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Localization of Glycogen in the Haemocytes of the Ixodid Ticks, *Hyalomma dromedarii* and *Hyalomma arabica* (Acari : Ixodidae)

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ABSTRACT. The glycogen content of the haemocytes of the ixodid ticks, *Hyalomma dromedarii* and *Hyalomma arabica* was investigated in males, females and nymphs of both tick species. Granular haemocytes, plasmatocytes and spherulocytes have a large glycogen content, but the oenocytoids and adipohaemocytes have very few glycogen granules, while the prohaemocytes are devoid of glycogen. All haemocytes of unfed ticks of both species, with the exception of granular haemocytes lacked glycogen granules. The glycogen granule counts of the haemocytes of fed female ticks of both species were much larger than those of fed male ticks. The role of glycogen synthesis and its utilization is discussed.

Cells circulating in the haemolymph of ticks and other arthropods are called haemocytes (Brinton and Burgdorfer 1971 and Balashov 1983) and are equivalent to vertebrate leucocytes (Gupta 1979, 1990, 1991, Al-Khalifa and Siddiqui 1985 and Farkas and Zithan 1989). They were recently shown in insects to be important sites of intermediary metabolism of glycogen (Khan *et al.* 1990). Glycogen synthesis and its utilization by haemocytes is dependent on the feeding status of the insect (Liu and Davies 1972, and Bardoloi and Hazarika 1992).

Even in insects, haemocyte glycogen metabolism has only been studied in very few species (Gupta 1979), while such studies in ticks do not exist. Hence, in the present study an attempt has been made to determine the glycogen content of haemocytes of both of *Hyalomma dromedarii* Koch and *Hyalomma arabica* Pegram, Hoogstraal and Wassef.

Materials and Methods

Colonies of both tick species have been established in our laboratory for some time (Al-Khalifa and Siddiqui 1993) at 28°C and 75% humidity and their various instars have been maintained on New Zealand rabbits in capsules according to method of

Varma (1964). One leg of either male, female or nymph of either tick species was amputated and drops of haemolymph were smeared onto clean microscope slides. The smears were fixed in methanol, washed in tap water and stained with Periodic Acid Schiffs reagent (PAS, Pearse 1980). Additionally, some of the smears were first treated with diastase for 30 min at 37°C and then stained with PAS as controls. Hanging drop preparations of haemolymph grown for 24 - 72 h in a short-term culture (Al-Khalifa and Siddiqui 1993) were stained with Lugol's iodine to determine glycogen in living haemocytes.

Results and Discussion

Six types of haemocytes, prohaemocytes, plasmatocytes, granular haemocytes, spherulocytes, oenocytoids and adipohaemocytes have been reported in the haemolymph of *H. dromedarii* and *H. arabica* (Al-Khalifa and Siddiqui 1993). Glycogen in the form of microgranules was observed in the cytoplasm of all types of haemocytes except prohaemocytes in the nymphs and adults of both tick species; the number of PAS positive granules in the two species, as well as among the various types of haemocytes in the same species varied. The haemocytes of the similar stages and sexes of *H. dromedarii* showed higher counts of glycogen granules in their respective types of haemocytes than those of *H. arabica*. This difference could well be due to species variations and could reflect enviromental adaptation of *H. dromedarii* that thrives in harsh desert conditions (Hoogstraal *et al.* 1981), while *H. arabica* is only found in Al-Sarawat Mountains of Taif and Asir where conditions are very amiable (Pegram *et al.* 1982, Hoogstraal *et al.* 1983, Al-Khalifa *et al.* 1986, and 1987).

The glycogen granule counts (GGC) in female haemocytes were generally higher than that in those of males. Moreover, the various haemocytes have different levels of GGC; the highest occurred in granular haemocytes followed by plasmatocytes; spherulocytes contained only one glycogen granule and prohaemocytes, as well as 50% of adipohaemocytes and oenocytoids were devoid of glycogen granules (Table 1, Figs. 1and 2). The occurrence of GGC was inversely proportional to the unfed state of the tick. Glycogen granules were rare in granular haemocytes and plasmatocytes of both tick species (Table 1) and were totally absent from all other types of haemocytes of unfed instars of both tick species. *In vitro* prepartions of haemocytes were devoid of glycogen, possibly due to absence of glycogen formation in artificial media. The higher GGC in females of both species might suggest a higher need for nutrients by females, possibly for oogenesis as have been reported in insects by Ahmed and Khan (1988).

The larger quantities of glycogen in plasmatocytes and granular haemocytes might indicate the active role of both haemocytes in metabolic transport and in the synthesis of glycoproteins in the haemolymph as have been reported in insects by Arnold and

Sohi (1974). The absence of glycogen granules in prohaemocytes could be due to the presence of large quantities of glycogen proteins in their cytoplasm which are needed for their transformation into other types of haemocytes as have been observed by Liu and Davies (1972) in insects.

Table 1. Glycogen granule counts per cell in the haemocytes of *Hyalomma dromedarii* and *Hyalomma arabica*

Haemocytes	*Hyalomma dromedarii*						*Hyalomma arabica*					
	Fed			Unfed			Fed			Unfed		
	♂	♀	Nymph	♂	♀	Nymph	♂	♀	Nymph	♂	♀	Nymph
Prohaemocyte	0	0	0	0	0	0	0	0	0	0	0	0
Granular haemocytes	5	6	4	0	1	0	2	4	3	1	0	0
Plasmatocyte	4	5	3	1	2	1	2	3	2	0	2	0
Spherulocyte	1	1	1	0	0	0	1	1	1	0	0	0
Oenocytoid	1	1	1	1	1	0	0	1	1	0	0	0
Adipohaemocyte	1	1	0	0	0	0	1	1	1	0	0	0

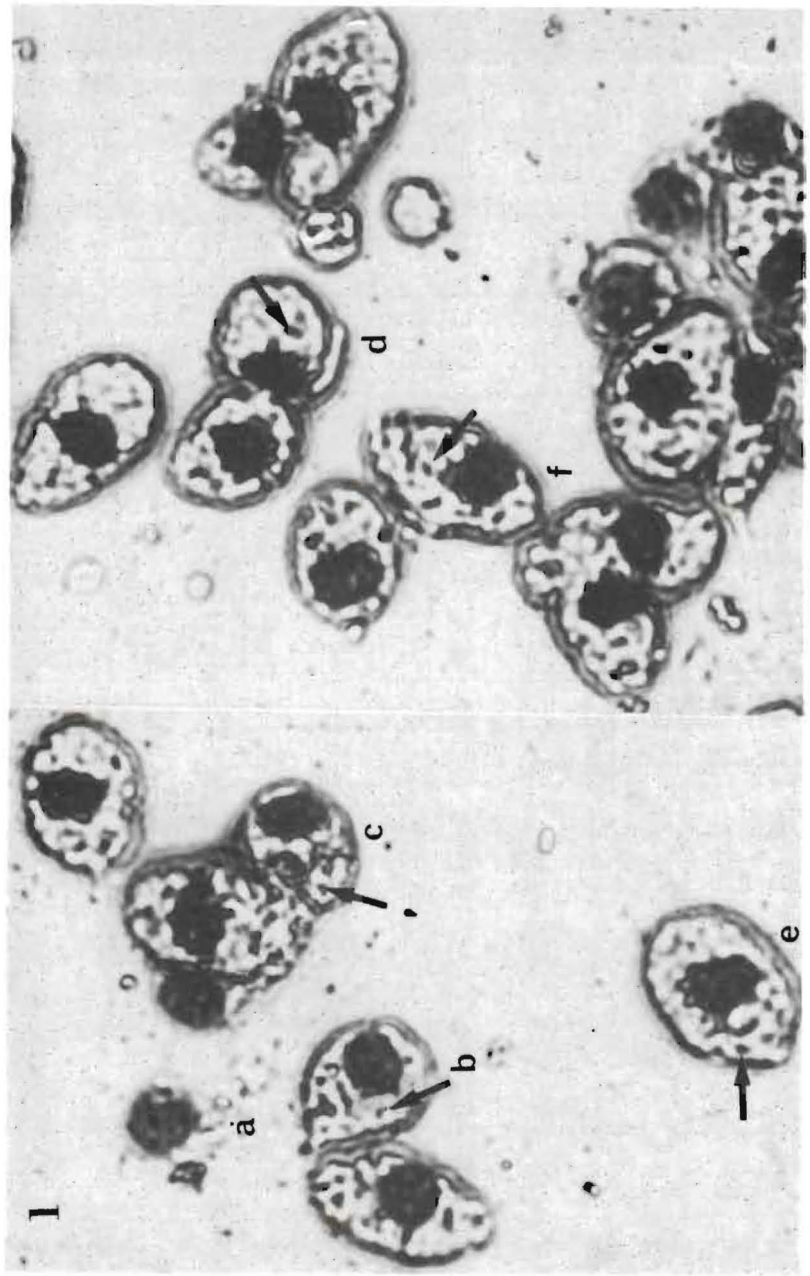

Fig. 1. Photomicrographs of glycogen granules (Arrows) in the haemocytes of *Hyalomma dromedarii* : a, Prohaemocyte; b, plasmatocyte; c, granular haemocyte; d, spherulocyte; e, oenocytoid, f, adipohaemocyte; (PAS, X1000)

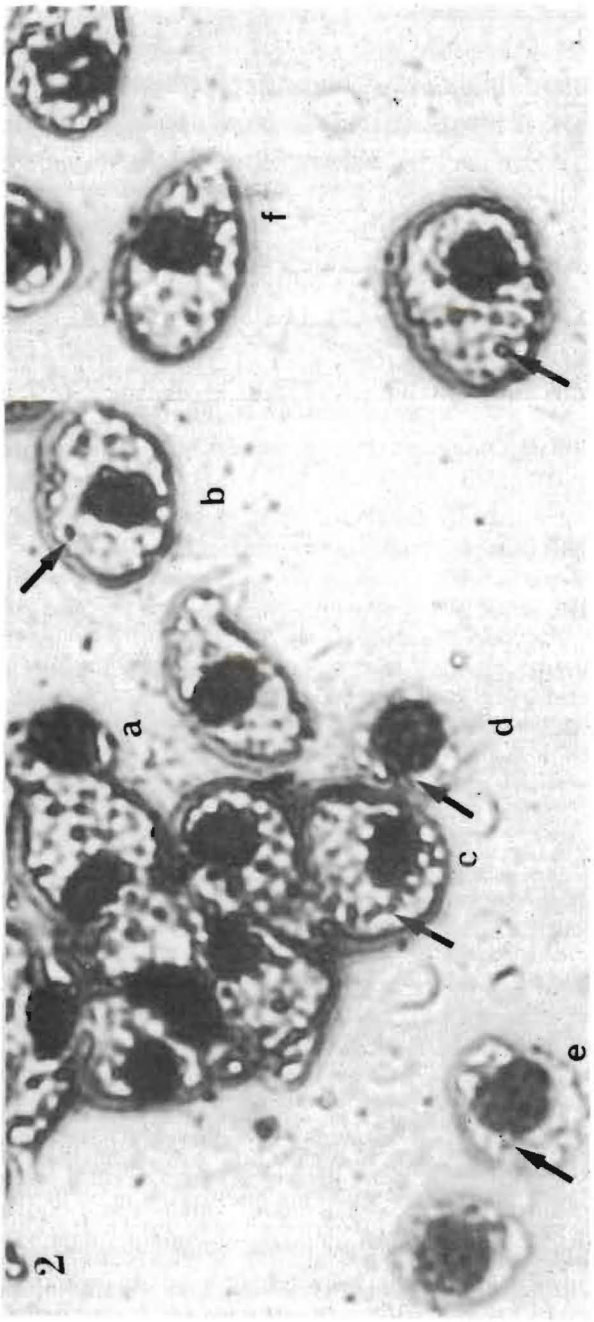

Fig. 2. Photomicrographs of glycogen granules (Arrows) in the haemocytes of *Hyalomma arabica*. a, Prohaemocyte; b, plasmatocyte; c, granular haemocyte; d, spherulocyte; e,oenocytoid; f, adipohaemocyte; (PAS, X1000)

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دراسة توزيع النشاء الحيواني في خلايا الدم الحارة للقراد الجامد نوعي *Hyalomma dromedarii* و *Hyalomma arabica*

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تم جمع قراد الماعز والضأن العربي من جبال السروات في منطقة مكة المكرمة وقراد الابل من الثمامة في منطقة الرياض والتي استعملت لإنشاء مستعمرات حيوانية مخبرية للنوعين بقسم علم الحيوان ـ كلية العلوم ـ جامعة الملك سعود بالرياض.

لقد تم جمع قطرات من دم كل من قراد الماعز والضأن العربي *Hyalomma arabica* وقراد الابل *Hyalomma dromedarii* عن طريق بتر رجل من كل منهما، ومن ثم عُملت سحبات على شرائح زجاجية نظيفة، ثبتت بواسطة كحول ميثيلي، ثم غسلت في ماء الصنوبر وبعد ذلك صبغت في صبغة دليل البريوديك شيف الحمضي (PAS).

لقد درست مكونات خلايا الدم من النشاء الحيواني (Glycogen) في القراد الجامد من نوعي قراد الماعز والضأن العربي وقراد الابل في كل من الذكور والاناث والحوريات. ووجد أن خلايا الدم المحببة (Granular haemocytes) وخلايا الدم البلازمية (Plasmatocytes) وخلايا الدم الكروية (Spherulocytes)

تحتوي على كميات كبيرة من النشاء الحيواني بينما خلايا الاينوسيتويد (Oenocytoids) وخلايا الدم الدهنية (Adipohaemocytes) تحوي كميات قليلة من تلك المادة ولكن خلايا الدم الأولية ليس بها نشاء حيواني على الاطلاق. لم تشاهد أية حبيبات نشوية في خلايا الدم لأطوار نوعي القراد البالغة وغير البالغة الغير متغذية ما عدا حبيبات قليلة من هذه المادة وجدت في خلايا الدم الحبيبية. بينما لوحظ ان الخلايا الدموية للاناث المغذاة تحوي على كمية أكبر من النشاء الحيواني اذا ما قورنت بخلايا دم الذكور. ولقد تمت مناقشة وظيفة النشاء الحيواني واستهلاكه بواسطة القراد.