



Influence of Age, Parity, and Fetuses' Number on Blood Parameters in *Rembi* Ewes during Late Pregnancy in Semi-Arid zone, Algeria

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ABSTRACT

Several nutritional problems can be observed in ewes with advanced pregnancy when energy demands related to fetal development increase, then can hurt ewes and their lambs during this period. The aim of our study was the assessment of hematological and some biochemical parameters changes in ewes during late pregnancy and the influence of age, parity, and fetuses number on the latter. In this study blood samples were collected from forty ewes of the *Rembi* breed during the end of pregnancy, housed in the region of Tiaret, aged 2 to 6 years old. The choice of these ewes was made randomly. Our results recorded a decrease in the values of red blood cells (RBC), hemoglobin (Hb), hematocrit (Ht), Iron (Fe), and glucose (Glu), plus an increase in total white blood cells (WBC) compared to the range of reference. Statistical analyzes showed a significant influence of age on the RBC and Fe values with a highly significant influence of parity on the mean corpuscular volume (MCV) value. Our results recorded a positive correlation between (RBC, Hb, and Ht) and Glu value, whereas a negative correlation was found between WBC and platelets (PLA). Metabolic diseases due to nutritional problems are very important in ewes at the end of pregnancy, and then it is necessary to highlight the changes that can adversely affect lambing and lamb viability in this physiological stage.

Keywords: Biochemical parameters, Ewes, Hematological parameters, Late pregnancy

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INTRODUCTION

In Algeria, sheep farming contains around 28.69 million heads (FAOSTAT, 2018), which plays a very important role in the economy of the country. It is the first source of production of red meats followed by bovine, caprine, and camels at 60%, 30%, and 10%, respectively (MADRP, 2019). *Rembi* sheep is one of the important breeds in Algeria, reputed to be rustic, resistant to cold and drought, and adapt to living in the highlands conditions (Laoun *et al.*, 2015).

Late pregnancy is a critical period of the reproductive cycle of the ewe. It represents a physiological load on the ewes' body, which activates adaptation mechanisms to maintain normal homeostasis (El-Bassiouny *et al.*, 2018). During this period several changes in blood parameters can manifest such as hypoglycemia (Mohammadi *et al.*, 2016; Berkani *et al.*, 2018), hypoproteinemia (Ismaeel *et al.*, 2019), and hypocholesterolemia (Darwish & El Ebissy, 2019). This metabolic disorder can cause some of these diseases in this period (Greguła-Kania *et al.*, 2020).

Hematological changes observed during pregnancy due to the increase in energy demand caused by fetal development and lactogenesis, endocrine and metabolic changes, and preparation of the body for parturition and lactation are

among the indicators used to assess animal nutrition, health, and adaptation (Greguła-Kania *et al.*, 2020).

Erythrocytes and their hemoglobin contents play an important role in the maintenance of gestation and fetus' survival through to their role in oxygen transport (Greguła-Kania *et al.*, 2020). Iron is an essential element for erythropoiesis, the formation of hemoglobin, myoglobin, metalloenzymes, and cytochromes in mammals. Pregnant females are highly sensitive to iron deficiency in reason to their rapid expansion in body fluids and increased fetal growth (Wu, 2018).

Several studies showed that red blood cells decrease particularly during late pregnancy due to the hemodilution effect. It is the phenomenon that improves the diffusion of nutrients and oxygen toward the fetus (Habibu *et al.*, 2018). When this physiological anemia occurs during the last third of gestation, it results in a decrease in the number of red blood cells, a variation in the mean corpuscular volume, and an increase in the absolute value of the mean corpuscular hemoglobin concentration (Bezerra *et al.*, 2017).

Economically, twin pregnancies are more important than single ones, but twins are associated with decreased oxygen supply to the fetuses and them born with lower weight, resulting in higher mortality and lower postnatal growth (Sales *et al.*, 2018). On the other hand, Singh *et al.*, (2022) suggest that parity plays an important role in the variation of blood parameters of ewes at different physiological stages.

Therefore, this paper aims to present changes in hematological and some biochemical parameters in *Rembi* ewes during late

pregnancy, as well as the influence of some factors such as age, number of fetuses, and parity on the variations on these parameters.

MATERIALS AND METHODS

Animals

The herein study was carried out in the Tiaret region, when 40 ewes (14 primiparous and 26 multiparous) of *Rembi* Breed reached the last trimester of pregnancy, clinically healthy, with an average age of (4, 1 ± 1, 75) year, were chosen for the experiment work. After lambing, we reveal the number of fetuses of each ewe studied.

Samples

Blood samples were collected by vein puncture of the jugular vein during the morning before ewes feeding. Blood samples were collected in Ethylen Diamine Tetra Acetic Acid (EDTA) tubes for hematological analysis, and tubes containing heparin for biochemical analysis. They were transported to the Medical Hematology-Biochemistry Laboratory of Tiaret Institute of Veterinary Sciences, within approximately 2 hours.

Laboratory analyzes

The analyzed hematological parameters consisted of the following measurements: red blood cell (RBC), hemoglobin (Hb), hematocrit (Ht), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular volume (MCV), platelets (PLA), total white blood cells (WBC). These parameters were performed using a mythic 18 automat (Orphée ®). Blood smears for each studied subject were prepared on slides previously cleaned and then stained

with (RAL 555®) and observed under the immersion microscope (x100) (OPTIKA ®) for the determination of the leukocyte formula including lymphocytes (Lym), monocytes (Mono), neutrophils (Neu), eosinophils (Eos) and basophils (Baso). While biochemical parameters consisted of glucose (Glu), Iron (Fe), total protein (TP), and cholesterol (Chol), and they were measured by spectrophotometer (Optizen®) using (Spin React®) kits.

Statistical analysis

All results were expressed in terms of means, standard deviation (SD), maximum (Max), and minimum (Min) values of each hematology and biochemical parameters of the studied ewes. All of these data are subjected to a statistical analysis using the STATISTICA software (version 7, Statsoft, Tulsa, OK). The comparison of means was performed using the one-way ANOVA test. The latter was supplemented by the Duncan test. A value of $P \leq 0.05$ was retained as the significance level. Pearson's simple correlation method was used to determine the correlations between some parameters. The level of significance was set at $P \leq 0.05$.

RESULTS AND DISCUSSION

The means of RBC, Hb, and Ht obtained in our study were lower compared to the reference range of Kramer *et al.*, (2006). In contrast, the WBC mean was higher when compared to these references. Our results showed that the Cholmean of ewes at the late pregnancy greatly increased compared to the reference values as long as TP and Glu were within the reference range (Table 1).

Table 1. Hemato-biochemical parameters values of ewes during late pregnancy

Parameters / (n=40)	Min	Mean	SD	Max	References
WBC ($\times 10^3$ /mm ³)	3500	23822,5	11540,8	59100	4–12 ^a
Lym ($\times 10^3$ /mm ³)	262	3632,9	2222,8	9761	2–9 ^a
Mono ($\times 10^3$ /mm ³)	1200	7330,5	4120,1	25680	0–0.75 ^a
Neu ($\times 10^3$ /mm ³)	1125	11551,8	7818,1	34050	0.7–6 ^a
Eos ($\times 10^3$ /mm ³)	0	793,4	611,7	2916	0–1.0 ^a
Baso ($\times 10^3$ /mm ³)	0	322,9	317	2256	0–0.3 ^a
N/L ($\times 10^3$ /mm ³)	0,32	3,17	5,94	37,5	/
RBC ($\times 10^6$ /mm ³)	5,01	8,08	1,2	14,14	9–15 ^a
Hb g/dl	4,9	8,44	1,45	15,8	9–15 ^a
Ht %	18,1	26,59	3,72	47,7	27–45 ^a
MCV fl	28,3	32,99	1,83	37,8	28–40 ^a
MCHC %	27,1	31,56	1,2	38,2	31–34 ^a
PLA (mm ³)	244	759,08	292,49	2091	100–800 ^a
Glu (g/dl)	0,18	0,44	0,2	0,89	0.50 - 0.80 ^b
TP (g/dl)	4,95	6,13	0,76	10,01	6 - 7,9 ^b
Fe (mg/dl)	0,02	0,14	0,04	0,33	0.33 - 0.45 ^b
Chol (g/dl)	0,34	1,11	0,36	2,33	0.52 - 0.71 ^b

Key: a= references of Kramer *et al.*,(2006), b= references of Kaneko *et al.*, (2008).

This study recorded a significant influence (P ≤ 0.05) of age on RBC, TP, and Fe, while the parity registers a highly significant

difference (P < 0.001) on MCV, PLA, and Glu values (Table 2).

Table 2. Mean and P-value of age, parity, fetuses' number, and viability on hemato-biochemical parameters of ewes during late pregnancy

Parameters	Age (year)			P value	Parity			No of fetuses		
	Mean				Mean			Mean		
	2 (n=14)	4 (n=10)	6 (n=16)		Primiparous (n=14)	Multiparous (n=26)	P value	1 Fetus (n=34)	2 Fetuses (n=6)	P value
WBC (×10 ³ /mm ³)	17643	28370	28063	NS	17643	27150	0,046	23588	25150	NS
Lym (×10 ³ /mm ³)	2599,7	4133,5	4157,6	NS	2599,7	4189,2	NS	3843,3	2440,8	NS
Mono (×10 ³ /mm ³)	7310,1	10915	6312,8	NS	7310,1	7341,5	NS	7506,4	6333,8	NS
Neu (×10 ³ /mm ³)	6674,1 a	12677	15687 b	a,b 0,02	6674,1	14178	0,014	10788	15879	NS
Eos (×10 ³ /mm ³)	774,5	728,3	900,88	NS	774,5	803,62	NS	897,94	201,17	0,041
Baso (×10 ³ /mm ³)	284,43	237,5	426,88	NS	774,5	803,62	NS	337	243,5	NS
RBC (×10 ⁶ /mm ³)	8,6621 a	7,2630 b	8,12	a,b 0,05	8,66	7,77	NS	8,25	7,16	NS
Hb g/dl	8,78	7,62	8,69	NS	8,78	8,26	NS	8,58	7,67	NS
Ht %	27,15	24,56	27,34	NS	27,15	26,28	NS	26,93	24,65	NS
MCV fl	31,371 a	33,950 b	33,637 c	a,b 0,0012 a,c 0,0030	31,37	33,86	0,00039	32,71	34,58	0,05
MCHC %	32,14	30,85	31,63	NS	32,14	31,25	NS	31,68	30,88	NS
PLA (×10 ³ /mm ³)	1059,0 a	642,30 b	580,19 c	a,b 0,0018 a,c 0,0006	1059	597,58	0,00018	792,41	570,17	NS
Glu (g/l)	0,57 a	0,31 b	0,40 c	a,b 0,004 a,c 0,044	0,57	0,37	0,006	0,47	0,31	NS
PT (g/l)	5,65 a	6,07	6,51 b	a,b 0,05	5,66	6,39	0,04	6,18	5,83	NS
Fe (mg/dl)	0,16 a	0,16	0,11 b	a,b 0,047	0,16	0,13	NS	0,15	0,13	NS
Chol (g/l)	1,18	0,89	1,13	NS	1,18	1,08	NS	1,18	0,74	0,026

Key: (P < 005) significant, (P < 001) very significant, (P < 0001) highly significant

In this study's ewes, we found a positive correlation between (RBC, Hb, and Ht) and Glu values. On the other hand, a negative

correlation was recorded between PLA and MCV and between PLA and WBC (Table 3).

Table 3. The correlation between parameters using Pearson's correlation test

	RBC	Hb	Ht	MCV	MCHC	PLA	WBC	Lym	Mono	Neu	Eos	Baso	Glu	PT	Fe	Chol	
RBC	r	1	,9533	,9561	-,1969	,6007	,1427	,3226	-,0215	,3000	,2586	,2973	,2036	,5334	-,0035	,0590	,1942
	P		p=0,00	p=0,00	p=,223	p=,000	p=,380	p=,042	p=,895	p=,060	p=,107	p=,062	p=,208	p=,000	p=,983	p=,718	p=,230
Hb	r	1	,9829	,0440	,6909	-,0589	,4075	-,0356	,3375	,3679	,2837	,2490	,4257	-,0207	,0693	,2589	
	P		p=0,00	p=,787	p=,000	p=,718	p=,009	p=,827	p=,033	p=,020	p=,076	p=,121	p=,006	p=,899	p=,671	p=,107	
Ht	r	1	,0957	,5518	-,0848	,4276	,0135	,3336	,3816	,2990	,2890	,4545	,0011	,0394	,2346		
	P		p=,557	p=,000	p=,603	p=,006	p=,934	p=,035	p=,015	p=,061	p=,071	p=,003	p=,995	p=,809	p=,145		
MCV	r	1	-,2149	-,7433	,3192	,0716	,1227	,3800	-,0138	,2857	-,2802	-,0394	-,0804	,1034			
	P		p=,000	p=,000	p=,006	p=,071	p=,035	p=,015	p=,061	p=,071	p=,003	p=,995	p=,809	p=,145			

	P		p=,183																		
	r	1	,1115	,2430	-,2155	,2865	,2434	,1567	,0088	,1467	-,1677	,1848	,2344								
MCHC	P		p=,494	p=,131	p=,182	p=,073	p=,130	p=,334	p=,957	p=,366	p=,301	p=,254	p=,146								
	r	1	-,3447	-,0376	-,0726	-,4510	-,0232	-,2085	,1785	-,1022	,2164	-,1221									
PLA	P		p=,029	p=,818	p=,656	p=,003	p=,887	p=,197	p=,271	p=,531	p=,180	p=,453									
	r	1	,2198	,7965	,9243	,5365	,5840	,0376	-,1670	-,0745	-,2890										
WBC	P		p=,173	p=,000	p=,000	p=,000	p=,000	p=,000	p=,818	p=,303	p=,648	p=,070									
	r	1	-,0607	-,0219	,5612	,2817	,0520	,2112	,0917	-,0226											
Lym	P		p=,710	p=,893	p=,000	p=,078	p=,750	p=,191	p=,574	p=,890											
	r	1	,6396	,3690	,4493	,1041	-,2960	,0021	-,2865												
Mono	P		p=,000	p=,019	p=,004	p=,523	p=,064	p=,990	p=,073												
	r	1	,3008	,4508	-,0648	-,1690	-,1462	-,2743													
Neu	P		p=,059	p=,004	p=,691	p=,297	p=,368	p=,087													
	r	1	,4942	,1019	-,0250	-,0020	-,1299														
Eos	P		p=,001	p=,532	p=,878	p=,990	p=,424														
	r	1	,1588	-,0650	-,0758	-,1939															
Baso	P		p=,328	p=,690	p=,642	p=,231															
Glu	r	1	,1069	-,1786	,2701																

	P		p=.511		
	r		1		
PT				p=.270	
	P				p=.092
	r				
Fe				p=.430	
	P				p=.238
	r				
	P				p=.698
	r				
Chol					1
	P				

Statistical significance of correlations was recorded at (P < 005)

The mean of RBC reported in the present study was lower than the reference range of Kramer *et al.*, (2006), but it approaches that of Plaza Cuadrado *et al.*, (2019) in Colombia in ewes at the end of gestation of the *Pelo Criollos* breed. El-Sayed *et al.*, (2020) in Egypt recorded a very low mean of RBC in *Barki* ewes during the last stage of pregnancy compared to the mean reported in our results.

The Hb value recorded in ewes at the end of gestation in our study shows a decrease compared to the value reported in ewes during late pregnancy in the study by El-Malky *et al.*, (2019) in Egypt in the *Barki* and *Ossimi* breed, while Kandiel *et al.*, (2016) recorded a lower mean of Hb in the *Barki* breed compared to the mean obtained in this study. We recorded a lower mean of Ht in pregnant ewes in the last stage compared to the mean obtained by Sharma *et al.*, (2015) in India in the breed of *Himalayan Gaddi* and Zvonko *et al.*, (2015) in Croatia in breed *Dubrovnik* and breed *Zeta zuja*.

The decrease in the values of RBC, Hb, and Ht in this study could be due to hemodilution. This is a physiological response to the decrease of blood viscosity, to improve blood supply to small vessels and those newly formed in the maternal uterus and placenta (Yaqub *et al.*, 2021). This agrees with the study of Greguła-Kania *et al.*, (2020) in Poland, which recorded a significant decrease (P ≤ 0.05) in RBC and Ht values in ewes during late gestation compared to the pre-gestation period. The decrease of red blood count values in *Rembi* ewes during late pregnancy in our study could also be due to the metabolic diseases which can be installed in ewes during this period, such as the clinical and sub-clinical pregnancy toxemia, the study of Marutsova and Marutsov, (2015) in Bulgaria in *Lacaune* ewes with clinical and subclinical pregnancy toxemia recorded a significant decrease (P ≤ 0.05) in red blood counts (RBC, Hb, and Ht), which could be associated with hypoglycemia and ketogenesis linked to this pathology. This was very clear in our study in which we recorded a decrease in RBC, Hb, and Ht with a positive correlation between these values and glucose (**Table 3**). This indicates that the hypoglycemia encountered in ewes at the end of gestation following the increase in energy

requirements could be the cause of anemia in these ewes during this period.

In our study, the values of MCV and MCHC were within the standard references of Kramer *et al.*, (2006) regardless of the decrease in the values of RBC, Hb, and Ht. MCV means obtained in this study were lower than that of Zvonko *et al.*, (2015), and higher than that of Bezerra *et al.*, (2017). According to the study by Greguła-Kania *et al.*, (2020), Hb, MCV, and MCH values were significantly elevated (P ≤ 0.05) during the peripartum period despite the decrease in the number of erythrocytes. The authors related the increase of hemoglobin concentration in erythrocytes to the organism's compensation for RBC deficiency, which is a physiological response that preserves oxygen content in the blood (Greguła-Kania *et al.*, 2020).

In our ewes, the MCV value was influenced by the number of fetuses and highly influenced by age and parity with a positive correlation with the WBC and Lym values (**Table 3**).

In addition, the PLA value was highly influenced by age and parity (P < 0.001) registering a negative correlation with WBC and Neu, it also registered another negative correlation with MCV. Along with this result, the mean of PLA recorded in ewes during late pregnancy in our study was lower than that obtained by Plaza Cuadrado *et al.*, (2019), and was in the reference value of Kramer *et al.*, (2006). We found that this means was highest compared to that obtained by Sharma *et al.*, (2015) in ewes in the last third of their pregnancy period. According to Plaza Cuadrado *et al.*, (2019), the thrombocytosis encountered in pregnant ewes could be due to a physical activity that stimulates the secretion of epinephrine by causing splenocontraction, and consequently, mobilization of the stored platelets can be toward the blood circulation. This could be due to a hyperproduction of thrombopoietin which occurs after or in association with a bleeding process, particularly when bleeding causes iron deficiency anemia. Also, in case of serum iron losses caused by gastrointestinal parasites, chronic anemia can set in these animals causing secondary thrombocytosis. Therefore, we can link the average platelets encountered in ewes at the end of pregnancy in our study to the decrease in iron which marked a much-reduced mean.

The value of WBC obtained in our study was higher than the reference value, and than those obtained in pregnant ewes by Soliman (2014) in Egyptian ewes during the late pregnancy of *Ossimi* Breed, Bezerra et al., (2017) and Greguła-Kania et al., (2020). Among the different types of leukocytes, the mean of neutrophils was highest in our study, followed by the mean of monocytes. The relationship between granulocytes/lymphocytes can be used as a physiological indicator of stress. Our study recorded a high ratio of neutrophils /lymphocyte (N / L) in pregnant *Rembi* ewes, and this is consistent with the study of Soliman (2014), and Sharma et al., (2015), and Greguła-Kania et al., (2020).

According to Soliman (2014), the increase in the N / L ratio could be because the stress of gestation stimulates the anterior pituitary gland to secrete ACTH, which in turn induces the adrenal cortex to produce glucocorticoids, by causing mobilization of neutrophils from the body pool to the peripheral circulation. Greguła-Kania et al., (2020) also attributed the decrease in lymphocytes and the increase in granulocytes at the end of gestation and postpartum to the release of cortisol in case of metabolic stress related to gestation and lactation period. We recorded a significant influence of parity on the WBC values, it was very lower in multiparous ewes compared to the primiparous.

Glycemia mean obtained in pregnant *Rembi* ewes in the last stage of pregnancy was greatly reduced compared with those obtained in ewes at the end of gestation by Berkani et al., (2017) in Algeria in the *Ouled djellal* breed, Mohammadi et al., (2016) in Iran in the *Mekouei* breed, Ismaeel et al., (2018) in Iraq among the *local* breed. According to these authors, the decrease of glycemia in ewes during late gestation seems to be related to the increase in the need for the fetoplacental unit in glucose, because 70% of fetal growth takes place during the last six weeks of pregnancy. This is contrasting with the study of Kandiel et al., (2016) in Egypt which recorded an increase in the serum glucose value in pregnant ewes during this period. Kandiel et al., (2016) explained this increase in glucose by the decrease in insulin levels which could be granted to the negative energy balance, and this promotes lipolysis and displacement of energy substrates away from the stores of adipose tissue. This is because the mobilization of stored energy at the end of gestation is essential to assemble the needs of fetal growth and mammary gland development. In our results, the influence of parity on the blood glucose value was very significant ($P < 0.01$). It was significantly reduced in multiparous compared to primiparous, these results agree with those obtained by Damián et al., (2020) in Uruguay.

The mean value of TP in ewes at the end of gestation in this study was within the reference standards of Kaneko et al., (2008). This value was higher than that of Ismaeel et al., (2018) in Iraq, and lower than that of Mohammadi et al., (2016) in Iran. According to Ismaeel et al., (2018), hypoproteinemia in ewes during late pregnancy could be due to embryonic growth, which leads to the consumption of large amounts of maternal amino acids to maintain fetal muscle development and the transmission of immunoglobulins from the blood plasma to the mammary gland for the production of colostrum during the last trimester of gestation. Mohammadi et al., (2016), however, linked the increase in serum TP concentration to the formation of globulin which is triggered in the three to four weeks before parturition to produce

colostrum rich in antibodies. In our study, plasma TP concentration was significantly ($P < 0.05$) increased in ewes aged 6 years compared to those having 2 years, and significantly increased in multiparous compared to primiparous ewes.

The recorded iron mean in our ewes was lower compared to the reference value, and to those obtained by Teleb et al., (2019) in Egypt in pregnant ewes in the last thirds of the *Saidi* breed, and Cihan et al., (2016) in Turkey in ewes of the *Ivesi* breed. Cihan et al., (2016) have associated the decrease in plasma iron concentration at the end of gestation with fetal consumption of this mineral. According to the study by Darwish and El Ebissy, (2019) in Egypt, iron concentration was significantly reduced in healthy pregnant *Barkie* ewes at the end of gestation compared to ewes from the control group (not pregnant), and compared to those with pregnancy toxemia. These authors have linked hypoferrremia and hypoferritinemia (depletion of iron stores) in ewes during late pregnancy to increased fetal demand and increased red mass at this moment. While they attributed the hyperferremia in ewes with pregnancy toxemia to liver damage associated with this disease and the dependent release of Kupffer cells that store iron, this hypothesis was confirmed by the positive correlation recorded between liver enzymes and iron in their study. Whereas in our study the iron value was significantly influenced ($P \leq 0.05$) by age of ewes, when it was decreased in old (6 years) ewes compared to those having 2 years (**Table 2**).

Our results showed that the plasma cholesterol mean recorded in ewes at the end of gestation was higher compared to the range of references, and to that recorded in ewes at the end of gestation by Berkani et al., (2018) in Algeria. However, it was lower than the value recorded by El-Bassiouny et al., (2018) in Egypt in *Barki* ewes in the last third of their pregnancy.

The increase in cholesterol in this study could be linked to the decrease in the sensitivity of adipose tissue to the action of insulin which decreases in ewes at the end of gestation (Mohammadi et al., 2016). The study of El-Malky et al., (2019) in Egypt did not record a significant influence of the physiological stage on cholesterol. In our results, the cholesterol values were significantly decreased ($P \leq 0.05$) in ewes with 2 fetuses compared to those having a single fetus.

CONCLUSION

The decrease in RBC, Hb, Ht, and iron values in this study indicates that *Rembi* ewes in late gestation are subjected to develop anemia. The energy deficit that develops in ewes at the end of pregnancy can induce some changes in hematological parameters and thus causing a decrease in the indices of red blood count. In our study, age and parity are the factors that can most influence the hematological and biochemical parameters in ewes during late gestation. To prevent the apparition of anemia in pregnant ewes, the energy status of these ewes should be improved in the quantity and quality of distributed food. It is also essential that the various parasitic infections that can worsen anemia in ewes during this period are tackled.

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ETHICS STATEMENT: All experiments were complied with the Algerian legislation (Law number 95-322/1995) inherent to protect animals designed for experiments or other scientific purposes, also the guidelines of the Algerian Association of Experimental Animal Sciences (AASEA approved under the agreement number 45/DGLPAG/DVA/SDA/14).

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