: 8.12 g/dL, MCH: 8.74 pg and MCHC: 30.70 g/dL).

Thrombocyte parameters

Platelets Counts. Overall mean for Platelets Counts was 577.3 ± 179.8 x10/^{3}mm^3, ranging from 227.5 to 978.5 x10^3/mm^3 (Table 3), with no significant (P>0.05) difference among months of the year. This mean is within the range (50-865 x10^3/mm^3) reported by Vugrovečki et al. (2017) in the Lika Pramenka ewes in Croatia, and (42.1-725.7 x10/^{3} mm^3 in Sangsari sheep in Iran (Ahmadi-Hamedani et al. 2015).

MPV. Overall mean for MPV was 3.99 ± 0.49 fL, ranging from 3.35 to 5.1 fL (Table 3), with no significant (P>0.05) difference among months of the year.

Despite the low number of animals used as indicated above, the merit and strength of the current study that it lasted for one year with collections of hundreds of blood samples and assessment of 13 different parameters. Using haematological parameters as useful tools in animal production and health, Aruwayo et al. (2005) evaluated the haematological parameters (PCV, HGB, RBC, WBC, Neutrophils, LYM and MON, Eosinophils) in 2 blood samples collected from 20 Uda lambs fed 5 experimental diets (4 lambs/treatment) before the feeding trial and at the last week of the experiment which lasted 84 days. In another study, nine sheep were divided into three groups (3 animals/group) for 45 days, to study the effect of heat stress on 4 blood parameters (WBC, RBC, HGB and PCV) (Rana et al. 2014) and concluded that heat stress had significant changes on some blood parameters (RBC, HGB and PCV).

Finally, the haematological values obtained in the current study are within the sheep reference values reported by Jackson and Cockcroft (2002).

Conclusion

For the first time in Syria, this current study successfully defined and established reference values for 13 essential haematological parameters in Syrian Awassi rams throughout the year. However, as the number of this preliminary study is low, more animals and other parameters should be considered in future studies.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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