



Scorpion Hemocyte- Plasmocyte

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ABSTRACT: The plasmocytes in five different scorpions- *Mesobuthus tumulus tumulus*, *M. tumulus concanesis*, *Orthochirus bicolor*, *Heterometrus xanthopus*, *H. phipsoni* were studied in the present investigation. The hemocytes are polymorphic and their population is about 70 – 80%. The length varied from 6- 12 μm and the width is 6- 12 μm in *M. tumulus tumulus*, *M. tumulus concanesis* and *O. bicolor*. In *H. xanthopus* and *H. phipsoni* the length ranged between 10- 35 μm and width between 3- 11 μm . The cytological characteristics were studied in all five species of scorpions. The nucleus is basophilic and placed at the centre. The cytological detail of plasmocyte in *H. xanthopus* of was studied by TEM. It is polymorphic with rod shaped mitochondria and centrally placed nucleus.

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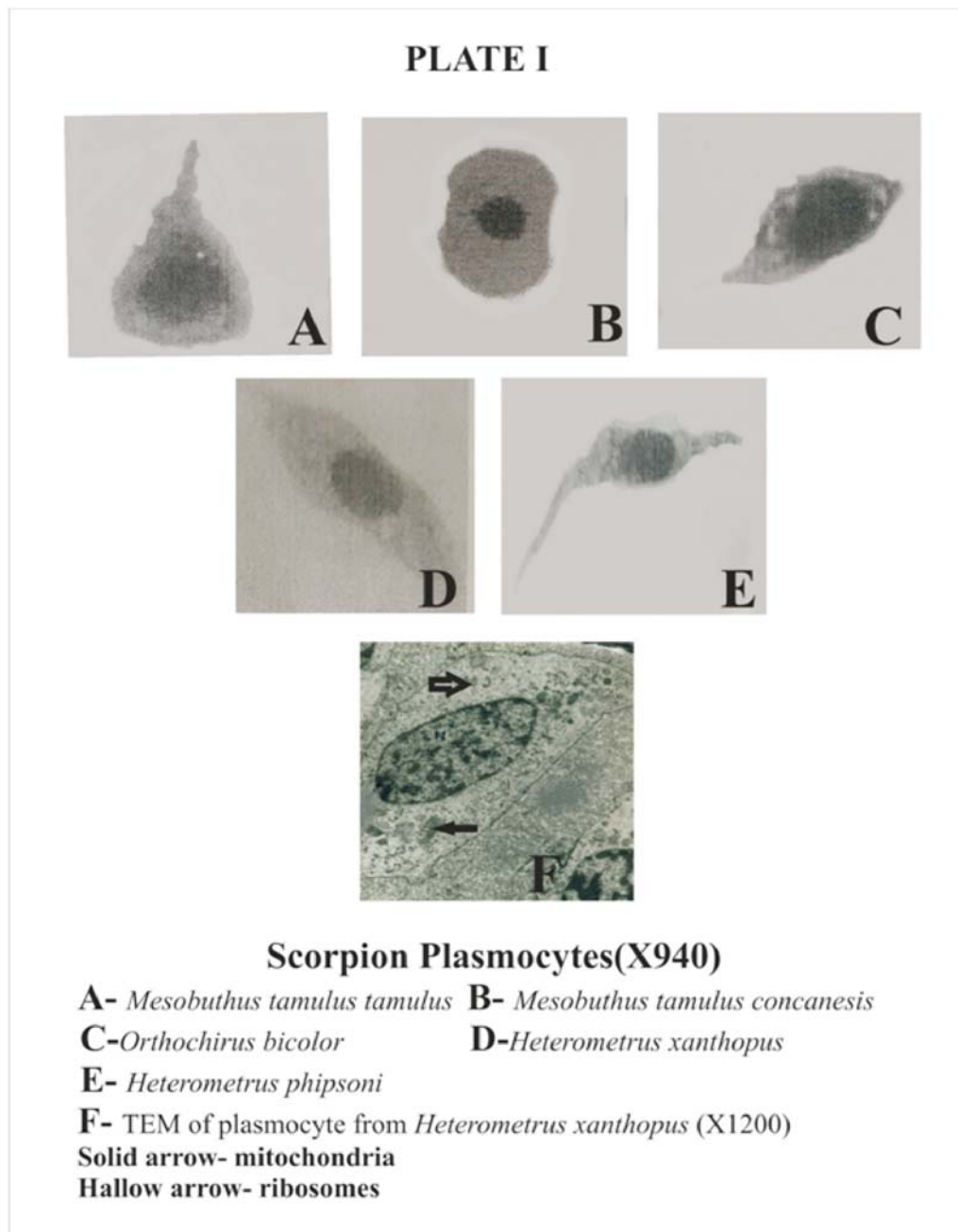
KEY WORDS: *Mesobuthus tumulus tumulus*, *M. tumulus concanesis*, *Orthochirus bicolor*, *Heterometrus xanthopus*, *H. phipsoni* cytological characteristics, TEM

Arthropoda is the largest phylum on the earth, inhabiting in air, water and on land. The study of hemocytes is the most fascinating area of research. Kollmann (1908) first reported the morphology and types of hemocytes- the blood cells, in arthropoda. Just like vertebrate blood cells, the hemocytes play important role in immune system, coagulation of hemolymph after rupture of cuticle, in wound healing etc (Millar and Ratcliff, 1989). The hemocytes are embryologically derived from intermediate mesoderm (Anderson, 1972). The morphology and nomenclature of hemocytes in different arthropods is confusing and varied. Jones (1962) and Price and Ratcliffe (1974), presented a new nomenclature system applicable for all arthropods. Though there is disagreement about the types of hemocytes, ultra structurally seven types of hemocytes are identified as- Prohemocytes (PRs), Plasmocytes (PLs), Granulocyte (GRs), Spherulocytes (SPs),

Adipohemocytes (ADs), Oenocytes (OEs) and Coagulocytes (COs) in almost all arthropods. A perusal of literature indicates that some groups of arthropoda have received detailed attention about study of hemocytes, are- insects, crustaceans and myriopods. Though some work was done on arachnids, scorpions – the living fossil, the oldest group in arthropods remain neglected. Ravindernath (1974) did a pioneer work about hemocytes of *Palamnaeus swammerdami*. In the present investigation the morphology of plasmocytes were studied in five different species of scorpion- *Mesobuthus tumulus tumulus*, *Mesobuthus tumulus concanesis*, *Orthochirus bicolor*, *Heterometrus xanthopus* and *Heterometrus phipsoni*.

During this investigation five different species of scorpions were collected from different localities

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from Maharashtra (India). *H. xanthopus*, *H. phipsoni* were collected from their natural burrows while *M. tumulus tamulus*, *M. tumulus concanensis*, *O. bicolor* were collected underneath the stones. The males and females were placed in separate perforated jars with hibiscus leaves and soil from its natural habitat. These were fed with cockroaches (Shah and Patil, 2011).

For collection of hemolymph the method employed as described by Padmanabha (1967) with some modifications. It is collected by aspiration through hypodermic needle inserted through arthroidal membrane of pedipalps. For morphological study Pappenheim'spanochrome, Giemsa stain and Leishman's stains were employed. Janus Green B was used to observe mitochondria while Sudan

Table 1. Plasmocyte Characteristics in Different Species of Scorpions

Sr. No.	Species of scorpion	Shape variation		Size variation (µm)		Nucleus-cytoplasmic ratio	Position of nucleus	Nature of staining reaction	
		cell	nucleus	cell	nucleus			cytoplasmic inclusion	nucleus
1	<i>Mesobuthus tumulus tamulus,</i>	P	R	7- 21♂ 4-11♀	3-5	35-45	C	AG	B
2	<i>Mesobuthus tumulus concanesis</i>	P	R	6- 18♂ 4-18♀	3-4	25-30	C	AG	B
3	<i>Orthochirusbicolor</i>	P	R	9- 20♂ 3-12♀	3-5	35-60	C	SG	B
4	<i>Heterometrusxanthopus</i>	P	O	10- 30♂ 3-10♀	4-10	40-50	C	SG	B
5	<i>Heterometrusphipsoni.</i>	P	E	11- 32♂ 4-11♀	4-10	45-50	C	SG	B

B- Basophilic C- Centric E- Elliptical O- Oval P- Polymorphic R- Round
AG- Agranular SG- Slightly Granular

Black B and PAS were used for cytochemistry. These techniques include light microscopy, phase contrast microscopy and transmission electronic microscopy (TEM). The quantitative methods include Total Hemocyte Count (THC) and Differential Hemocyte Count (DHC).

The morphological study was done in different five species of scorpion in both male and female. All morphological features of scorpion are similar to that of insect with slight differences. In this investigation Jones system of classification was followed.

In all five species of scorpion clearly differentiated seven types of hemocytes were observed- Prohemocytes (PRs), Plasmocytes (PLs), granulocytes (GRs), Spherulocytes (SPs), Adipohemocytes (ADs), Oenocytoids (ODs), and Coagulocytes (COs).

Plasmocyte is the unique type of hemocyte in scorpion due to its polymorphic nature. It is fusiform in *M. tamulus tamulus*, elongated in *M. tumulus concanesis*, irregular in *O. bicolor* and spindle shaped in *H. xanthopus* and in *H. phipsoni*. However these shapes are not characteristics of the species. During preparation of hemolymph smear PLs adhere the glass slide to produce broad cytoplasmic extensions. Due to this it was very difficult to measure the dimensions of the cell. This was overcome by using 2% versene fixative. In *M. tumulus tamulus*, *M. tumulus concanesis* and *O. bicolor* the length and width is 6- 12 µm. In *H. xanthopus* and in *H. phipsoni* the length ranged from 10- 35 µm while the width was 3- 11 µm (Table 1). The cytoplasm was agranular in *M. tamulus concanesis* and granular in *H. xanthopus*, *O. bicolor* and *H. phipsoni*. The granules are Sudan Black B positive indicating lipid material in it. The nucleus is centrally placed and measured about 3-

10 µm in diameter. It is round in *M. tumulus tamulus*, *M. tamulus concanesis*, and *O. bicolor*. It is oval in *H. xanthopus* and elliptical in *H. phipsoni* (Plate I).

The values PLs was 72% in *M. tumulus tamulus*, 65% in *M. tamulus concanesis*, 75% in *O. bicolor*, 82% in *H. xanthopus* and 79% in *H. phipsoni* as compare to that of other types of hemocytes.

Examination of scorpion hemocytes with TME was particularly useful in distinguishing the PLs from other hemocytes. In *H. xanthopus*, the plasma membrane of PLs clearly showed cytoplasmic projections. TEM photography also showed pinocytic vesicles indicating phagocytic nature of it.

The selection of proper technique becomes important since several methods are available for the study of arthropod hemocytes. During the course of this investigation, it was noticed that any one technique was not suitable for all hemocytes; therefore certain modifications were made with combination of some methods. In all five species the PLs represents bulk of hemocyte population. A common feature of PLs is presence of vacuoles which was not reported in all five species of scorpions. In fixed preparation PLs showed stiff spike like projections (plate I).

As described by Gupta (1985a), PLs are having large nucleo-cytoplasmic ratio, which is considered as the stem cell nature of the hemocytes (Srivastava and Richards, 1965) which gives rise to other types of hemocytes. In the present investigation the PLs of all species having common features to that of GRs. The granules in PLs are small and refractile as compare to PLs as described by Kollaman (1908).

The large number of population and presence of stiff projections might be because of its phagocytic

activity. The functional importance of the different hemocytes is still not clearly defined; hence there is ample scope to study scorpion hemocytes.

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