

## Serum progesterone level of camel (*Camelus dromedarius*) according to the physiological status

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**Summary** - The present work was planned in the Department of Animal Production, Higher School of Agriculture, Mateur. Analysis of progesterone level in blood samples can be a pathway to define the physiological status of females. The progesterone level measurement can indicate an early pregnancy. 13 females and 01 male were included in this study, out of which primiparous, multiparous and heifers. In total 364 blood samples were collected, along one year, every 10 days. The blood samples were used to establish the pregnancy diagnosis. Serum progesterone levels were determined by radioimmunoassay. For gravid and empty females, the averages of serum progesterone were respectively  $0.13 \pm 0.17$  g/L and  $3.99 \pm 1.26$  g/L respectively. For growing female (category 1), young pregnant females (category 2), empty multiparous females (category 3) and pregnant multiparous females (category 4) were respectively  $0.14 \pm 0.19$  g/L,  $4.25 \pm 1.31$  g/L,  $0.12 \pm 0.13$  g/L and  $3.55 \pm 0.87$  g/L. Progesterone hormone is even in growing females, and its concentration started to rise after successful mating. Statistical analysis, with General Linear Model procedure and Multiple linear regression models, indicates a significant effect ( $P < 0.001$ ) of age, physiological status, pregnancy status on serum progesterone level and significant differences in serum progesterone levels between category ( $p < 0.001$ ). The reproductive variables means value for serum progesterone in analyzed individuals, were significantly higher in the young pregnant females, pregnant multiparous females than those for growing female and empty multiparous females. Progesterone level for all analyzed empty female always low for all experimental period. It started to rise again after successful mating and remained high until the end of the pregnancy. Monitoring serum progesterone level can be a useful pathway to monitor pregnancy in camels.

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**Keys words :** Serum progesterone, pregnancy status, camels

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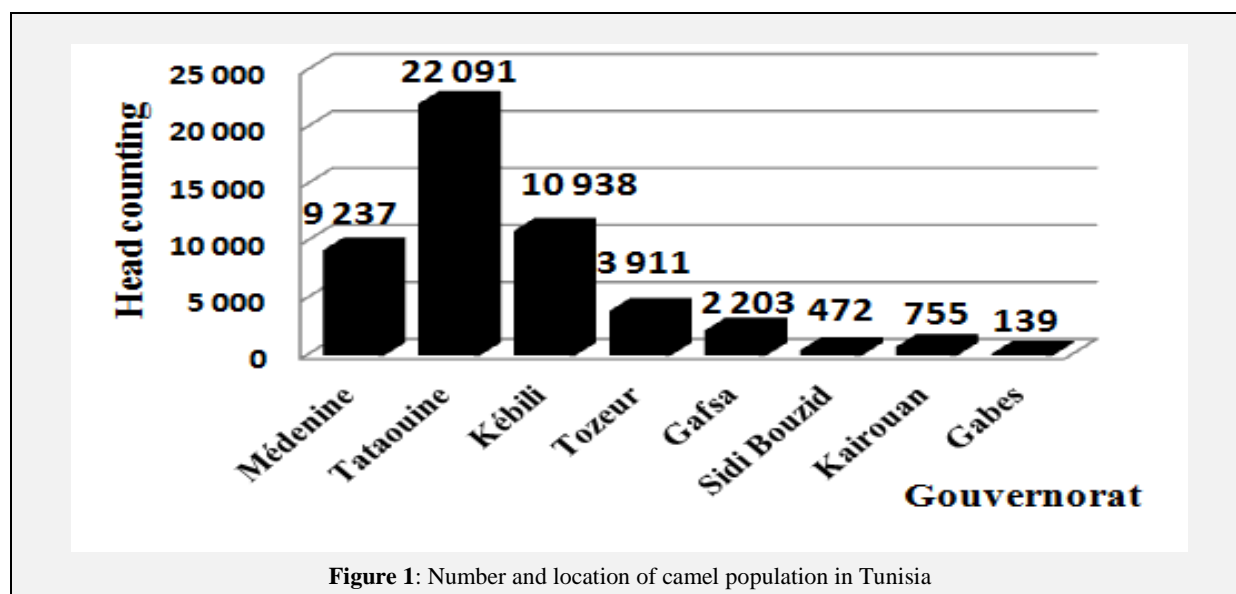


## 1. Introduction

The level of fertility in domestic animals results from a number of interacting factors, some of which have a genetic basis while others are environmental in origin. The systems reproductive physiology of some animals has attracted considerable studies, although certain aspects of reproductive performance for males and females, especially fertility, are inadequately understood. For camel, it is known that the female is an induced ovulation. Adult camel male is characterized by an intermediate penis type, between the vascular and fibrous types. Generally, dromedaries give one calf per birth (Kamoun and Wilson, 1994; Tibary et al., 1997; Nikjou et al., 2008). The reproductive physiology of camel has been incompletely characterized. Most literature describe general characteristic such as occurrence, growth, maturation, diets and pregnancy rate (Hammadi et al., 2001; Ayoub et al., 2003; Deen, 2008; Babiker et al. 2011; Kamoun and Jemmali, 2012 ). The dromedaries' reproductive efficiency is generally

characterized that low under natural conditions. The short breeding season, the long prepubertal period, the long gestation period, the prolonged period of lactation (anestrous of 8–10 months) and the long inter-calving interval are the principal causes for these low performances. (Babiker et al., 2011; Kamoun and Jemmali, 2012).

FAO statistics (2004), reported that the number of camel in world was about 19 million, this number was founded essentially in Africa and Asia with 15 million and 4 million respectively. African populations represent 79% of world camels. All of these animals were one humped and are concentrated in Nord-East of Africa. In Egypt, their numbers were previously estimated as 230.000 camels. In Tunisia, there are about 100.000 camels (*Camelus dromedarius*). This estimated number was distributed especially in southern and center of Tunisia (figure 1). Despite this important number, there is a little information on genetic diversity of this population and no clear classification for camels exists.



Camel known as a beast of burden in the desert is presently being transformed into a multi-utility animal providing draft power, patrolling international borders, production of commodities like hair, meat and milk. Study of low fertility level of camel can be a way to improve reproductive performances for this animal. The objective of this study is to estimate the serum progesterone level

according to physiological status of Maghrabi She-camels.

## 2. Materials and methods

### 2.1. Study site

These studies were undertaken at the experimental station of the High School of Agriculture at Mateur (37°02'50"N, 09°37'22"E) in northern Tunisia. Mateur is



situated 60 km north-west of Tunis, is a few meters above sea level, and has a typically Mediterranean climate characterized by a hot dry summer and a mild moist winter. Average rainfall is about 450 mm, 65% of which falls in October-February. Highest monthly maximum temperatures of 32.3°C occur in July, with a lowest maximum of 14.3°C in January. Lowest monthly minimum temperatures of 6.0°C occur in January with the highest monthly minimum of 19.1°C in August.

## 2.2. Animals

A total of 13 females and one fertile male camel (*Camelus dromedarius*) were used in this experiment to study the variation of serum progesterone level. All animals are of Maghrabi breed, born and raised in the experimental station (Table 1).

They are weighed every 14 days, linear measurements are carried out monthly and all events related to female reproduction are recorded. At blood sampling startup (9 July) there were six pregnant females, who gave birth during the experiment, five females had a successful mating amid of experience (in winter) and the remaining were 16 months old at the sampling end (July 7). For pregnant females, eight were in first-pregnancy, mated at 25.0±4.0 months old which corresponded to 68.8±7.5% of mature weight and three in the second pregnancy mated at 56.3±8.8 months

old which corresponded to 90.7±4.5 % of mature weight. Documents relating to the reproduction of these 11 female, mating and calving situated the average length of gestation at 367 ± 9 days. The animals characteristics are summarized in Table 1.

The samples belong to she-camels in different pregnancy stages, to young female (pre-pubertal or pubertal), but also nursing females. Thus Four groups were formed: growing female (category 1), young pregnant females (category 2), empty multiparous females (category 3) and pregnant multiparous females (category 4).

## 2.3. Blood sampling

Blood samples (10 ml) were collected individually from the jugular vein of all animals in the experiment morning before feeding every 10 days. Blood samples were placed on ice during collection and stored at + 4 °C for 6 hours in refrigerator, then serum was separated by centrifugation at 3000 rpm for 20 minutes and stored frozen at - 20 °C till analysis. Samples collected in August were badly frozen and lost for analysis, thus the progesterone was determined in three hundred sixty-four sample of blood serum taken from thirteen non-pregnant, early, mid, and late pregnant, as well as Pre-Pubertal she-camels, at regular intervals throughout one year.

Table 1: Characteristics of analyzed animal																	
Sampling		Characteristics	Female													Male	
Day	Date		106	204	205	304	305	402	403	501	503	504	506	601	602		
0	09-July	Age (months)	64,3	53,1	52,6	38,5	35,2	26,8	26,3	18,6	18,4	18,2	13,9	5,9	4,1	67	
		Weight (kg)	522	467	463	534	504	431	379	280	292	286	225	164	142	665	
		Status	Pregnant	Empty	Pregnant	Pregnant	Pregnant	Pregnant	Pregnant	Pregnant	Growth	Growth	Growth	Growth	Growth	Growth	
		Stage (day)	29	-	187	149	180	149	117	-	-	-	-	-	-	-	
		Age (months)	70,2	59	58,5	44,4	41,1	32,7	32,2	24,5	24,3	24	19,8	11,8	10	73	
180	4-January	Weight (kg)	573	482	441	598	558	557	487	349	350	359	286	261	244	655	
		Status	Pregnant	Pregnant	Suckling	Pregnant	Pregnant	Pregnant	Pregnant	Pregnant	Pregnant	Pregnant	Growth	Growth	Growth	Growth	in rute
		Stage (day)	208	1	9	328	359	328	296	26	27						
		Age (months)	76,3	65,1	64,5	50,5		38,8	38,2		30,4	30,1	25,8		16	79	
		Weight (kg)	532	546	431	488	Died-16/02	534	489	Died-06/02	481	429	336	Died-06/02	324	710	
363	07-July	Status	Suckling	Pregnant	Empty	Empty		Empty	Empty		Pregnant	Pregnant	Pregnant		Growth		
		Stage (day)	22	185	-	-		-	-		211	149	62		-		

### 2.4. Serum Progesterone RIA

Progesterone assay was performed using kits supplied by the Joint FAO/IAEA through the CRP D31017 titled "INTER-REGIONAL RESEARCH NETWORK FOR IMPROVING THE PRODUCTIVITY OF CAMELIDS". The sensitivity of this method was 0.09 ng/mL and the inter- and intra-assay coefficients of variation were 9.2 and 11.0%, respectively.

### 2.5. Statistical analysis

The means procedure in SAS (Version 9.1; SAS Institute, 2003) was used to determinate average serum progesterone level for each category. The multiple linear regression models was used to estimate difference if exist between level progesterone by category. The GLM procedure was used for statistical analysis. Differences in serum concentrations of progesterone were examined between she-camel females the by General Linear Model (GLM) procedures of the SAS. The effect of a probability value of 0.05 was used as an indicator of statistical significance among comparisons.

### 3. Results and discussions

The serum progesterone level was determined in three hundred sixty-four blood sample. Serum progesterone level mean detected according female physiological status was summarized in Tables 2 and 3. In total, 190 samples to different physiological stages of 13 empty females (Table 2), young (pre-pubertal and pubertal) but also older in postpartum period (lactating or dry), that the progesterone is undetectable in 9% of samples, these negatives assays coincides with postpartum periods. The empty postpartum camels have plasma progesterone concentrations varying between 0 ng/ml and 0.41 ng/ml and means varying from zero to  $0.38 \pm 0.04$ . Serum progesterone concentrations of the empty postpartum camels were varying between 0 ng / ml and 0.41 ng / ml with an average of  $0.38 \pm 0.04$ . Lactating status have a significant effect on serum progesterone level. Considered separately the mean plasma progesterone of five lactating females is  $0.17 \pm 0.12$  ng / ml, this rate is higher than that of non-lactating she-camels that had parturated ( $p < 0.01$ ).

**Table 2:** Plasma progesterone levels for different physiological status (pre-pubertal, pubertal and lactating empty camels)

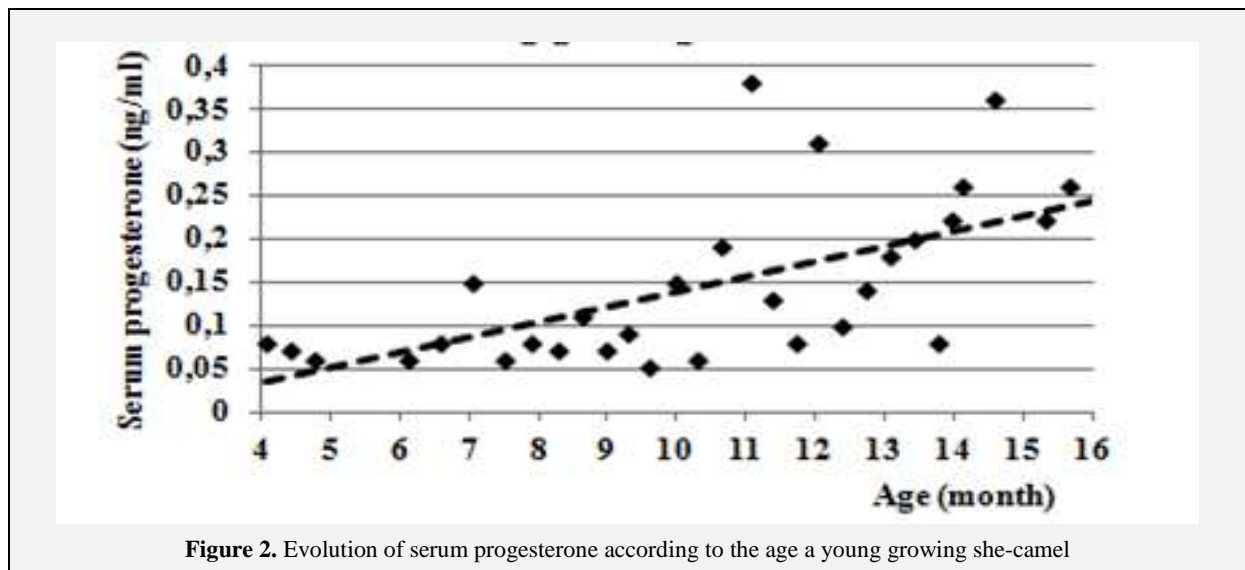
She-camel		Total samples	physiological stage	Serum progesterone (g/L)			
N°	Age (Month)			Mean	SD	Min	Max
106	75.92±0.35	3	Lactating	0.38	0.04	0.34	0.41
204	56.22±1.87	13	non lactating	0.01	0.02	0	0.07
205	61.41±1.85	18	Lactating	0.29	0.07	0.14	0.40
304	48.00±1.56	15	Lactating	0.06	0.03	0	0.11
305	42.16±0.36	3	non lactating	0	0	0	0
402	36.15±1.56	15	Lactating	0.08	0.06	0.02	0.17
403	36.44±1.11	10	Lactating	0.11	0.06	0.02	0.17
501	21.32±1.72	11	growing female	0.22	0.08	0.15	0.37
503	21.12±1.72	11	growing female	0.17	0.17	0.02	0.65
504	22.02±2.18	17	growing female	0.24	0.42	0	1.51
506	19.39±2.94	26	growing female	0.04	0.04	0	0.19
601	9.79±2.18	17	growing female	0.13	0.04	0.05	0.22
602	10.49±3.40	31	growing female	0.15	0.09	0.05	0.38
The Total	29.75±18.49	190	Empty	0.13	0.17	0	1.51

**Table 3:** Serum progesterone levels in camels after a successful mating and during pregnancy

She-camel		Total samples	Duration from the mating date (day)		Serum progesterone(g/L)			
N°	Age (Month)		Bottom	Top	Mean	SD	Min	Max
106	70.20±3.10	28	29	348	3.02	0.73	1.56	4.61
204	62.11±1.75	18	1	185	3.71	1.06	0	5.09
205	55.31±2.01	15	187	355	4.03	0.86	2.03	5.27
304	42.21±2.10	16	149	349	2.96	0.51	2.16	3.75
305	38.70±2.02	15	180	369	3.17	0.49	2.31	4.06
402	30.51±2.10	16	149	349	4.07	0.91	2.13	5.91
403	30.89±2.52	21	117	369	4.80	0.82	3.26	6.32
501	24.71±0.65	6	6	59	5.30	0.40	4.85	5.82
503	26.91±2.04	20	7	211	5.34	1.77	2.73	9.78
504	27.64±1.42	14	8	149	4.39	1.14	2.17	5.78
506	24.93±0.78	5	4	62	4.25	0.66	3.36	4.99
The Total	43.11±16.70	174	1	369	3.99	1.26	0.00	9.78

The serum progesterone concentrations tend to increase with age in younger she-camels (Figure 2). At this point the average level of progesterone, in she-camels N°602 is about

0.08±0.03 ng/ml at 7.6±2.0 month of age and 0.21±0.08 ng/ml towards 13.2±1.7 months of age with a highly significant difference ( $p < 0.01$ ).



On the other hand there are 174 samples taken from 11 she-camels after a successful mating and/or during pregnancy (Table 3), there is only one negative sample of female N ° 204 matching one day posts mating. Progesterone

assays indicated a significant rise ( $\geq 2.96$  ng/ml) in progesterone 2 days after successful mating in all female (Figure 3), and found its basic level at calving day (Figure 4).



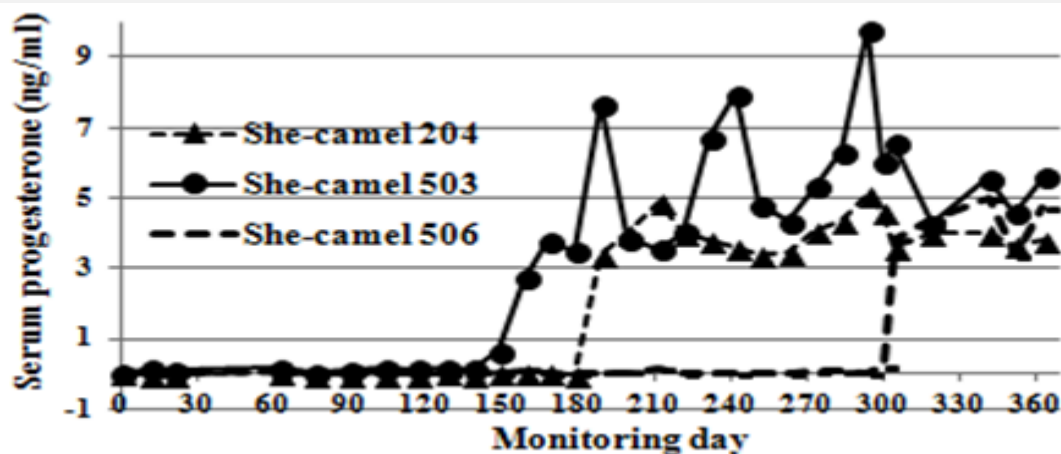


Figure 3. Progesterone level evolution after a successful mating of she-camel

Samples, from she-camel 402 and she-camel 403, showed serum progesterone

level rising from the fourth post parturition day (Figure 4).

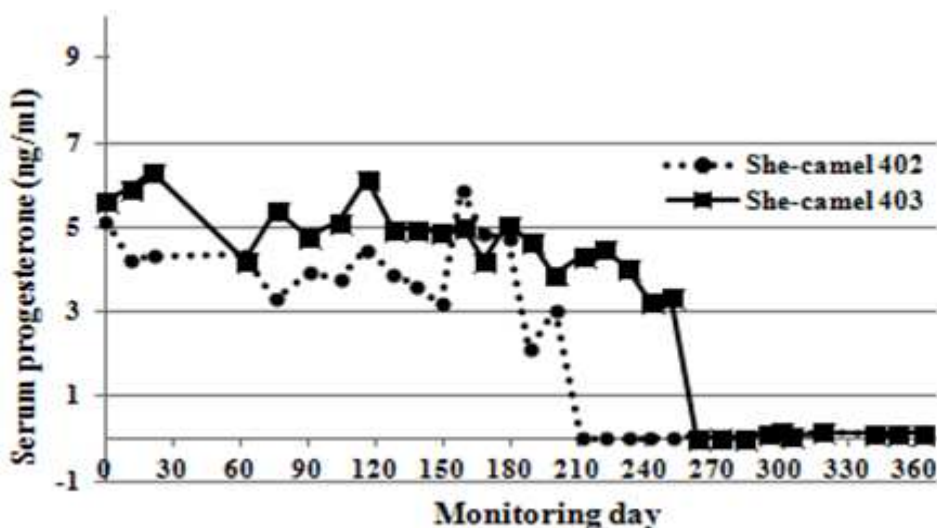


Figure 4. Progesterone level evolution after calving of she-camel

As regards to she-camel N° 504 (Figure 5) was successfully fertilized, the 214th experience day at 25 months age the progesterone level increased and remained at a high level until the end. But before this successful mating, monitoring brings up two peaks of 1.09 ng/ml

and 1.51ng/ml the 138th and the 179th monitoring days followed by a sharp decline at basal level. We would have suspected an embryonic mortality, but the reproduction records do not state a mating of she-camel N°504 at these dates.

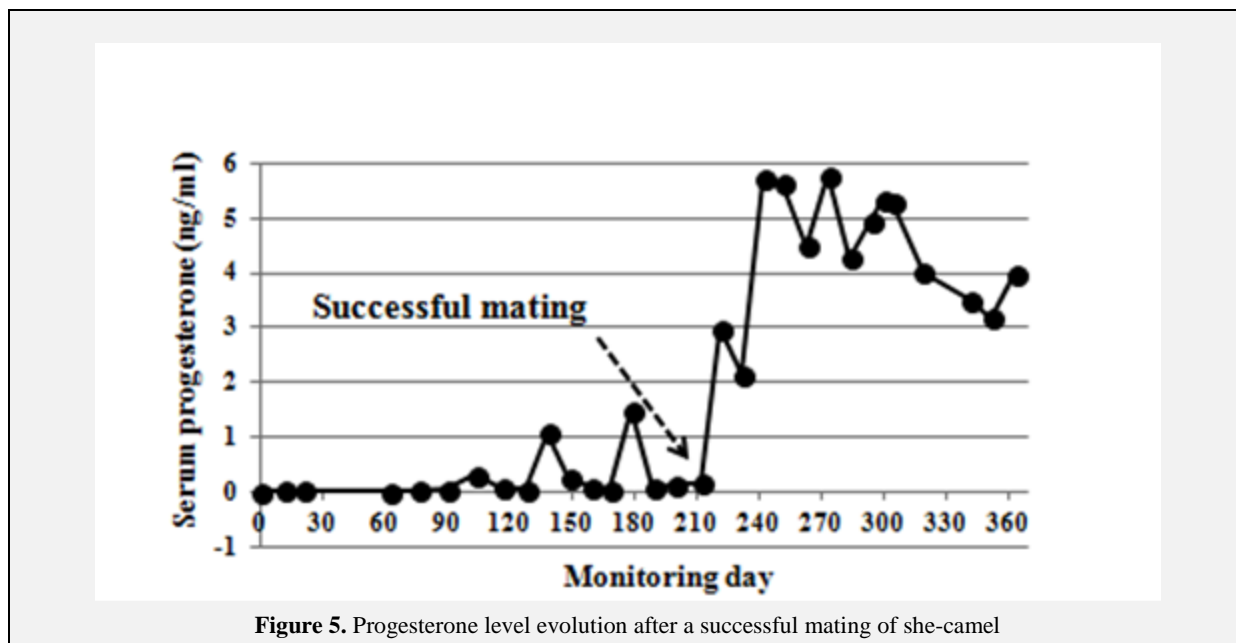


Figure 5. Progesterone level evolution after a successful mating of she-camel

The serum progesterone level change is in relation with the reproductive state and age of she-camel. Progesterone levels depend to reproductive status and age of analyzed she-camel. For all studied females the averages of serum progesterone are  $0.13 \pm 0.17$  g/L for non pregnant she-camel and  $3.99 \pm 1.26$  g/L for pregnant she-camel, the difference is highly significant ( $p < 0.001$ ). Age influenced progesterone concentration (Table 4). The

average progesterone concentration was slightly higher on growing females than the multiparous empty females  $0.14 \pm 0.19$  g/L vs.  $0.12 \pm 0.13$  g/L ( $p < 0.05$ ). This trend is amplified during pregnancy, thereby average progesterone concentration was significantly higher on young pregnant than the multiparous pregnant  $4.25 \pm 1.31$  g/L vs.  $3.55 \pm 0.87$  g/L ( $p < 0.01$ ).

Table 4. Serum progesterone levels and average age for analyzed category of age.

Category	Characters	observation	Mean	Std Dev	Min	Max
Category 1	Progesterone (g/L)	113	0.14	0.19	0	1.51
	Age (month)	113	16.26	5.89	4.08	25.12
Category 2	Progesterone (g/L)	115	4.25	1.31	2.13	9.78
	Age (month)	115	32.14	6.65	23.67	52.96
Category 3	Progesterone (g/L)	77	0.12	0.13	0	0.41
	Age (month)	77	49.55	11.47	33.8	76.27
Category 4	Progesterone (g/L)	59	3.55	0.87	1.56	5.27
	Age (month)	59	64.57	6.44	52.6	74.83

The plasma samples of pregnant females were used to reconstruct the critical points on progesterone serum profile for the Maghrabi she-camels (Figure 6). The profiles show a significant increase in progesterone beyond the

first day after successful mating, the high level of progesterone maintained throughout pregnancy falls at calving to reach the basic minimum.



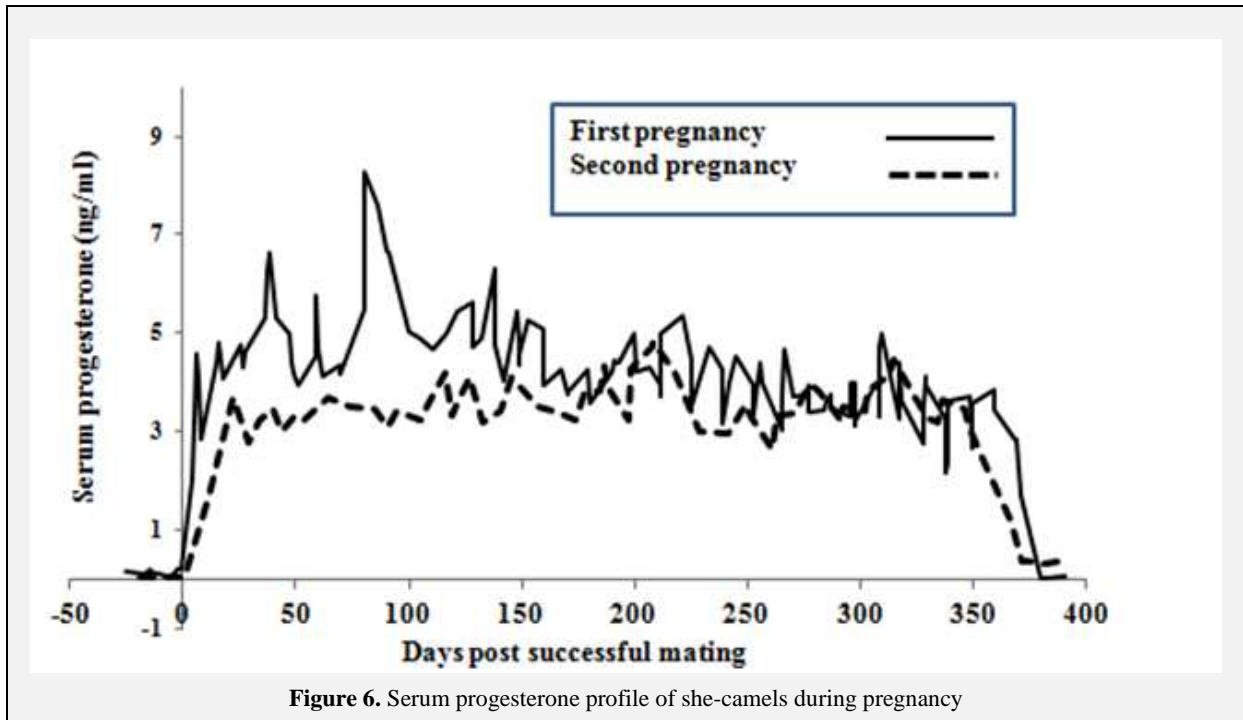


Figure 6. Serum progesterone profile of she-camels during pregnancy

The overall mean of progesterone concentration was slightly higher during pregnancy first half that the second half  $4.37 \pm 1.38$  ng/ml vs.  $3.70 \pm 0.96$  ng/ml ( $p < 0.05$ ). Average progesterone level for pregnant female was slightly higher for first than second half with respectively  $4.37 \pm 1.38$  ng/ml and  $3.70 \pm 0.96$  ng/ml ( $p < 0.05$ ). This change is quite visible ( $p < 0.001$ ) in the young growing she-camel where progesterone

concentration goes  $4.82 \pm 1.40$  to  $3.75 \pm 0.99$  between the first and the second pregnancy half. The she-camels in second pregnancy did not exhibit any definite trend (Table 5). The progesterone profile of first pregnancy is higher compared to the second pregnancy (Figure 6). Differences are highly significant ( $p < 0.001$ ) at first half of gestation  $4.82 \pm 1.40$  than  $3.51 \pm 0.85$  but were weakly significant in the second half  $3.75 \pm 0.99$  vs.  $3.59 \pm 0.89$ .

Table 5. Variation of progesterone level according to stage of pregnancy progress

camel categories	Gestation period	Observation	Mean	Std Dev
First pregnancy	First half	54	4.82	1.40
	Second half	61	3.75	0.99
Second pregnancy	First half	29	3.51	0.85
	Second half	30	3.59	0.89
All pregnancy	First half	83	4.37	1.38
	Second half	91	3.70	0.96

This study suggests that the progesterone levels exceeding 1,5 ng/ml are reached only if mating is fertile. Plasma progesterone level is high during pregnancy and it drop at parturition. Progesterone concentration starts to rise after mating and during pregnancy and falls at parturition

Progesterone hormone level in females is a very useful tool to monitor pregnancy in camels (Alfurajji, 1998). The primary source of progesterone in the female camel is the corpus luteum (CL). The placenta does not contribute to progesterone secretion, and all camelids depend entirely on progesterone from

the CL to maintain their pregnancy (Skidmore, 2005).

In general, the pregnancy status have a significant effect on serum progesterone level ( $P < 0.01$ ). The measurement of progesterone levels is still the most reliable method to detect estrus, ovulation and the ovarian function in animals. This is not the case for camel, because only pregnancy status can be identified.

Our results indicate a gradual increase, for serum progesterone level, during this study. The serum progesterone level started to increase at March and continue high during July (Babiker et al., 2011). Ovulation in camel is induced by mating stimulus. If successful mating exists, serum progesterone level started to rise, else blood progesterone quantity still low (Ayoub et al., 2003; Skidmore, 2005; Ghazi, 2007). Sexual behavior of male can explain serum progesterone level variation in camel during this period. The present findings are similar to those of Musa and Abusineina (1978). Camel's species have seasonal sexual behavior affected by long daylight of the year (Chen and Yuen 1979; Sumar, 2000).

Statistical analysis, with General Linear Model procedure, indicates a significant effect ( $P < 0.001$ ) of age on serum progesterone level. These results were confirmed by regression procedure. The use of multiple linear regression models showed significant differences between category for the serum progesterone levels ( $F_{9,178} = 5.05$ ,  $P < 0.001$ ,  $SEM = 0.32$ ; adjusted  $R^2 = 0.16$ ). The reproductive variables means value for serum progesterone in analyzed individuals, were significantly higher in the young pregnant females, pregnant multiparous females than those for growing female and empty multiparous females. During pregnancy progesterone hormone is found to be high and declined just after parturition (Alfuraiji, 1998; Skidmore, 2005). In empty female, progesterone level remained low and it increase again after successful mating. As demonstrate by different studies ovulation in camel is induced by mating stimuli (Ayoub et al., 2003; Skidmore, 2005; Ghazi, 2007). Progesterone level indicated a significant rise ( $>5$  ng/ml) in progesterone 2 days after successful mating in all female (Abdel Rahim et El-Nazier, 1993). A Rapid drop of the progesterone concentration, to values of  $< 1$  ng/ml on the day before, or the day of parturition, is recorded (Skidmore et al., 1996).

Mobarak and El-Wishy (1971) reported that from 5 pregnancy months until parturition there to be a gradual decrease for this hormone. These results indicate a significant rise in progesterone two days after successful mating of all young she-camel.

#### 4. Conclusion

Serum progesterone concentrations of the empty postpartum camels were varying between 0 ng / ml and 0.41 ng / ml with an average of  $0.38 \pm 0.04$ . Lactating status have a significant effect on serum progesterone level. A significant variation between progesterone level for lactating females and non lactating female was found ( $p < 0.01$ ). The serum progesterone value was affected significantly and tends to increase with age in younger she-camels.

Progesterone quantities rise after a successful mating. This significant rise was  $\geq 2.96$  ng/ml two days after mating in all female. These levels decrease and tend to basic level at calving day. Embryonic mortality can be a considerable cause for serum progesterone decline. The serum progesterone level depends to the reproductive state and age of she-camel. For all studied females the averages of serum progesterone are  $0.13 \pm 0.17$  g/L for non pregnant she-camel and  $3.99 \pm 1.26$  g/L for pregnant she-camel, the difference is highly significant ( $p < 0.001$ ). Age influenced progesterone concentration. The average progesterone concentration was slightly higher on growing females than the multiparous empty females  $0.14 \pm 0.19$  g/L vs.  $0.12 \pm 0.13$  g/L ( $p < 0.05$ ). This trend is amplified during pregnancy, thereby average progesterone concentration was significantly higher on young pregnant than the multiparous pregnant  $4.25 \pm 1.31$  g/L vs.  $3.55 \pm 0.87$  g/L ( $p < 0.01$ ). The profiles show a significant increase in progesterone beyond the first day after successful mating, the high level of progesterone maintained throughout pregnancy falls at calving to reach the basic minimum. The overall mean of progesterone concentration was slightly higher during pregnancy first half that the second half  $4.37 \pm 1.38$  ng/ml vs.  $3.70 \pm 0.96$  ng/ml ( $p < 0.05$ ). Average progesterone level for pregnant female was slightly higher for first than second half with respectively  $4.37 \pm 1.38$  ng/ml and  $3.70 \pm 0.96$  ng/ml ( $p < 0.05$ ). This change is quite visible ( $p < 0.001$ ) in the young growing she-

camel where progesterone concentration goes  $4.82 \pm 1.40$  to  $3.75 \pm 0.99$  between the first and the second pregnancy half. The she-camels in second pregnancy did not exhibit any definite trend. The progesterone profile of first pregnancy is higher compared to the second pregnancy. Differences are highly significant ( $p < 0.001$ ) at first half of gestation  $4.82 \pm 1.40$  than  $3.51 \pm 0.85$  but were weakly significant in the second half  $3.75 \pm 0.99$  vs.  $3.59 \pm 0.89$ .

Statistical analysis, with General Linear Model procedure, indicates a significant effect ( $P < 0.001$ ) of age on serum progesterone level. These results were confirmed by regression procedure. The use of multiple linear regression models showed significant differences between category for the serum progesterone levels ( $F_{9,178} = 5.05$ ,  $P < 0.001$ ,  $SEM = 0.32$ ; adjusted  $R^2 = 0.16$ ).

The reproductive variables means value for serum progesterone in analyzed individuals, were significantly higher in the young pregnant females, pregnant multiparous females than those for growing female and empty multiparous females.

Progesterone level of for all studied dromedaries is low in empty female. It started to increase if successful mating exist and continue to be high until the end of the pregnancy.

Progesterone hormone level is a very useful tool to monitor pregnancy in camels.

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