A. Techniques of the Study :

During the present investigation, the hemocytes were studied by employing a combination of techniques. As suggested by Jones (1962), none of the individual methods for studying the morphological types of hemocytes is entirely satisfactory for all insects or for all types of cells within a given insect. Differences in the techniques have often led to conflicting views. Each procedure provides a somewhat different view of the cells. Arnold and Hinks (1979) discuss that all methods are empirical to some extent, because of variability in the character of both, the hemolymph and the hemocytes in different species. Realising these difficulties a combination of techniques were devised, modified and employed in the present investigation.

Unfixed wet films give better understanding of the coagulocytes. These cells cannot be identified solely in the fixed and stained preparations. The coagulocytes are detected by their function. They are known to bring about coagulation of the hemolymph. In fixed and stained monolayers, there is usually no much morphological difference between the GRS and COS. Motile nature of the PLS can only be studied in unfixed wet films. The PLS when get adhered to a glass surface, show amoeboid movement and exhibit polymorphism. This phenomenon cannot be detected in the fixed and stained slides.

Phase contrast microscopy is useful to know and avoid the possible artifacts produced by classical methods of fixation and staining.

The method of smear preparation used by Shapiro (1966) is considered to be typical and is employed by many workers (Jones, 1967 a, 1967b; Gupta and Sutherland, 1968; Nappi and Streams, 1969). Presently the monolayer preparation is done by applying a very thin layer of the adherent medium to the glass slide. It is advantageous over a classical method of smear preparation, where the possibility of disrupting the cells cannot be excluded. Similarly without adhesive, many cells wash away in further staining and dehydration procedure.

Although vertebrate blood can be prepared for microscopy by placing a drop directly on a slide and drawing it out with another slide to dry quickly in air. Insect blood treated in this way, usually agglutinates before the film can be made or else shows much cellular distortion. For this reason, the hemolymph is fixed before preparing the film. It is accomplished in two ways : (1) by a suitable degree of heat fixation or (2) by dilution of the blood in a fixative. Heat fixation has been employed by submerging the insect(s) into hot water. The method is considered satisfactory (Shapiro, 1979), since hemolymph can be obtained easily and it does not agglutinate. The hemocytes do not alter their size, shape and other morphological peculiarities. Hence, this method has been

used by many workers (Yeager, 1945; Shapiro, 1966, 1967, 1968; Jones and Liu, 1969; Kitano, 1969; Arnold and Hinks, 1976). Studies by Arnold and Salkeld (1967), on the effectiveness of several preservatives or fixatives led to eliminate most of the standard fixatives. This study opened the door to consider some of the anticoagulants as the possible preservatives. The dilution of the hemolymph with aqueous fixatives is also tried and gives satisfactory results. Various anticoagulants tested include the disodium EDTA (Versene). It proved to be the best fixative (Arnold and Salkeld, 1967). In the present investigation the fixation is done by heat treatment followed by the chemical treatment. This "double fixation" has been employed for the first time in the studies on the insect hemocytes. It proved to be advantageous. Because if, the heat treatment failed to fix the hemocytes, the chemical fixation completed the process. Further, the characteristics of the various hemocytes are to be well preserved. In addition, the heat treatment has one more advantage, that it brings more cells in the circulation, which provides more cells in the monolayer for the differential count.

The hemocytes are stained with three different staining techniques. The chemical variability of the hemolymph and hemocytes modifies the staining reaction. Hence, it is advantageous to employ more than one staining procedures. This reveals, the intracellular details clearly and helps to

distinguish the cell types with greater ease. Fading of the stain was observed when graded alchol is used for dehydration. To avoid this disadvantage, preparations were dehydrated with acetone which proved to be the better dehydrating agent over ethyl alcohol.

While doing differential cell count 500 cells are classified for every preparation of each species. Many workers (Jones, 1967 b; Vinson, 1971; Arnold and Hinks, 1976) have examined ~~only~~ a minimum of five smears and have counted 100 cells per smear. Counting of more cells every time, statistically helps in minimizing the possible errors and more accurate data is obtained. The THC is done only in nine species. Many factors did not permit to do the THC in all the insects studied presently. It is mainly because of the unavailability of the sufficient hemolymph. The THC, however, is done to determine the effects of the starvation, dehydration and a combination of starvation and dehydration. This has yielded in providing beautiful results which are discussed at a later part of this Chapter.

B. Categories of the Hemocytes :

The extensive survey of hemocytes from 27 species belonging to 13 different orders reveals six categories of the hemocytes. The results obtained are concurrent with Price and Ratcliffe (1974). Classifying the cells into six types as

prohemocytes, plasmatocytes, granulocytes, spherulocytes, coagulocytes and oenocytoids have several advantages over the existing schemes. It is more simpler than the previous ones, which divided the hemocytes into nine or more types (Jones, 1962). It is wholly based on our own observations and those of Price and Ratcliffe (1974). Most previous classifications are the attempts to waive the results obtained by different workers in one species or species belonging to the same order. In such work, it is rather difficult to clarify the terminology and homologies without the personal observations rather than relying on the descriptions with inadequate photographs. Present study places great emphasis on clearly illustrating and describing the cell types and the variations there in. For this reason the present study included adequate number of insects from 13 orders.

As argued by Arnold (1979), it seems strange that after almost a century of research in insect hemocytology, there is still no consensus on one of its most basic questions: Do the different forms of cells we find in the hemolymph represent transient morphological varients of one kind of cell or constitute a number of distinctive and immutable types ? This question cannot be answered to everyone's satisfaction.

The status of the hemocyte types is inseparable from hemocyte classification. Unfortunately the hemocyte classification is constantly in disrepute. Hemocytes are classified in

different ways by different authors, by different terminologies, based on different criteria, and for different purposes. In such a jargon, new workers are forced to create new systems, that further complicate the problem. Most classification schemes are based on some aspect of cell morphology, often also with an attempt to relate cell structure to cell processes or functions. These relationships can often be envisaged by the cell shape, by the chemistry of the inclusions or by the reactions of the cell to particular conditions. Some authors view such a classification (based on cell form and function) as the ultimate scheme. No doubt it has considerable merits for some purposes, but it leads to confusion, where a hemocyte type is multifunctional or where a particular function is served by different cells in different insects.

Using multiple criteria, the hemocytes of the different insects have been classified into two to nine distinctive types (Yeager, 1945; Jones, 1962). In some insects the differences among the cells are obscured or are minor. This has led those insect hematologists (who have worked with only one species) to conclude that only one or possibly two types of hemocytes exist in all insect orders. In some other species, many stricking differences appear. This fact has led other hematologists to think that there are many different types present among the insects.

There is no need to recount here the history of

hemocyte classification or to dwell on disputes about the hemocyte nomenclature and synonymies. Jones (1962, 1964), Gupta (1969) and Arnold (1976) have reviewed the matter at length. The hemocyte classification of Jones (1962) is considerably less complex than that proposed by Yeager (1945). Jones (1962, 1964) noted that nine seemingly valid types of hemocytes had been described from all insects. He still holds this view (Jones, 1977), with an addition of the granulocytophagous cells, which he finds only in Rhodnius. The nine types proposed by Jones (1962) are prohemocyte, plasmatocyte, granular hemocyte, cystocyte, spherule cell, oenocytoid, adipohemocyte, podocyte and vermiform cell (nematocyte). His classification is the pooled result of different workers, who employed different techniques, used different criteria and for different functions. The extensive study of fifteen insect orders carried out by Price and Ratcliffe (1974), reveals only six cell types. These are prohemocyte, plasmatocyte, granular cell, spherule cell, cystocyte and oenocytoid. They have adopted the terminology used by Jones (1962). Regarding podocytes and vermiform cells, they remark that no distinction could be made between plasmatocytes, podocytes and vermiform cells. Hoffmann (1967), Devanchelle (1971) and most other workers consider them to be homologous. Adipohemocytes are not classified as a separate cell types. Wigglesworth (1956) considers them to be indistinguishable from typical insect fat body cells. The results of the present investigation support his views.

Six different types of hemocytes exist in insects and any single species may have all the six types or atleast four of them (with PRS, PLS and GRS common to all). The hemocytes described are prohemocytes (PRS), plasmatocytes (PLS), granulocytes (GRS), spherulocytes (SPS), coagulocytes (COS) and oenocytoids (OES). The terminology used by Gupta (1979) has been used presently. Cells like vermicytes (Rhodnius and Hydaticus) and podocytes (Lepisma) described by other workers are also observed. But their morphological details are most similar to PLS. More over, plasmatocytes are described as polymorphic cells. Otherwise, pyriform cells, spindle cells, stellate cells and many other forms have to be given a separate status. Which has little significance than to complicate the classification. Further, the percentage of such variants is too low as compared to the six generalized hemocyte categories (less than 1%). Arnold (1979), comments that the two of Jone's nine types, the podocytes (POS) and nematocytes (NES or VES) should be recognised as peculiar to only one or two species. They are considered to represent species - specific morphological variants. They seem similar to (but more exaggerated than) some forms found in Euxoa species (Arnold and Hinks, 1975; Arnold, 1976). Gupta and Sutherland (1966) have suggested the transformation of plasmatocytes in to podocytes. This seems to be supported by Rizki's (1962) observations in D. melanogaster. Whitten (1964), however, question the concept of POS. Devauchelle (1971) considers them as the variant form of PLS. Vermicytes

are said to be derived from PLS (Gupta and Sutherland, 1966). Lea and Gilbert (1966) think them as a variant form of PLS. According to Arnold (1976), they occur mainly just prior to pupation, but never in large numbers.

Adipohemocytes (ADS) cannot be given a separate status for the following reasons. They are filled with lipid droplets and possess stricking similarity with the typical fat body cells. Similar views have been putforth by Wigglesworth (1955) and Price and Ratcliffe (1974). They consider them to be indistinguishable from typical insect fat body cells. It has been noted (Arnold 1952 a,b; Raina, 1976), that GRS in some insects accumulate lipid droplets progressively to the point, where they can be referred to as ADS. This reason, however, cannot justify the separate status for them. Many workers never recognise the ADS as a separate type (Wittig, 1968; Akai and Sato, 1973; Costin, 1975; Francois, 1975; Goffinet and Gregoire, 1975; Boiteau and Perron, 1976; Raina, 1976; Beaulaton and Monpeyssin, 1977).

Coagulocytes are evident only in the wet films, where they are recognized with an assurance only when they erupt and cause gelation of hemolymph in the immediate vicinity. Some authors have identified the cystocytes (= coagulocytes) by ultramicroscopy. Hoffmann et al. (1968) and Devauchelle (1971), however, find little distinction at the ultramicroscopic level between COS and GRS.

It has been discussed by Arnold (1979), that PRS, PLS, GRS, SPS and OES are quite distinctive *in vivo* and *in vitro* in the fixed and stained blood films and ultramicroscopically. Separate status of these cells is quite clear. Yet apparently there are no cytological characters that distinguish each with certainty in all insects, not at least, to the satisfaction of different authors. Nor their activities are exclusive to any one type in all insects. One may suspect whether the types are mere illusions or else are not homologous in different groups of insects.

The controversies about hemocyte types stem from many sources, most important perhaps from the inherent differences in the hemocyte complex in different taxa, the inadequacy of our definition of the hemocyte type and undoubtedly also from the proclavity of hemocytes to react drastically to any change in their environment and thus, to the different techniques used to prepare them for observation. Controversial views symbolize the two main concepts on the status of hemocyte types : (1) The hemocyte types are morphological representations of phases in the life of the blood cell, each phase with a different function. (2) There are several distinct types of hemocytes, usually about five, which are immutable and serve different roles in insect physiology. These are termed "single-cell theory" and "multiple-cell theory", respectively.

Many transitional cell forms have been observed during

the present study. Transitional forms between PRS and PLS, PLS and GRS, GRS and SPS as well between GRS and COS are evident in many insects. It often created problems in classifying them in the former or later group. These observations support the "single-cell theory". The concept is also supported by many general observations and by a number of ultramicroscopic and cultural studies on hemocytes from different species (Moran, 1971; Scharrer, 1972; Olson and Carlson, 1974; Ratcliffe and Price, 1974; Crossley, 1975; Landureau and Grellet, 1975 and Beaulaton and Monpeyssin, 1977).

Fundamentally, the above theory takes into account the seemingly transitional forms of hemocytes that share characteristics of more than one tpe. It also accounts for the general shift in the relative number of the different types during larval life, from an early complex that favours PRS and PLS, to a complex near the end of larval development that is composed mainly of GRS and SPS.

The "multiple-cell theory" differs from the earlier mainly on the basis that each of the differentiated types is a separate cell line and that the cell grows, matures and degenerates without loosing its identity. The theory is supported by a number of ultrastructural, histochemical and cultural studies (Wittig, 1968; Akai and Sato, 1973; Lai-Fook, 1973; Neuwirth, 1973; Zachary and Hoffmann, 1973; Francois, 1974 and Raina, 1976).

The six types of hemocytes, if evaluated in the light of transient forms, single-cell theory and functional attributes, only three distinct categories are represented.

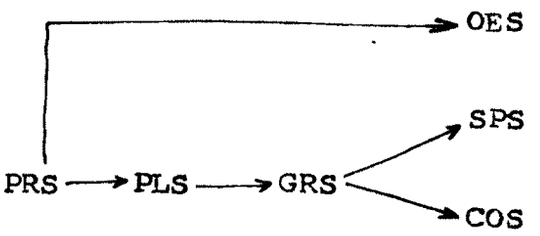
Type I Cells : Prohemocytes, cells with undisputed germinal characteristics.

Type II Cells : PLS, GRS, SPS and COS; cells with transitional forms indicative of transformation.

Type III Cells: Oenocytoids, cells with controversial origin and function(s).

Gupta (1979), reports that the GRS are the plesiomorphic hemocytes. It has been reported by several authors (Ratcliffe and Price, 1974; Crossley, 1975; Landureau and Grellet, 1975; and Beaulaton and Monpeyssin, 1977) that one hemocyte type can and does differentiate into another type. From this, it is conceivable that during evolution the plesiomorphic GRS differentiated into other hemocyte types. It can be postulated also that the GRS originate from the so called PRS or stem cells and go through the PLS before becoming distinct GRS type. In taxa that are reported to possess other types besides PRS, PLS and GRS, the last perhaps is further differentiated into SPS and COS. However, OES might have originated separately from PRS. Further more, in some taxa any of the types might have suppressed. The main differentiation pathway as postulated

above may be represented as shown below :



It is obvious from the foregoing discussion that the various criteria for distinguishing hemocyte types do not resolve the question of the status of the types. It would be possible to find the answer from the knowledge of the origin of the types in the major groups of Insecta. Validity of the types will be debated until the origin and course of development of one or several hemocyte lines are clearly demonstrated.

In conclusion, it is not of utmost importance whether the categories of the hemocytes are distinct and immutable cell types or merely developmental stages of a single cell type. Significance of the types lies in assessing their role in insect physiology. The different developmental stages of a single cell type may be capable of distinct functions, just as truly different cell types are. Even if, categories may be mere expressions of a single cell type, their morphological identity is important in knowing functional attributes.

C. Comparison of the Hemocyte Picture :

Almost a century of research in insect hematology has

yielded to provide a fairly intricate but still incomplete picture. Taking into account, the number of species studied in various orders Lepidoptera, Hymenoptera, Coleoptera and Diptera seem to be the most extensively studied groups. Only few species have so far been studied from Heteroptera, Homoptera and Odonata. The reports on the remaining orders are poorly available.

C.1. Thysanura :

Bruntz (1908), Price and Ratcliffe (1974) and Francois (1974, 1975) have worked on the hemocytes of Thysanura. Francois (1974, 1975) describes four types of hemocytes in Thermobia domestica. These are PLS, GRS, SPS and COS. Price and Ratcliffe (1974) report the occurrence of five cell types (PRS, PLS, GRS, SPS and OES) in Petrobius maritimus. In the present study five types of cells have been identified and their percentage is also calculated. The cells identified and are counted as PRS-02 %, PLS-07%, GRS-77%, SPS - 03% and OES-11 %. These figures are from the hemolymph of L. saccharina. The hemocyte complex in P. maritimus and L. saccharina seems to tally at least with the 5 types of cells. In L. saccharina the granulocytes form the major cell type. The coagulocytes occur in T. domestica (Francois, 1974, 1975). These cells however, are lacking in L. saccharina. Absence of these cells has been also reported by Price and Ratcliffe (1974) in P. maritimus. From the above studies it can be concluded that

in Thysanura four or five types of hemocytes occur.

C.2. Odonata :

Very poor information is available on the blood cells in Odonata. Gupta (1979) describes only two types, namely plasmatocytes and granulocytes. In Aeshna grandis, Price and Ratcliffe (1974), have described PRS, PLS, GRS, COS and OES. While in Libellula quadrimaculata PRS, PLS, GRS, SPS and COS are identified. Present study on Aeshna sp. yielded following composition : PRS - 04%, PLS -80%, GRS - 06%, SPS - 02% and OES - 08%. From the above it is evident that in the same genus, in one study the COS are described and in another they are reported to be absent. Instead SPS are described. Whether it accounts for the inherent differences in the nature of the hemolymph remains to be answered. Considering, even only three species as noted above, all the six types of hemocytes PRS, PLS, GRS, SPS, COS and OES are evident in the order Odonata.

C.3. Orthoptera :

Studies on J. subgullata, L. migratoria, S. gregaria (Price and Ratcliffe, 1974) reveal four types of cells (PRS, PLS, GRS and COS). Sharma and Dutta (1976), have listed nine types of cells in Crotogonus trachypterus and Acrida exaltata. They are PRS, PLS, GRS, COS, SPS, OES, POS, VES and ADS. In Crotogonus the hemocyte composition is reported to be as follows : PLS - 52-57 %, PRS -26 -32 %, GRS and COS -11 to 14%

and rest of the types form only 1-4 % of total free hemocytes in circulation. Total number of hemocytes/mm^3 ranges from 1110-3020 and 1900-3326 cells in Crotogonus and Acrida respectively. In L. migratoria (Costin, 1975) six categories of hemocytes i.e. PRS, PLS, GRS, SPS, COS and OES are reported. In the present study only four types of cells could be identified (PRS, PLS, GRS and COS) in two species and five types (PRS, PLS, GRS, SPS and COS) in the remaining species. H. nigrophlexus has PRS - 02%, PLS - 82%, GRS -08%, SPS - 04% and COS - 04 %. Its THC is 25,625 $\pm$ 427 cells/mm^3. In G. bimaculatus the 4 types are PRS - 06%, PLS -63%, GRS - 23% and COS - 08%, whereas in G. fossor following preportion exists : PRS - 02%, PLS - 11%, GRS - 81 % and COS -06 % with THC 15,562 $\pm$ 329 cells/mm^3. Foregoing results thus, show that in Orthoptera 4 to 9 types of hemocytes are described.

C.4. Dictyoptera :

Extensive survey of blood of cockroach reveals only five hemocyte types. Arnold (1972 b) describes the hemocytes of 16 species of cockroaches. Data are given on hemocyte size and numbers by him. Each family is represented in his studies. In all of the species hemocyte complex comprises of PLS, GRS, in addition to mainly germinal elements-PRS. Spherulocytes, a distinct form of GRS occur in most 'modern species' (S.F. blaberoidea), but absent in more primitive group (Blattoidea). Taxonomic relationship at the order levels are also reflected;

particularly in the morphological features of GRS. Although the hemocyte complex alone offers an insufficient evidence for systematic arrangement, it, however, provides an additional character for classifying the species. Price and Ratcliffe (1974), give the hemocyte picture of four species, Blaberus cranifer; Leucophaea maderae; Periplaneta americana and Sphodromantis bioculata and concluded that all the species contained 5 types (PRS, PLS, GRS, SPS and COS). An evidence indicating that the PLS, GRS and SPS represent a developmental series originating from PRS has been given by Ratcliffe and Price (1974). But at what stage the cystocytes (= COS) exactly arise is unkwnon. Six types of hemocytes (PRS, PLS, GRS, SPS, ADS and OES) are described during post embryonic development of Nauphoeta cinerata (Kochetova, 1978). When their entire postembryonic development is studies, it became evident that their ratios changed permanently. These changes are most likely due to the morphological and physiological changes in the organism (Kochetova, 1978). Fernandez (1977), describes six types of hemocytes in P. americana. They are PRS, PLS, GRS, OES, Cystocytes (= COS) and lamellocyte-like cells. In this study lamellocyte-like cells are probably the variant form of PLS. During the present study only five types could be identified in the following proportion : PRS - 03%, PLS - 53%, GRS-40 %, SPS - 01 % and COS -03%. Its total count is 22,291 $\pm$ 262 cells/mm^3.

C.5. Dermaptera :

Dermaptera is the most poorly studied group. Gupta (1979) described 5 types of hemocytes in his review, without mentioning the source. The hemocytes mentioned by him are PRS, PLS, GRS, SPS and COS. Price and Ratcliffe (1974) described five types as PRS, PLS, GRS, SPS and Cystocytes (=COS) in the blood of Forficula auricularia. The present study on Labidura riparia resulted in identifying 5 types of hemocytes : PRS-01%, PLS - 79 %, GRS - 10%, SPS - 07% and COS - 03%. The occurrence of oenocytoids has not been mentioned by any of the workers in Dermaptera.

C.6. Isoptera :

In Zootermopsis nevadensis, Price and Ratcliffe (1974) described five types of hemocytes PRS, PLS, GRS, SPS and Cystocytes (=COS) . Microtermes sp. (adult worker) possesses also five types. Their percentage is as follows : PRS - 02%, PLS - 28%, GRS - 49%, SPS - 14% and COS - 07 %. Further, studies on different species are needed for the confirmation of exact number of the morphological types of hemocytes in this order.

C.7. Heteroptera :

As far as is known, most of the work being confined to a few species in the order Heteroptera (Poisson, 1924; Hamilton, 1931; Khanna, 1964; Wigglesworth, 1955, 1956; Jones, 1965;

Laifook, 1970; Zaidi and Khan, 1974). Most of them described five types of hemocytes (PRS, PLS, GRS, ADS and OES) with light microscopy and four types (PRS, PLS, GRS and OES) with transmission electron microscopy.

Studies in D. singulatus (Zaidi and Khan, 1974) reveal five types (PRS, PLS, GRS, ADS and OES). In the same species, however, Price and Ratcliffe (1974) identify only four types (PRS, PLS, GRS and SPS). Other two species studied by them were Nepa cinera (5 types : PRS, PLS, GRS, SPS and COS) and Rhodnius prolixus (four types : PRS, PLS, GRS and COS).

The order Heteroptera in the present study is represented by four species. In R. prolixus five types are recorded (PRS - 01 %, PLS - 69 %, GRS - 27%, COS -01 % and OES - 02 %). C. rotundatus contains PRS - 01%, PLS - 18%, GRS - 36 %, SPS - 20% and OES -25 %. Notonecta sp. has PRS - 02%, PLS - 66%, GRS -27% SPS - 02 % and OES - 03%. However, R. sordidula shows only four types, namely PRS - 02%, PLS - 92%, GRS - 03% and OES-03%.

From the above, it is evident that the coagulocytes occur in Rhodnius and Nepa only; whereas all the other heteropterans studied have PRS, PLS, GRS and OES. In Dysdercus, the OES are reported to be absent (Price and Ratcliffe, 1974). The SPS are described in Dysdercus, Nepa, Cimex and Notonecta spp.

C.8. Homoptera :

Granados and Meehan (1973), Boiteau and Perron (1976)

and Behura and Dash (1978) investigated the hemocytes in Aphididae. Six types of hemocytes are identified in Agallia constricta (Granados and Meehan, 1973). They are PRS, PLS, GRS, SPS, ADS and OES. Whereas five types are identified (PRS, PLS, GRS, SPS and OES) in Macrosiphum euphorbiae (Boiteau and Perron, 1976). Rhopalosiphum maidis (Behura and Dash,1978) contains 4 types PRS, PLS, POS and Cystocytes (=COS) . There are five cell types in Magicicada sp. Their names and percentage are PRS - 13%, PLS -41%, GRS - 23%, SPS - 01% and COS - 22%. A given species of order Homoptera thus, may contain four to six types of hemocytes. If podocytes in R. maidis are considered as a variant form of the PLS, then the number is reduced to three (PRS, PLS and COS). Determination of exact blood picture in Homoptera requires further studies.

C.9. Neuroptera :

Data on the hemocyte types are poor in Neuroptera. Gupta (1979) mentions six types (PRS, PLS, GRS, SPS, ADS and OES) in his review. Sialis lutaria (Price and Ratcliffe, 1974) has five types of hemocytes PRS, PLS, GRS, SPS and Cystocytes (= COS) . The larvae of Croce filipennis studied presently, shows five types (PRS - 07%, PLS -48%, GRS - 42%, SPS - 01% and OES - 02%). From the account presented above it can be said that the Neuroptera, probably have five distinct cell types like PRS, PLS, GRS, SPS and OES.

C.10. Lepidoptera :

Although sufficient information is available on the lepidopteran hemocytes, there is no concensus on the exact number of cell types. The hemocyte types reported vary widely. Two to seven types of hemocytes have been reported in different species. Majority of the reports, however, reveal five distinct categories. According to Arnold (1976), all the categories occur only in Prodenia (Yeager, 1945; Jones, 1959). Five categories of hemocytes are described in Malacosoma disstria (Arnold and Schi, 1974). The cells described as : PRS, PLS, GRS, SPS and OES. By using ultrastructural characters, hemocytes of 5th instar larvae of Bombyx mori (Akai and Sato, 1973) are divided into five groups : PRS, PLS, GRS, SPS and OES. Arnold and Hinks (1976) described five types as : PRS, PLS, GRS, SPS and OES during hemopoiesis of Euxoa declarata. Five types of hemocytes (PRS, PLS, GRS, SPS and OES) have also been described in Pectinophora gossypiella (Raina, 1976), Calpodes ethlius (Lai-Fook, 1973), Choristoneura fumiferana (Dunphy and Nolan, 1980), Lambdina fiscellaria (Boiteau and Perron, 1977), Bombyx mori, Galleria mellonela and Pieris brassicae (Price and Ratcliffe, 1974). Breugnon and Berre (1976), however, report the occurrence of COS instead of OES as reported earlier by Price and Ratcliffe (1974) in P. brassicae. Controversy also exist with regard to the number of hemocytes in G. mellonela. Four hemocyte types has been observed (PLS, GRS, SPS and OES); in larval stage by Neuwirth (1973). Previous reports tell us

the occurrence of PRS, PLS, GRS, SPS and OES. Four types of the cells, PRS, PLS, spindle-shaped cells and vermiform cells are distinguished in the hemolymph of Papilio demoleus (Narayanan and Jayaraj, 1976). Nine types designated as PRS, PLS, ADS, OES, COS, POS, SPS, Rhagamtocytes and degenerating cells occur in the full grown larvae of Alabama argillacea (Habib, 1977).

Two species studied during present work contain five categories of hemocytes, namely PRS, PLS, GRS, SPS and OES. In the larvae of Clania sp. cell count is : PRS - 04%, PLS - 70%, GRS - 15%, SPS--06% and OES - 05% and THC as 16,375 ±216 cells/mm^3. Whereas in Papilio demoleus PRS -02%, PLS - 69%, GRS - 16%, SPS - 08 % and OES -05 % with THC 17,187 ± 237 cells/mm^3.

THCs for many Lepidopterans are available in the literature Prodenia eridania larvae $\bar{x}$ 17,200 (Rosenberger and Jones, 1960); Trichoplusia ni larvae 14,000-25,000/mm^3. (Laigo and Paschke, 1966); Heliothis zea larvae 25,000-31,000/mm^3. (Shapiro et al., 1969); Papilio demoleus larvae $\bar{x}$ 21,541 (Narayanan and Jayaraj, 1976). The THC reported by Narayanan and Jayaraj (1976), tallies with the THC done presently in the same species.

In conclusion it may be said that the Lepidoptera possess five distinct categories of hemocytes (PRS, PLS, GRS, SPS and OES). The occurrence of COS is reported in only one insect and hence, seems to be the exception.

C.11. Diptera :

Many dipteran species have been studied for hemocyte morphology and functions. Four categories of cells (PLS, POS, crystal cells (=OES) and lamellocytes (=PLS) are described in the larvae of D. melanogaster (Rizki, 1957, 1962). In D. euronotus (Nappi, 1970) the cells occur in the form of PRS, PLS, OES and lamellocytes. The hemocytes of Sarcophaga bullata are studied in detail by Jones (1956, 1967b). He reports the presence of PRS, PLS, GRS and SPS. Zachary and Hoffmann (1973), describe the hemocytes of larvae and young pupae of Calliphora erythrocephala, three lineages are distinguished: PLS, OES and thrombocytoids. However, Price and Ratcliffe (1974) describe five types : PRS, PLS, GRS, SPS and OES for C. erythrocephala. Six types occur in Hypoderma. Murray (1972) describes PRS, PLS, GRS, ADS and granulo-adipohemocytes along with a new type of hemocyte which he has not named. Ultrastructural studies in D. melanogaster (Shreshtha and Gotteff, 1982) reveal 2 principal cell types in the hemopoietic lobes. These are prohemocytes and proplasmatocytes. In addition there are different developmental stages of crystal cells (Procrystal cells). Prohemocytes differentiate into proplasmatocytes. After their release in the hemolymph, they transform into the plasmatocytes, podocytes and lamellocytes. Procrystal cells differentiate into crystal cells. The hemolymph has thus PRS, PLS, POS, lamellocytes and crystal cells. Presently the PLS, POS and lamellocytes are

grouped into a single category. Hence, in D. melanogaster only three types : PRS, PLS and Crystal cells (= OES) exist. Kaaya and Otieno (1981) describe four categories of hemocytes as PRS, PLS, GRS and spindle cells; in addition thrombocytoids occur but they are included in PLS. The cell categories are described in Glossina morsitans morsitans and G. pallidipes. Culex tarsalis, Psorophora ciliata, Aedes aegypti and A. atlanticus (Hall and Avery, 1978) show three classes : PLS OES and GRS. More recently PRS have also been observed in anal papillae of A. aegypti (Hall, 1983).

Adults of C. fatigans contain 5 types. These are - PRS - 01%, PLS - 88%, GRS - 08 %, SPS - 01 % and OES - 02%. The adult M. domestica show PRS -02%, PLS - 35%, GRS - 60% and OES - 03%. Whereas larvae of L. sericata has six types : PRS - 11%, PLS - 14%, GRS - 53%, SPS - 15%, COS - 03% and OES - 04%.

Foregoing discussion clearly indicates a great diversity in the blood picture of Dipterans. From two to six types are identified in different species. Most studies reveal PLS, GRS and OES as main types. While COS are recorded in only a few species.

C.12. Coleoptera :

Although number of species studied in the order are numerous, diverse results have been presented by different

workers. Two to seven types have been described. DHC of Tenebrio larvae are done by Jones (1950). According to him PLS GRS (=coagulocytes) are the principal hemocytes in it. In Melolontha larvae great variability in different types has been noticed by Collin (1963). He observed PRS, PLS, GRS, SPS, COS and OES. Price and Ratcliffe (1974) describe six types as PRS, PLS, GRS, SPS, COS and OES in Dytiscus marginalis and T. molitor. In the adult red pumpkin beetle (Mall and Gupta, 1979) enumerate five categories (PRS, PLS, ADS, GRS and OES).

Present study included five species of beetles. Out of them four species have six types of hemocytes. The remaining species shows five types. In P. belli following 5 types have been recorded : PRS - 06%, PLS -72 %, GRS - 15%, SPS - 62% and COS - 05%. The count in the Heliocopris sp. is PRS - 02%, PLS - 04%, GRS - 50%, SPS - 38%, COS - 03% and OES - 03%. Adult Z. pustulata contain PRS - 24%, PLS - 35%, GRS - 18%, SPS - 19%, COS - 03% and OES - 01%. Adult H. vittatus contains PRS - 01%, PLS - 76%, GRS - 17%, SPS - 02%, COS - 02% and OES - 02%. Whereas D. indicus possesses PRS - 01%, PLS - 91 %, GRS - 04%, SPS - 01 %, COS - 01 % and OES - 02%.

THC obtained by Collin (1963) in the larvae of Melolontha is 4,000/mm^3 and in Tenebrio larvae 48,000/mm^3 (Jones, 1950). Schwartz and Townshend (1968) indicate about 25,000 cells per cubic mm in P. japonica. Following are the values of THC done presently in the Coleopterans : Adult

P. belli 13,375 ± 240 cells/mm^3. Larvae of Heliocopris sp. 13,125 ± 161 cells/mm^3 and adult Z. pustulata 25,375 ± 331 cells/mm^3.

Most Coleopterans seem to have all the six types of hemocytes. From the remaining many of them have at least five types. Only few species possess less than four types.

C. 13. Hymenoptera :

Four to six types of hemocytes are described in different species of this order. Zapol 'Skikh (1976) has classified hemocytes of the larvae, pupae and adult workers of Apis mallifera into PRS, PLS, OES and SPS. But Price and Ratcliffe (1974) describe PRS, PLS, GRS and SPS for the same species. In Formica rufa and F. fusca Zapol' skikh et al.(1981) recognise four types of cells: PRS, PLS, SPS and OES. A similarity is noticed in the composition of hemocytes in ants and bees. Hemocytes of Sawflies, Diprion pini; D. similis; D. sertifor; Gilpinia frutetorum have been studied by Stroganova and GulII (1973). They distinguished six types as proleucocytes, micronucleocytes, macronucleocytes, basophilis, phagocytes and oenocytoids. Zapol' skikh (1976) describe proleucocytes, macronucleocytes, phagocytes, oenocytoids, macrophagocytes and chromocytes in the sawfly Heterarthrus acropodae. Chromocytes according to him are the analogue of erythrocytes. Green color of the cytoplasm may be due to chlorocruorin, a green pigment found in some invertebrates.