Chapter 1
Introduction
1.1 General Introduction

Nature is too great and complex to be conquered by man and in their battle to outclass each other, nature and man have never been able to strike a trace. Man's interference with the balance of nature in any manner immediately creates a disturbance. The continuous large scale interference by man has created serious problems for the very existence of mankind and other animals (CCIRG, 1991; Baur et al., 1993; Beerling et al., 1994).

Study of living organisms would not be completed without probing their relationship with environmental entities. Environment is the sum of many abiotic and biotic factors, interacting constantly. The organism not only exists in this dynamic, fluctuating complex but also it is a part and parcel of it (Crawford-Sidebothen, 1972; Diaz et al., 1998). The environment is neither homogenous nor unchanging certain minimum conditions such as sufficient warmth and food are necessary for the continued existence of living organism in any environment (Chatfield, 1976; Magnuson et al., 1979; Hulme P., 1994; Ledergerber et al., 1997). The animal is in constant confrontation against the exogenous factors for its survival. Because of the restraining influence of the ambient factors the organisms are not uniformly distributed over the earth; this is the recurring conclusion drawn by the environmental physiologists (Edward et al., 1987; Gates, 1993).
Climatic changes were observed in the last few years since the last glacial maximum, has affected the composition and structure of plant and animal communities (Peters and Lovejoy, 1992) but two elements make the changes in climate presently predicted different from those that occurred in the past. The increase in the concentration of CO$_2$ in the atmosphere is now much faster than that in the past, as is the rate of predicted warming as a consequence of the greenhouse effect (Houghton et al., 1996). Temperature is considered as a critical environmental factor in the ecology of most of the organism (Precht et al., 1973; Magnuson et al., 1979; Vianey-liaud, 1982; Ahmed and Raut, 1991; Raut et al., 1993). It can act as both a trigger for the commencement of a biological process and as a threshold essential for its continuation.

It is predicted that climate change will have an important impact on the dynamics of plant and animals and communities (Solomon and Shugart, 1993; Graves and Reavey, 1996). A forecasted increase in winter temperatures, and an increase in anticyclonic activity, potentially leading to an additional 20% rainfall during summer, more summer thunderstorms (CCIRG 1991, 1996; Beerling and Woodward, 1994) are likely to have a considerable effect on ectotherm animals such as snails and slugs (Crawford-Sidebotham, 1972). Land mollusc population composition and abundance could change, not only because
of physiological responses to a new climate, but also as a consequence of a shift in plant community composition and structure.

The climate manipulations produce changes in the composition and structure of the plant community in the grassland (Clarke, 1998; Sternberg et al., 1999). These changes had a significant effect on the relative abundance and activity of terrestrial gastropods. Snails and slugs consistently responded to changes in the physical environment. Furthermore, changes in the vegetation altered the distribution of food availability. The increase in the number of gastropods collected in warmed and watered quadrates when the treatments were operating, demonstrated their capacity to respond to changes in local climatic conditions (Webley, 1964; South, 1992).

The differential response of snail and slug species to the climate manipulations enables a prediction of what might occur to mollusc populations in the grassland in a changing climate. Warmer winters will probably result in a general increase of mollusc activity and herbivore. If the increase in temperature is associated with an increase in summer rainfall that results in a closed sward dominated by perennial grasses, the population size of gastropod species such as *D. reticulatum* or *C. intersecta*, which feed mainly on seedlings of annual forbs and legumes, will decline. Under drought conditions, we predict an increase in decaying leaf material and litter cover (Sternberg et al., 1999) and a
consequent increase in the populations of gastropod species that feed on dead plant material, such as *M. cantiana*.

In a changing climate, the calcareous grasslands is not straightforward, because distribution and abundance are strongly influenced by intrinsic population characteristics such as growth rates and relative densities, by interactions with other organisms through competition, herbivory and predation, and by abiotic changes that occur simultaneously (Graves and Reavey, 1996, Morton, 1999).

The terrestrial mollusks mainly face the water scarcity problems in the environment of variable humidity and temperature. This problem is solved by various physiological processes that is controlling permeability of water, temperature regulation and excretion. The physiological and biochemical changes in the unfavorable conditions have been well studied by Florkin and Scheer (1972). The effect of climate change on land mollusc populations have been experimentally studied in enriched CO$_2$ chambers (Ledergerber et al., 1997; Diaz et al., 1998; Sternberg, 1999, 2000).

The organism in a harsh environmental complex, more survival is the animal level of performance, but for the perpetuation of the species, more highly integrated processes leading to reproduction must be operational. Irrespective of the levels of performance demanded of an organism a constant interchange of energy take place between the
organism and its ambient environment (Webley, 1964; Masters et al., 1998).

To be functional, the organism must have energy input followed by effective utilization of energy. Over and extended period of time, energy input must equal energy utilization if an organism has to maintain itself. When excess of energy is available, the animal may either undergo rapid growth and or store the excess energy for future use. The bioenergetics of an animal is completely dependent on an input of energy and chemical necessities in the form of food. The taking in of food in walls separates processes: 1) Perception 2) Capture and 3) Ingestion of food. Perception of food depends upon the ability of the organism to be stimulated by some external cue. This cue may be a chemical entity or it may have a physical basis. The relative dependence of feeding on specific physical and or chemical mechanism is associated with the ecology and mode of existence of the species.

A living organism is both structurally and functionally adjusted to the environment in which it is living. It must respond to external stresses in such a way that its internal environment is maintained in the optimum condition for the continuation of its metabolic reactions (Peters and Lovejoy, 1992; Baur et al., 1993). The animals in diverse environments are confronted with difference problems conversely in the same environment different animals react to stress in different ways.
The organism has to face a variety of environmental factors like water, organic food, oxygen, carbon dioxide, light, pressure, radiation and temperature (Gibson, 1986; Buckland, 1994; Ledergerber et al., 1997; Diaz et al., 1998).

Acclimation refers usually to the compensatory change in an organism under maintained deviation of a single environmental factor (usually in the laboratory). If acclimation is complete a measured rate function is the same under one environmental condition as under another. Acclimation refers to those compensatory changes in an organism undergoing multiple natural deviation of milieuclimatic, physical and biotic. The majority of living organisms live in an environment where temperature fluctuates both diurnally and seasonally (Clarke, 1998). Temperature is the most important environmental factor limiting the distribution of living organisms. Terrestrial animals are subjected to too much greater fluctuations in the temperature than aquatic ones, and their body temperature is closely related to their water balance. The rate of chemical reaction increases as the temperature rises (Uvarov, 1931; Getz, 1959). Generally speaking organism acclimatized to low temperature (cold/latitudes) have a higher metabolic rate than similar organism acclimatized to a higher temperature (Summer condition/equatorial latitude). If an animal is kept at the altered temperature for many days its rate functions often show some
compensation, that is it becomes acclimated. The nature of physiological adaptation of poikilotherms to constant temperature has been investigated to some extent (Bullode, 1955; Prosser, 1955, 1958; Fry, 1958). Lymnaea are adapted to a wide range of temperature (Prinsloo and Van Eeden, 1969). If it is now returned to the original temperature the rate function does not returned to the original level but rather to a higher or lower value according to the direction of acclimation (Wells, 1944; Getz, 1959). Not only can acclimatization compensate for changes occurring within viable limits but they can also extend these limits of resistance of extreme temperature. This form of compensation is usually called resistance adaptation.

Slug is depressed transiently by a rise in temperature and stimulated temporarily by a fall in temperature. Animals from cold water have an upper lethal temperature, which may be below the low lethal temperature of similar animals from warm water. For example *Aurelia* from Halifax had a high lethal of 26.8°C to 28°C while in those from Tortugas and Florida it was 40°C Hamml et al., (1955).

The range of the species is determined through natural selection by its limits of tolerance. One environmental factor, such as salinity, may limit the distribution of one group; another factor, such as temperature may limit another group. Over an ecological range individuals may vary within limits set by the genotype, a phenotype results from the balance of genetic and environmental factors.
Three general time scales can be distinguished in response to an environmental alteration:

1) There is a direct reaction to the environmental changes. These may be mediated by stimulation of sense organs, which initiated chains of responses, or there may be direct effect on metabolic reactions. Changes in ions, temperature, oxygen and foodstuffs may lead to alteration in enzymatic reaction rates. Frequently if the environmental change is abrupt or great, as with cooling or warming, the animal shows an initial overshoot or series of oscillatory metabolic reactions before setting down to a new stabilized rate. The time for the direct response or new rate is usually in minutes or hours.

2) The time period in the response of an organism to an environmental alteration may require days or weeks. This is a period of acclimation or compensation and the magnitude of the physiological adjustment varies with the amount of environmental change. In seasonal change the compensatory alteration may develop gradually. Slow salinity shifts in estuaries and seasonal changes in temperature and available food may involve migrations or changes in hormone balance and in reproductive activity.

3) A third type of response to environmental change is that of regulation of internal state with respect to environmental parameters.

Usually prolonged period of thermal extremes both high and low
temperature have resulted in the mass mortality of organisms. Lethal
temperatures are alterable according to acclimation temperature. In
addition to changes in lethal temperature, poikilotherms show alterations
in various rate functions, in behavior and in biochemical activity in
accordance with temperature of acclimation. When the temperature is
raised or lowered abruptly, most poikilotherms shows an initial
overshoot or shock reaction. Part of the initial metabolic response is
due to increased motor activity resulting from sensory stimulation but
there may be also a general cellular effect. Slugs are most active in
response to a falling temperature in the range 4°C to 20°C it is known,
whether the specific surface theromoreceptors or whether responses
result from changes in temperature of the nervous system (Stellen and

Both a snail (deroceras) and an insect (carausicus) show no
acclimation in low temperature and both are dormant in low temperature
and winter. Species that become dormant seasonally may show some
temperature acclimation during their active phase; failure to take
torpidity into account at low temperature has diminished the value of
many acclimation.

Slugs are members of the phylum mollusk. Mollusks are the
animals, which have come on land but are still dependent on the moist
environment and are inactivated by draught and low temperature. Like
all animals, as long as the mollusks are not considered by man to be either useful or destructive. The mollusca have been divided into 7 classes. Out of these, Gastropods being the biggest class comprising of nearly 80,000 known species distributed to all over the world (Hyman, 1967). Gastropod being ectotherms shows variations in physiological processes with varying environmental conditions. Also the variation in the form and physiology of mollusks are greater than in any other invertebrate phylum (Ghirette, 1966) and all the metabolic processes of mollusca are in some way or the other dependent on the fluctuation in the environmental parameters. Gastropods are divided into three subclass prosobranch, opisthobranchia and pulmonata. The terrestrial snails and slugs are all pulmonates. The slugs and snails are much like some insects in their biology.

Thousands of kinds of land slugs exist in the world, varying in size, and color (Coldrey, 1987). Some slugs are extremely small, less than one inch in length, while others are over ten inches. Black, brown, white, yellowish and gray are all hues that are attached to certain slugs. Slugs are extremely similar to snail except slugs do not possess the large shell that is attached to snails. Slugs do, however, have a shell that is hidden by the fleshy mantle on their backs (Olkowski et al., 1991).

The slug’s body is a very interesting system that allows the organism to thrive in its environment (White, 1953; Webley, 1964;
South, 1992). Slug move slowly with the help of soft foot. Foot secretes mucus. The waves of muscular contractions allow the slug to travel. Joined to this foot is the slug’s head where two pairs of retractile tentacles are located. The upper pair serves as optical tentacles. These “eyes” allow the slug to determine day or night and see dim shapes (Coldrey, 1987). The lower pair of tentacles is usually shorter and serves as sensory tentacles, which feel the environment of the slug. They have a remarkable ability to elongate and contract the body. When fully extended in travel the body is elongated, narrow and nearly cylindrical. When contracted the body is shortened to less than 1/4th of its former length, becomes thick and stout (Lewis, 1969). The tentacles are retracted and the head is concealed beneath the mantle. Also on the head of the slug is the mouth. The mouth contains a tongue like radula that can move back and forth carrying food to be eaten by the slug’s tiny teeth. The rest of the slug’s body is a lump covered by the mantle, a thin strip of skin. The mantle cavity lies between the mantle and the main body. This cavity allows slugs to take in water and air for respiratory purposes and excrete wastes. Slugs are covered with a tough, slippery skin that is not waterproof and must be kept wet for survival. Slugs prevent themselves from drying out by secreting mucus to protect their bodies (Key, 1902; Kulkarni, 1970).

Movement of plant material facilitates the dispersal of these pest
snails and slugs (Dundee, 1977). As the slugs mature, they become functional males and then true hermaphrodites (Self fertilization has been observed in some species). Older slugs are females (Paraense, 1955; Karlin, 1961; Peterellis and Dundee, 1969; Nagabhushanam, 1963).

Slugs live in a variety of environments but in general, the environments must be damp and cool (Wells, 1944). Semperula is mainly concentrated in the Australian region. *Semperula maculata*, *Semperula parva* and *Semperula wallaces* are found in the Australian region, close to the Wallace line, with the exception of the latter which can also be found on the island of Upolu, much further to the east (Gomes and Thome, 2004). Slugs are nocturnal. During the day slugs hide in damp, shady places under stones, logs, rotting leaves, or between crevices. At night, when the environment is cool and damp, and there is no danger of their bodies drying out, slugs leave their hiding places to forage for food. In very cold weather, like other animals, slugs hibernate underground until warmer weather returns (Webley, 1964; Coldrey, 1987).

Slugs are apparently not repelled by light, but are repelled by rising temperatures. As temperatures rise, slugs crawl down to their hiding places on the soil surface to rest and absorb water through their skin. As temperatures start to fall, slugs actively begin foraging. Thus slugs may be active during the day after a cooling shower as long as the
temperatures decline or remain steady. Slugs are very sensitive to ambient temperature and can detect temperature changes as gradual as 2°F per hour. Slugs prefer to remain at 17°C to 18°C (Chernin, 1967; Prinsloo and Van Eeden, 1969; El-Hassan, 1974). Slugs can withstand slight freezing temperatures although their tendency to take shelter in cold weather protects them from freezing. Slugs try to escape from temperature higher than 21°C. Slugs are also sensitive to air currents. Gently breezes elicit a positive response in which the slug turns toward the source and extends its antennae. As the breezes become more brisk, the slugs turn away from the source evidently to escape dehydration. Slugs can withstand brief periods of immersion under water, although they drown after several hours.

Most of the slugs are herbivores feeding on fungi, lichens, green plants, shoots, roots, leaves, fruits, vegetables and flowers. Slugs also play an important role as scavenger eating decaying vegetation, animal feces and carrion (Chatfield, 1976; Rees et al., 1992; Anzeijer Fur Schadling Skude, 2002).

As in other gastropods, pulmonates serve as intermediate host of trematodes. Iles (1959) reported 22 species of Cercariae corneus and Lymnea stagnalis. The slugs are the most successful stylommatophoran pulmonates as far as their adaptability is concerned (Kulkarni, 1970). They can withstand adverse climatic conditions.
Slugs and snails are economically very important. Since they cause damage in agriculture, horticulture and forestry (Collinge, 1921; Dundee et al., 1965; Gilmore, 1982; Diesler et al., 1984).

Slugs are also more of a problem with root crops, buried seeds, and seedling. There are reports of slug damage to many ornamentals like Aconitum, Chrysanthemum, Coleus, Dahlias, Hibiscus, Ivy, Orchids, Poppies, Primroses, Roses and Zinnias. Bulbs and tuber crops such as Irises, Calla lilies, Lilies, Narcissus, and Tulips young conifer trees could be damaged or killed by aggressive slug attacks (Dundee et al., 1975; Martin, 1991). Pulmonate gastropod slugs form serious horticultural pests resulting in low yields of crops such as Coffee, Cereals, Rubber, Potato etc. Hunter, (1969b); Runham and Hunter (1970, 1971). Slugs are transmitters of various plant diseases like fungi.

The slug *Arion hortensis* not only consuming all the above surface parts of plants but burrowing down to get at the roots. The Slug *Milax budapestensis* specialists at root destruction, attacking crops such as potatoes carrots and beet roots, slug *Cryptozona bistrialis* feeds rarely on tender part of rubber plants as reported in central Nursery of Rubber Board, Karikkatooor, Kottayam District (Wester et al., 1966).

Slugs may transmit plant pathogens (Wester et al., 1966). The knowledge regarding their role as intermediate hosts of disease
parasites of animals (Chichester and Getz, 1968) is limited. Most important to man are the planorbid snails that serve as intermediate host for schistosomes, very serious human parasites. Pulmonates are economically important as intermediate host of strongyloid nematodes of mammals. *Achatina fulica*, can serve (Cheng, 1973) the intermediate host carrier eosinophillic meningo-encephalitis. Lymnaeids have drawn the attention of a large number of researchers because of their role as hosts for larval stages of the helminth parasites which cause disease in man and domestic animals (Liston and Soparkar, 1918; Rao, 1933; Chatterjee, 1952; Malek and Cheng, 1974; Ghosh and Chauhan, 1975; Godan, 1983; Burch, 1985; Raut, 1986). Considering their involvement in regulating the life-cycle of the worm parasites various aspects of the biology and ecology of these freshwater gastropod snails were studied by Seshaiya (1927), Noland and Carriker (1946), Kendall (1953), McCraw (1970), Berrie (1965). Van der Steen et al., (1973), Raut et al., (1992), Raut and Misra (1993), Misra and Raut (1993) with a view to developing methods for control. Although additional information on ecology of lymnaeids is available (Jong-Brink, 1990, Moens, 1991).

Slugs also carry helminthes parasites to poultry. Snails are vectors of trematod parasites also (Jong-Brink, 1990). Indian snails *Indoplanorbis exustus* and *Gyrulus convexiusculus* serve to be
intermediate hosts for trematode parasites *Echinostomamusi* and *E. ilocanum* respectively. The second trematode parasite mentioned is well known parasite of dogs, rats and man. List of such vector snails and their respective trematode parasites is given by Rao, 1933; Cheng, 1973; Godan, 1983; Raut, 1986, Raut and Misra, 1991, 1993.

Furthermore they are of importance in medical and veterinary practice, since they serve as intermediate host for certain parasitic worms of man and his domestic animals (Sturrock, 1972; El-Hassan, 1974; Appleton, 1977).

Slugs have occupied an important position in the ecosystem of the world. Like other organisms, slugs are not as much in danger of being prey because of its sticky, slimy mucus (Coldrey, 1987). However predators of slugs exist. They include invertebrates and vertebrates like Hedgehog, Badger, Shrew, Mole, Mouse, Frog, Toad, Snake, carnivorous beetle some birds and some small mammals (Slugs: The organism WEB page; Pelseneer, 1935; Stephenson et al., 1966). Insect specially coleoptera and diptera, are among the serious predators of pulmonates.

Minor relationship to human include the eating of *Helix pomatia* and other large helicids in Europe, and of the strophocheilidae by Indians in south America and the production of humus by land snails.
This indicates that slugs have an important place in the ecosystem and the food web of the environments in which it lives. Some organisms depend upon the slugs, in turn, depends upon other organisms.

Although some may think that the extinction of the slugs would be "good riddance" but the presence of the slugs is very important. Ecologist Teearu states "Each and every being on earth has its place (a 'niche') in the ecosystem together it works". Teearu, et al. If the slug was to suddenly disappear the ecosystems and food webs that contained the slug would be drastically affected (Van Der Steen et al., 1973). Ecologist Amos explains, "the removal of any single species of animal from a food web has effect throughout the web."

Predators of the slug would have to search for new food sources others, too, would be affected by the disappearance of the slug. Wildlife ecologist Studenroth State, "In ecosystem slugs are important in breaking down decomposing materials faster and helping to release nutrients back into the overall system quicker". Soil would lose important nutrients, without slugs recycling of decaying and fecal matter. Thus plants and crops would not grow as well. This of course would create a domino effect, affecting the entire ecosystem and food web of organisms.

Teearu clarifies an important point "slugs may seem slimy and horrible to us, but we need them just as much as we need cow for milk"
and chicken for eggs”, while many people think that the removal of the land slug species from the earth would be beneficial, but the disappearance of slugs would have a negative impact on humans. In addition to making up the biodiversity of this world, slugs play a crucial role in the ecosystem that has a direct impact on humans. For example, if the slugs did not perform their important role as a decomposer, there would be domino effects, which would reach humans. As Studentroth explains “nutrients would be locked up in dead organisms longer and unavailable to living organisms.” This “locking up of nutrients” would affect the fertility of soil (Carrick, 1942; Barnes et al., 1944, 1945; Burch, 1955). Fertile soil is very important for human existence. As Tina Teearu states, “at the end of the end, absolutely everything we humans eat can be tracked back to soil” While gardener and farmer may think that the disappearance of slugs would improve their garden and crops from being destroyed by the pest slug, they are wrong. If slugs become extinct, soils would be less fertile and as Amos states “other species would be obliged to compensate for their absence”. It means that other organisms deemed as “pests” would take the place of the slugs. Even master Gardner Monica, Strom, DTM admits that “slugs have a very important purpose”.

Slugs are also important to humans in lesser known ways currently; the slug is being studied at the University of Washington. The
biochemical properties and cellular mechanism of the slugs' mucus are under investigation. Researchers at the University believe that learning more about slug mucus could help in treating or curing human deficiencies involving mucus. Deyrup Olsen explained, “This (the slug’s mucus chemical make up) has a potential relationship to human diseases in which mucus formation is abnormal (Newton, 1955; Malek and Cheng, 1974; Misra and Raut, 1993). An example is a genetic based disease cystic fibrosis” If the slugs were to be become extinct, it would halt this ongoing research.

In conclusion, land slugs are an important species on the earth. They play significant role in their ecosystem and serve humans in many ways. Through Shep investigation on the slug, about biodiversity on earth although an organism may appear disgusting and insignificant, every organism plays an extremely important role on earth. Extinction of one life form will affect many life forms and every one must realize this and strive to support biodiversity. The perusal of literature indicates that the studies on various aspects of slugs are of great importance because of their role in food web that is ecosystem.

1.2 Temperature tolerance

Temperature tolerance limits the distribution of animals and at the same time determines their rate of activity. In general, life activities occur within a range of about 40°C. However, most animals live within
much narrower limits. It has been well established that the fish becomes more increasingly tolerant to heat as it becomes acclimatized to higher temperatures within the range of thermal tolerance of its species, and that its acclimation to low temperature entails a loss of heat-tolerance (Doudoroff, 1942; Brett, 1944, 1946). When compared with the fish the literature on the temperature tolerance in the invertebrates is very scanty. In many cases the lethal point was determined at the temperature at which death of the animal occurred when it was kept to slowly increasing temperatures (Huntsman and Sparks, 1924; Gowanloch and Hayes, 1926; Afanasjev et al., 1997). But the results thus obtained could not be compared by various authors due to the raising of temperature at different rates and because the time for increase of the temperature has got a marked effect on the final lethal point. Animals, which were maintained at constant temperatures provided an accurate determination of the lethal point for a period of time and it, helped much for a comparison among the species (Prosser, 1955).

Acclimatization to low and high temperature has been described in the animals on the basis of laboratory acclimatization. Dehnel and Segal (1956) in a laboratory study on American cockroach, Periplaneta americana, demonstrated higher rate of oxygen consumption in nymphs and adults after acclimatization of these to
10°C. When compared with the animals acclimatized to 26°C, Ohsawa and Tsukuda (1956) worked on the periwinkle, *Nidolittorina granularis*, observed seasonal acclimation of response to temperature. Microgeographical acclimation has been demonstrated in the gastropod, *Acmaea limatula* (Segal, 1956).

Another facet of acclimatization is its effect on the temperature tolerance of a species. A previous temperature history at the upper levels of the physiological temperature range is known to raise the thermal resistance. On the other hand, acclimatization to low temperature will decrease the tolerance to high temperatures. Edwards and Irving (1943), ORR (1955) reported the thermal death point of the sand crab, *Emerita talpoida*, to be about 10°C higher in summer than in winter. Marlier (1949) found indications that the lethal temperature of larvae of the caddis fly, *Hydropsyche angustipennis*, increased from 31°C in the spring to 32°C in early summer (Prosser and Brown, 1955). Effect of temperature, and salinity on heat tolerance in two grapsid crab, *Hemigrapsus nudus* and *Hemigrapsus crenegensens* (Todd et al., 1960; Kulkarni, 1978).

In general, animals that become torpid or lethargic in the cold show much less capacity for acclimation than those that remain active over a wide temperature range.

Relatively little appears to have published about the temperature
relations of the slugs. Segal (1961); Eyvor (1967); Mc Mahan et al., (1991) worked on the American slug, *Limax flavus*, mentioned that animals acclimatized to the cold have a higher rate of oxygen consumption than those acclimatized to the warm, when both are measured at the same temperature. Southward (1958); Nagabhushnam and Kulkarni (1970); Kulkarni and Tankar (1984); Kulkarni et al., (1985); Mc Mahan et al., (1994, 1995); Panigrahi, (1998) studied the temperature tolerance in the slug. Maintenance of zebra mussel under laboratory condition (Nichols, 1992). Respiratory response to temperature and temperature tolerance of some intertidal Gastropods has been reported by Eyvor, 1967. In the present study, it has been observed that the slugs. *Semperula maculata*, are abundant in moist soils the temperature of which reaches to about 40°C. In view of its convenient size, longevity and availability, the temperature relations of *Semperula maculata* are of considerable interest not only to the ecologist but also to the physiologist concerned with the problem of explaining the mechanism of acclimatization to temperature.

### 1.3 Neurosecretory cells

Since (Scharrer, 1928, 1933) reported the occurrence of secretory cells in the hypothalamus of various vertebrates. Many investigators (Gabe, 1955; Van Mol, 1960, 1962; Antheunisse, 1963; Wigdenes et al., 1980) paid attention to neurosecretory phenomena.
The results of these studies with vertebrates showed that hormones are produced in neurosecretory cells of hypothalamus and transported via the axons of these cells to the neurohypophysis (Baranyi and Salanki, 1963). In this organ the hormones are stored and eventually released into the blood (Bargmann, 1949). In case of vertebrates the presence of neurohormones was ascertained. Such a system with neurohormonal factors and a central position of neurosecretory cells in the brain have been found also in various groups of invertebrates.

The products of neurosecretory cells are known for coordination and integration of various body processes such as growth, metabolism, reproduction and thus help the animal to maintain harmony with the environment. A modern definition of neurosecretory system would include a neurosecretory with neuronal and glandular morphologies, neurosecretory material, a release site for neurosecretory product and a target (Boer and Joosse, 1975).

In pulmonates, nervous system has several advantages, such as giant size of neurons, simplicity of structure, complexity of function and easy availability as compared to the rest of the molluscs. Some aspects of neurosecretion have been studied histologically in a number of pulmonates. Detailed maps of the neurosecretory systems are available for three species of Stylommatophorans that is in slugs, *Arion hortensis* and *Deroceros reticulatum* and in the snail, *Helix aspera*.
(Wigdenes et al., 1980) and Onchidium verruculatum (Deshpande, 1980).

Among the molluscs, several observations have been made concerning the neurosecretory cells of gastropods, but rather few about the central nervous system of other groups. In gastropods, the presence of neurosecretory cells in various ganglia were reported by Gabe (1951, 1953a, b); Lever (1957) described five types of neurosecretory cells in a freshwater pulmonate, Ferrissia sp. Herlent-Meewis and Van Mol (1959) and Van Mol (1960 a, 1962) described two types of neurosecretory cells in Arion. Krause, (1960) described cell of Types I and II in the cerebral and visceral ganglion of Helix pomatia and suggested that types II cells of the visceral ganglion might produce a hormone which controls hibernation. While Krause, (1963) distinguished only two cell types in Helix. The presence of neurosecretory cells in the central nervous system of lamellibranchs was reported by Gabe, (1955). He observed these cells only in the cerebral and visceral ganglia of 20 species, all belonging to one cell type. Lubet (1955) also described one cell type in the cerebral and visceral ganglia of Mytilus and Chlamys. On the other hand, Fahrmann, (1961) and Nagabhushanam, (1963) reported two types of neurosecretory cells in Unio tumidus and Crassostrea virginica respectively.
The histological structure of the neurosecretory cells in the nervous system of stylommatophoran pulmonates like *Vaginulus* and *Ariphanta ligulata* were studied in detail by Nagabhushanam and Swarnamayee (1963, 1969). Lane (1964a, 1964b and 1964c) observed the fine structure of secretory cells in the optic tentacles of *Helix*. Quattrini (1964, 1965) identified elementary neurosecretory granules in the neurones of the slugs like *Milax* and *Vaginulus*.

Thus most of the observations so far made on the neurosecretion in pulmonates deal with description of cell types while the work on their functional aspect is very little. Pelluet and Lane (1961) showed the relation between neurosecretion and cell differentiation in the ovotestis of *Arion ater, A. subfuscus* and species of *Milax*.

In lamellibranchs some investigators have studied neurosecretory phenomena. Gabe (1955) published a table with 20 species of marine lamellibranchs, in which neurosecretory cells were observed. These species included not only protobranchs and filibranchs but also pseudolamellibranchs and eulamellibranchs. Small neurosecretory cells were found only in cerebropleural and visceral ganglia. Lubet (1956) found a relation between the neurosecretory activity and reproduction in *Mytilus* and *Chlamys*. Gabe and Rancurel (1958) found distinct neurosecretory cells in the cerebropleural and visceral ganglia of teredinids but not in the pedal ganglia.
Clark (1959); Fahrmann (1961); Shukla et al., (1979) studied neurosecretory phenomena in the ganglia of the freshwater lamellibranch, *Unio tumidus*. Phenomenon of neurosecretion though studied in several pulmonate gastropods (Boer and Joose, 1975).

Rozsa and Nagy, (1967) studied physiological and histochemical evidence for neuroendocrine regulation of heart activity and Joose (1963, 1964) observed dorsal bodies and neurosecretory cells of cerebral ganglion in *Lymnaea stagnalis*. Nagabhushanam and Lomte (1981), made some observations on the cytology and cytochemistry of the neurosecretory cells of *P. corrugata*. Effects of environmental parameters on the neurosecretory activity in molluscs have also been extensively studied. Experiments were performed by various authors in different species of molluscs such as *Lymnaea stagnalis* (Lever and Joosse, 1965), *Cryptozona semirugata* (Mantale, 1970), *Lymnaea auricularia* (Hanumante et al., 1978) and *Laevicaulis alte* (Kulkarni, 1982).

In the present investigation, study was undertaken in the slug, *Semperula maculata* in relation to neurosecretory cells with reference to thermal aspects on them.

### 1.4 Biochemical profiles

Since organic constituents acts as a key substrate for metabolism. The animals living in a relatively constant thermal environment do not have to face with such extremes and their metabolic processes may show a compensatory shift ([Vernberg, 1962](#)). In the terrestrial environment temperature causes adverse effect on terrestrial organisms at cellular or molecular level and ultimately leads to disorder in biochemical composition. It is to be expected that the ability to exist at an environmental temperature be expressed in the physiological and biological responses of the animals. Biochemical correlation with acclimated temperature ([Langridge, 1963](#); [Rao, 1967](#); [Praveenkumar and Umadevi, 1998](#)).

May (1932, 1934) made a detailed study on galactogen in *Helix pomatia*. Meenakshi (1956a) worked on the seasonal variations in the glycogen and fat content in *Pila virens*. The biochemical studies in relation to hibernation were made by Meenakshi (1955, 1956a) in *Pila*, Thiele (1959) in *Helix pomatia* and Mitra and Sur (1989) in *Achantina fulica* and *Pila globosa*. Studies on carbohydrate metabolism in the land slug, *L. alte* were carried out by Kulkarni
(1973); Nanaware (1974). Ramamurthi and Subramanyam (1976) reported the effect of starvation on organic composition of haemolymph hepatopancreas and fat muscle of the *Cryptozona semirugata*. Studies on Seasonal variation in the glycogen content of fresh water Basommatophoram snail, *Indoplanorbis exustus* were carried out by Kulkarni and Shinde (1992). Effect of photoperiod and temperature on metabolic contents of ovotestis of a snail *Cerastus moussonianus* was observed by Magare and Kulkarni (1993).

Animal exposed to temperature disturbs the physiological and biochemical process with in the organisms following the exposure to them. Exposure to different temperature affects biochemical constituents of slug (Blackmore, 1969; Giese, 1969; Kulkarni et al., 1992) and other terrestrial animals and this is the current topic of interest because of the changing ecological parameters day to day.

Exposure to temperature causes stress on the non target organism, which induces the changes in them. Any change to overcome the stress needs energy. Normally various sources of energy metabolism are required by the organism to encounter the stress (Horiguchi, 1956; Marques et al., 1976). In this process the metabolic cycles involved in the interchange of organic constituents that are responsible for production of energy, undergo a drastic change. It is this change that determines whether the organism develops the
necessary potential to counteract the stress or otherwise. Some general physiological aspects like neuroendocrine control of carbohydrates and protein metabolism and aerobic, anaerobic and glyconeogenic metabolism are left virtually untouched as these aspects have been studied only in *Lymnea stagnalis* (Geraerts and Mohamed, 1976).

In molluscs, hepatopancreas plays a very important role in the physiological process. It is an important digestive gland and storage depot. It is comparable to the vertebrate liver. This hepatopancreas is the hot bed for various biochemical reactions like the vertebrate liver. Metabolite reactions are also found in foot. Hence these organs were taken for biochemical study. As carbohydrates are the source of ready energy for organism to overcome stress in cold glyconeogenesis decreases hence glycogen content increases as compared to warm acclimation. The stored carbohydrate that is glycogen in the tissue like hepatopancreas, muscles can give the clue of the utility. Hence, tissue glycogen was estimated.

Proteins are the most characteristics organic compound found in the living cell. Protoplasm of cell is composed of proteins. So proteins have a major role in the process of interaction of cellular mechanism. Proteins have a dual importance for the organism as building material and as a source of energy. The protein of the cell may be considered as an important tool for evaluation of physiological stress, there is greater
energy demand for performing enhanced metabolic activities. This energy may be obtained from protein. As enzymes, proteins participate in intricately balanced sub cellular function (Sekeres et al., 1970). Protein synthesis increases in cold than warm. Kulkarni et al. (1983); Shinde et al. (1993) have done quantitative studies on protein and glycogen in slug and snail. Salquddin et al. (1970); Magare and Kulkarni (1994) studied the effect of starvation, aestivation and pH on biochemical contents of the land snail Cerastus moussonianus. The levels of the tissue protein are determined by their rate of synthesis and degradation both processes are well regulated (Segal, 1976). The tissue protein of the body under temperature stress plays a pivotal role in the activation of compensatory mechanism. Hence it becomes essential to study about protein.

Lipid is also a major source of energy after carbohydrate in animals, as it yields highest amount of energy (9.3cal/gm), which is more than double the energy obtained from carbohydrates and proteins. Considerable work has been carried out on the lipid contents of different organisms indicating that there is influence of various factors such as age, temperature, toxicity etc. on lipid content of the organisms. Lipid and lipid metabolism have important role in the metabolism under stress condition. The storage lipids are primarily located in the hepatopancreas and muscles, lipidic changes that are likely to occur
after exposing the animal to high and low temperature. Lipid synthesis in cold increases. Total lipids lowered since more energy is needed to overcome any stress condition, the observed decrease in the lipid is reported by various workers. It might be due to the decrease in glycogen content of the same tissue, which is an immediate source of energy during stress condition, indicative of induced gluconeogenesis in muscle. The decrease in the lipid content of tissues indicates the pronounced lipolysis and its utilization during exposure to temperature.

Plants and poikilotherms maintained at lower temperatures have lower melting point of fats than the members of the same species living at higher temperature. Meenakshi (1956); Lawrence and Giese (1969); Vassallo (1973); Ansell (1974); Kulkarni (1981) studied changes in lipid content. Decrease in lipid content with the rise in temperature observed in goldfish. Lipids in the economy of marine invertebrates have been studied by Giese, 1966b; Barnes et al., 1973. Lipids in an intertidal polychaeta and their relation to the maturation of the worm (Pocock et al., 1971). Effect of temperature on lipid content of slug has been scarcely studied hence it is desirable to observe the changes in the levels of total lipids.

Comparatively scant, attention has been given to nitrogen excretion of terrestrial Pulmonates (Bayne and Friedl, 1968). Earlier studies on marine mussels Modidlus demissus, Crossostrea virginica,
Mya arenaria, Mytilus edulis and fresh water mussel, Parreysia corrugata have shown also to excrete some urea. The types of nitrogenous waste product excreted by an animal are dependent on the availability of water (Prosser and Brown, 1961; Campbell and Bishop, 1969; Campbell and Davis, 1975) and a correlation is reported to exist between the degree of terrestriality of an animal and its dependence of ureotelism (Hochachka and Somero, 1973). Of the three terminal product of nitrogen metabolism namely ammonia, urea and uric acid, the lamillibranchs, mostly excrete ammonia and two some extent urea. The result of the experiments of Lum and Hammen (1964) on the mussel, Modiolus demisus and the oyster, Crassostrea virginica lead to the conclusion that these two species are ammonotelic. A part of the nitrogen is also given out as urea in these species. Florkin (1966) made a review of the work on excretion in Mollusca. Allen and Garrett (1971) worked out excretion of ammonia and urea in Mya arenaria. The amphibious snail Pila globosa reported uricotelism during aestivation and accumulates uric acid in its nephridium (Lal and Saxena, 1952; Shylaja et al., 1974; Mudkhede, 2002). The weight specific ammonia excretion rates reported by Sprung and Borchending, (1991), David et al., (1995). Accordingly it was thought that a study of a physiology of excretion in the slug Semperula maculata would yield interesting data. No more work has been done on the role of
temperature on the accumulation of uric acid in the slug. Hence, the present study deals with the uric acid accumulation in different tissues of *Semperula maculata* at different acclimated temperature.

Considering the impact of temperature on metabolism on terrestrial animals, the present work was undertaken.

### 1.5 Enzymalogical profiles

A large system of enzyme is functional at various metabolic pathway and any changes it gets reflected as functional disorder, hence enzyme assays have been proposed as valid biochemical means to evaluate the effect of acclimated temperature. Temperature can produce metabolic changes at cellular level by influencing enzyme system. Studies about the influence of temperature on enzyme activities are necessary because enzymes play a key role in all vital activities of living organisms.

The presence of glycogen and alkaline phosphatase in the germinal epithelium was reported by Pelluet and Watts (1951) in the slugs of the genera *Arion refus* and *Deroceros* and by Laviolette (1954) in *Arion refus*. In *Philomycus caralinianus* (Kugler, 1985) was found that glycogen and alkaline phosphatase are associated together throughout epithelium of the reproductive tract. In cell differentiation (Raven, 1972) the cell constitutes also differ in their contents depending upon their functional aspects. For instance, alkaline
phosphatase, a part from its occurrence in ciliary cells, is especially found at places where secretory or excretory processes are taking place, the shell gland and mantle fold. The statocyst during the secretion of the statolith, the wall of the nephric channel in the larval kidney, the wall of the foregut in the region of the chitinous jaws, the odontoblasts and supraradular epithelium which takes part in the hardening of the teeth by their impregnation with either calcium or iron salts with proteins alkaline phosphatase and calcium.

The alkaline phosphatase is non-specific phosphomonoesterase having pH specificity which hydrolases various phosphate esters and liberate phosphate. It is known to involve in variety of metabolic activities such as permeability (Seth et al., 1969). Protein synthesis and general maturation (Shaffi et al., 1974) steroidogenesis (Guraya and Sidhu, 1975) certain transaminases were reported to be indicators of protein degradation in salmoinds (Bell, 1968). The nervous system has a profound influence on the functional activity of enzymes (Bajusz, 1965). Acid and alkaline phosphatase activity of skeleton muscle of the frog Rana cynophylactis during low acclimation studied by Sridhara 1979; Talesra et al., 1977.

Available literature shows that the knowledge about the effect of acclimated temperature on activities of certain enzymes of Semperula maculata is meager. In present chapter an attempt has been made to
Illustrate the investigation results of different acclimated temperature on some of the enzymes like acid phosphatase and alkaline phosphatase.

*Semperula maculata* is most commonly found slug in Vidarbha region and it is abundantly available in the field and gardens. Now a day scenario is gradually changing. Manson failure and seasonal uncertainty bring in famine, flood and excess warmth. Forest cover has dwindled to about 11% of the landscape. All these destruction to a great extent is manmade particularly industries have accelerated this process. The above influence on ecosystem rise in temperature, fall in temperature or availability of food affects on population of slug.

This fact provided an incentive to undertake the present investigation that is ‘Studies on some aspects in the slug *Semperula maculata* with special reference to the ecological parameters’.