

Variations in some haematological parameters and gonadal indices of *Uromastyx aegyptius* during hibernation and active periods

Kamar M Said¹ and Hussein K Hussein^{2*}

1. Biology and Geology Department, Faculty of Education, Alexandria University, Egypt.

2. Zoology Department., Faculty of Science, Alexandria University, Egypt

ABSTRACT

The red blood cell count, packed cell volume, haemoglobin content and the level of blood glucose of the lizard, *Uromastyx aegyptius* were measured during summer and winter periods. During hibernation, the studied blood parameters were decreased by at least 33 %. The gonadal indices also varied considerably during hibernation and activity periods, reaching maximum values in summer. The onset of winter induces gonadal regression, reduced testis size, fewer eggs in ovaries and decreased numbers of large oocytes.

KEYWORDS: *Uromastyx aegyptius*, hibernation, gonadal indices, blood parameters

INTRODUCTION

In reading much of the literature about hibernation one gets the impression that ectotherms become dormant in winter simply because they are unable to maintain normal metabolic rates at reduced ambient temperatures occurring at that time of year (El-Masry & Hussein 2001). The utilization of endogenous energy reserves by reptiles under hibernation conditions has received little attention despite the fact that these reserves in certain reptiles undergo pronounced changes throughout the year (Gillett & Dacruz 1981 and Said & Hussein 1992). A number of ecologists have investigated lizard reproductive cycles, but until quite recently, relatively little has been written about reproductive activity during hibernation and activity periods (El-Ghazaly *et al.* 1987_a; Said & Hussein 1992; Madkour *et al.* 1999).

On the other hand, there is little information about the effect of hibernation and/or natural hypothermia on the haematology of lizards, in addition to the conditions at which reptiles frequently exhibit marked seasonal fluctuations in their various blood constituents.

The purpose of this study is to show that hibernation is a rather complex phenomenon in the agamid lizard, *Uromastyx aegyptius*. It presents information dealing with gonadal activity and discusses one ecological phenomenon that tends to modify that activity. To do this, we investigated variations in red blood cell counts, packed cell volumes, haemoglobin contents and blood glucose levels. Such blood components give a measure of the state of reproductive activity and other physiological activities of the animal.

MATERIALS AND METHODS

Adult male and female *Uromastyx aegyptius* were collected from different locations in the Burg El-Arab desert region, 50 kilometer west of Alexandria, Egypt. Lizards were captured twice in the year, in the active season (July, 1999) and during hibernation (January, 2000). Ten animals (5 males and 5 females) during summer and ten animals (5 males and 5 females) during winter (snout-vent length: 180–220 mm; tail: 80–130mm; total length: 265–340 mm; mass : 257–270 gm.) were kept in a large terrarium with 30 cm depth of sand in the laboratory. The animals were decapitated with a sharp blade and blood was collected into heparinized tubes. Red blood cells were counted by the classical method using a Thoma

* Address for Correspondence

haemocytometer. Blood haemoglobin was estimated using a Sahli haemometer. The packed cell volume was determined in microhaematocrit tubes. Blood glucose levels were determined according to the method of Hjelm & DeVerdier (cited in Varley 1963).

The results obtained were statistically analyzed statistically according to the method of Parker (1979). Main effects of season and sex different parameters were analyzed by two-way ANOVA. $P < 0.01$ was taken as the level of significance.

Animals were dissected for the gonadal activity studies. The left testes and left ovaries were extracted from all animals and immediately fixed in Bouin's solution, dehydrated in series of alcohol, and embedded in paraffin wax. They were serially sectioned at 5μ , stained with Ehrlich's acid alum haematoxylin and counter stained with eosin. A number of measurements were taken, including testis volume, testis diameter, seminiferous tubule diameter, number of eggs in the ovary, number of large oocytes and the diameter of large oocytes. All measurements were made by using an ocular micrometer (20 mm diameter, 10 mm divisions into 200 parts).

RESULTS

The blood constituents of *Uromastix aegyptius* during winter hibernation and summer activity (Table 1) revealed marked variation in all investigated parameters. There were significant decreases in red blood cells, haematocrit percentage, haemoglobin content and glucose level of the blood in winter, significantly more in females than males ($P < 0.01$).

Testis size of males, varied considerably during hibernation and activity seasons. In winter, lizards remain sequestered in their burrows and become dormant because they are unable to maintain normal metabolic rates at the reduced ambient temperatures occurring at that time of the year. In this period, the testes were much smaller than in summer (Table 2). Specimens sacrificed in summer had larger tubule diameters (Table 2, Fig. 1). The walls of the tubules consisted of larger cell layers and all spermatogenic stages were clearly represented, and the beginnings of spermatozoon formation were also observed (Fig. 1). The seminiferous tubules were completely inactive (Table 2, Figs.1 & 2). Their lumina were small and virtually filled with numerous layers of spermatids in all stages of maturation, although earlier stages predominated, and the interstitial space was filled with interstitial cells (Fig. 2). Specimens sacrificed in summer had larger tubule diameters (Table, Fig. 1).

In females, the egg characteristics in the ovaries are shown in Table 2. There were a great number of small oocytes and complete gonadal regression in winter (Fig. 3), but in summer the number of eggs in the ovary as well as number and diameter of large oocytes were increased and enlarged eggs were present in the ovaries (Fig. 4).

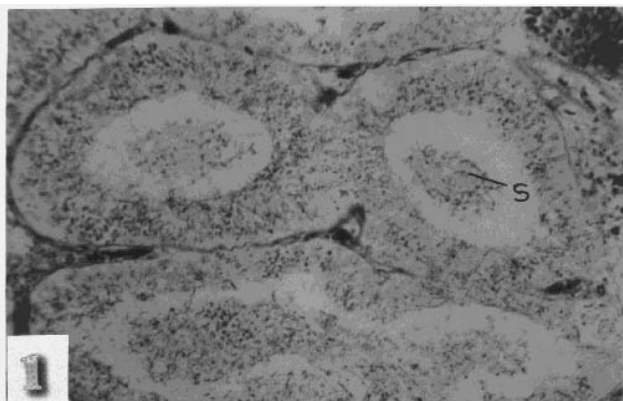


Fig. 1: T.S. in the testis of the adult lizard, *Uromastix aegyptius* during the activity period. The seminiferous tubule lumina are filled with mature spermatozoa (S). X= 300.

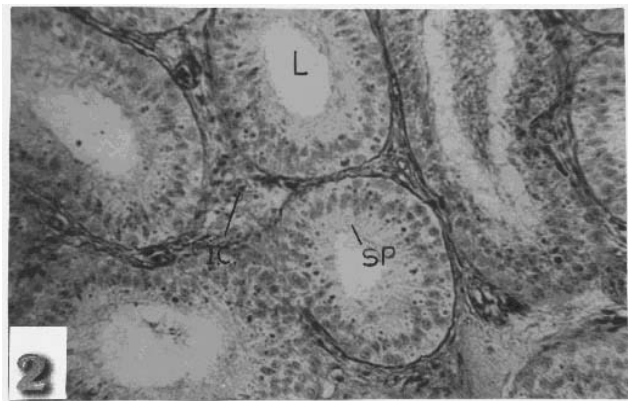


Fig.2: T.S. in the testis of the adult lizard, *Uromastix aegyptius* during the hibernating period. Seminiferous tubules with large luminae (L), empty from any spermatozoa and metamorphosing spermatids (SP) only is present. Note interstitial space is filled with interstitial cells (I.C.). X= 300.

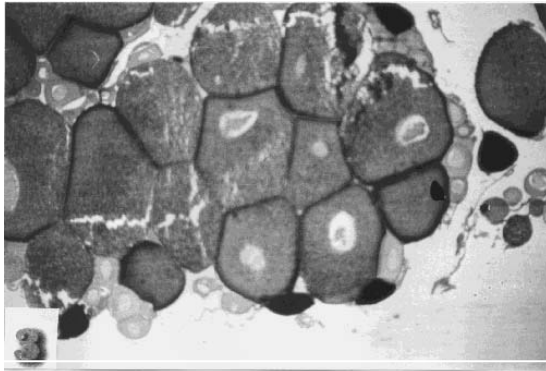


Fig. 3: T.S. in the ovary of the adult lizard *U. aegyptius* during the activity period showing an increase in number of large oocytes. X300.

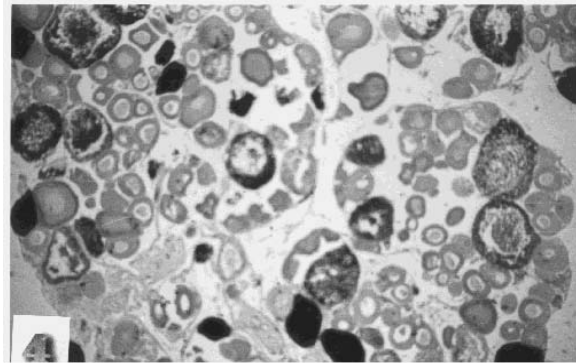


Fig.4: T. S. in the ovary of the adult lizard *U. aegyptius* during the hibernating period showing an increase in number of small oocytes. X= 300.

Table 1: Variations in some blood parameters of *Uromastix aegyptius* during winter hibernation and summer activity seasons. Data are expressed as mean \pm S.E. of five animals; Anova test: Between species (df=1, F=3.22, P<0.01); between seasons (df=1, F=5.44, P<0.01).

Parameter	Sex	Season		Changes	Changes calculated as percentage of the summer value (%)
		Summer	Winter		
Erythrocytes/ mm ³	Male	1,489,281 \pm 51.12	912.275 \pm 71.70	-577.006	-38.74
	Female	1,225.196 \pm 46.40	866.146 \pm 66.12	-359.056	-29.31
Packed cell volume (%)	Male	32.02 \pm 1.77	21.12 \pm 1.96	-10.90	-34.04
	Female	30.46 \pm 0.64	20.67 \pm 1.05	-9.79	-32.14
Haemoglobin (g/100 ml)	Male	11.55 \pm 0.31	7.76 \pm 0.46	-3.79	-32.81
	Female	10.62 \pm 0.64	6.84 \pm 0.39	-3.78	-35.59
Glucose (mg/100 ml)	Male	84.23 \pm 5.26	39.66 \pm 3.69	-44.57	-52.91
	Female	72.16 \pm 4.97	39.47 \pm 3.10	-35.69	-49.46

Table 2: Changes in gonadal indices of *Uromastix aegyptius* during hibernation and activity periods. Data are expressed as mean \pm S.E. of five animals.

Measurements	Season		Changes	Changes calculated as percentage of the summer value (%)
	Summer	Winter		
Testis volume (mm ³ x10 ³)	9.9 \pm 0.26	6.3 \pm 0.11	-3.6	-36.36
Testis diameter (μ)	1217 \pm 6.55	844 \pm 7.17	-373	-30.65
Seminiferous tubule diameter (μ)	195.2 \pm 0.12	145.3 \pm 0.35	-49.9	-25.56
Number of eggs in the ovary	49.8 \pm 2.12	32.7 \pm 2.77	-17.1	-34.33
Number of large oocytes	30.3 \pm 1.12	12.2 \pm 1.43	-18.1	-59.74
Diameter of large oocytes (μ)	9.2 \pm 0.96	5.1 \pm 0.35	-4.1	-44.57

DISCUSSION

The present results showed a decrease in the number of erythrocytes, haematocrit value, haemoglobin content and blood glucose content of *Uromastix aegyptius* during winter. An animal in hibernation can not maintain itself in an active condition, so a decline in these parameters seems logical. Since its oxygen demands become very low (El-Masry & Hussein 2001). Our results are in agreement with those of Maclean *et al.* (1975) who have reported that the number of erythrocytes, haematocrit value and the haemoglobin content were related to the habit of the animal, being low in comparatively sluggish forms and high in active ones. They also added that at low temperatures, the metabolic rate and energy turnover in reptiles are low, since all physiological processes are slow down. It was also noticed that as the

temperature increases, the physiological processes such as metabolic rate and energy turnover are increase in proportion to the oxygen consumption (Taha *et al.* 1986; Hussein & Said 1991).

The present findings contradict those reported for other reptilian species such as snakes (Carmichael & Petcher 1945) and turtles (Hutton & Goodnight 1957) where increases in the number of erythrocytes and haemoglobin content during hibernation were reported. On the other hand, Hutton (1960) reported that the haematocrit value of the red-eared turtle did not show any consistent seasonal variation. A great decrease in blood sugar content of the experimental animal *U. aegyptius* during the winter season, as in other findings (Otis 1973; Taha *et al.* 1986). In winter, hypoglycaemia has been shown to occur in alligators (Coulson & Hernandez 1953), in lizards (Khalil & Yanni 1959) and in snakes (Al-Badry & Nuzhy 1983), which may be responsible for the depression of activities in the nervous system during hibernation (Haggag *et al.* 1966). Thus, a fall in blood sugar level seems to be a common feature of hibernation where the activities of the body are lowered. A significant decrease in all blood parameters of females as compared to that of males was noticed throughout hibernation and activity periods. This might be attributed to the differences in requirements of nutritional elements and physiological needs of both sexes in converting assimilated calories into production (Smith 1976).

As for gonadal activity *U. aegyptius*, the pattern of change during activity and hibernation appears to follow that of *Anolis carolinensis* (Fox 1958), *Sceloporus orcutti* (Mayhew 1963), *Urosaurus ornatus* (Asplund & Low 1964), *Chalcides ocellatus* (El-Ghazaly *et al.* 1987 a), and *Scincus officinalis* (Said & Hussein 1992). Winter induces gonadal regression characterized by reduced testis size, fewer eggs in the ovaries and decreased number of large oocytes. However, the type of seasonal change in *U. aegyptius* differs considerably from that found in many species of lizards so far. For example, *Sceloporus undulates* (Altland, 1941), *Sceloporus occidentalis* (Wilhoft & Quay 1961) and *Uta stansburiana* (Asplund & Lowe 1964) emerge from hibernation with testes and ovaries almost at maximum size. The gonads of these species become greatly reduced in size during summer season, followed by tremendous increase again just before hibernation. These pattern were not found in *U. aegyptius*. The blood parameters can be correlated with reduced metabolic rates and less oxygen demand during hibernation, a condition that probably leads to a low rate of erythropoiesis. In this respect, similar conclusions were reported for different reptilian species during winter (Haggag *et al.* 1966; Maclean *et al.* 1975; Hussein & Said 1991; Bashandy *et al.* 1996).

On the other hand, the annual reproductive cycles in temperate zone squamate reptiles appear to be controlled by environmental stimuli with temperature as playing the major role. The majority of studies on the environmental control of the sexual cycles of squamate reptiles have been done on lizards and snakes (Hawley & Aleksiuik 1976; Pearson *et al.* 1976; El-Ghazaly *et al.* 1987_b). According to their conclusions, exposure of individuals to low temperatures prevents completion of spermatogenesis and oogenesis while exposure to high temperature permits or stimulates complete gametogenesis. It could be argued that in such desert environment, it is starvation that inhibits testicular recrudescence and ovarian development, since individuals would not possess enough energy to maintain gametogenesis as well as other bodily functions.

REFERENCES

- Al-Badry KS & Nuzhy S (1983) Haematological and biochemical parameters in active and hibernating sand vipers. *Comp. Biochem. Physiol.* 74 (A): 137- 141
- Altland PD (1941) Annual reproductive cycle of the male fence lizard. *J. Elisha Mitchell Sci. Soc.* 57: 73-83

- Asplund KK & Lowe CH (1964) Reproductive cycles of the iguanid lizard *Urosaurus ornatus* and *Uta stansburiana* in Southeastern Arizona. *J. Morph.* 115: 27-23.
- Bashandy MA, Kawashti IS & Abdel-Raheem AMA (1996) A comparative study of general metabolic rates in the rat and lizard under low and high temperatures. *J. Egypt. Ger. Soc. Zool.* 2 (A): 459-472.
- Carmichael EB & Petcher PW (1945) Constituents of the blood of hibernating and normal rattlesnake, *Crotalus horridus*. *J. Biol. Chem.* 16: 693-696
- Coulson RA & Hernandez T (1953) Glucose studies in crocodilia. *Endocrinology* 53: 311- 320
- El-Ghazaly NA, Moursi AA & Hussein HK (1987a) Seasonal changes in testicular histology of the Egyptian lizard, *Chalcides ocellatus*. *Delta J. Sci.* 11 (2): 951-975.
- El-Ghazaly NA, Moursi AA & Hussein HK (1987b) Effect of the light and temperature on the testicular recrudescence of the lizard, *Chalcides ocellatus*. *Com. In. Sci. & Dev. Res.* 19:181-196.
- El-Masry AA & Hussein HK (2001) Thermal relations, metabolism and winter dormancy of the sand lizard, *Acanthodactylus boskianus*. *Pak. J. Biol. Sci.* 4 (4): 492 – 497.
- Fox W (1958) Sexual cycle of the male lizard, *Anolis carolinensis*. *Copeia* 1959: 22- 29.
- Gillet MPT & Dacruz MEM (1981) Seasonal variation in plasma and hepatic lipids in relation to nutritional status and vitellogenesis in male and female lizards, *Ameiva ameiva* (Teiidae). *Comp. Biochem. Physiol.* 70 : 313 –316.
- Haggag G, Raheem KA & Khalil F (1966) Hibernation in reptiles. II. Changes in blood glucose, haemoglobin, red blood count, protein and non-protein nitrogen. *Comp. Biochem. Physiol.* 17: 335 –339
- Hawley AWL & Aleksyuk M (1976) The influence of photoperiod and temperature on seasonal testicular recrudescence in the red-sided garter snake, *Thamnophis sirtalis parietalis*. *Comp. Biochem. Physiol.* 53A: 215-221
- Hussein HK & Said KM (1991) Effect of temperature and feeding on body weight and some physiological parameters of the blood of the lizard, *Agama stellio*. *J. Egypt. Ger. Soc. Zool.* 6 (A): 337-348.
- Hutton KE & Goodnight CJ (1957) Variations in the blood chemistry of turtles under active and hibernating conditions. *Physiol. Zool.* 30: 198- 207.
- Khalil F & Yanni M (1959) Studies on carbohydrates in reptiles. *Z. Vergl. Physiol.* 42: 192 – 198.
- Maclean GS, Lee AK & Withers PC (1975) Haematological adjustments with diurnal changes in body temperature in a lizard and a mouse. *Comp. Biochem. Physiol.* 51 A: 241-249.
- Madkour G, El-Desouki N & Talaat H (1999) Comparative histological and histochemical structure of some reptiles. *J. Union Arab Biol.* 11 (A): 325-343.
- Mayhew WW (1963) Reproduction in the granite spiny lizard, *Sceloporus orcutti*. *Copeia* 1963 (1): 144-152.
- Otis VS (1973) Haematological and serum chemistry parameters of the African puff adder, *Bittis orietans*. *Herpetologica* 29: 110-116
- Parker RE (1979) Introductory statistics for biology. 2nd edition edited by Edward Arnold, London.
- Pearson AK, Tsui HW & Licht P (1976) Effect of temperature on spermatogenesis, on the production and action of androgens and on the ultrastructure of gonadotropic cells in the lizard, *Anolis carolinensis*. *J. Exp. Zool.* 195: 291- 304.
- Said KM & Hussein HK (1992) Seasonal fluctuation in the fat storage of the lizard, *Scincus officinalis* and its adaptive significance to gonadal activity. *J. Egypt. Ger. Soc. Zool.* 7 (A): 1- 15.
- Smith GC (1976) Ecological energetics of three species of ectothermic vertebrates. *Ecology* 57: 252-264.
- Taha HM, Al-Badry KS, Fathi MM & Sakran A (1986) Haematologic parameters in terrestrial and aquatic snakes. *Proc. Zool. Soc. A. R. Egypt* 11: 197 –212.
- Varley H (1963) Practical Clinical Biochemistry. (4th Ed.) pp. 83-84, White Friars Press, London and Tonbridge.
- Wilhoft DC & Quay WB (1961) Testicular histology and seasonal changes in the lizard, *Sceloporus occidentalis*. *J. Morph.* 108: 95 – 106.

