

# Earthworm Resources and Vermiculture



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1993

# **EARTHWORM RESOURCES AND VERMICULTURE**

*Edited by  
The Director, Zoological Survey of India*



सत्यमेव जयते

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## **PREFACE**

The soil ecosystem supports the most vital components for human development by way of agricultural production, forests and vegetation. India with a vast area under wastelands has taken up a national policy for reclamation of such land area which invariably will involve restoration of soil condition. While soil management has been traditionally dependent on water and fertilizer, role of biotic components has not been fully evaluated and integrated in the management strategy. The earthworms constitute one of the vital groups of any faunal element in the soil and the role of earthworms in keeping the fertility of soil system has never been increasingly acknowledged. India has a large earthworm resource both in terms of faunal diversity and in terms of numbers. However, rearing of earthworm, for practical application in restoration of degraded soil system or improving of existing condition has never been attempted on a commercial basis. The technology of vermiculture as such needs to be standardised and result oriented.

The Zoological Survey of India during its last seven and a half decades has been acting as the national organisation to study the faunal diversity and also to assist the management of biological resources. With a tremendous development-impact in the post independence period this Survey has become the right partner in national efforts to restore and conserve vital ecosystems. In order to disseminate the knowledge acquired through field and laboratory researches, the Survey also organised periodic training programme on some topics of priority. During its Platinum Jubilee celebrations, Zoological Survey of India has organised 3 special training programme viz., (i) Snails, Flukes and Man, (ii) Snakes and Human Welfare and (iii) Earthworm Resources and Vermiculture.

The present publication is based on the last of the above named programme which was conducted at the High Altitude Zoology Field Station of Zoological Survey of India at Solan (Himachal Pradesh) in September, 1990. It is expected that the contents of the presentation will be useful to the users community. I would like to put on record my sincere thanks to Dr. J.M. Julka, Scientist-in-Charge of HAZFS, Solan and his colleagues for initiating the training programme on "Earthworm Resources and Vermiculture" and for the major textual contents in the present volume.

**Dr. A. K. Ghosh**  
DIRECTOR  
*Zoological Survey of India*

# Earthworm Resources and Vermiculture

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## **Earthworms and Vermiculture : An Introduction**

**Mohammad Shamim Jairajpuri**

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Earthworms are perhaps the best known of all soil-inhabiting animals which possess cylindrical body and well marked external and internal metameric segmentation. They lack any appendages and suckers but have a few hook-like chaetae for gaining hold on the substratum. Hence, the name Oligochaeta (Gk. *Oligoi*, few; *chaite*, hair), a group of the phylum Annelida to which they belong. Earthworms are hermaphrodite and sexually mature worms have a distinctive epidermal ring-shaped area called, the clitellum, which has gland cells that secrete materials to form the cocoon.

Earthworms vary greatly in size though not in shape. In India, some peregrine species like *Microscolex phosphoreus* (Duges), *Dichogaster saliens* (Beddard) and *Bimastos parvus* (Eisen) are even less than 20 mm long, while some endemic geophagous forms, such as *Drawida nilamburensis* (Bourne) and *Drawida grandis* (Bourne) may reach up to one metre in length. *Megascolides australis* (McCoy) from Australia is reported to attain a length of over 4 metres. The world's largest known worm *Microchaetus microchaetus* (Rapp) which is found in South Africa has a length of about 7 metres.

Ordinarily earthworms have to be dug out of the soil, but during rains they may be found crawling actively on the soil surface. Generally, they occur in top 30-40 cm layers of soil which is moist and has plenty of organic matter. Some species burrow deeper in search of moisture to avoid desiccation during periods of prolonged drought in summer and spring months. The species, *Drawida grandis* was collected from a depth of 2.5 to 3 metres in the Nilgiri hills during summer.

### **General Activity**

Earthworms occur in diverse habitats. Organic materials like manure, compost, litter, humus, effluents and kitchen drainage are highly attractive for some species. They are also found in very hydrophilic environs close to both the fresh and brackish waters. Some species can survive under snow and a few are arboreal inhabiting accumulated detritus in the axils of banana, palm and bamboo trees. Earthworms are omnivorous but they mostly derive nutrition from dead organic matter, which generally does not occur abundantly in the soil. As a result, they are adapted to swallow large quantities of soil for extracting sufficient nourishment from it. The soil-inhabiting protozoans, nematodes, rotifers, bacteria, fungi, etc. have been recorded from the contents of their gut. Earthworms are capable of withstanding considerable starvation with water loss of up to 70% of their body weight. *Agastrodrilus* Omodeo and Vailaud, a carnivorous genus of earthworms from the Ivory Coast of Africa has been reported to feed upon other earthworms of the family Eudrilidae (Lavelle, 1983). The quantity of food taken by a worm varies from 100 to 300 mg/g body weight/day according to Edwards and Lofty (1977).

The main activity of earthworms, however, involves the ingestion of soil, mixing of different soil components and production of surface or subsurface castings. The earthworms consume the soil organic matter and convert it into humus within a short period of time and thereby increase

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the soil fertility. Within 24 hours they can pass soil almost equivalent to their own weight through the alimentary canal. They have therefore rightly been called the nature's ploughman. Thus, the soil is being constantly and continuously turned over and over again by these worms and the amount brought to the surface is quite considerable. Annual worm cast production has been estimated to be between 1.4 and 77.8 tonnes/ha at some Indian sites (Roy, 1957; Dash and Patra, 1979) as compared to 18-40 tonnes/ha in English pastures (Darwin, 1881). Larger quantities of 2100-2600 tonnes/ha have been reported in Africa (Edwards and Lofty, 1977). Different species produce structurally distinct and taxonomically significant casts : heterogeneous masses, spheroidal to oval-shaped individual pellets, small towers of coiled tubes, short threads and beaded-strings (Julka, 1988). A species of *Tonoscolex* in Burma produces casts that may reach up to 20-25 cm in height.

Reproduction and cocoon production is possible throughout the year, although maximum cocoon production by Indian species of worms in pasture soils has been recorded in late October and early November (Das and Senapati, 1982). The incubation period varies from species to species. It may be 14-30 days for some Indian species as compared to 8.5 to 30 weeks in some European species. Usually one or two young worms hatch from a cocoon, but they may be as many as six in number in *Eisenia fetida* and *Bimastos parvus*.

Earthworms possess limited means of active dispersal. Mountains, deserts and oceans are effective physical barriers for their migration. Some species are able to move actively for considerable distances during or after heavy rainfalls. In some areas of Western Himalaya, a few litter dwelling species emerge on a mass scale towards the end of monsoons and migrate for short distances in search of suitable environs to tide over unfavourable winter conditions (Julka, 1988). Passive dispersal through stream drift and in mud on the feet of animals and birds has been recorded. Over the years man has also played a significant role, though unintentionally, in transporting some species in soil around roots of plants. It is quite possible that all the lumbricid and a few other species have been brought to India in this manner.

The activity of most earthworms is interrupted during dry periods or under high temperatures. To overcome the adverse period they usually move into the deeper soil layers and may undergo 'diapause' or transform into a quiescent stage. During this period the worm stops feeding and constructs a spherical chamber lined with mucus within which it usually rolls into a tight ball or a loose knot.

### **Environmental Requirements**

With adequate supply of food and availability of moisture the earthworms can thrive very well in all kinds of soils. The type and amount of food influences their population size, diversity, growth rate and fecundity. Tolerance of soil pH varies from species to species. Usually they can live in soils with pH ranging from 4.5 to 8.7, but neutral soils have greater densities of earthworms as compared to alkaline or acidic soils. The soil temperature and moisture are other two important factors that influence their seasonality and distribution. In sub-tropical climate like that of India, they are active and abundant mainly during summer rains. Prolonged drought decreases their numbers significantly. A period of about two years is generally required for populations to recover upon the return of favourable conditions. Fluctuations in temperature influence their overall activity, metabolism, respiration, growth as well as reproduction. The ultra-violet rays are injurious and extreme temperatures may often be fatal for the earthworms.

### **Effects of Pesticides**

Large quantities of insecticides, herbicides and fungicides are usually applied to soils for controlling different kinds of pests. Some of these chemicals are general biocides that may also kill earthworms besides the target organisms. By and large they are not very susceptible to

pesticides at normal dosages, but at higher concentrations these toxic substances are absorbed into earthworm's tissues as the soil passes through their intestine while feeding. Residues of heavy metals like cadmium, lead, zinc and nickel have also been recorded in their bodies. The accumulation of toxic chemicals in earthworm tissues is very significant ecologically, because these animals are important components in the food chain of several species of birds and mammals.

### **Economic importance**

To a common man earthworms are rather insignificant animals which generally come out on the soil surface during the rains and serve as bait for angling. The role of earthworms in enhancing soil fertility was, however, known to even ancient farmers. But with the advent of modern agricultural practices, during the last 2 to 3 decades, and the use of artificial fertilizers their significance faded. In recent years the farmers are once again realizing the worth of these highly beneficial animals and are making all possible efforts to culture and subsequently release them in fields and gardens. The beneficial effects of earthworms in increasing soil fertility was documented since the time of Darwin (1881). Soil with higher densities of worms remains loose and has a greater capacity to retain air and moisture. The earthworms by making tunnels while burrowing aerate the earth which helps in increasing the air-holding capacity of soil. In the act of depositing their castings on the surface at night, they bring the sub-soil to the top and expose it to bacterial action. The bacteria help in the decomposition of cellulose which otherwise does not breakdown easily. The earthworms also take the rich humus from soil surface to plant roots and thereby help in maintaining soil pH. Worm castings contain more water stable aggregates which keep the soils well drained. Soil nitrogen is generally bound in organic complexes and as such is not readily available to plants. This bound nitrogen is converted into available forms like ammonia, nitrates and nitrites as it passes through the digestive tracts of earthworms. Compared to parent soil the worm casts contain more available nitrate nitrogen, calcium, magnesium, phosphorus and potassium. Organic matter ingested by the worms is pulverized in their alimentary canal, and excreted as a colloidal humus which is rich in plant nutrients. Also, a large number of worms die during unfavourable periods when the chemical demand in the soil is maximum due to growing plants. The microbial decomposition of dead worms releases considerable amount of locked up nitrogen and thereby making it available to the plants.

Increase in the organic wastes mainly due to growth of human population, agriculture and industry is a global problem and a serious constrain in the maintenance of a clean and healthy environment. Because of their food and feeding habits, the earthworms should be considered nature's most useful converters of these waste products. Experiments with worms have successfully been conducted for recycling the utilisable organic wastes arising out of household garbage, city refuse, sewage sludge, and paper, food and wood industries (Mitchell and Horner, 1980).

Earthworm tissues comprise high amounts of proteins which after proper processing could benefit the livestock and aquaculture by augmenting or replacing traditional feeds. There are also reports of worms being eaten by Maoris of New Zealand and the natives of New Guinea (Edwards and Lofty, 1977). In Indian Unani system of medicine, the external application of preparations made from the dried worms is used in treating wounds, piles, chronic boils, sore-throat, hernia, etc., and when taken internally for curing respiratory ailments, jaundice, rheumatic pains, etc.

On the other hand, certain habits of earthworms are considered harmful. Some species seize leaves of growing plants and pull them into their burrows, often killing the plants. Their extensive burrowing activity sometimes retards germination, growth and root development of paddy and some vegetable crops. There are some reports which suggest that earthworms contribute to soil erosion because they bring fine soil particles to the surface. The earthworms are also known to help in the spread and development of some parasites and diseases of both animals and

plants. The foot and mouth viral disease of domestic animals is transmitted by earthworms (Edwards and Lofty, 1977). They also act as intermediate hosts of certain parasitic protozoans, cestodes and nematodes. A number of species of nematodes of interesting and phylogenetically primitive type are found as parasites of earthworms.

### Vermiculture

Vermiculture, the technology of producing rich bio-fertilisers and animal protein by using earthworms, has established itself commercially in many developed countries. It has been estimated that one million worms can convert about 120 tonnes of organic wastes into bio-fertilisers in about one month's time. According to conservative estimates over 2,000 million tonnes of solid and liquid excreta of animals and human beings, and another 200 million tonnes from crop straws are available as wastes annually in the country. Besides there are vast quantities of domestic garbage and industrial wastes. Thus production of bio-fertilisers through vermiculture has a bright future in India. But it is very essential to select suitable species for vermiculture. These should be capable of living in rich organic matter, be stress resistant, efficient decomposers, and having high fecundity and growth rates.

It is not very difficult to raise and maintain earthworms. They can be reared in small containers filled with compost, cow-dung and kitchen refuse. The rainy season seems to be best for culturing them. Sufficient soil moisture and adequate organic residues are considered ideal for their growth and multiplication. Within a period of about one year, if the culture is properly maintained, the multiplication may be more than 50 times. The worms may be taken from culture as and when needed, and can be introduced in the desired fields, gardens, etc. If used in orchards, the worms may be released in the pits in which the trees or plants are growing. The earthworms provide excellent conditions for the build up of a number of useful micro-organisms and consequently the soil with its teeming millions of organisms becomes highly suitable for plant growth.

Earthworms along with some soil micro-arthropods are important for the formation of humus which is the end product of organic matter in top soils. This organic matter is cycled and recycled, so that the humus formation is natural and a continuous process. The worms, no doubt, accelerate this process, when multiplied and released through cultural practice. The enrichment of soil, thus helps in agriculture and forestry. By planning and managing agricultural operations, supplemented with inputs from the earthworms, the farmers can get significant increase in crop yields. Countries like U.S.A., U.K. and Japan have realized this potential of earthworms and are therefore taking vermiculture quite seriously as an aid to farming. Young and healthy live worms of those species which are more suitable for a particular type of crop and soil of a given area are raised. These are mixed in soil free medium with a shelf-life of about 4 weeks. The worms in the medium are spread around roots of plants of agri- and horticulture, or may be used as a source of food for poultry or fisheries.

As part of bio-technology, vermiculture has attracted attention, since it is an entirely natural process which maintains the environmental balance and leaves no adverse effects. The earthworms feed on decaying organic matter in the soil and after its assimilation in the alimentary canal, excrete the soil as 'cast' which is rich in the nutrients. Vermicast contains various amino acids, minerals and micro-organisms which humify the organic matter in the surrounding soil and act as a bio-fertiliser for plants.

According to Bhole (*pers. comm.*) the soil with worm casts, in comparison to soil without these, has 5 times more nitrogen, 7 times more phosphorus, 11 times more potassium, 2 times more magnesium and calcium each. All these along with other trace elements and soil nutrients, soluble in water, are released during vermiculture, and are readily available to the root systems. The same applies to the *Actinomyces* bacteria also which are 7-8 times more in the cast soils than in the surrounding soils. Different kinds of organic wastes could be utilised for enriching soils through vermiculture. The earthworm feeding helps in the quality of composting. This

vermi-compost is mixed with soil and is recommended by the vermiculturists in the ratio of 30 : 70 v/v. To avoid leaching of chemical fertilisers, it can also be used in 40 : 60 proportion.

Considering the potential of vermiculture in enriching soils and in turn in the increase of crop yields, the practice needs more and more support. The fact that it does not harm the ecology in anyway, is an additional advantage. There is much scope to expand vermiculture, its trade and export, in the third world countries, which is still sustained on traditional organic farming.

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## Collection, Preservation and Study of Earthworms

**J. M. Julka and R. Paliwal**

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Earthworms are found in all types of soils provided there is sufficient moisture and food. They occur in forests, grasslands, arable lands, gardens, orchards, plant nurseries and green houses. They have been living in caves and axils of tree leaves. Organic materials like compost, manure, forest litter and humus, municipal dumps, soils wetted with effluents and kitchen drainage are highly attractive to some species. Some earthworms are very hydrophilous and a few species can live under snow on high mountains. They are soft-bodied and require special methods of collection, narcotisation, fixation and preservation for their morphological, taxonomic and ecological studies.

### Morphology and Taxonomy

#### *Collection and Preservation*

The best method for collecting earthworms is by digging soil with a shovel or spade or any other suitable implement. For a comprehensive survey of earthworms of an area, they should be collected from different ecological niches, viz., litter, kitchen drainage, manure heaps, different types of soils, margins of freshwater bodies, pastures, grasslands, forests, cultivated fields, etc.

For morphological studies it is essential to narcotise live worms before fixation. Several narcotising solutions are effective, and 5-10% ethyl alcohol or 1% propylene phenoxetol are amongst the most convenient. Live worms brought from the field are placed in a suitable flat-bottomed container with little freshwater. Anaesthetic solution is gradually added to the container till the worms become motionless. When the worms no longer respond to probing they should be transferred to a flat dish containing the fixative solution for at least 24 hours. The most suitable fixative for normal morphological and taxonomic studies is 5-10% formalin depending upon the size of the worm. A 10% solution of Dowicil 100 (proprietary name for 1-(3-chlorally) 5,7-triazol-1-azoniaadamantane chloride) is also an excellent fixative for earthworms. Specimens fixed in dowicil solution retain their shape and remain quite flexible, showing none of the brittleness often associated with formalin preserved material. Both formalin and dowicil solution are slightly acidic and can be neutralized with limestone or marble chips without affecting the preservative action. Nephridia are best studied by dissecting a freshly narcotised worm in normal saline (0.75% solution of Sodium chloride) and fixed *in situ* by covering the whole worm with Bouin's fluid or with acetic bichromate.

Histological studies of earthworms require special attention. The main difficulty encountered in sectioning is the presence of soil in their gut. To flush out the soil, worms are fed on wet blotting paper for several days until their faeces contain no trace of soil or blotting paper. Vail (1972) fed worms on sphagnum moss for about 7 days to ensure the voiding of solid particles from the gut. He found the use of sphagnum moss for the maintenance of worms in good condition for a longer period than did wet blotting paper/filter paper, wet paper towels, wet leaves or just water in a dish. Worms with voided guts should be narcotised and fixed in Bouin's fluid (75 cc saturated aqueous solution of picric acid, 25 cc formalin and 5 cc glacial acetic acid) or AFA : alcohol-formalin-acetic acid (10 cc formalin, 10 cc glacial acetic acid, 30 cc of 95% ethyl alcohol, 50 cc distilled water) for microtome sections of an entire worm or its various organs.

Worms are killed by dropping them in 70% ethyl alcohol for taxonomic studies. When the movement stops, they are removed from alcohol and placed on a piece of blotting paper or any other absorbent paper in a straight position. They are then transferred to a flat-bottomed container with 10-15% formalin for fixation for a period of at least 24 hours. It is essential that worms are straight because curled and twisted specimens are difficult to handle during dissection. The specimens are stored in suitable sized vials or bottles filled with 70% ethyl alcohol or 5-10% formalin. A label with locality and altitude data, name of collector and date of collection is to be added to each vial. For best results, the preservative should be changed within a week, especially for large worms. Sometimes for lack of adequate time in the field it is not possible to follow this programme, it is then recommended to preserve the specimens directly in 4-10% formalin depending on the size of the worm. Fixation of specimens in alcohol is not desirable as they become soft and macerated, and are unsuitable for dissection. Some workers anaesthetize and relax the worms before fixation by placing them in a container filled with water and gradually adding alcohol to it. The main disadvantage of this method is that length of the relaxed specimens may be twice, thrice or even more than the contracted specimens as obtained by dropping them directly into alcohol or formalin. For taxonomic description of a species, the latter condition is preferable because uniform contraction is often more easily obtained than uniform relaxation of a worm.

Various methods of estimating earthworm populations and habitat preference are being used. Their effectiveness varies with the species and habitat. No one method is equally suitable for all species and habitats.

### **Digging, Hand Sorting and Wet Sieving**

Though laborious and time-consuming, this method has been widely used for sampling earthworms with best results (Edwards and Lofty, 1977; Reynolds, 1977). Sometimes specimens are liable to be damaged during digging. With the help of suitable digging tool, cores or quadrats of soil of exact dimensions are taken for accurate population estimates. Usually 16 sample units of 25 cm<sup>2</sup> with 20 cm depth provide adequate estimation of medium-sized worms. For bigger species and deep burrowers, large areas of deeper samples are required. The dug out cores or quadrats are gently broken and the worms are handsorted and preserved in 5-10% formalin. For better results the broken soil is washed with a jet of water through a series of sieves for collecting smaller worms and cocoons. The sieved samples are stirred with magnesium sulphate (specific gravity 1.2) solution and a stream of air is blown simultaneously into the solution. After sometime, the liberated worms and organic debris float on the surface. In this way earthworm cocoons can be easily collected. Thus, all stages of the population can be sampled by this method.

### **Chemical extraction**

Various types of chemical extractants have been used for studying earthworm population dynamics. The standardized technique employed for quantitative extraction is based on 0.25 m<sup>2</sup> of soil surface. A solution of 0.55% formalin (25 ml of formalin in 4.5 litres water) is sprinkled over each quadrat (Fig. 1) taking care to avoid its run off. The earthworms that surface in 10 minutes following the application of the expellant are collected and preserved in 5-10% formalin for studies in the laboratory. For wet biomass studies, the worms should be washed in freshwater and soak-dried over a blotting paper before weighing. Other chemical extractants like mercuric chloride solution (1.7-2.3) litres of solution containing 15 cc mercuric chloride in 18.25 litres water), potassium permanganate solution (1.5 g/litre at the rate of 6.8 litre per m<sup>2</sup>) and Mowrah meal have also been used. The advantage with a chemical extraction method is that the sampling time and labour are reduced, a well-defined sampling area may be chosen, and there is minimum disturbance of habitat. But the disadvantages are that only active surface dwelling species are collected because aestivating or hibernating individuals or some species do not respond to these

extractants. It is also difficult to collect and quantify cocoons by this method. A suitable quadrat for chemical extraction is shown in Fig. 1.

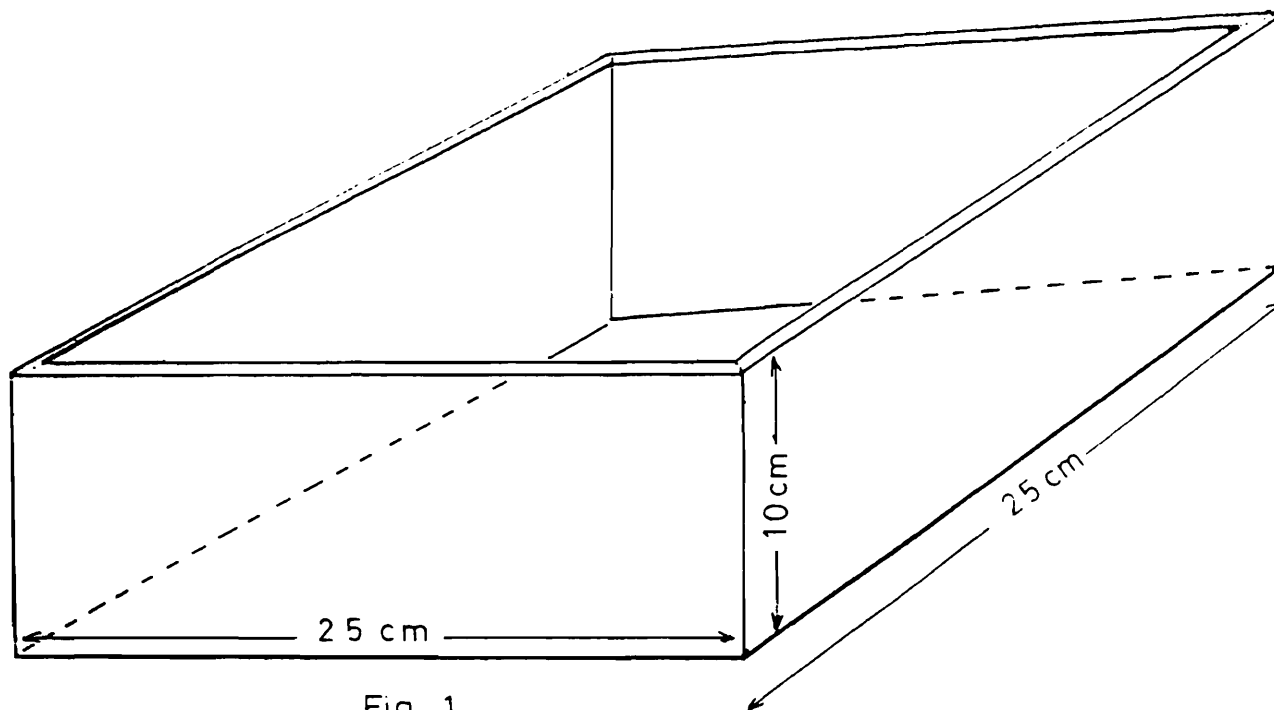


Fig - 1

Fig. 1. Quadrat for chemical extraction.

### Electrical extraction

A current of electricity passed through the soil also acts as an expellant (Walton, 1933). Deoksen (1950) used an electric current of 220-240 volt at 3-5A through 75 cm long electrode for expelling worms from the soil. The only advantage with this method is minimal disturbance to the habitat. The disadvantages are in defining the exact volume of soil treated and variability of physico-chemical properties of the soil. For example the current penetrates deep into moist soil which brings deep dwelling species to the surface. There is some danger of too much current killing the worms near the electrodes. The response of different species to electricity may also vary.

### Heat extraction

This method operates on the principle of Baermann funnel and may be useful in obtaining small surface dwelling species that are difficult to hand-sort. Reynolds (1977) employed Tullgren funnel and incandescent lights for extracting worms from soil samples brought from the field. This method is again time consuming and has limited use in earthworm sampling.

### Vibration method

The mechanical extraction by vibration methods are currently limited to south-eastern United States. Mechanical stimulation by vibrations seems to have very little effect on Lumbricidae but is extremely successful for some Acanthodrilidae and Megascolecidae species. This often takes the form of a vibrating flexible rod with a bow. The advantages of the mechanical extraction are minimal habitat destruction and the reduced sampling time required for each sample. The disadvantages are the difficulty of defining the exact volume of soil treated, the effects of the

variability of the physical and chemical properties of soil and the variable response of the different species.

Several workers have compared the relative efficiency of extracting earthworms by two or more of these methods. The hand sorting or washing give the best result for most species but are very time consuming. The formalin method seems to be the best compromise for species with burrows. Formalin extraction followed by handsorting to find animals that had not been extracted (Bouche, 1969) seems to be most suitable method for ecological studies.

### Method of study

Earthworms cannot be identified without resorting to dissection since their generic or even suprageneric identification is dependent on internal characteristics. Before dissecting a worm, its various external characters like shape of prostomium, location of genital and nephridial apertures, and form and extent of clitellum should be recorded. It is then pinned in a dissecting dish, containing water, by fine entomological pins at the anterior and posterior ends, taking care to avoid injury to the prostomium. Using a fine scissor or scalpel or even a sharp shaving blade, the body is cut open longitudinally slightly to the left or right side of the mid-dorsal line in order to avoid damage to dorsal pores. By carefully cutting septa, the flaps of the body wall are slowly pinned out with fine forceps, preferably first at the post-prostatic region and then continuing forward, care being taken to record exact location of missing and delicate septa in the gizzard region. To determine the presence of calciferous lamellae and openings of calciferous glands, it is necessary to slit open the oesophagus along the mid-dorsal line. The beginning of intestine and form of typhlosole can be determined by giving a slit just below the mid-dorsal line on one side of the intestine. Penial and copulatory setae are easily removed along with their enlarged follicles from inside, they cannot be pulled from outside without some damage to them. After cleaning the adhering tissue, the setae are mounted on a slide in glycerine or any other media provided the refractive index is sufficiently different from that of the setae. Canada balsam is not satisfactory for this reason, unless the setae are stained. For the study of epidermal setae, a small portion of the body-wall is cut off with a pair of scissors and treated with 40% solution of hot potassium hydroxide (KOH) for 15–30 minutes. The skin is washed in water and mounted directly in glycerine after proper dehydration and clearing.

To study digestive system, gizzard should be cut into two by a longitudinal incision for observing thickness of its wall and its cuticular lining. In the same way a portion of intestine and rectum is also opened from the ventral side to study the morphology and limits of the typhlosole, which lies along the mid-dorsal line of the intestine. Excretory system should be examined after fixation of the nephridia *in situ* with Bouin's fluid; a septum with attached nephridia is dissected out with needles under a binocular dissecting microscope in order to separate individual nephridia, care being taken to keep funnel intact on each nephridium. These are stained and mounted in balsam. Pharyngeal nephridia are studied by tracing their ducts in the pharynx region. Integumentary nephridia are picked up with the forceps and are studied by mounting on a glass slide in glycerine. In a similar manner holonephridia and megameronephridia are taken out from the parietes with the help of forceps for study under microscope. Care should be taken to keep funnel intact. Septal excretory canals and the supra-intestinal excretory ducts are best studied in well fixed and preserved specimens. Preparations of complete septa show the septal excretory canals, while the supra-intestinal excretory ducts can be dissected out with needles from the roof of the intestine. The opening of these ducts into intestine are seen only in sections (Bahl, 1950).

For the purpose of sectioning, worms with voided gut and fixed in Bouin's or AFA are used. The dehydrated specimens are cleared with a toluene-terpineol mixture (3 : 1) and then used alone and embedded in a hard paraffin (melting point 60-62° C). The use of xylol and a softer paraffin give unsatisfactory results, and many of the sectioning problems (e.g. excessive static elasticity, failure of ribbon formation, shredding of individual sections) are eliminated with the use of

toluene, terpeneol, and hard paraffin. Embedded tissue is sectioned at 10  $\mu$ , and the sections are stained with Harri's hematoxylin and alcoholic eosin.

In order to obtain cocoon, it is best to select a piece of ground showing castings in April-June or in September-October. Put heap of earth in a sieve and stir the earth while keeping the sieve in a bucket of water. The earth passes through the sieve while cocoons remain in the sieve along with some pebbles and stones. Cocoons are easily picked up with a camel-hair brush. Cocoons are opened in normal saline by using needles under dissecting microscope. Embryos of all ages can be obtained and studied by making whole mounts or by sectioning.

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## **Rearing and Culturing Earthworms**

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Earthworms feed upon a variety of organic material and could be raised commercially for recycling biodegradable organic wastes, production of biofertilisers and animal protein for poultry and fish feed. Vermiculture is feasible in suitable containers or specially designed boxes, since they are omnivorous, able to withstand environmental changes and resistant to many diseases. The technology involved is very simple and can easily be adopted in India, especially in the rural areas. It is possible to culture worms both indoors and outdoors depending upon the local climatic conditions.

The culture boxes or containers should be non-porous to minimise loss of moisture from culture medium. The boxes should be made up of light weight materials like plastic, wood, tin etc., which could easily be carried from one place to another. The size of the containers may vary according to the need. Reynolds (1977) considers a specially designed wooden box to be more convenient and useful (Fig. 1). It measures 50 cm in length, 35 cm in width and 15-20 cm in depth. The bottom of the box is provided with a few holes of 50 mm diameter. Plastic window screen is placed on the inside bottom with a burlop (or jute cloth) lining on top of the screened sides before the culture medium is added. This prevents the culture medium from sticking to the box and escape of worms through the holes but allows the excess of water to drain. Top of the box is covered with a burlop (or jute cloth) frame. Earthworms can be cultured in commonly available glazed earthen pots, plastic tubs or even discarded wooden cases, etc., each being covered with a lid made up of plastic or iron window screen. Plastic tubs are considered to be advantageous because these are more durable, lighter in weight and could easily be arranged one above the other in vertical rows on concrete shelves in limited space (Fig. 2).

Various combinations of soil and organic matter have been tried for raising worms. A mixture of 1/3 soil and 2/3 organic matter is considered to be more useful in culture containers by Reynolds (1977). Beds in plastic or discarded wooden cases are prepared by spreading a sand layer of 2-4 cm in height over which another layer of equal thickness of soil is added. Organic matter is placed on one side of the container. Water is added to the culture medium so as to hold 25-30 per cent of moisture. Indoor cultures are preferably kept in a cool building at a temperature between 10°C and 15°C for the lumbricids (e.g. *Eisenia fetida*) and about 20°C for tropical species (e.g. *Eudrilus eugeniae* and *Perionyx excavatus*). Sources of common organic materials are : decayed leaves, hay, straw, rice or wheat bran, vegetable wastes, cow dung, poultry droppings, biogas sludge, etc. Kale (1986) carried out trials of various mixtures of organic matters to study the dietary influence on the biomass and size of population in *Eudrilus eugeniae*. Young worms fed upon a feed combination of dung and gram bran gained maximum population and dry weight biomass after 3 months of their introduction into the culture medium.

The following precautions should be taken for vermiculture :

1. The culture medium must have sufficient organic material to avoid its formation into a soggy mass.
2. Moisture of the medium should be maintained at required levels by sprinkling water regularly. Overwatering affects the culture adversely.

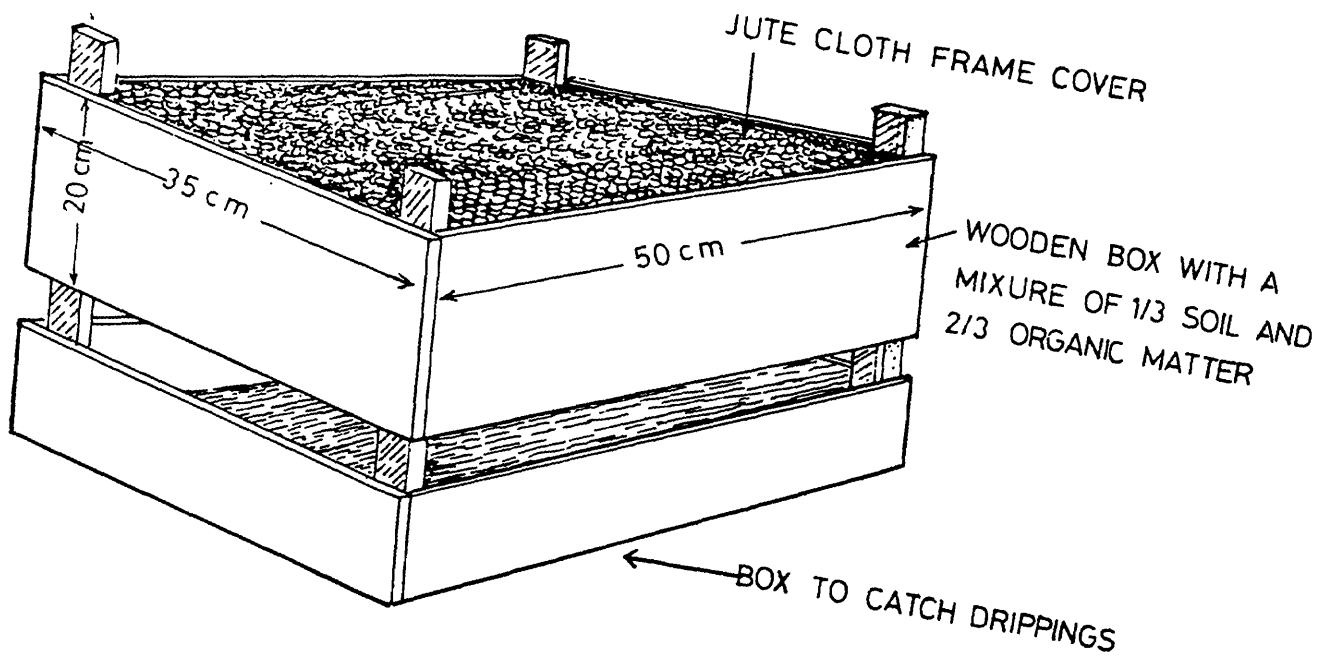


FIG-1

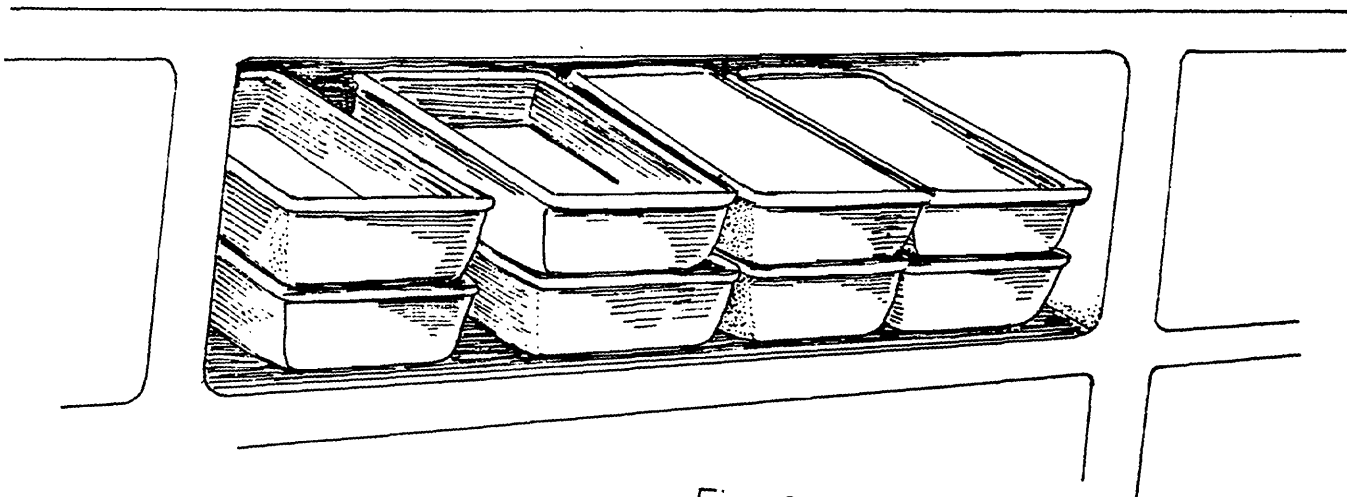


Fig -2

- Fig. 1. A design of vermiculture wooden box (after Reynolds, personal communication).  
 Fig. 2. A diagrammatic representation of indoor vermiculture in vertical rows of plastic tubs.

3. Presence of a low watt light will prevent the worms from crawling out of boxes.
4. Outdoor cultures at places with low temperature in winters should be covered with suitable insulation materials like wheat straw, dry hay or weeds, manure, compost, etc.

Large outdoor vermiculture beds of convenient dimensions may also be established on waste lands. An outdoor culture bed is generally prepared with a bottom layer of 10 cm high gravel over which plastic window screen is placed with its edges raised up to 20 cm in height. A layer of 2-4 cm sand is laid over the window screen layer. A mixture of 1/3 soil and 2/3 organic matter is spread over the sand layer. The bed is slightly raised in the middle which allows drainage of excess of water on sides during the rains. The bottom layers of gravel and sand also help in maintaining the water content in the culture. The window screen prevents the escape of worms.

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## General Morphology and Characters of Taxonomic Importance

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Earthworms are defined as terrestrial annelids with external and internal metameric segmentation throughout the body, without any appendages and suckers but possessing few setae on all segments except the first and last ones. They are hermaphrodite with few gonads in definite segmental locations. They possess a true coelom and closed vascular system. In sexually mature worms, a precisely located epidermal thickening, the clitellum, secretes a cocoon in which ova and spermatozoa are deposited and which are fertilized and develop without a free larval stage.

### External Structure

Earthworms are elongate and vermiform in shape. They are usually circular in cross-section but some forms may be squarish or trapezoidal. An arboreal species of *Perionyx* has flattened ventral surface. The length and thickness of worms are of limited taxonomic importance, since these characters vary considerably within a species. Amputation, regeneration and methods of preservation also affect their body dimensions. A few species of *Bimastos* (Family Lumbricidae) and *Dichogaster* (Family Octochaetidae) are less than 20 mm in length, whereas some deep burrowing representatives of *Drawida* (Family Moniligastridae) exceed 1000 mm. Different types of colours of worms like rich brown, light to dark red, grey, purple, etc. are due to deposition of pigments in the circular muscles of their body walls. The colour should be recorded when a worm is alive since strong fixing fluids generally destroy the pigment. Litter dwellers are deeply pigmented as compared to inhabitants of top soil and deep burrowers.

The entire body is divided externally into a series of distinct segments by furrows. External segmentation of the body corresponds to an internal segmentation. In some forms, segments may be superficially subdivided into two or three or more annuli by secondary and tertiary grooves. The number of segments vary intraspecifically and this character can be of taxonomic value only when its limits of variations have been determined in a large number of individuals of each species. For a taxonomic description of a species, segments are numbered by convention in roman numerals i.e. i, ii, iii .... (capitalized by some authors) beginning with the peristomium. Intersegmental furrows are designated by the number of segments on either side of a furrow as 1/2, 2/3, 3/4, etc. The first segment with a crescentic opening, the mouth, is the peristomium. It is provided with a small fleshy lobe the prostomium which is located above the mouth. The different shapes of the prostomium (Figs. 1-8) are sometimes of taxonomic importance. In mature worms, a conspicuous cylindrical band of glandular tissue known as the clitellum is present at some distance from the anterior end. Its shape may be either annular (extending all round the body) or saddle-shaped (restricted to dorsal and lateral sides of the body). The location of clitellum varies between families/genera/species. *Drawida* spp. (Family Moniligastridae) have the clitellum extending over segments x-xiv and include male genital pores (Fig. 9). In Megascolecidae, Acanthodrilidae and Octochaetidae the clitellum begins at or in front of xiv, and posteriorly it may include male pores (Figs. 10-13). Lumbricidae have the clitellum behind male pores beginning on segments xxii-xxviii, and extending over four to ten segments posteriorly (Fig. 14).

Characteristics of all earthworms are the short hook-like retractile chaetae or setae embedded in the skin with which they hold gain on the substratum during burrowing and locomotion. The

positions of setae provide significant reference points for describing location of taxonomic characteristics like genital and nephridial pores, grooves, genital markings, etc. Often setae in the region of genital tumescences, male thecal pores are modified in size and shape. Those associated with genital tumescences are known as genital setae, those with male/prostatic pores as penial setae and those with spermathecal pores as copulatory setae. The arrangements of setae according to their number are expressed as lumbricine (8 setae per segment in 4 pairs, e.g. *Drawida*, *Octochaetona*, *Eutyphoeus*, etc.) or perichaetine (more than 8 setae per segment, e.g. *Amyntas*, *Metaphire*, *Perionyx*, *Lampito*, etc.). Rarely, setal arrangement may be lumbricine in anterior and middle regions, and perichaetine in posterior region of the body as in a few species of *Wahoscolex* from Coorg area of Karnataka (Julka, 1988). In taxonomic descriptions, individual setae are designated by italicized letters, i.e. in the lumbricine arrangement by *a,b,c,d* beginning with the most ventral one and in the perichaetine arrangement by *a,b,c,d,e,.....* beginning with the most ventral setae and *z,y,x,.....* beginning with the most dorsal one irrespective of the actual number in the ring (Julka, 1988).

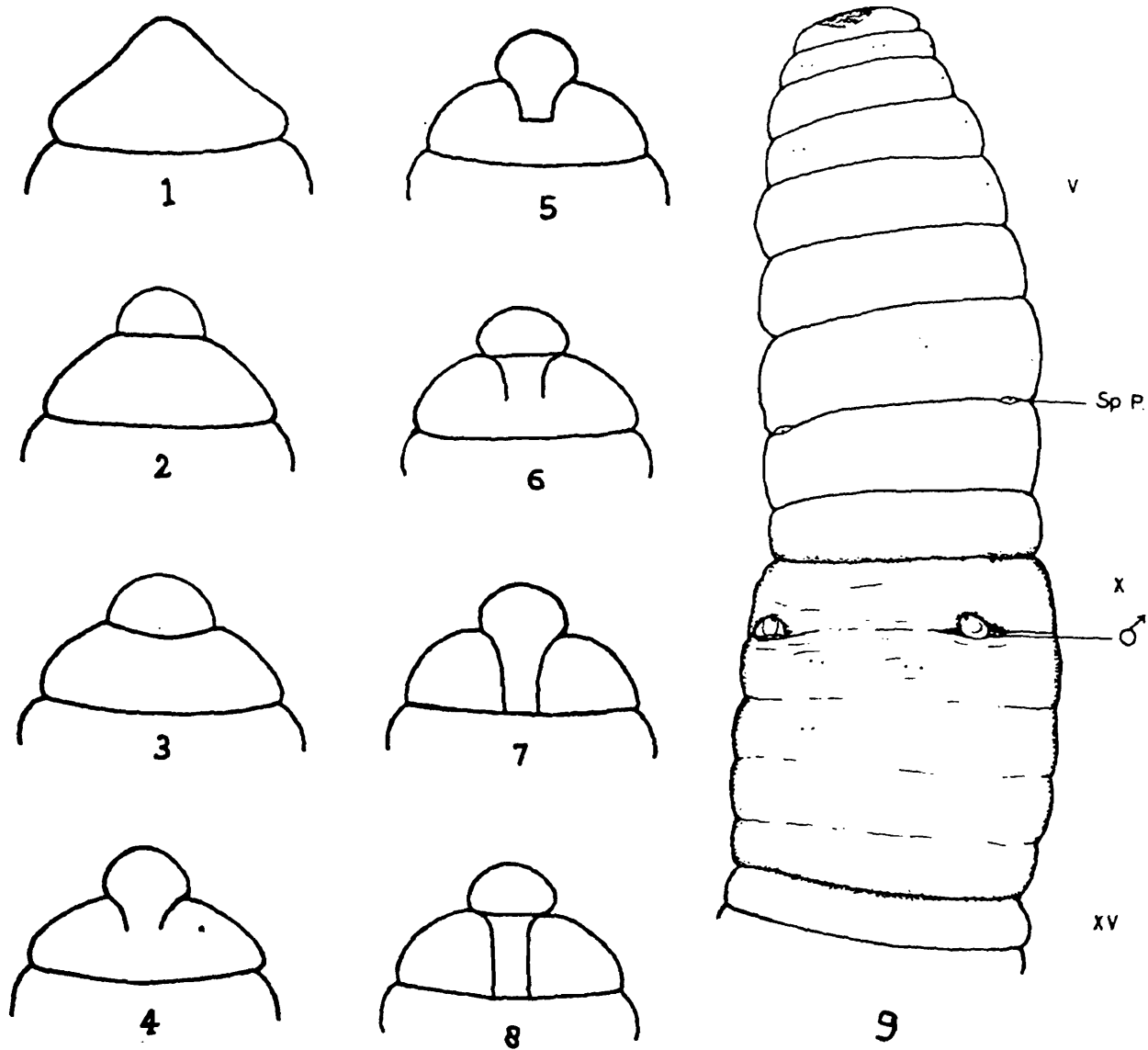
A series of tiny openings, the dorsal pores, are located along the mid-dorsal line in the intersegmental furrows. These pores lead directly into the body cavity. The location of first dorsal pore varies intraspecifically. Dorsal pores are usually absent in worms with aquatic or subaquatic habitats (*Drawida* spp. and most of the ocnerodrilids). Different types of genital pores are located on the ventral surface of earthworms. The position and size of these have long been employed as taxonomic characters. In the Ocnerodrilidae, Acanthodrilidae, Octochaetidae and Megascolecidae, the male pores are associated with the prostatic pores (openings of the ducts of prostates, accessory reproductive glands). The prostatic and male ducts may open to the exterior either separately or as combined pores. The basic conditions of these openings are : acanthodriline (male pores on xviii, prostatic pores on xvii and xix, all pores in seminal grooves), microscolecine (prostatic pores alongside or combined with male pores on xvii), balantine (prostatic pores alongside or combined with male pores on xix) and megascolecive (prostatic pores alongside or combined with male pores on xviii). Male pores in some forms are located on papillae of various shapes or at tips of intromittant organs. In the lumbricid worms the male pores are often located on segment xv, and in the Moniligastridae these pores are one or two pairs in intersegmental furrows 10/11, 11/12 or 12/13. The female pores are most commonly a single pair, either in an intersegmental furrow or on a segment. They are tiny in size and their position is often diagnostic of a particular family. Thus, they are on segment xiv or its homoeotic equivalent in the Lumbricidae, Octochaetidae, Ocnerodrilidae, Acanthodrilidae and Megascolecidae, and in the Moniligastridae they are either in the groove 11/12 or on segments xiii or xiv. Sometimes the female pores are united into a single median pore.

The location and number of spermathecal pores vary between families and species. They may be paired or sometimes combined to form single median series of pores. In some species (*Bimastos parvus*), they may be absent. A few species like *Eisenia fetida* and *Ocnerodrilus occidentalis* may have athecal morphs. In *Polypheretima elongata*, spermathecae are more than one pair in each segment. The openings of the integumentary meronephridia (nephridiopores) are microscopic apertures and cannot be easily recognised. But nephridiopores in some holonephric species are quite obvious and their axial position provides important distinguishing characters.

Certain epidermal areas on the ventral surface of sexually mature worms are sometimes modified in the form of markings, tumescences, ridges, pits, tubercula pubertatis, etc. (Bahl, 1950; Edwards and Lofty, 1977; Julka, 1988).

### Internal Structure

The body wall consists of an outer thin non-cellular membrane of cuticle, epidermis, circular and longitudinal muscle layers, and coelomic epithelium, which separates body wall from the coelom. The coelom or body cavity is filled with a fluid and is divided at each segment by a



Figs. 1-8. Various forms of prostomium in earthworms, 1. zygalobitic, 2. prolobitic, 3. proepilobitic, 4. open epilobitic, 5. closed epilobitic, 6. combined proepilobitic, 7. tanylobitic, 8. combined protanylobitic.

Fig. 9. Ventral view of *Drawida nepalensis*. Sp. P.-spermathecal pore.

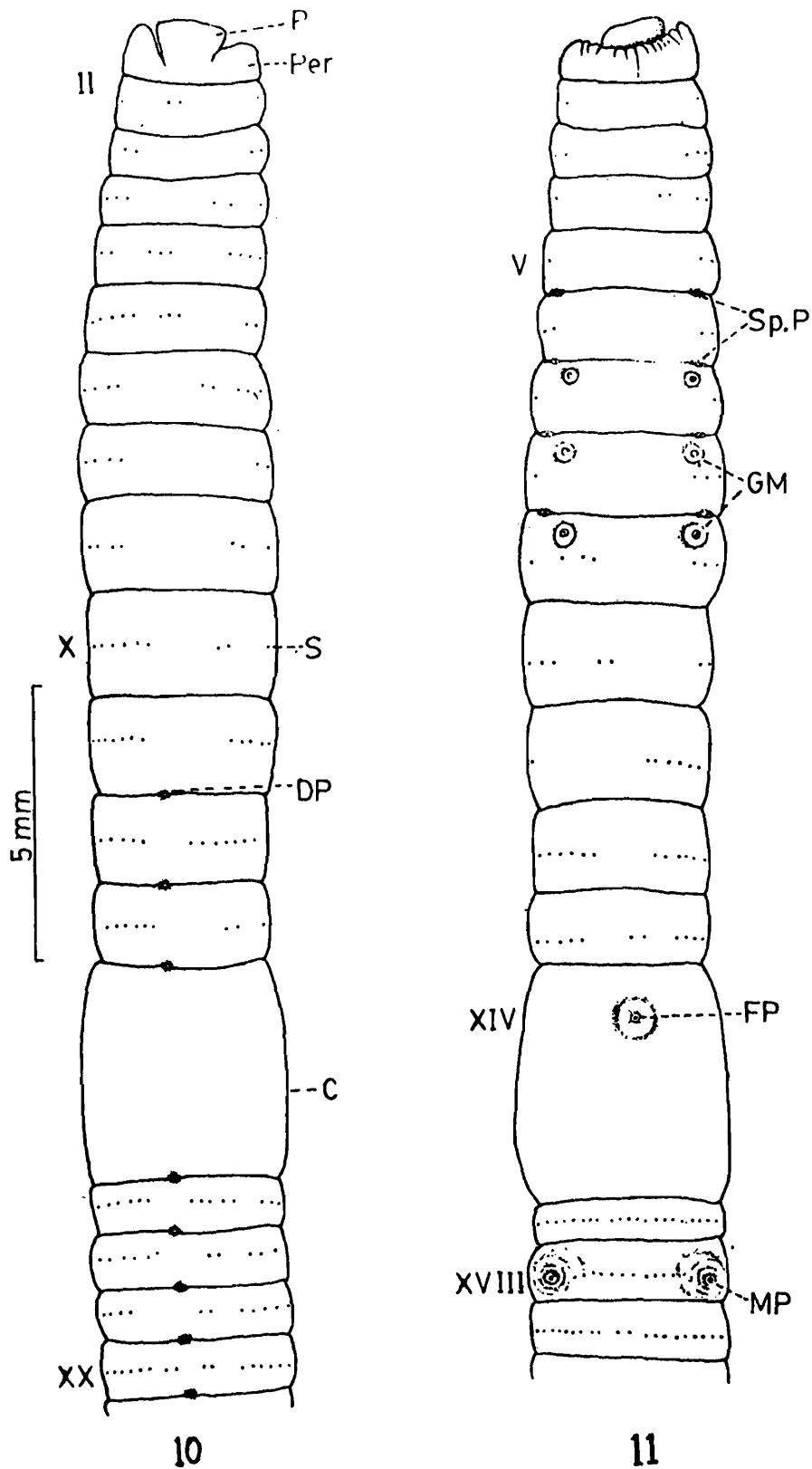


Fig. 10. Dorsal view of *Amynthes diffringens*. c-clitellum, DP-dorsal pore, P-prostomium, Per-peristomium, S-setae.

Fig. 11. Ventral view of *Amynthes diffringens*. FP-female pore, GM-genital marking, MP-male pore, Sp. P-spermathecal pore.

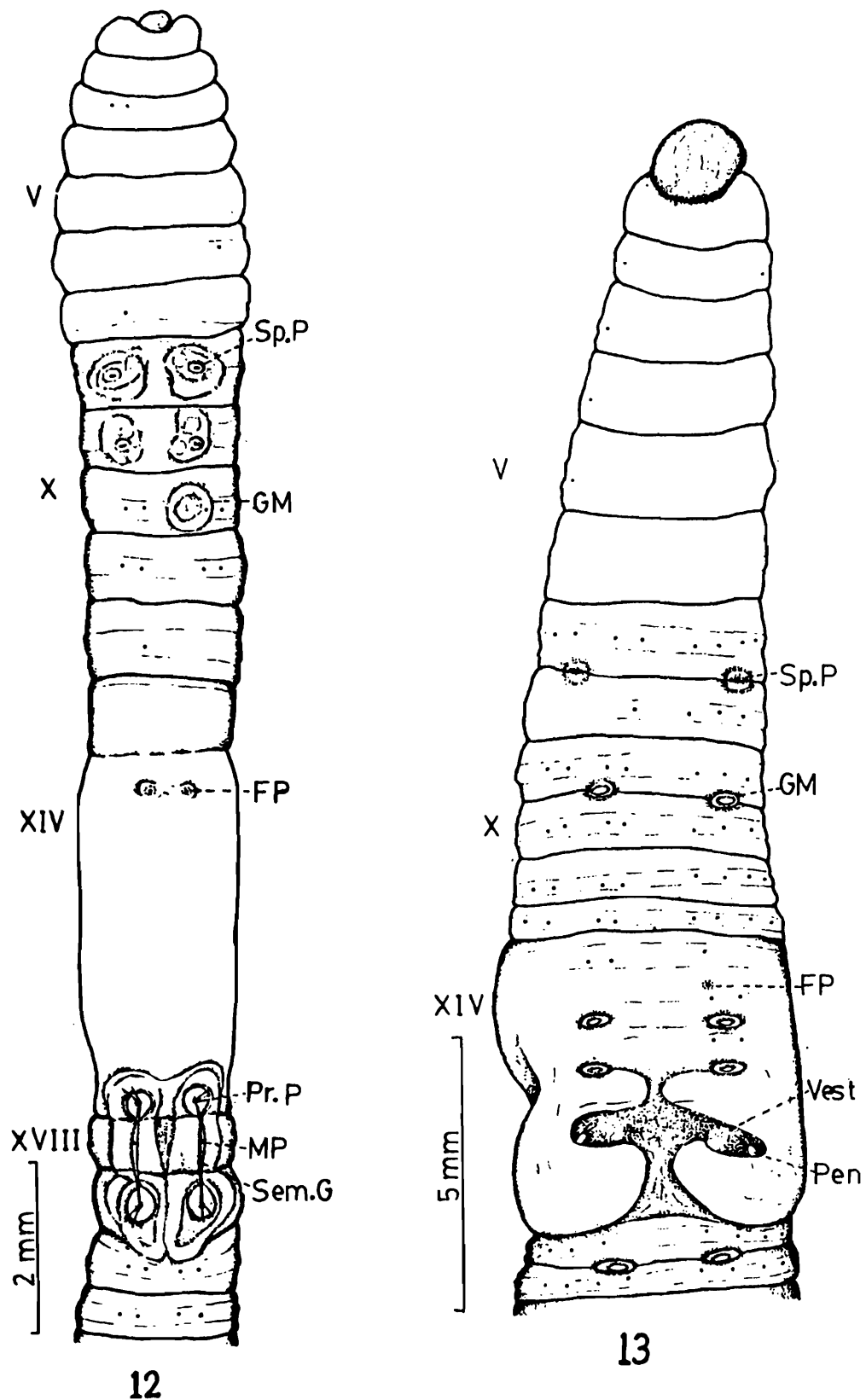


Fig. 12. Ventral view of *Pellogaster bengalensis*. FP-female pore, GM-genital marking, MP-male pore, Pr. P-prostatic pore, Sem. G-seminal groove. Sp. P-spermathecal pore.  
 Fig. 13. Ventral view of *Eutyphoeus waltoni*. FP-female pore, GM-genital marking, Pen-penis, Sp.P-spermathecal pore, Vest-vestibulum.

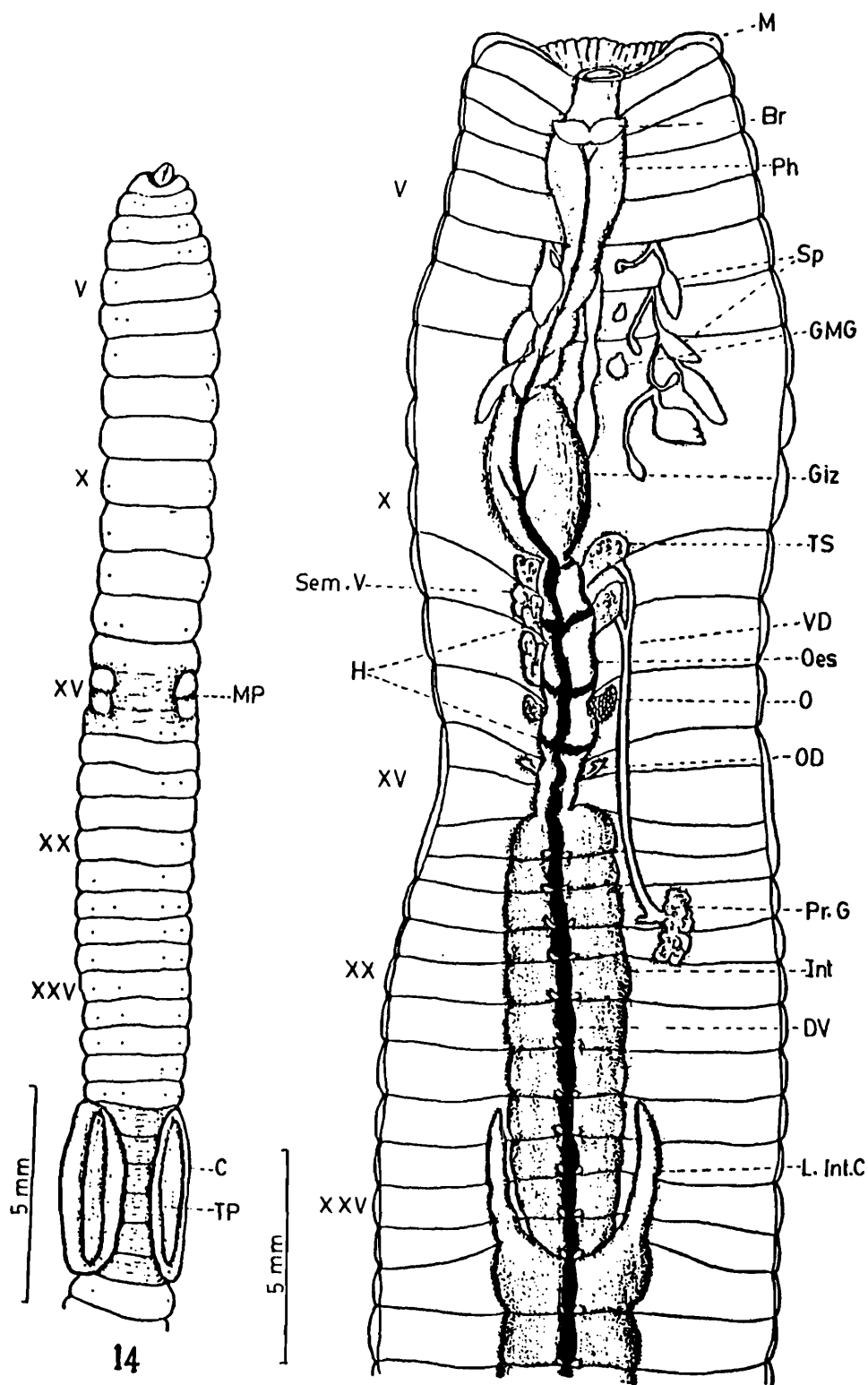


Fig. 14. Ventral view of *Octolasion tyrtaeum*. C-clitellum, MP-male pore, TP-Tubercula pubertatis.

Fig. 15. Internal organs of *Aynthas diffringens*. Br-brain, Dv-dorsal vessel, Giz-gizzard, GMG-genital marking gland, Int-intestine, L.Int.C-lateral intestinal caecum, M-mouth, O-ovary, Oes-oesophagus, OD-oviduct, Ph-pharynx, Pr. G-prostatic gland, Sem. V-seminal vesicle, Sp-spermathecae, TS-testis sac, VD-vasa deferentia.

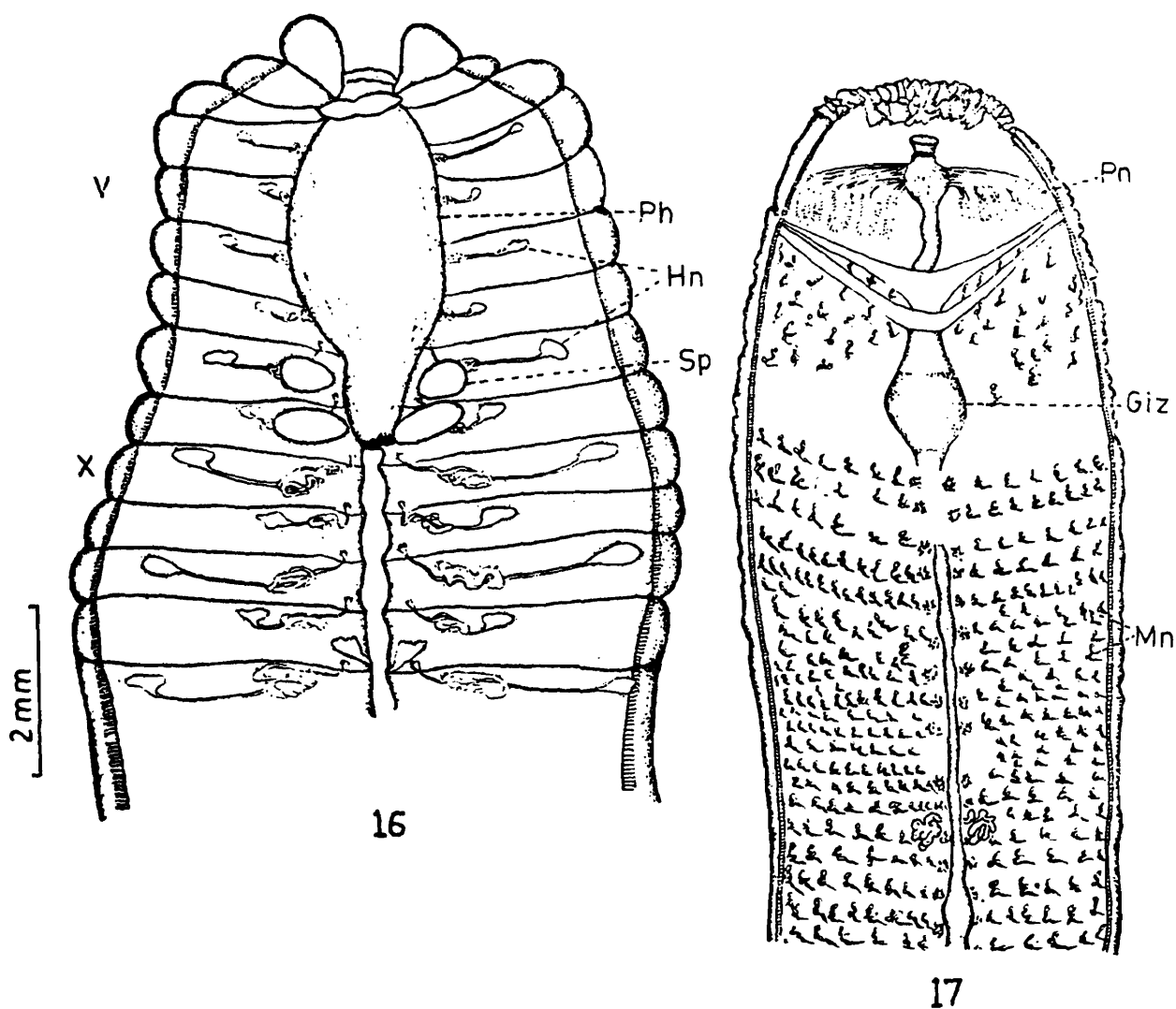


Fig. 16. Holonephric excretory system in *Perionyx sansibaricus*. Hn-holonephridia, Ph-pharynx, Sp-spermathecae.  
 Fig. 17. Meronephric excretory system in *Eutyphoeus nicholsoni*. Giz-gizzard, Mn-meronephridia, Pn-pharyngeal nephridia.

septum at the intersegmental furrow. Most of the septa are provided with minute apertures which permit the coelomic fluid to pass freely between segments. Some of the septa in the gizzard region in some species may be much thickened or absent. The presence or absence of septa in the anterior region is of taxonomic importance.

The digestive system (Fig. 15) comprises a straight alimentary canal extending from mouth to anus, and associated caeca and glands. The anteriormost part of the canal consists of a short but muscular buccal cavity, followed by a pharynx. The dorsal surface of the pharynx is thick, muscular and glandular. The worm sucks food by the action of pharynx. A short narrow tube, the oesophagus, passes posteriorly from the pharynx. In most of the megascolecids, octochaetids and acanthodrilids, oesophagus is modified to form a very prominent oval structure, the gizzard, in any of the segments from v to viii. The gizzard is a highly muscularized organ for pulverising the food material. The number and position of gizzards have been used in distinguishing the genera, but their position should be determined carefully as some of the septa in this region are either absent or very delicate which may break as the worm is opened. The gizzard may be rudimentary in some worms, e.g. *Perionyx* spp. There are two oesophageal gizzards in some octochaetid genera like *Dichogaster*, *Eudichogaster* and *Barogaster*. In the Moniligastridae, usually more than 3-5 gizzards are present in xii and posterior segments. A thin-walled storage chamber, the crop, is located at the posterior end of the oesophagus and in front of the gizzard in the family Lumbricidae. Various types of calciferous glands are associated with oesophagus in some earthworms. They are highly vascular organs provided internally with lamellae. Their shape, number, segmental position as well as stalked or sessile, paired or unpaired and extramural or intramural provide useful distinguishing characters. The rest of the alimentary canal is the intestine in which most of the digestion and absorption of food takes place. The internal surface of the intestine is sometimes increased by a large dorsal fold, the typhlosole, which may be in the form of a simple, bifid or even trifid lamella. Its presence or absence, and anterior and posterior limits are important taxonomically. Some species have small tubular outgrowths of intestinal caeca; their shape, position, number and whether single or paired are of systematic value. Several pairs of glands, the supra-intestinal glands, are sometimes located on the dorsal wall of the intestine in successive segments at the posterior end of typhlosole (e.g. *Eutyphoeus*).

The blood vascular system comprises three main vessels extending almost the entire length of the body. These are : a dorsal vessel, closely associated with the mid-dorsal line of the alimentary canal; a ventral vessel between alimentary canal and nerve cord; a subneural vessel between the nerve cord and the body wall. A supra-oesophageal vessel is present on the dorsal wall of the gut in anterior segments. Paired extra-oesophageal and latero-parietal vessels may be present in some earthworms. The dorsal and ventral vessels are connected in each segment by paired commissures, which in some of the anterior segments are enlarged as contractile 'hearts'. The segmental location of last pair of hearts is of taxonomic importance. These may be in segment xi (e.g. Families Ocnodrilidae and Lumbricidae) or in xii or xiii (e.g. Families Acanthodrilidae, Octochaetidae and Megascolecidae). The dorsal vessel may be aborted anteriorly in some species of *Eutyphoeus*, while the subneural is absent in several octochaetid genera.

There is no formalized respiratory mechanism in earthworms. Exchange of gases takes place through highly vascular moist epidermis. Respiration occurs in air but worms can exist for long periods in highly oxygenated water (Reynolds, 1977).

The excretory organs of earthworms are series of coiled tubes called nephridia. They are of various kinds and have recently gained importance in earthworm taxonomy. Different types of nephridia may be found within a species. They may be one pair (holonephric; Fig. 16) or more than one pair (meronephric; Fig. 17) in each segment. Either type of nephridia may be open (stomate, furnished with a ciliated nephrostome) or closed (astomate). Nephridia are either exonephric with their ducts opening directly to the exterior or enteronephric with ducts discharging into the alimentary canal. Astomate enteronephric holonephridia are not yet known. Meronephridia are either very small (micromeronephridia) or relatively conspicuously enlarged into

megameronephridia. Ectal ends of the ducts of holonephridia are sometimes dilated to form nephridial bladders or vesicles of various shapes.

The central nervous system comprises a bilobed cerebral ganglion on the dorsal surface of the pharynx, a pair of subpharyngeal ganglia, a pair of circumpharyngeal connectives and a ventral nerve cord. The cerebral ganglion is connected to the subpharyngeal ganglia by the circumpharyngeal connectives. The ventral nerve cord runs beneath the alimentary canal close to the body wall from the subpharyngeal ganglia to the last body segment. Superficially the ventral nerve cord appears to be single, it is actually made up of two longitudinal fused cords. Behind the fourth segment, the nerve cord is swollen in each segment to form a ganglion, from which arise three pairs of segmental nerves that extend around the body wall. At present characteristics of the nervous system have not been used in the earthworm taxonomy.

The reproductive organs (Fig. 15) consisting of testes, ovaries, seminal vesicles, spermathecae and prostatic glands have long been used as the main source of taxonomic characters. The basic arrangement of the gonads in the megascolecid worms (Families Ocnero-drilidae, Acanthodrilidae, Octochaetidae and Megascolecidae) is paired testes in segments x and xi on posterior faces of septa 9/10 and 10/11, paired male funnels on anterior faces of septa 10/11 and 11/12, and paired ovaries in xii and xiii (hologyny). The number of testes may be reduced to a single pair (meroandry); a condition with one pair of testes in segment x is termed as proandric, and when in segment xi it is called as metandric. The number of ovaries may also undergo reduction i.e. one pair of ovaries in segment xii (progyny) or in xiii (metagyny). The testes and male funnels may lie free in their segments or enclosed in special coelomic chambers, the testis sacs. Septa of the testis and ovarian segments may be evaginated to form the seminal vesicles and ovisacs respectively. The shape, size, number and segmental location of these structures are of systematic importance. The male funnels open into straight or coiled male ducts, the vasa deferentia. In holandric forms, the anterior and posterior male ducts on each side extend backwards and may unite with each other before opening to the exterior or may discharge independently on the body wall. They may open directly on the body wall (as in *Octochaetona*) or through the prostatic glands (as in *Amynthas*, *Metaphire*). The posterior end of the vas deferens is sometimes enlarged into an ejaculatory bulb (e.g. *Hoplochaetella*). Accessory reproductive organs, the prostates, are associated with the posterior ends of vasa deferentia in most families of earthworms. Prostates are tubular in shape with a central canal as in the Octochaetidae, Ocnero-drilidae and Acanthodrilidae or are of racemose shape without a central canal as in the Megascolecidae. In the Eudrilidae, these glands are in the form of outgrowths from the male ducts and are called as 'euprostates'. In the Moniligastridae, the prostates (also termed as male atria) have an outer and an inner glandular, and a middle muscular layer, the latter forming a prostatic capsule. Prostatic glands are absent in the Lumbricidae. The spermathecae are sac-like organs opening ventrally in some of anterior segments, and receive the sperm of the other worm during copulation. A spermatheca, typically, consists of an ental sac-like ampulla, a duct by which it opens to the exterior and one or more diverticula usually arising from the duct. Spermathecae in some worms may be absent or adiverticulate. The structure, arrangement and position of reproductive organs in other families of earthworms are different as compared to the megascolecid worms. One or two pairs of testes and male funnels are enclosed in intraseptal sacs in the Moniligastridae. Spermathecae are one or two pairs with long tubular ducts in this family.

Internally accessory glands may be associated with genital markings, tubercula pubertatis, copulatory and genital setae. The shape and size of these glands are of taxonomic value.

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## **Distribution Pattern in Indian Earthworms**

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Earthworms possess limited means of active migration; but passive dispersal has distributed some species over wide areas. Most of them cannot survive marine environments which are, therefore, effective barriers for extension of their natural distribution. Endogeic species have, in general, a poor capacity to migrate (Bouché, 1983). Some of them are very sensitive to desiccation and do not leave the soil. Anecic worms are able to leave their burrows under certain conditions and colonize a few metres per annum (Mazaud and Bouché, 1980). Epigeic forms are most mobile and travel some distances (sometimes on a mass scale) in search of food or shelter. Man is also responsible for transporting a few species for culture and in soil around roots of exotic plants. It is well known that anthropochorous species are able to colonize disturbed environments which may be created by deforestation, intensive cultivation of new area, pesticides, waste disposal, etc. Natural destruction of earthworm habitats may be due to glaciation, tectonic disturbance and changes in sea-levels. Presence of anthropochorous or peregrine species in an area sometimes obscures the distributional pattern of earthworms. These species, therefore, must be disregarded for the distributional considerations of earthworms.

Earthworms of the world belong to not more than a dozen families. According to Wallwork (1984), this is a remarkably small number for a group of invertebrates that has been around for a very long period, and successfully spread to soils throughout the world. Possibly, these organisms have not been able to exploit diverse food niches in an evolutionary sense.

The evolution of earthworms is obscured due to paucity of fossil records. Nevertheless, their origin has been inferred from studies on the distribution, ecology and comparative anatomy of the extant species. On the basis of their food and feeding habits, Stephenson (1930) believes that they appeared in the Cretaceous when dicotyledenous plants came into existence. Michaelsen (1903) and Arldt (1908) estimate their origin much earlier during the upper Jurassic and upper Triassic periods respectively. Recently, Bouché (1987) traces their origin in the Palaeozoic period. Sims (1980) assumes that the ancestors of present day earthworms were wide spread in the undivided paleocontinent of Pangaea, which existed during the Palaeozoic.

### **Physiographic and Zoogeographic Regions of India**

Considering the past geological history of the Indian subcontinent, Wadia (1973) divided the present day India (excluding Burma, Nepal and Pakistan) into six well-defined physiographic regions depending upon topography, climate and vegetation. These are : Western Himalayas, Eastern Himalayas and Northeast Ranges, Indo-Ganga Plains, Central Highlands, Peninsular Plateaus and Western Ghats including Sahyadri and Nilgiri Hills. The insular areas of the Andaman and Nicobar form a distinct region having geological and biotic affinities with Burma and Malayasia.

Different zoogeographical divisions of the Indian subcontinent have been proposed from time to time. Recently, Menon (1990) has critically reviewed these and followed the system as proposed by Smith (1931-43) keeping in view the present day distribution of freshwater fishes. Smith's zoogeographical areas of the subcontinent includes the Indo-Chinese subregion. These divisions are further divided into distinct geographical areas. The Indian subregion comprises : the Desert area of

North-west India (North-west frontier Province, Punjab, Western Rajasthan as far as the Aravallis and Sind); Kashmir and the Western Himalayas upto Nepal; the Indo-Ganga Plains (from the Indus valley in Sind to the right bank of Brahmaputra in Bengal); Central India (between the Ganga Plains and the Deccan, bounded on the west by the Aravalli, and on the east by the Chota Nagpur Plateau); the Deccan (most of the Peninsular India between 10°N and 20°N); mountains of the Malabar Tract and Sri Lanka; the Chota Nagpur area. The Indo-Chinese subregion is represented by Eastern Himalayas (from Western border of Nepal to the bend of the Brahmaputra in Arunachal Pradesh); Trans-Himalayan Mountains (hills east of the Brahmaputra, whole of Burma except lowlands in the south and the northern part of Siam); Annam; Peninsula of Indo-China; Tenasserim and Peninsular Siam.

### Distribution of Earthworms in the Indian Subcontinent

#### *Family Moniligastridae*

It is a group of primitive earthworms with endemicity in eastern and southern Asia. Some species are common in arable soils, but most are found in primary forests. Most of the moniligastrids prefer soils with high per cent of moisture. A few species exceed one metre in length. Of the 5 genera known in the world, 4 are found in our subcontinent. These are : *Desmogaster*, *Drawida*, *Hastirogaster* and *Moniligaster*. *Desmogaster* and *Hastirogaster* are restricted to Burma. A single record of *Desmogaster ferina* from Arunachal Pradesh may be due to its transportation from Burma. *Drawida* is the largest genus in terms of number of species. Its natural distribution extends to the Indian Peninsula, Eastern Himalayas and northeast Ranges, Burma and as far as Korea, Japan, China, Manchuria, and Phillipine Islands, Sumatra and Borneo. Occurrence of pregrine species, *Drawida japonica* and *Drawida nepalensis*, in the Western Himalayas may be due to recent transportation. *Moniligaster* comprises 9 species, which are restricted in their distribution to the southern region of the Western Ghats.

#### *Family Criodrilidae*

Its indigenous range is confined to the southwestern Palaearctic but some species have been carried to other regions, probably with water plants. Immature specimens of the widely distributed *Criodrilus lacuum* from our region belong to *Glyphidrilus* (Family Almidae).

#### *Family Lumbricidae*

The family is endemic throughout the Palaearctic region and eastern North America. A number of species have been introduced into almost all the zoogeographical regions of the world. At places they have been able to compete with them, especially in cultivated soils. Stocks of one species, *Eisenia fetida*, have been carried from one place to another all over the world for commercial culture. Lumbricids possess inherent ability to colonize wide range of soil types and microclimates. Peregrine lumbricids have acquired domicile in India at hill resorts with a temperate-like climate in the Himalayas, and Nilgiri and Palni Hills in the Peninsula. They are more widely distributed in Western Himalayas, and form a dominant group in certain habitats at some places.

#### *Family Glossoscolecidae*

This family forms the dominant group in tropical South America. It is represented in the Indian subcontinent by a circummundane species, *Pontoscolex coreithritus* which is now widely distributed in the Indian Peninsula and northeast region.

#### *Family Almidae*

Members of this family are mud dwellers or found in freshwater habitats. In this region, it is represented by a single genus, *Glyphidrilus* with endemicity in Burma, Northern Ranges, Ganga

Plain, Western Ghats, Peninsular Plateaus and Sri Lanka. The genus is also indigenous to Africa, Malayasia and Indonesia. An African origin of the genus has been assumed.

#### *Family Ocnerodrilidae*

It comprises two subfamilies : Ocnerodrilinae and Malabarinae. Ocnerodrilinae is represented in this subcontinent by one or two circummundane species of *Gordiodrilus*, *Ocnerodrilus*, *Eukerria* and *Nematogenia*. They were possibly transported from Africa or South America where the subfamily is endemic. Monospecific *Curgiona* of this subfamily is found only in South India. Though its origin is uncertain, its home range is also believed to be in Africa.

The subfamily Malabarinae is constituted by 3 genera : *Deccania*, *Thatonia* and *Malabaria*. They occur mainly in the peninsular India, but some species have spread into the Himalayan hills and Burma.

#### *Family Acanthodrilidae*

Endemics of the family are found in South America, some parts of North America, Africa, southeast Asia and Australasia. Three genera occur in the Indian subcontinent. *Pontodrilus* and *Microscolex* are represented by the peregrines *P. bermudensis* and *M. phosphoreus* respectively. *Plutellus* has endemicity mainly in the Western Ghats and Annamalai Hills in the Peninsula, Eastern Himalayas and Northeast Ranges and Burma. One species has been recorded from the Ganga Plains. The genus *Plutellus* has been restricted by Jamieson and Nash (1976) to include only Australian species. Jamieson (1977) referred the Oriental species of the genus to the Australasian *Diporochaeta*, a view which is not acceptable on morphological and geographical grounds.

#### *Family Octochaetidae*

The family occurs in New Zealand, Australia and spreads through the tropics of America and Africa, including Madagascar, with the Asian representatives extending outward from peninsular India.

Twenty six octochaetid genera are known from the Indian subcontinent. Of these, twenty five are indigenous and form more than 50% of the known endemic genera of earthworms in the subcontinent. These can be referred to two distinct groups : a small north and northeast group of *Eutyphoeus*, *Scolioscolides*, *Bahlia* and *Calebiella* and the rest forming a larger southern group. The southern group is confined to the Western Ghats (including the coastal plain), Nandi Hills, Central Highlands and Eastern Plateau of the Peninsula. A few peregrine species of the *bolawi* group of the Ethiopian genus *Dichogaster* have domiciled at various parts of subcontinent.

#### *Family Megascolecidae*

The distributional range of the family extends between the warm-temperate Asia and Australasia. Two genera of the pheretimoid group, *Amyntas* and *Metaphire*, are endemic in Burma, and Andaman and Nicobar Islands, but are represented by peregrine species in other parts of the region. Two genera of the group, *Pithemera* and *Polypheretima* are exotic in the subcontinent. The other endemic megascolecids in our area are : *Comarodrilus*, *Lampito*, *Kanchuria*, *Megascolex*, *Lenoscolex*, *Nelloscolex*, *Notoscolex*, *Perionyx*, *Tonoscolex* and *Troyia*. Of these *Comarodrilus*, *Lenoscolex*, *Notoscolex*, *Troyia* and *Lampito* (excluding widely distributed *L. mauritii*) are confined to the southern portion of the Western Ghats. *Megascolex* is widely spread in the Western Ghats and adjoining peninsular plateaus. Both *Megascolex* and *Notoscolex* have also endemic species in Australasia. It is suspected that the Australasian species are not congeneric with the Indian forms. *Nelloscolex* and *Tonoscolex* occur in northeast Hills, the latter's range extending to the Eastern Himalayas. Both these genera are also endemic in Burma. Another allied genus,

*Kanchuria*, is restricted to the Garo and Khasi Hills. *Perionyx* (excluding widely distributed *P. excavatus* and *P. sansibaricus*) is found in the Peninsula, Eastern and Western Himalayas and Burma.

### Conclusions

Majority of earthworm genera in the Indian subcontinent have endemicity in the Deccan Peninsula (including Sri Lanka), northeast India and Burma. The Peninsula was once a part of an ancient supercontinent Gondwana and has never been submerged under the sea. Excepting the Malayan Moniligastridae and Ethiopian Almididae, most of the indigenous earthworm genera presumably evolved and developed in the Deccan Peninsula. *Plutellus*, *Megascolex* and *Notoscolex* also have endemic species in Australasia. These genera are considered to be polyphyletic congeries and their relationships with Indian species are yet to be determined, and should be disregarded for any biogeographical considerations.

*Nelloscolex*, *Tonoscolex*, *Eutyphoeus* and *Scolioscolides* in the northeast region, and *Bahlia* in the Ganga Plains do not occur in the peninsula. According to Gates (1972), these genera are the culmination of megascolecid and octochaetid lineages that evolved in the northern half of the peninsula. The route for the migration of their ancestral stock was probably over a Miocene land bridge across the present day Rajmahal Gap between the Satpura Hills of the Peninsula and northeast ranges. *Perionyx* and *Plutellus* are found in the northeast region and in the Peninsula and migrated to the northeast across the Rajmahal gap. The Malayan moniligastrids possibly followed this route to reach the Peninsula. The endemics of *Amyntas* and *Metaphire* occurring in Burma have closer affinities with the corresponding Malayan and Chinese fauna.

The most interesting feature of earthworm distribution in the Western Himalayas is pauper representation of endemic species; only four species belonging to *Perionyx*, *Eutyphoeus* and *Glyphidrilus* have been recorded at isolated places. It seems that the endemics in this region were exterminated during the last Quaternary glaciation which affected the area most than the other regions of the subcontinent (Wadia, 1973). Alternatively the wave of westward migration of endemic earthworms from the east could not reach this region with full force.

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## Chromosomal Pattern in Earthworms

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Earthworms play a major role in the formation of soil structure and its fertility. They are also an important link in the food chain of soil fauna. As source of animal protein they contain essential amino acids, and have been added to poultry and pet food in some countries. It is estimated that there are nearly 90,000 earthworm ranchers, who rear and sell worms in the United States of America. Many families in the world are profiting by utilising worms for the improvement of soil, disposal of organic wastes and producing worm casts as biofertilizers.

There had been a rapid advancement of chromosome findings in many groups of animals but such studies remained totally lacking on earthworms. A perusal of the literature reveals that only a fraction of earthworm species is known cytologically when compared to the vast taxonomical available data. Reasons for scanty information on the cytology of earthworms could be due to small size of their gonads, clumping tendency in their chromosomes, low mitotic and meiotic indices, high degree of polyploidy resulting in an uneasy counting and incomplete analysis of chromosome structure, existence of parthenogenesis, etc. Earthworms are hermaphrodites and are believed to have originated from some ancestral polychaete having separate sexes. These animals are characterised in the basic chromosome numbers of 11, 16, 17, 18, 19 and frequent occurrence of polyploidy in many species. Some of earlier reports show that polyploidy and aneuploidy are common but their evolutionary significance has not yet been realized. Further, the extent of polyploidy is too limited for any assessment of its role in speciation.

The Indian subcontinent is rich in earthworm fauna with about 500 known species belonging to about 65 genera. Handa (1969) initiated cytological studies on Indian earthworms and brought to light some interesting chromosomal information. As a result, he reported chromosome number in 13 species of worms of the families Lumbricidae and Megascolecidae.

**Table 1** Number of Cytologically known species

Family	No. of spp. Cytologically worked out	No. of spp.* Known	%
Tubificidae	2	147	1.36
Enchytraeidae	71	500	14.20
Lumbricidae	54	302	17.80
Megascolecidae (including Acanthodrilidae and Octochaetidae)	24	1912	1.25
Total	151	2861	

(\* Source : Brinkhurst and Jamieson, 1971; Sims, 1982)

Table 2 Details of chromosomal studies by different workers

Author	Year	Chromosome number and related information
Calkins	1895	Spermatogenesis <i>Lumbricus terrestris</i> , $2n = 32$ , $n = 16$
Foot	1898	Oogenesis of <i>Allolobophora foetida</i> , $2n = 22$ , $n = 11$
Gathy	1900	Oogenesis of <i>Tubifex</i> , somatic cells-100, oocyte-1-110
Bugnion & Popoff	1905	<i>Lumbricus agricola</i> , $n = 16$
Foot & Strobell	1905	<i>A. foetida</i> -11 bivalent with tetrad, rod, rings and cross-figures, $2n = 22$ , $n = 11$
Meek	1913	<i>Lumbricus</i> sp., $2n = 32$ , $n = 16$
Muldal	1949	30 spp., polyploidy in 5 genera (3n, 4n, 10n)
	1952	$n = 18$ in <i>Lumbricus</i> ; 16,18 in <i>Allolobophora</i> ; 18 in <i>Octolacium</i> , <i>Eiseniella</i> ; 11, 18, 20 in <i>Eisenia</i> ; 17 in <i>Dendrobaena</i> ; 10 in <i>Bimastus</i>
Makino	1951	For details see Atlas on chromosomes
Walsh	1951	Reported variations in <i>Lumbricus terrestris</i>
	1954	No multiplication of chromosome set beyond $2n$ in testes and seminal vesicle
Omodeo	1952	Lumbricidae, 30 spp.
	1955	$2n = 114, 72, 54, 36, 32, 30, 180$ (10n), $22$ ( $n = 11$ ), 34, 102, 112-124, 108
Christensen & Nielsen	1955	Worked on 7 Danish genera. $2n = 120-140$ , in some 180-200; $n = 13-17, 21, 32$
Murchie	1967	11 spp. of <i>Diplocardia</i> (Megascolecidae), $n = 22$ , highest no. as 198
Handa	1969	Studied 13 spp. of land earthworms
	1971	Studied 5 spp. of aquatic earthworms
	1974	Described $2n, n$ , aneuploidy, polyploidy
	1975	Aberrant behaviour, low mitotic index
	1977	Chiasma in Lumbricidae and its absence in Megascolecidae
Dulout <i>et al.</i>	1971	$2n = 30$ in <i>Pheretima californica</i> Kinberg (in testes, seminal vesicles and ovaries, somatic tissues)
Snadra & Renata	1972	Gametogenesis in 3 spp. of worms of Iceland, i.e., <i>Dendrobaena</i> and <i>Lumbricus</i> . In <i>D. rubida</i> , hexaploid parthenogenesis and octoploid amphigonic strains.
Sharma <i>et al.</i>	1976	Cytogenetical relationship, etc.
Mihailova & Subchev	1981	$n = 7, 2n = 14$ in <i>Branchiobdella astaci</i>
Handa & Kumari	1984	Chromosomes of <i>Eutyphoeus incommodus</i> and <i>E. waltoni</i>

## Techniques for the Study of Earthworm Chromosomes

### *Location of Reproductive Organs*

The oligochaetes are protandrous, hermaphrodite and the number, position and arrangement of the testes, ovaries and seminal vesicles vary greatly between families and genera. In the order Moniligastrida with a single family Moniligastridae, paired testes and seminal vesicles are intraseptal (septum 9/10 in *Drawida*), and ovaries are elongately band like, with large, yolky oocytes, paired in segment XI (*Drawida* spp.). The order Haplotaxida with major families of Lumbricidae, Acanthodrilidae, Octochaetidae and Megascolecidae possess 2 pairs of testes in segments X and XI, one pair of ovaries in XIII and seminal vesicles in all or some of IX-XII. One of the paired testes may be absent in some genera/species.

The individuals are dissected with the help of a fine blade, a pair of forceps and needles. Testes and seminal vesicles are removed for chromosome preparations.

**Dissecting mediums** : The specimens are dissected separately in either 0.7% saline (Baker, 1944) or 1% Sodium citrate (French, Baker and Kitzmiller, 1962) medium. The dissecting medium 0.7% saline yields better results.

**Hypotonic treatment** : In order to get good spreading of chromosomes and clear morphological structures, various hypotonic treatments are used resulting in the swelling of cells as well as avoiding clumping of chromosomes.

**Saline treatment** : An aqueous salt solution recommended by Darlington and Lacour (1960) is tried for 30-40 minutes. The concentrations applied are 0.7% and 0.9%. The results obtained by this method are, however, not very satisfactory.

**Sodium citrate** : Different concentrations of sodium citrate solutions used can be 0.9% (Ford and Hamerton, 1956), 0.7% (Fredga, 1964) and 1.12% (Sparano, 1961). Best results are obtained by using 0.9% sodium citrate solution for 60 minutes at 37°C.

**Fixation** : After pretreatment, the material is fixed in one of the freshly prepared fixatives. Carnoy's fixative (3 parts absolute alcohol : 1 acetic acid-45% acetic acid) (Vaartaja, 1963) i.e. Carnoy's without chloroform for 60 minutes at room temperature gives good results.

### *Preservation of Material*

The material is proceeded for staining soon after fixation. Sometimes, it is even preserved in 70% alcohol at 4°C for 1-2 days, which may be stained later on after washing it 4-5 times in distilled water.

### *Chromosome Techniques*

The following techniques can be employed for the preparation of chromosomes.

1. **Squash techniques** : The various stains used for the squash preparations are : Gomori's haematoxylin (Gomori, 1941; Melander and Wingstrand, 1953), Aceto-caramine (Rattenburry, 1956) and 2% aceto-orcein (Darlington and Lacour, 1960)

**For squashing** : Take a small piece of the tissue (Gonads and seminal vesicle or any growing tissue) which is already lying in the stain (15-30 min.) on a slide and put a cover glass on it. Apply uniform pressure on the cover slip with the help of thumb through folds of a blotting paper. Invert the slide with cover glass (after squashing) in a covered tray containing tertiary butyl alcohol and keep till cover glass is detached. Mount the cover glass and slide separately in euparal. Slides stained in Gomori's haematoxyline produce good results to some extent.

2. **Air drying technique** : This technique is employed as recommended by Crozier (1968). A small piece of material is taken on chemically clean slide and on it 2-3 drops of 60% acetic acid is poured. This is followed by vigorous tapping of tissue with forceps or needles so that cells are loosened. The slides are then allowed to dry and stained in Carbol-fuchsin stain (Carr and Walker,

1961) for 24-48 hours. Differentiation is done by just giving a dip in rectified alcohol. For dehydration, the slides are kept in n-butyl alcohol for 40-50 minutes and mounted in euparal. Excellent results are obtained by this technique.

### *Giemsa Staining*

*Preparation of Giemsa stain* : Giemsa stock-380 mg/Giemsa powder is dissolved in 25 ml of glycerol and kept for overnight. To this is added 25 ml of methanol and stored.

#### *Sorrenson's buffer*

Solution A	-	0.9078 g of $\text{KH}_2\text{PO}_4$ /100 ml
Solution B	-	1.1876 g of $\text{Na}_2\text{HPO}_4$ /100 ml
Solution C	-	50.8 ml of solution A and 49.2 ml of solution B

*Working solution* : From 100 ml of solution C, 47 ml is taken and to it 3 ml of Giemsa stock is added. This makes the working Giemsa stain with pH 6.8.

*Method of staining* : The air dried slides are kept in working Giemsa stain for half an hour and after washing in distilled water, these are dried and mounted in DPX.

### **Chromosomal Pattern in a few selected species**

Handa (1969-1984) in a series of papers has given chromosome information on 13 species of Indian earthworms. Some of the papers refer to only chromosome number and level of polyploid cells in them while others relate to detailed aspects of chromosomal characteristics. In the present account, it is not possible to give details of chromosomal pattern of species studied so far. However, an attempt is made to give chromosomal pattern in some of selected species so that future workers in this field are able to know the type, shape, size and behaviour of chromosomes in earthworms. The species reported here belong to the families Lumbricidae (3 species), Octochaetidae (1 species), Megascolecidae (3 species).

Family : Lumbricidae

#### *Bimastos parvus* (Fig. 15)

Lumbricidae is one of the most widespread and investigated families of the Oligochaeta. Its species from the Indian subcontinent had remained totally neglected. Handa (1969, 1971) for the first time reported diploid number in *Bimastos parvus* as  $2n = 18$  with no morphological distinction between autosomes and sex chromosomes. The meiosis is conventional but for the occurrence of a diffused stage after pachytene. Several spermatogonial metaphases showed the presence of  $3n$ ,  $4n$ ,  $8n$ ,  $10n$ ,  $17n$  and  $18n$  chromosomes in them.

#### *Eisenia fetida* (Figs. 4, 8, 14, 18, 25, 28-29)

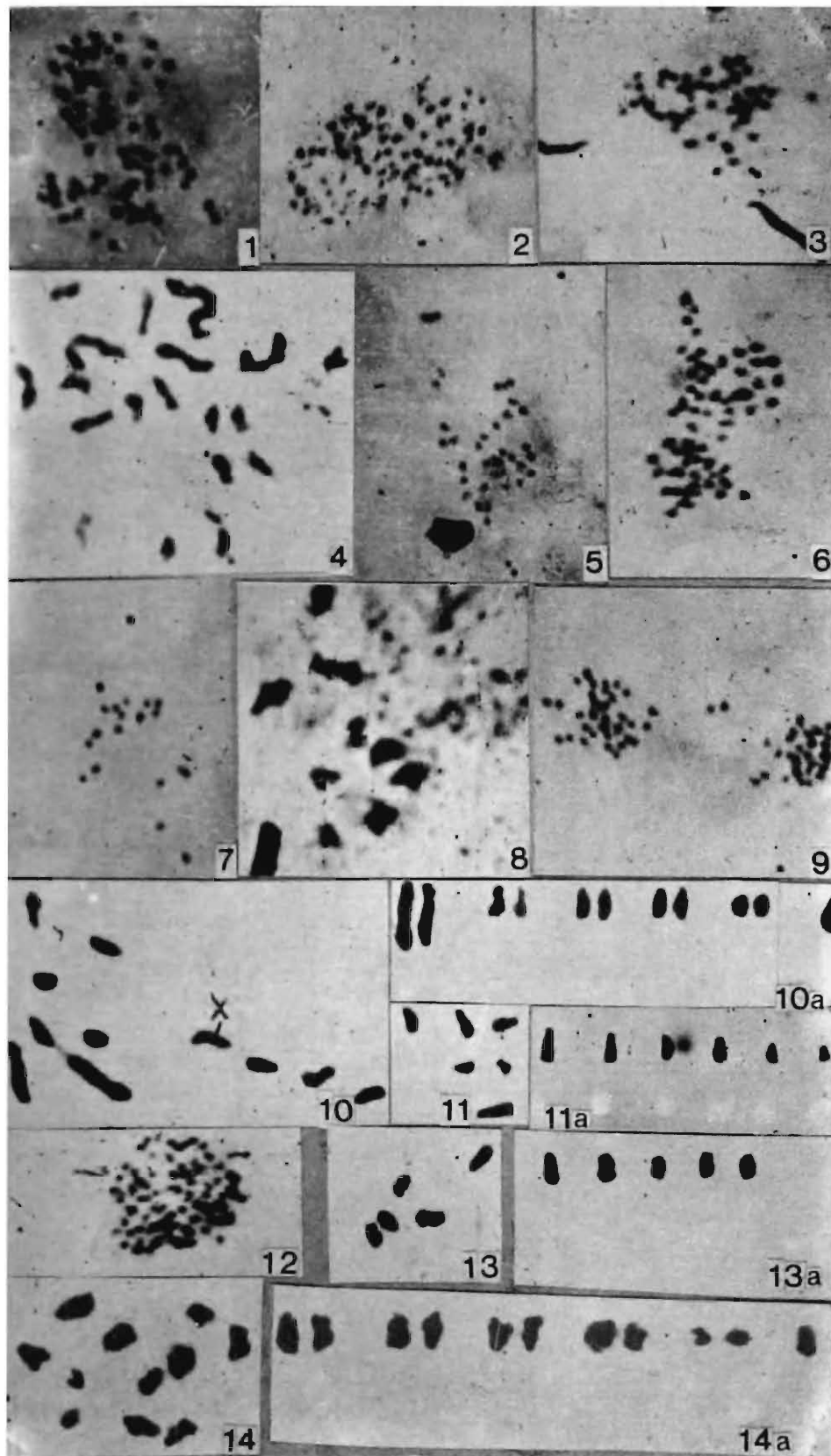
The Indian population of this species (Handa, 1969; 1975) carry a diploid number of 11 chromosomes while 22 have been recorded for this species from Europe. Location of centromere in various chromosomes cannot be marked out with certainty. Aneuploid cells with chromosome number 5,6,9 are frequent. The availability of polyploid cells is common and they are mostly at the mitotic metaphase stage with  $4n = 22$ .

#### *Aporrectodea* (= *Allolobophora*) *trapezoides* (Fig. 10, 11, 13, 17, 24, 26, 31)

This species carry  $2n=11$  and chromosomes can be arranged in their descending order in a karyotype. The aneuploid cells with chromosome number 5, 6, 8 ( $n = 5, 6$ ) are also met with. Polyploid cells at  $4n$  level ( $4n = 22, 26$ ) are quite common.

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**Note** : Explanation to Figures does not include details of each Figure. These are available in the original publications thesis.



Figs. 1-3, 5-7, 9,12. *Pheretima morrisi*. Figs. 4, 8, 14-14a. *Eisenia fetida*. Figs. 10-10a, 11-11a, 13-13a. *Aporrectodea* (=Allolobophora) *trapezoides*.

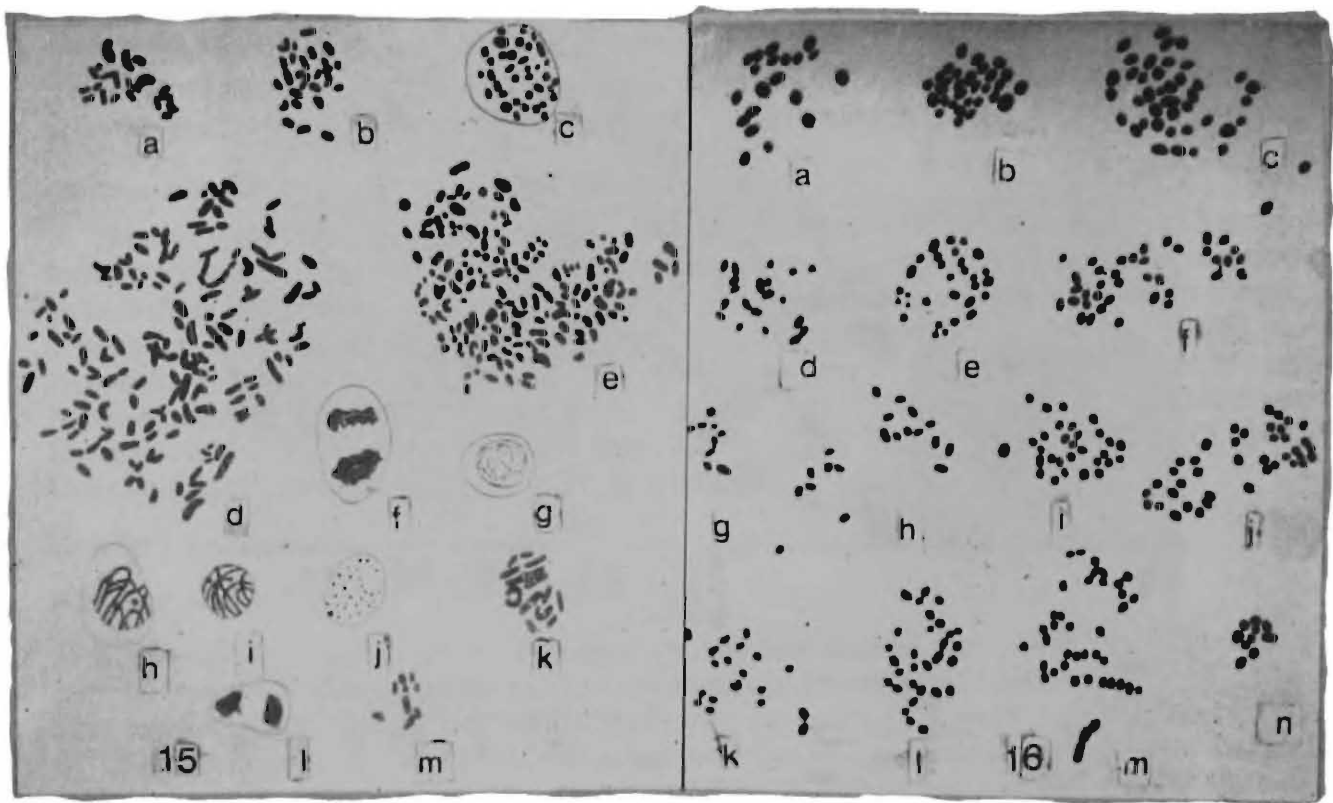
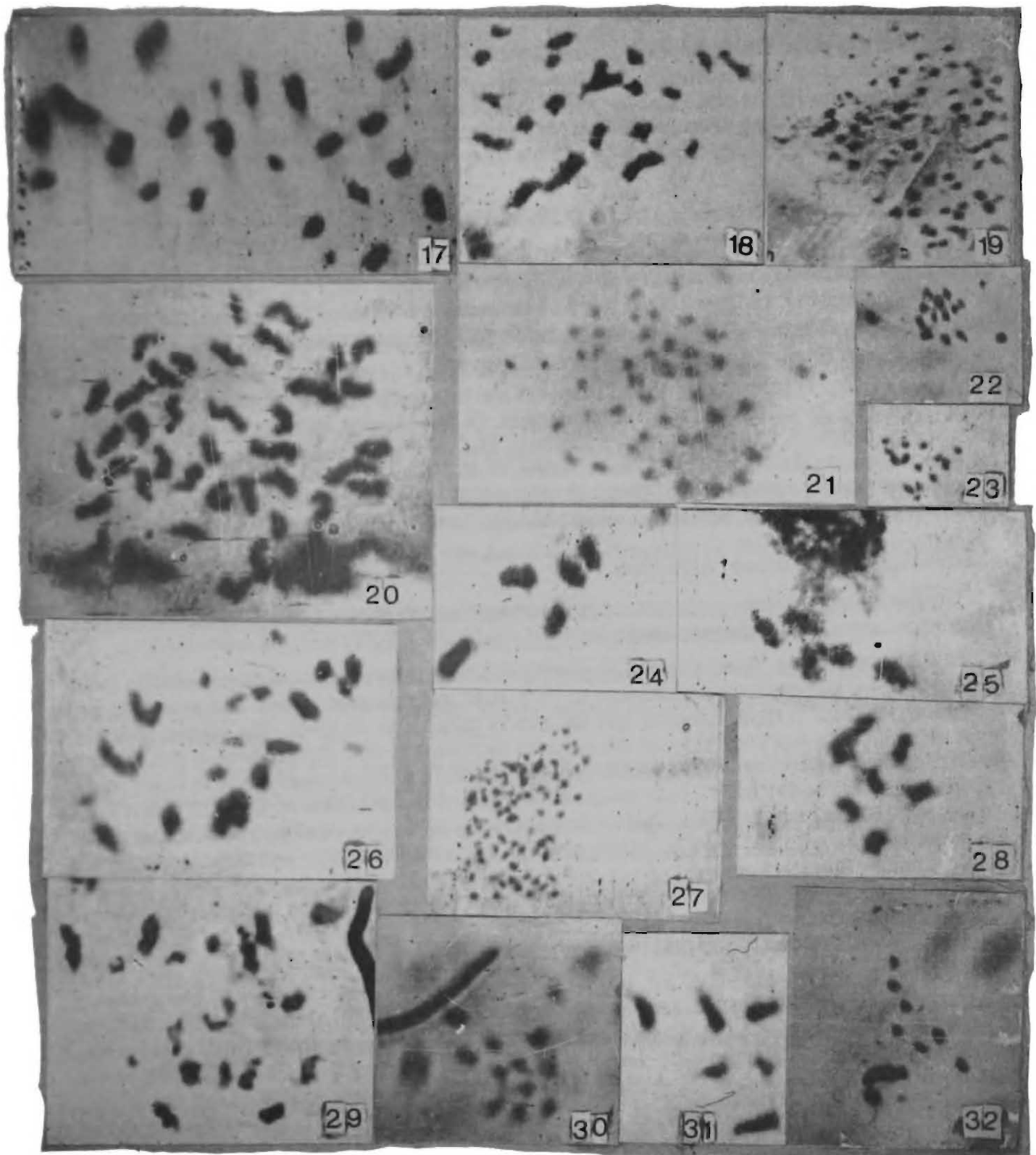


Fig. 15. *Bimastos parvus*. Fig. 16. *Megascolex konkanensis* (a-g); *Megascolex* sp. (h-n).



Figs. 17, 24, 26, 31. *Aporrectodea trapezoides*. Figs. 18, 25, 28-29. *Eisenia fetida*. Figs. 19, 22-23, 27. *Pheretima morrisi*. Figs. 20-21, 30, 32. *Eutyphoeus incommodus*.

### Family Octochaetidae

#### *Eutyphoeus incommodus* (Figs. 20-21,30,32)

Handa (1969) reported the chromosome number and variations in its spermatogonial and seminal vesicle cells. The  $2n$  appears to be either 20 or 22 with  $n$  as 10 or 11 chromosomes. Cells with  $4n$  number of chromosomes at mitotic metaphase are also available. This species on reinvestigation by Handa and Parveen (1984) made them to change their earlier findings on the diploid number depending upon availability of the maximum number of diploid cells. According to them, it appears that actually the  $2n$  is 44 and not 22.

### Family Megascolecidae

#### *Pheretima morrissi* (Figs. 1-3,5-7,9,12,19,22-23,27)

Handa (1969) reported the  $2n$  number in this species as either 20 or 22 with  $4n$  level of polyploidy cells. When this species was reinvestigated in 1984 (data unpublished) it was found that this species carried a  $2n = 16$  and  $n = 8$  chromosomes. Variations in chromosome number is quite extensive. Polyploid cells at the  $4n$ ,  $5n$ ,  $8n$ ,  $9n$  and  $10n$  are also available.

#### *Megascolex konkanensis* and *Megascolex* sp. (Fig. 16)

Handa (1969,77) reported the chromosomes of the two species of the genus *Megascolex*. Both of these possess  $2n = 20$  and  $n = 10$  chromosomes. In *Megascolex konkanensis*, chromosomes vary in size, cells with  $3n$  and  $4n$  are quite common. The presence of pseudo-trivalents and inverted 'V'-shaped pseudo-tetralents are quite common. In *Megascolex* sp., however, chromosomes are alike in appearance with  $2n = 20$ . Polyploid cells are frequent. The chromosomes form pseudo-trivalents and pentavalents, besides the usual bivalents, in various metaphase I cells.

### Conclusion and Future Attention

The literature reveals that only three families of earthworms *Lumbricidae*, *Megascolecidae* and *Octochaetidae* are known chromosomally.

In the Lumbricidae, there exists a constancy in the diploid number of chromosomes in its various species, and chromosomes appear as 'V's, rods and spherical bodies. The occurrence of aneuploidy in them is quite common and the ploidy goes up to  $4n$  level. The mitotic index is, however, quite low and possibility of the chiasma formation is also there. Many of the workers have given the  $n$  number of chromosomes varying from 6-27 with 18 as occurring in many of the species. Earlier workers on *Eisenia fetida* have reported the presence of  $2n = 22$  and  $n = 11$  chromosomes. The findings of Handa from this laboratory on this species have, however, revealed the presence of  $2n = 11$  and  $n = 5,6$  chromosomes. Polyploid cells upto  $4n$  level in this species are quite common. Species *Bimastos parvus* with  $2n = 18$ , and  $n = 9$  (Handa, 1969, 1971) is a new cytological record for this family. Similarly, studies on *Aporrectodea* (= *Allolobophora*) *trapezoides* (unpublished) is another new cytological addition to this family having  $2n = 11$ ,  $n = 5,6$ . The polyploid cells at the  $4n$  level are also present.

From the family *Megascolecidae*, various workers have reported variations in chromosome number, wherein, the diploid number varies from species to species. The  $2n$  may be 16,20,22 as reported by them with  $n=8,10,11$ . The chromosomes are generally in the form of spherical bodies. Polyploidy (cells only) is quite extensive in the family and it varies from  $4n$  to  $10n$  level at the mitotic metaphase stage. Out of 23 species, Muldal (1952) reported six species of this family in a polyploid state. The present findings, however, depicted the presence of only polyploid cells but none of the individuals in a polyploid state.

It is suggested that extensive studies should be carried out on the cytology of Indian earthworms which is still a virgin field. About 3300 species of earthworms are known in the world, but only 151 have so far been reported cytologically. It is hoped that future workers would lay emphasis on the following aspects of cytological studies on Indian earthworms.

1. Chromosome constancy/inconstancy. 2. Polyploidy and its role in speciation. 3. Chiasma formation and chromosome behaviour in cell cycle. 4. Configurations of chromosomes at mitosis and meiosis, and its causes. 5. Sex chromosomes, if any. 6. Amino-acid analysis by different methods. 7. Protein pattern by electrophoresis, etc., in order to work at the biochemical level for evaluating the differentiation of the various species.

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## **Know-how of Earthworms - The Soil Macrofauna**

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Earthworms belonging to class Oligochaeta form the major terrestrial and soil inhabiting organisms of the phylum Annelida. The lumbricid worms have dominated in their distribution in temperate soils and the megascolecid worms predominate sub-tropical and tropical soils. The intensity of competition is high in temperate regions with a narrow niche where litter forms the primary food. As a result, earthworm populations concentrate on soil surface with little behavioural diversification. To avoid competition for space, large sized worms have become épianécic and anécic that live deep in burrows and surface out for collecting litter as food. On the other hand, niche is enlarged with greater food diversity for tropical worms and they show much variation in size and behavioural patterns. This has primarily resulted in preferential feeding habits.

### **Ecological Strategies**

The contrasting climatic conditions of the tropical and temperate zones have made it difficult to compare the role of this major soil fauna. Between extremely high and low temperature and evenly distributed rainfall have favoured worms of temperate region to have a prolonged active phase with intermittent rest period. The lower temperature also favours accumulation of litter and humus due to slow microbial activity, which in turn provides rich source of food for worms. This fits into the reports of Bouché (1975) where he has reported that 88.1% of earthworms are detritivorous and only 13.1% of them are geophagous in a permanent pasture in France. Based on the nature of their vertical distribution in soil strata, their feeding and defecation activity in temperate soils, three different categories of worms can be distinguished ecologically (Bouché, 1977). The épigéic worms inhabit the litter heaps or any other degrading organic matter on soil surface; the endogéic forms inhabit mineral soil horizons and feed more on soil than on organic matter; the anécics live in highly complicated burrow systems. The anécic worms surface out to feed on dead leaves which they normally drag into their burrows before feeding. Majority of endogéic worms have intrinsic factors that would trigger them into the state of seasonal diapause. The anécics which are at the advantage of using the qualities of both épigéics and endogéics pack the burrow walls with their excrements, thus forming stable tunnel in soil. The anécics help in regular mixing of surface matter to lower strata and also loosening the soil. Such a demarcation could not be made with respect to earthworms of tropics and majority of them are geophagous i.e. the endogéics, and only a few are detritivorous (Kale and Krishnamoorthy, 1978; Dash, 1978). Lavelle (1979) also noted lack of anécics among the dominant species in this studies in tropics and suggested that unlike the Lumbricidae of temperate regions where 50-75% of total biomass is anécis, in tropics they form a minor component. Probably, posterior positioning of gizzard and clitellum in lumbricids has allowed for the development of strong musculature which may be responsible fo the occurrence of more anécic species among lumbricids. In our studies in uncultivated marshy lands of Bangalore, *Polypheretima elongata* was found to form complicated firm burrows upto a depth of 90 cm and considered as an example of anécic magascolesid worms (Kale and Krishnamoorthy, 1982).

Table 1. Relative abundance of earthworm species and their distribution pattern (In ratios in different agro-ecosystems (after Bano and Kale, 1988)

Family	Name	Coastal region	Malnad region	Plains
Acanthodrilidae	Plutellus timidus	●	■	
Almidae	Glyphidrilus annandalei	● ▲ 1:2	▲	
Glossoscolecidae	Pontoscolex corethrurus	●	● Y 7:12	● Y 2:10
Ocnereodilidae	Gordiodrilus elegans			●
	Curgiona narayani	Y	●	●
	Priodoscolex montanus			Y
	Octochaetona rosea	Y ▲ 2:2		
	Octochaetona beatrix			Y 1:1
	Octochaetona albida		Y	
	Mallehulla indica		■	
	Karmiella karnatakensis		■	
Octochaetidae	Howascolex stephensoni		Y	
	Hoplochaetella suctorica	■		
	Hoplochaetella kempii	● Y 2:7 1:0		
	Hoplochaetella sp.	● Y 1:1	■	
	Dichogaster modigliani			▲ 2:1
	Dichogaster saliens			▲ ● 1:7 1:0
	Dichogaster curgensis			Y
	Dichogaster bolauii	●	Y ■ 10:4	Y 2:1
	Dichogaster affinis			● ■ 4:3
Megascolecidae	Polypheretima elongata		●	● ● ▲ 1:1:7
	Peilonyx excavatus			▲ 3:1
	Metaphire houletii		▲ ■ 2:2	● ▲ 1:15
	Megascolex lawsoni		■	● Y 3:5
	Megascolex konkanensis	■ ▲ 7:12	● ▲ Y 2:10:7	
	Megascolex insignis	● ▲ ■ 3:3:0		
	Megascolex filliciseta			Y
	Lampito mauritii	● Y 2:10	■ ● 2:10	● ● ▲ 1:2:10
	Drawida sp. (nov.)	●		
	Drawida sulcata	● Y 2:10		
	Drawida scandens			▲ ● 1:4
	Drawida pelludida pallida		●	
	Drawida pellucida		■ ● 2:14	
	Drawida paradoxa	Y ● 2:7	Y	●
	Drawida mysorensis			
Moniligastridae	Drawida modesta			■
	Drawida lennora			▲ ● 2:2
	Drawida kanarensis		■ ● 10:2	▲ ● 1:2
	Drawida ghatensis		Y	
	Drawida ferina	Y ● 1:10	■ ● 1:10	
	Drawida fakira			●
	Drawida calebi		Y	●
	Drawida barwelli impertusa			● ▲ 2:5
	Drawida ampullacea	●		▲

<b>Scale</b>		<b>Agro-ecosystems</b>
□ < 100 worms/100 sq.m	□□□□□ 3000-5000 worms/sq.m	● Arable land
□□ 100-500 worms/100sq.m	□□□□□ > 5000 worms/100sq.m	▲ Fallow land
□□□ 500-1000 worms/100sq.m		■ Orchards/Plantations
□□□□ 1000-3000 worms/sq.m		Y Forests

**Species Diversity and their Role**

In subtropical and tropical regions, there is wider size variation in species rather than species richness. Species richness is more in stable and predictable environments. Unstable and unpredictable environments accommodate more robust communities and as such, the conditions lead to species poverty. This kind of species association could be observed with respect to earthworm fauna of Karnataka - a part of southern Deccan plateau (Table 1). As moisture and available organic matter are important limiting factors for their distribution, species diversity is more in heavy rainfall areas with large plantations and forests than in the plains with dry farm lands which experience unpredictable and poor rainfall. Earthworm species in heavy rainfall areas are larger in size than those found in the plains. *Pontoscolex corethrurus* and *Lampito mauritii* find wider choice of habitats because of their adaptability to various niches (Kale and Krishnamoorthy, 1978; Bano and Kale, 1988 - in press). Moreover, in a subtropical country like ours, the worm activity is limited to a short period, and not much importance has been laid on their role as components of a soil ecosystem. But the scanty and scattered sources of information show how

**Table 2 Cellulose and Lignin utilization in Earthworm worked soils**

Sample Type		Metabolic ratio measured in terms of H <sub>2</sub> SO <sub>4</sub> utilized	Total microbial population x 10 <sup>7</sup> /g	Cellulolytic organisms x 10 <sup>4</sup> /g	Lignolytic organisms x 10 <sup>3</sup> /g	Residual cellulose mg/g	Residual lignin mg/400 ml
Control soil	(A)	18.68 <sup>d</sup>	4.60 <sup>b</sup>	1.96 <sup>b</sup>	1.40 <sup>a</sup>	465.00 <sup>e</sup>	71.15 <sup>e</sup>
without worms	(S)	14.35 <sup>c</sup>	3.15 <sup>a</sup>	1.15 <sup>a</sup>	1.90 <sup>b</sup>	461.50 <sup>e</sup>	70.75 <sup>e</sup>
Soil worked by <i>E. eugeniae</i>	(A)	3.21 <sup>b</sup>	6.68 <sup>d</sup>	4.00 <sup>d</sup>	2.73 <sup>c</sup>	110.00 <sup>d</sup>	36.42 <sup>d</sup>
	(S)	1.68 <sup>a</sup>	7.68 <sup>e</sup>	5.00 <sup>e</sup>	1.72 <sup>b</sup>	56.00 <sup>a</sup>	20.51 <sup>a</sup>
Soil worked by <i>Perionyx excavatus</i>	(A)	3.64 <sup>b</sup>	4.30 <sup>b</sup>	3.13 <sup>c</sup>	2.83 <sup>c</sup>	97.50 <sup>c</sup>	31.75 <sup>b</sup>
	(S)	1.96 <sup>a</sup>	5.90 <sup>c</sup>	4.23 <sup>c</sup>	2.03 <sup>b</sup>	68.25 <sup>b</sup>	17.39 <sup>a</sup>
'F' Test		**	**	**	**	**	**
LSD at P = 0.01		0.63	0.66	0.45	0.36	0.11	3.32

\*\* In Each column means with same alphabets do not differ significantly at P = 0.01  
 A = Agricultural waste, S = Sugarcane trash

much these worms help in annual soil turn over in tropics by their selective feeding and casting activity even during restricted periods. Dash (1978) has reported that earthworms assimilate at least 13% of their net annual primary production in our grasslands and this in fact is the contributory

factor in breaking down of fallen litter and enhancing of microbial activity in ingested soil and litter. Improved water holding capacity of cast and drainage due to their movement in soil strata form a positive factor for plant growth (Dash, 1978). The intestinal mucus excreted by earthworms may add upto 20% of total organic matter content in a Mexican tropical pasture (Lavelle *et al.*, 1983). The quality of the soil organic matter acts as a regulating factor of mechanical activity of earthworms. The nature of soil organic matter, particularly the soluble fraction, determines the intensity and nature of microbial activity in their guts. This factor is of importance for recycling organic and mineral matter in the ecosystem wherein earthworms ingest 1200 tonnes dry soil/ha/yr (Lavelle, 1978). Similar dependence of microflora and earthworms on the nature of available organic matter was reported by Bhat (1974), where in his cultures establishment of *Azotobacter* colonies varied with the organic feed mix used in the medium. Favourable physical conditions due to worm activity and addition of energy rich assimilable organic matter provide scope for microorganisms to increase their activity and digest more complex soil organic matter, a situation also observed by Kale *et al.* (1988-in press) under laboratory conditions for cellulose and lignin digestion in given period of time by known biomass of worms (Table 2).

### Cast : Production and Physico-chemical Properties

The cast production by worms during short periods of favourable conditions has shown that sufficient soil turn over takes place due to worm activity. Worm casts are produced in large quantities on soil surface during rainy season (Gates, 1961; Dash and Patra, 1979). The cast production of *Pheretima hupiensis* during active phase averaged 3.8 kg/m<sup>2</sup> (Watanabe, 1975). A maximum 31 tonnes/dry weight/acre/year of cast production was reported by Dash and Patra (1979). In Thailand, the annual cast production in grasslands was estimated to be 13.26 kg/m<sup>2</sup> to 22.48 kg/m<sup>2</sup> or 132.6 to 224.9 tonnes/ha from May to late November (Watanabe and Ruaysoongnern, 1984).

The size and shape of the released cast vary with species but this cannot be taken as criteria for identifying the worms. *Lampito mauritii* deposits granular cast of moderate size on soil surface whereas the cast released by *Pontoscolex corethrurus* and *Polypheretima elongata* are thick and sticky mounds. Pellet-like and thread-like casts are produced by *Metaphire postuma* and *Perionyx millardi* respectively. The biggest recorded casting of *Notoscolex birmanicus* from Burma weighed 1.6 kg after drying for 4 months. The largest cast of *Pheretima* species in northern Thailand was reported to be 35 cm in height, 5 cm in diameter and weighed 975 g (Watanabe and Ruaysoongnern, 1984). Surface casting *Perionyx excavatus* and *Eudrilus eugeniae* release fine and loose granular casts on soil surface.

A higher level of carbon, nitrogen, magnesium and calcium observed in cast than in surrounding soil was attributed to selective feeding of worms (Watanabe, 1975; Kale and Krishnamoorthy, 1978). From the observations of Nijhawan and Kanwar (1952), it could be deduced that physical characteristics and chemical properties of worm casts vary with different species but normally have better percolation and dispersion coefficient than the parent soil. Earthworms utilise organic matter as food and release part of carbon as CO<sub>2</sub> during the metabolic process. The production of mucus and nitrogenous excrements enhance the level of nitrogen which helps in bringing down the ratio of carbon to nitrogen. This process is most essential in the humification process. Under laboratory conditions nearly 31% reduction in the C/N ratio was observed in cultures of *Octochaetona surensis* in a period of 25 days (Senapati *et al.*, 1980). An integrated study on mechanisms involved in the process of habit selection, feeding, cast production and physico-chemical properties of soils on inhabitation of earthworms are required to be assessed and categorise different groups of tropical worms. By encouraging organic farming and amelioration by introducing hardy and more adaptable species in various agro-ecosystems, it is

feasible to develop a better biocenosis. This, in long run, would build up a more stable and healthy soil.

### **Effect of Biocides**

Worms in general are highly resistant to many pesticides. Senapati and his team of workers are involved in studies on the effect of various biocides commonly used in this country for sanitation purpose and also in fields on the life and activity of earthworms. The question rests on the adoption of a standard method to assess toxicity of any biocide. It is very difficult to use worms as standard laboratory test material because of their capabilities for physiological, behavioural and biochemical adaptations.

Even in some of preliminary studies carried out on the effect of the carbamate Sevin on *Pontoscolex corethrurus*, it has been shown that it could overcome the effect by restricting its movements, increasing mucus secretion level of the pesticide (Kale and Krishnamoorthy, 1979). Earthworms are also known to concentrate pesticides and heavy metals in their tissues (Edwards and Lofty, 1977) and accumulation of pesticides is considered to vary with soil properties. With little knowledge, a few of the agricultural scientists think of controlling earthworm population in some arable lands since they believe them to be responsible for the decline in the crop yield either directly or indirectly. Hence, a careful thinking is very much needed on the use of chemicals. On the one hand, normal admissible dosages of these chemicals have little effect on them and on the other hand, the weak and senile worms affected by these chemicals may help in establishment of pathogenic groups (Dash *et al.*, 1979 a,b). Studies are still to be made under tropical conditions to establish the role of earthworms in detoxifying the soil.

### **Effect on Plant Growth**

The improved growth in pastures and crops like rye and barley has been attributed to the richness of earthworm fauna and linked with their chemical exudates and associated microbes. Beneficial influence of worm cast has been related to biological factors like gibberellins, cytokinins and auxins released due to metabolic activity of microbes harboured in the cast. The presence of compounds allied to IAA in earthworm tissues have been observed. Chemical exudates of worms and those of microbes in the cast influence rooting of layers or shoots. Antibacterial activity of coelomic fluid of earthworms is directed against highly pathogenic soil bacteria. These pathogenic bacterial strains were found to possess at least one common antigen with the sheep red blood cells. It has been demonstrated that the inhibitory effect of coelomic fluid on bacterial strains which have a common surface antigen with sheep red blood cells. Earthworms, apart from encouraging the establishment of beneficial microorganisms, can also inhibit soil borne pathogens.

### **Species for Biotechnology**

Since emphasis is being laid on organic farming and use of earthworm for conversion of organic waste into organic manure, a question is often raised on the use of suitable species for waste degradation. From available information and our experiments with different species of worms, it is observed that some selected species are preferred for this technology. The endogéic worms feeding on soil rich in humic substances fail to live for long under seminatural conditions and with a higher gut transit time are poor feeders and have little preference for rich organic matter with high nitrogen content. Epigéic species are fast breeders and are active feeders on rich organic matter which is high in nitrogen level. But they may avoid animal waste and vice versa. Depending on the type of organic matter to be utilized for the purpose, appropriate species should

be selected. *Eisenia fetida*, *Eudrilus eugeniae*, *Perionyx excavatus* and *Lumbricus rubellus* are used all over the world for waste degradation, and are found to be very successful functionaries for manure and biomass production. In this country also, many of the entrepreneurs have collected the know-how of technology from within and outside the country, and are involved in culturing worms like *Eisenia fetida* for compost production. At the University of Agricultural Sciences, Bangalore, *Eudrilus eugeniae* has been maintained as culture for production of worm biomass and vermicompost. Compost application studies have been in progress in several wet and dry lands. Both these species are exotic and it would be pertinent to develop indigenous technology based on them.

It is a usual practice to introduce some exotic species of both plants and animals for commercial culture all over the world. Among earthworms, *Perionyx excavatus*, from this country has been carried for culture in United Kingdom and Germany, and from the latter to South Africa especially to study its efficiency in waste degradation. Earthworms from time immemorial might have been entering this subcontinent, from different parts of Eurasia. But today we fail to observe all the species that are found distributed in these regions in the southern peninsular India which formed the gateway for many of the species. Most of the species have failed to establish themselves. This was proved in the survey conducted for one year in the farm yard garden in Bangalore where different plants brought from within and outside the country has only three earthworm species namely *Polypheretima elongata*, *Lampito mauritii* and *Pontoscolex corethrurus*. (Kale and Krishnamoorthy, 1982). Even when intentional introduction of worms into open fields and pastures was tried in New Zealand, they take several decades to establish. Hence, the worms that are being deployed for waste degradation can be cultured without any fear or favour and without any adverse effect which has been proved beyond any doubt in this laboratory.

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## **Earthworm Resources of India and Their Utilization in Vermiculture**

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Oligochaetes a group of the phylum Annelida, dwell in all types of aquatic and terrestrial niches with sufficient moisture and food. On the basis of size and habitat, they are often distinguished into two convenient groups : Microdrili (small, mainly aquatic worms including the terrestrial family Enchytraeidae) and Megadrili (larger, mostly terrestrial worms and their aquatic representatives). It is the latter group that consists of earthworms and corresponds to the order Moniligastrida and Haplotaxida including suborder Tubificina.

Earthworms are familiar to almost everyone. They are one of the most popular forms of live bait for fishing all over world including various parts in the Indian subcontinent. Farmers consider them as their friends and hold them in high esteem as nature's ploughmen (Darwin, 1881). Some species play a significant role in organic matter decomposition and mineral cycling (Edwards and Lofty, 1977) which is an important criterion for selecting these organisms in waste utilization and vermicomposting. A global problem for the disposal of wastes has arisen due to astronomical growth in human population and increased industrial and agricultural activities. Earthworms with their peculiar food and feeding, and burrowing habits are nature's most useful converters of wastes. A great deal of work has been carried out on commercial earthworm farming in other countries (Shields, 1971), primarily for producing worms as bait for fishing. But these worms could also be used profitably for recycling biodegradable wastes by vermicomposting techniques for augmenting not only an alternate source of organic fertilizers but also for providing cheap animal protein in the fish and poultry feed.

More than 4200 species of oligochaetes are known in the world. Of these, 280 are Microdrili and remaining about 3200 belong to Megadrili (earthworms). In the Indian subcontinent, earthworms also form bulk of the oligochaete fauna. They are represented by 509 species and 67 genera, indicating a high degree of diversity in this region as compared to other areas. Though majority of the forms have specific habitat preference, a few ubiquitous species also occur. It is, therefore, important to critically assess existing diversity in Indian earthworms and their ecological requirements for evolving suitable vermicomposting techniques.

### **Earthworm Diversity**

The first records of earthworms in the Indian subcontinent were provided by Robert Templeton in 1844, when he discovered a new species of *Megascolex* (*M. caeruleus*) from Sri Lanka. Subsequently, several species have been discovered from the subcontinent by various workers notably Rosa (1894), Michaelsen (1909), Stephenson (1914, 1920, 1921), Gates (1929, 1930, 1931, 1932, 1933, 1945) and Julka (1976a, 1976b, 1978, 1981). The collective studies of Indian oligochaete taxonomists have resulted in the publication of three well-documented taxonomic monographs on these organisms : Fauna of British India and adjacent countries on Oligochaeta by Stephenson (1923), Burmese earthworms by Gates (1972) and Fauna of India or megadrile Oligochaeta (earthworms), Family Octochaetidae by Julka (1988). These monographs are of great utility for the identification of majority of earthworm species of our region.

Table 1. Earthworm genera and species (no.) in the Indian subcontinent (including Andaman & Nicobar Isls.) Genera marked with an asterisk (\*) are peregrine in the region

Family/Genus	No. of spp.	Family/Genus	No. of spp.
Acanthodrilidae		Ocnerodrilidae	
<i>Microscolex</i> *	1	<i>Curgiona</i>	1
<i>Plutellus</i>	32	<i>Deccania</i>	1
<i>Pontodrilus</i> *	1	<i>Eukerria</i> *	1
Almidae		<i>Gordiodrilus</i> *	2
<i>Glyphidrilus</i>	4	<i>Malabaria</i>	4
Criodrilidae		<i>Nematogenia</i> *	1
<i>Criodrilus</i> *	1	<i>Ocnerodrilus</i> *	1
Eudrilidae		<i>Thatonia</i>	5
<i>Eudrilus</i> *	1	Octochaetidae	
Glossoscolecidae		<i>Bahlia</i>	1
<i>Pontoscolex</i> *	1	<i>Barogaster</i>	3
Lumbricidae		<i>Calebiella</i>	1
<i>Aporrectodea</i> *	3	<i>Celeriella</i>	7
<i>Bimastos</i> *	1	<i>Chaetocotoides</i>	1
<i>Dendrobaena</i> *	1	<i>Dashiella</i>	1
<i>Dendrodrilus</i> *	1	<i>Dichogaster</i> *	5
<i>Eisenia</i> *	3	<i>Eudichogaster</i>	6
<i>Eiseniella</i> *	1	<i>Eutyphoeus</i>	45
<i>Lumbricus</i> *	4	<i>Hoplochaetella</i>	18
<i>Octolasion</i> *	2	<i>Karmiella</i>	1
Megascolecidae		<i>Konkadrilus</i>	4
<i>Amyntas</i>	33	<i>Kotegeharia</i>	1
<i>Comarodrilus</i>	1	<i>Lenogaster</i>	6
<i>Kanchuria</i>	4	<i>Mallehulla</i>	1
<i>Lampito</i>	8	<i>Octochaetoides</i>	1
<i>Lenoscolex</i>	2	<i>Octochaetona</i>	15
<i>Megascolex</i>	33	<i>Octonochaeta</i>	1
<i>Metaphire</i>	26	<i>Pellogaster</i> .	3
<i>Nelloscolex</i>	2	<i>Priodochaeta</i>	1
<i>Notoscolex</i>	11	<i>Priodoscolex</i>	1
<i>Perionyx</i>	53	<i>Ramiella</i>	6
<i>Pithemera</i> *	1	<i>Rillogaster</i>	2
<i>Polypheretima</i> *	2	<i>Scolioscolides</i>	1
<i>Tonoscolex</i>	16	<i>Travoscolides</i>	4
<i>Troyia</i>	1	<i>Wahoscolex</i>	9
Moniligastridae			
<i>Desmogaster</i>	8		
<i>Drawida</i>	79	Total	509
<i>Hastirogaster</i>	2		
<i>Moniligaster</i>	9		

At present, earthworm fauna (megadrile) in the Indian subcontinent comprises 509 species placed in 67 genera and 10 families (Table 1). Majority of them are endemic and belong to 47 genera. *Amyntas* and *Metaphire* have endemicity in Burma and Andaman and Nicobar Islands, but they alongwith other pheretimoids like *Pithemera* and *Polypheretima* are peregrine in the Indian and Sri Lankan regions. Fauna of the Andaman and Nicobar Islands is more closely related to that of Burma and Malayasia than to the Indian mainland. About 68% of known species of earthworms in the subcontinent belong to ten endemic genera with the break up as *Drawida* (79), *Perionyx* (53), *Eutyphoeus* (43), *Megascolex* (33), *Amyntas* (33), *Plutellus* (32), *Metaphire* (26), *Hoplochaetella* (18), *Tonoscolex* (16) and *Octochaetona* (15). The remaining 57 genera are either monospecific or represented by less than 10 species. Excluding a few widely distributed species, thirty eight of the endemic genera are only found in this region (Table 2). The rest of seven genera have distribution in other parts of the world (species of *Plutellus* and *Megascolex* from Australian region possibly are not congeneric with Indian species).

A few peregrine forms have also been introduced presumably in soil around roots of exotic plants. The peregrine genera are distributed among 8 families : Lumbricidae (8), Ocnerodrilidae (4), Megascolecidae (2), Acanthodrilidae (2), Eudrilidae (1), Glossoscolecidae (1), Criodrilidae (1) and Octochaetidae (1). Successful colonization of peregrine species is mainly due to their tolerance to a wide range of ecological conditions and to some extent parthenogenetic mode of reproduction in most of them. The extent of colonization of lumbricids has become so extensive in the Western Himalayas that they now dominate over the endemic species at several places.

### Selection of Suitable Species in Vermicomposting

Though faunal resources of Indian earthworms are very rich, it is very important to select most suitable species for vermicomposting. It is well established that different species of earthworms have different life styles which were formally classified into three categories by Bouché (1977).

1. **Épigés** : Litter or dung dwellers; small in body size; tolerant to disturbance; high rate of cocoon production; short life cycle; uniform colouration.
2. **Endogés** : Dwellers of top organo-mineral soil and construct horizontal and branching burrows; tolerant to some disturbance; moderate to high rate of cocoon production; life cycle intermediate; small to large in body size; weakly pigmented.
3. **Anéciques** : Deep burrowers that construct vertical burrows, cast at surface and emerge from burrows at night to draw down organic material; intolerant to disturbance; low rate of cocoon production; long life cycle, large in body size; slightly pigmented at anterior and posterior ends.

The most suitable species in vermicomposting which can be economically cultivated on a large scale should have following characteristics : ability to inhabit and feed upon high percentage of organic matter, tolerance to disturbance and fluctuations in environmental parameters, high rate of cocoon production and short duration of life cycle. The ecologically classified species as épigé or endogé possess many of the features associated with this type of selection. In our region, some peregrine as well as endemic species with these ecological characteristics are available for utilization in vermicomposting. These are :

1. Family Lumbricidae : *Bimastos parvus*, *Dendrobaena rubida*, *Eisenia fetida*, *Eisenia hortensis*.
2. Family Eudrilidae : *Eudrilus euginiae*.

3. Family Megascolecidae : *Amyntas diffringens, Lampito mauritii, Metaphire anomala, Metaphire birmanica, Perionyx excavatus, Perionyx sansibaricus.*
4. Family Octochaetidae : *Dichogaster bolau, Dichogaster saliens, Ramiella bishambari, Hoplochaetella khandalaensis, Hoplochaetella suctoria*
5. Family Ocnerodrilidae : *Ocnerodrilus occidentalis*
6. Family Moniligastridae : *Drawida willsi, Moniligaster perrieri*

Table 2 Endemic earthworm genera in the Indian subcontinent

Restricted to Indian subcontinent (excluding widely distributed sp.)	Extra – Indian subcontinental distribution
	Acanthodrilidae
	<i>Plutellus</i>
	Almidae
	<i>Glyphidrilus</i>
	Megascolecidae
<i>Comarodrilus, Kanchuria, Lampito, Lennoscolex, Nelloscolex, Perionyx, Tonoscolex, Troyia</i>	<i>Amyntas Megascolex Metaphire Notoscolex</i>
	Moniligastridae
<i>Moniligaster</i>	<i>Desmogaster Drawida Hastirogaster</i>
	Ocnerodrilidae
<i>Curgiona, Deccania, Malabaria, Thatonia,</i>	
	Octochaetidae
<i>Behlia, Barogaster, Calebiella, Celeriella, Chaetocotoides, Dashiella, Eudichogaster, Eutyphoeus, Hoplochaetella, Karmiella, Konkadrilus, Kotegeharia, Lotegeharia, Lennogaster, Mallehulla, Octochaetoides, Octochaetona, Octonochaeta, Pellogaster, Priodochaeta, Priodoscolex, Ramiella, Rillogaster, Scolioscolides, Travoscolides, Wahoscolex</i>	

## Conclusions

Our subcontinent is rich in earthworm resources in having both peregrine and endemic species for developing vermicomposting as a potential industry. Some of the peregrine species, which have been successfully utilized for this purpose in other countries, are also present. These are : *Eisenia fetida* (an European temperate species) and *Eudrilus eugeniae* (an African tropical species). The agricultural scientists at University of Agricultural Sciences, Bangalore have developed adequate know-how on vermicomposting under Indian conditions by utilizing *Eudrilus eugeniae*. They have also released a vermicompost (Vee comp E.83 UAS) for field trials in south India with tropical climate. Potentiality of *Eisenia fetida* in this regard, especially in the mountainous regions with temperate like climate, is yet to be explored. An endemic compost worm, *Perionyx excavatus*, with a wide range of distribution in the subcontinent, is an excellent waste decomposer.

Its potentiality in developing indigenous vermiculture for waste control and protein production could be exploited under natural conditions. As compared to other compost dwelling earthworms such as *Eisenia fetida* and *Eudrilus eugeniae*, it has fast maturation rate and produce cocoons at much earlier stage (Hallet *et al.*, 1990). Though this species has a lower mean growth rate and produces fewer cocoons than *Eudrilus eugeniae* and *Eisenia fetida*, it has much shorter life cycle and ability to reproduce without copulation (parthenogenesis) which could be a great advantage under certain circumstances. Being a tropical and endemic species, *Perionyx excavatus* possesses enough potentiality for its use in vermicomposting in our country.

Ecological and biological studies in several litter dwelling endemic species are lacking. It is, therefore, very important to undertake extensive studies on these aspects before they are exploited for waste disposal. Experiments should be conducted to develop appropriate methods for their culture in different types of organic matter under different climatic conditions.

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## **Population, Biomass and Secondary Production in Earthworms**

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### **Synopsis**

Organism interacts and establishes itself in the ecosystem through population. Population dynamics, biomass and production studies on Indian earthworms has been scanned. Methodology, progress, prospects and challenges on the topic have been dealt. Detail comparison has been made among three peregrine Indian earthworms; *Dichogaster bolau* (Michaelson); *Drawida willsi* Michaelson and *Polypheretima elongata* Perrier, belonging to three major ecological category on the basis of their biological strategy. Out of 374 earthworms reported till now hardly some information is available on about 40 species. Knowledge on biodiversity and biological strategy is important for academic and economic application of the species. Predictable models should be developed for utilisation by vermiculture based industries and for their management in field. There is immense prospective of this approach among with problems in methodology simplification and research objectives relating to worm activity in wide spectrum of habitat, climate species, waste biomass and human impact on them.

### **Understanding the Organism through Population, Biomass and Production Studies**

Population is the assemblage of individuals belonging to a particular species. Population biology provides the first basic information about any species and is regulated both by abiotic and biotic factors. Assessment of relative importance of a population in an ecosystem may be possible if the information on population density, dynamics, distribution, growth and metabolic rate of different age classes are available. Individual organism acquires essential resources from its immediate environment and process through its metabolic activities utilising stored energy. Assimilated energy is allocated to growth, repair, defence and reproduction. The organism interact and established through the population. Thus the population remains dynamic with constant input. Resource acquisition and allocation at the organismal level is given in Fig. 1. Population dynamics and biomass production studies indicate : (i) dynamics of the number, tissue content and their distribution over time and space, (ii) population growth and distribution of different age classes (iii) impact of different environmental factors (biotic and abiotic), (iv) differential allocation of matter and energy into growth and reproduction compartments and (v) mortality and natality.

### **Population, Biomass and Secondary Production Studies on Earthworm**

Population biology of earthworm has drawn attention since the time of Darwin (1881).

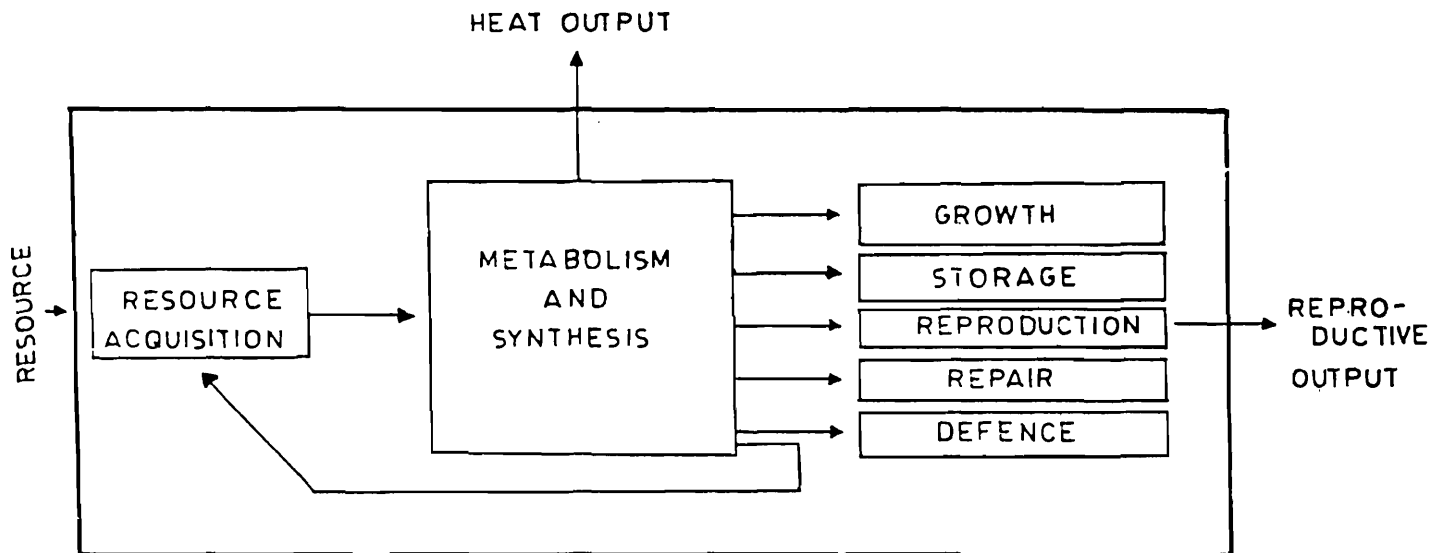


Fig. 1. Resource acquisition and allocation at organismal level.

Quantitative figures are available from the work of Morris (1922), Eaton (1942), Evans and Guild (1947 a, b & c) and onwards. Several research publications including review papers, books and monographs have been published by the European workers. Some of them are by Satchell (1967), Bouche (1972), Edwards and Lofty (1972), Atlavinyte (1975) and Lavele (1978). The latest review on the biology of earthworm is made by Lee (1985). In India, Ali *et al.* (1973), Dash *et al.* (1974), Dash and Patra (1977) are the pioneer to indicate detail study of earthworm dynamics in grassland ecosystem followed by a series of papers from Life Sciences Centre of Sambalpur University. Before 1977 scattered and mostly qualitative information are available from the works of Stephenson (1923), Bahl (1926), Roy (1957), Tembe and Dubash (1961) and Gates (1972). Several papers have now been published dealing with seasonal activity, population density and biomass of earthworms in many Indian ecosystems (Kale and Krishnamoorthy, 1978 a & b; Reddy and Alfred, 1978; Senapati *et al.* 1979 a & b; Chauhan, 1980; Dash and Senapati, 1980; Senapati and Dash, 1981; Ismail and Murthy 1985; Sahu and Senapati, 1986; Pani, 1987; Julka and Senapati, 1987; Krishnamoorthy & Ramachandra, 1988; Sahu *et al.* 1988; Bhaduria and Ramakrishnan, 1989; Ismail *et al.* 1990; Dash and Senapati 1991). However very scanty information is available on secondary productivity of earthworms from different world ecosystems including India (Lee, 1985).

### Population Dynamics, Distribution and Environmental Regulation

Dash and Patra (1972, 1977), Ali *et al.* (1973), Senapati *et al.* (1979), Dash and Senapati (1980) Senapati and Dash (1980, 1981, 1983, 1984), Mukherjee and Singh (1986) working on ecophysiological studies have reported methods of earthworm extraction, sampling, preservation and analysis suitable for Indian workers. They have certified a handsorting method combined with wet sieving with a sample size of 25 cm x 25 cm x 40 cm depth in 10 replicates for each sampling occasion at each site. Although this method is very much time consuming and expensive, but it is most efficient in comparison to other methods. TSBF (Tropical Soil Biology and Fertility

Programme) methodology recommends a minimum of five and preferably 10 samples of 25 cm x 25 cm by 30 cm deep where the macroinvertebrates (body length greater than 2 mm) are to be handsorted (Anderson and Ingram, 1987). TSBF certified method holds good in soil where the clay content is much less. It is also difficult to handsort juvenile and cocoons of small earthworms (like *Dichogaster bolau*). During the field work different types of materials required may be classified into seven groups : (i) measuring materials (scale, tape, quadrat, soil thermometer, weighing balance, altimeter, hygrometer, moisture meter, soil field kit for chemical analysis etc.), (ii) digging materials (flat digger, digging rod, narrow digger etc.), (iii) sample carrying materials (plastic packets, plastic bottles, rubber bands, field trays, sample carrying bags etc.), (iv) preserving materials (formalin 5%, ethyl alcohol 95%), (v) sieving and collecting materials (sieve 500 @ pore size, 2 mm pore size, white enamel tray, plastic bowl, white rexin sheet 2 m x 2 m, forceps small and large), (vi) labelling and recording materials (paper labels, field note book) and (vii) precautionary materials (first-aid and emergency medicines). Before starting the field work, proper care must be taken for experimental design and selection of the site. General characterisation of the study site could be analysed by using the standard methodology described in TSBF handbook of methods (Anderson and Ingram, 1987). Identification of different flora and fauna should be made as per the requirement of the experimental design. Although generally it is certified to go for 5 to 10 replicates and in 25 x 25 x 40 cm area, the number of replicates and sample area should be checked by species - area curve method (Mishra, 1968). This shows progressive stabilization of the mean number of species along with increase in area (to determine sample area) or with increasing number of replicates (to determine sample number). The replicates should be located at about 5 meter regular intervals along a line whose starting point and direction should be chosen randomly. Microhabitats such as foot of trees, surrounding of large bark, decaying logs and similar such situation should be avoided, which may justify a separate study (Lasebikan, 1974). In each sample, first sample area should be demarcated with a quadrat. Vegetation, litter and soil has to be removed outside the boundary of sampling area by digging a trench of about 20 cm width, around the example so as to keep the sampling area intact and undisturbed. This facilitates cutting of the sample into horizontal state and collecting the soil animals escaping from the block. All invertebrates larger than 10 cm length excavated from the trench should be collected. These are mainly large millipedes and earthworms. The sample block is then divided into layers of 0-10 cm, 11-20 cm, 21-30 cm and 31-40 cm. This will enable to have an idea about the vertical distribution or stratification of soil organisms alongwith physicochemical parameter. The soil could be deposited on the left side of the tray and progressively moved towards right side over the whole surface while handsorting soil animals, litters, roots etc. This enhances the handsorting processes. After removing large animals the soil has to be mixed, weighed and divided into subsamples. Subsamples could further be utilised for : (i) wet sieving method to assess cocoons and small juveniles, (ii) to determine moisture content and other parameters like soil respiration, microbial analysis, enzyme analysis etc. (iii) to air dry and to store for physicochemical analysis. Soil has to be sieved through 2 mm sieve before analysis. Soil animals could be preserved in 5% formaldehyde keeping earthworms separate from other groups for detailed study. At each stage of sampling and subsampling there must be proper labelling of site, date, sample and subsample number, depth and name of the workers. Climatic data of each sampling occasion of soil temperature has to be recorded for future use. It is not always possible to carry live earthworms sampled from the field for quantitative study because of rapid decomposition of damaged worms. So it is suggested to preserve the worms in 5% formalin. The loss of weight due to formalin preservation could be calculated as per the method suggested by Senapati and Dash (1980). Qualitative and quantitative analysis of soil animals including earthworms could be done in the laboratory. Earthworms and other soil organisms should be collected separately for taxonomic purpose. For this, healthy, intact adult

worms should be washed in water. Then the worms are to be narcotised in a tray with small amount of water, to which ethyl alcohol (95%) could be added drop wise and slowly shaken till all the worms are motionless. Narcotised worms should be kept straight on 5% formalin drenched blott up paper. Dissection of straightened worm is much easier than coiled one (Julka per. cum.). After 24 hours, formalin preserved worms could be transferred to 70% alcohol, labelled and stored for taxonomic purpose. However in field condition where narcotisation and straightening are not possible, healthy adult worms are simply to be dropped into 5% formalin. In addition to this taxonomic classification, it is also important to divide them into functional categories like, epigeics, anecics and endogeics by analysing their habit (gut content analysis) and habitat (Stratification). Epigeic species live and feed on soil surface and these earthworms are important in the nutrient release but do not actively redistribute plant material. Besides earthworms, arthropods like Myriapods, Isopods also belong to this epigeic category. Anecic species of earthworms which remove litter from the soil surface through their feeding activities but most of the time remain under soil. Endogeic species which live in the soil and feed on organic matter and dead roots, mostly spent their life at lower depth of the soil. Here alongwith the earthworms one could consider humivorous termites (Anderson and Ingram, 1987). Characteristic features of functional categories of earthworms has been dealt in the paper on "selection of suitable earthworm species for vermicomposting under Indian condition" as a part of this publication. Each earthworm has to be identified into the taxonomic unit and to the respective age group (Table 1). Cocoons (live, dead and empty have to be recorded species wise also. Numerical analysis has to be used for population dynamics, mortality and natality etc. Population mortality figure should be quantified by using the formula developed by Nowak (1975).  $E = N_{t1} - (N_{t2} - V_t)$  where  $E$  = population mortality,  $N_t$  = density in fortnight and  $V_t$  = number of newly recruited individuals.

**Table 1. Age group classification in some Indian earthworms**

Worm type	Worm size	Worm age group		
		Non-clitellate		Clitellate
		Juvenile	Immature	Adult
<i>D. bolau</i>	Small	< 1 cm	> 1 < 2 cm	> 2 cm
<i>O. occidentalis</i>	Small medium	< 2 cm	> 2 < 4 cm	> 4 cm
<i>D. willsi</i>	Small medium	< 2 cm	> 2 < 4 cm	> 4 cm
<i>L. mauritii</i>	Large medium	< 4 cm	> 4 < 8 cm	> 8 cm
<i>O. surensis</i>	Large medium	< 4 cm	> 4 < 8 cm	> 8 cm
<i>P. elongata</i>	Large	> 6 cm	> 6 < 14 cm	> 14 cm

There are two main approaches to the species diversity : (i) Simpson's index (Simpson, 1949) and (ii) Shannon-Wiener index (Shannon-Wiener, 1963). Simpson index is calculated by the formula :  $D = N(N-1)/n(n-1)$  where  $D$  = diversity index,  $N$  = total number of individuals of all species,  $n$  = number of individuals of a species. Shannon-Wiener index is calculated by the formula:

$$H' = - \sum (n_i/N) \log_2 (n_i/N),$$

where  $H'$  = diversity of species,  
 $n_i$  = number of individuals in the 'i' the species and

**N** = total number of individuals of all the species,

**S** = total number of species collected.

Index of dominance (**c**) could be calculated as per Simpson (1949) using the formula :

$$c = \sum (n_i/N)^2$$

where  $n_i$  = number of individuals in the 'i' the species and  
**N** = total number of individuals.

Index of species richness or variety of species ( $d_1$ ) could be determined using the formula (Margalef, 1968) :  $d_1 = S-1/\ln N$

where **S** = total number of species,  
**N** = total number of individuals.

Index of evenness (**e**) could be quantified on the basis of the formula developed by Pielou (1966):  $e = H'/\ln S$  where  $H'$  = Shannon-Wiener index, **S** = total number of species, Ecological index (E.I.) has been proposed by Lavelle (1979) which summarises the life style of earthworm species and is defined as the product of maximum body weight (**W**) to the average depth (**p**) at which worms live in soil. This may be expressed as  $E.I. = W.p$ . Average depth of occurrence could be quantified by dividing depth interval in cm between which maximum adult population occurred divided by two.

For further analysis in different components relating to soil organisms the workers are suggested to refer Andrewartha (1961), Phillipson (1971), Lewis and Taylor (1974) and TSBF methodology by Anderson and Ingram (1987). Statistical analysis for calculation of average values with standard error of mean variance, test of significance, simple and multiple correlation and regression and analysis of variance, any standard text book could be referred such as Bailey (1964), Snedecor and Cochran (1967). For analysis of physicochemical parameters in soil standard books like Mishra (1968), Allen *et al.* (1974), Jackson (1973) may be referred.

Morphological variation of different Indian earthworms has been reported by Michaelsen (1909), Stephenson (1923) Gates (1972), Julka (1975, 1981, 1988), Julka and Senapati (1987) and Julka *et al.* (1989). Fauna of British India, Oligochaeta volume by Stephenson (1923) is being revised by Dr. J.M. Julka of Z.S.I.. The morphological characters are not only important on the point of taxonomy but also for ecological analysis. Population dynamics data reported by most of the Indian workers have not been analysed in the light of their ecological function excepting few recent studies (Dash and Patra, 1977; Senapati *et al.* 1979; Senapati and Dash, 1981; Kale and Krishnamoorthy, 1982; Mishra and Dash, 1984; Krishnamoorthy 1985; Sahu and Senapati, 1988, 1991; Krishnamoorthy and Ramachandra, 1988; Sahu *et al.* 1988; Sahu, 1989; Bhaduria and Ramkrishnan, 1989; Ismail *et al.* 1990; Senapati and Sahu, 1991). Looking into the objective of this publication which is largely for research workers who need very much methodology and the prospective of the study, and problems, the authors have chosen to compare population dynamics, biomass and secondary production in three Indian earthworms which are not only peregrine (wide occurrence) but also distinctly fall into the major ecological category on the basis of their habitat stratification (epigeic, anecic and endogeic earthworms by Bouche, 1971, 1977) and selection pressure ('r' - and 'K' - selected worms by Satchell, 1980; Bouche, 1977; Lavelle, 1978). They are *Dichogaster bolau* (Michaelsen), *Drawida willsi* Michaelsen and *Polypheretima elongata* Perrier. However other papers published on Indian earthworms dealing with population dynamics, biomass and production have been referred where ever necessary. Research workers are advised to refer individual papers for detail information with respect to their objective, species and habitat, etc. *D. bolau* is about 19-40 mm in length, 1-3 mm in diameter and is available in the litter zone. *P. elongata* is about 95-300 mm in length, 3-6 mm in diameter and mostly remain below 30 to 40 cm

of soil depth where as *D. willsi* (40-100 mm length, 1.5-3 mm diameter) stratify itself in the litter-soil transition zone with feeding and defecation at the surface. A high significant positive correlation ( $r > 0.8$ ,  $p < 0.001$ ) has been reported between body length and body weight. Other such regression models developed on the basis of morphometry is given in the section dealing with selection of earthworm for vermicomposting in this publication.

Indices of diversity of tropical earthworms indifferent agroecosystems of Orissa have been reported in Table 2. A maximum number of five species occur in natural grassland whereas in man interfered grassland and crop field one to three species has been reported. Index of dominance indicate a reverse trend than that of species diversity ( $H^1$ ) in natural ecosystems. This has been proved in other ecosystems in both plants and animals by Elton (1958), Odum (1971) and Smith (1980). Maximum ecological index (E.I.) of 112.0 has been calculated, for *P. elongata* with a length diameter ratio of about 85, maximum live weight of 4 gm and mean depth of activity at 28 cm. Mean depth of activity of *D. bolau*i and *D. willsi* is 5 cm and 9 cm respectively whereas ecological index came to be 0.2 and 0.2 and 0.4 respectively.

Table 2. Indices of diversity of tropical earthworms in different agroecosystems of Orissa, India

Site	Number of species (S)	Index of dominance (c)	Index of species richness ( $d_1$ )	Index of evenness (e)	Index of species diversity ( $H^1$ )	Reference
Upland grazed pasture	5	0.29	0.78	1.16	1.87	Senapati & Dash (1981)
Upland protected pasture	5	0.33	0.73	1.11	1.79	Senapati & Dash (1981)
Lowland irrigated paddy field	3	0.51	0.47	0.68	0.75	Pani (1978)
Plain protected pasture	3	0.39	0.33	1.10	1.21	Sahu (1989)
Upland irrigated paddy field	3	0.57	0.33	0.55	0.61	Pani (1987)
Upland grazed pasture	1	1.00	0.00	0.00	0.00	Sahu (1989)

Fortnightly dynamics of total earthworm population of *D. bolau*i, *D. willsi* and *P. elongata* in pasture ecosystems of Orissa have been represented in Fig. 2 Dynamics of different age class density of *D. bolau*i, *D. willsi* and *P. elongata* earthworms in pasture ecosystems of Orissa have been given in Table 3. Maximum peak population of 8038 for *D. bolau*i, 245 for *D. willsi* and 231 for *P. elongata* worms/m<sup>2</sup> have been reported during 1984-85 period (Sahu and Senapati, 1986, 1988, 1991; Sahu 1989). Total earthworm density ranged from 0-8038 for *D. bolau*i, 0-245 for *D. willsi* and 49-231 for *P. elongata* during 1984-85 period (Fig. 2 and Tab. 3). Population turnover (maximum density/average density) for *D. bolau*i, *D. willsi* and *P. elongata* were about 5.6, 2.7 and 1.7 respectively. Considering worm size such as length (mm), dry weight (mg) and biovolume (mm<sup>3</sup>) correlation coefficient values between worm size and turnover came to be negatively significant ( $r > -0.7$ ,  $p < 0.2$ ) indicating an inverse relationship. An unimodal peak for *D. bolau*i (during Aug. II week, 1984), bimodal peak for *D. willsi* (during Sept. II week 1984 and Feb. II week, 1985) and a multimodal peak for *P. elongata* (during Sep. IV week 1984, Nov. IV week 1984, Mar. IV week 1985 and July IV week, 1985) exhibit their respective niche segregation (Fig. 2 and Tab. 3). Epigeicity and 'r'-selected strategy is associated with unimodality where as endogeicity and 'K'- selected strategy is associated with multimodality (Bouche, 1971, 1977; Satchell, 1980;

Senapati and Sahu, 1991). Table 4 shows population density (nos/m<sup>2</sup>) and biomass (g live wt/m<sup>2</sup>) of earthworms in different tropical ecosystems of India reported by several workers. Sahu *et al.* (1988) have reported the highest population density of earthworms of 12617/m<sup>2</sup> in dung deposit site whereas maximum live biomass of 122 g/m<sup>2</sup> occurred in plain grassland of Sambalpur (Sahu and Senapati, 1991). However population density and biomass values by most of the Indian workers have been reported on the basis of handsorting method.

Table 3. Dynamics of different age class density (nos/m<sup>2</sup>/fortnight) of *D. bolau*, *D. willsi* and *P. elongata* earthworms in pasture ecosystems of Orissa, India

Month (week)	Year	<i>D. bolau</i>				<i>D. willsi</i>				<i>P. elongata</i>			
		J	I	A	Total worm	J	I	A	Total worm	J	I	A	Total worm
Jul.(IV)	1984	1193	1466	372	3031	—	—	—	—	—	—	—	—
Aug.(II)		1582	5585	871	8038	—	—	—	—	—	—	—	—
Aug.(IV)		1208	2571	644	4423	17	56	74	147	165	23	22	210
Sep.(II)		78	3242	1132	4452	0	63	73	136	149	55	16	220
Sep.(IV)		238	120	676	1034	46	102	97	245	169	40	22	231
Oct.(II)		118	473	474	1065	23	40	95	158	86	45	16	147
Oct.(IV)		0	40	80	120	6	45	85	136	56	74	16	146
Nov.(II)		—	—	—	—	—	—	—	—	—	—	—	—
Nov.(IV)		0	0	39	39	17	45	62	124	114	54	22	190
Dec.(II)		—	—	—	—	17	17	11	45	130	44	16	190
Dec.(IV)		0	39	0	39	0	29	51	80	84	58	16	158
Jan.(II)	1985	—	—	—	—	6	23	56	85	74	29	11	114
Jan.(IV)		0	0	39	39	0	29	50	79	80	23	11	114
Feb.(II)		—	—	—	—	68	62	74	204	34	34	11	79
Feb.(IV)		0	0	39	39	11	34	29	74	40	27	11	78
Mar.(II)		—	—	—	—	6	17	17	40	23	44	11	78
Mar.(IV)		0	0	0	0	0	12	17	29	107	40	16	163
Apr.(II)		—	—	—	—	0	0	12	12	61	34	10	105
Apr.(IV)		0	0	0	0	0	0	0	0	28	27	5	60
May.(II)		—	—	—	—	0	0	0	0	11	28	10	49
May.(IV)		0	0	0	0	0	6	23	29	34	32	10	76
Jun.(II)		0	40	80	120	6	34	17	57	45	38	10	93
Jun.(IV)		118	473	119	710	17	34	34	85	57	56	11	124
Jul.(II)		—	—	—	—	28	22	56	106	80	51	16	147
Jul.(IV)	1985	—	—	—	—	34	29	51	114	76	62	17	155

J = Juvenile I = Immature A = Adult — = Sampling not done

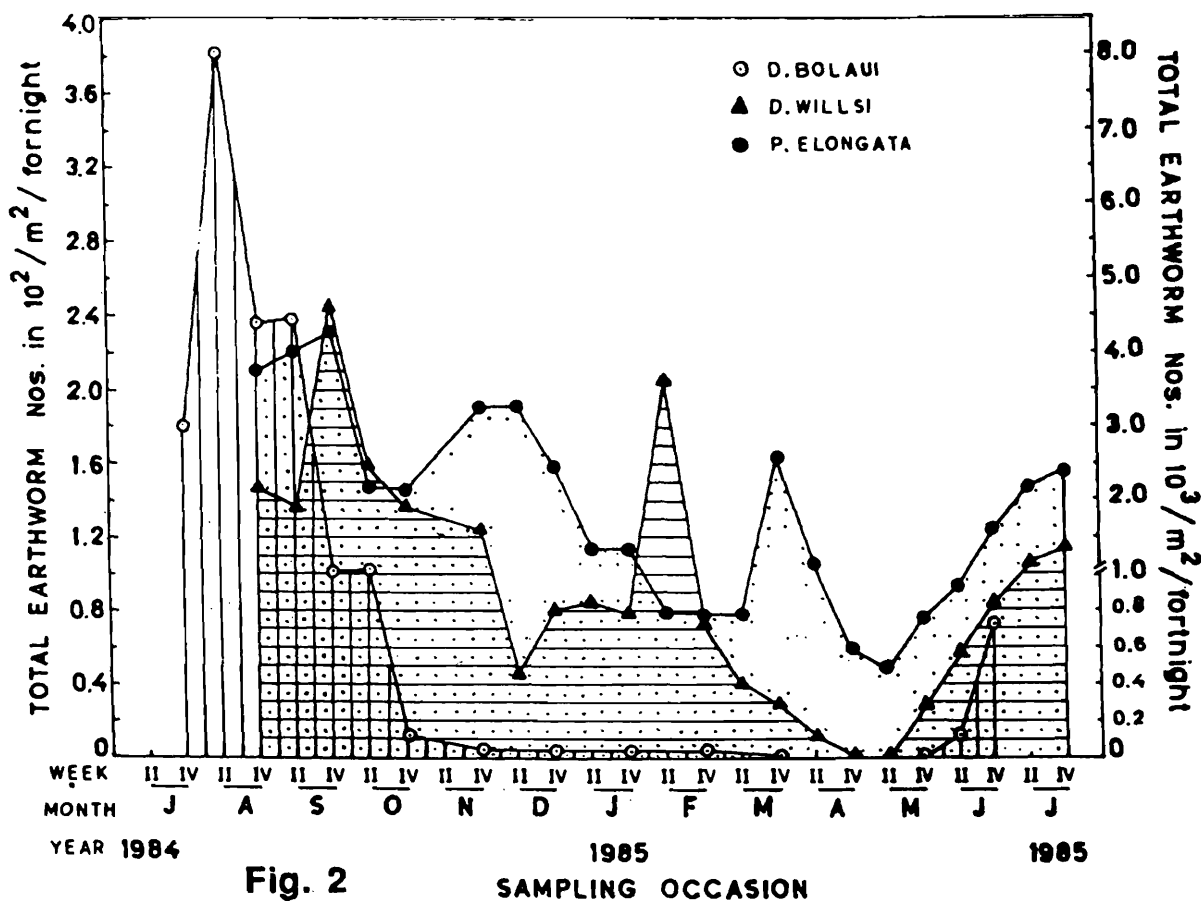


Fig. 2

PERCENTAGE OF OCCURENCE

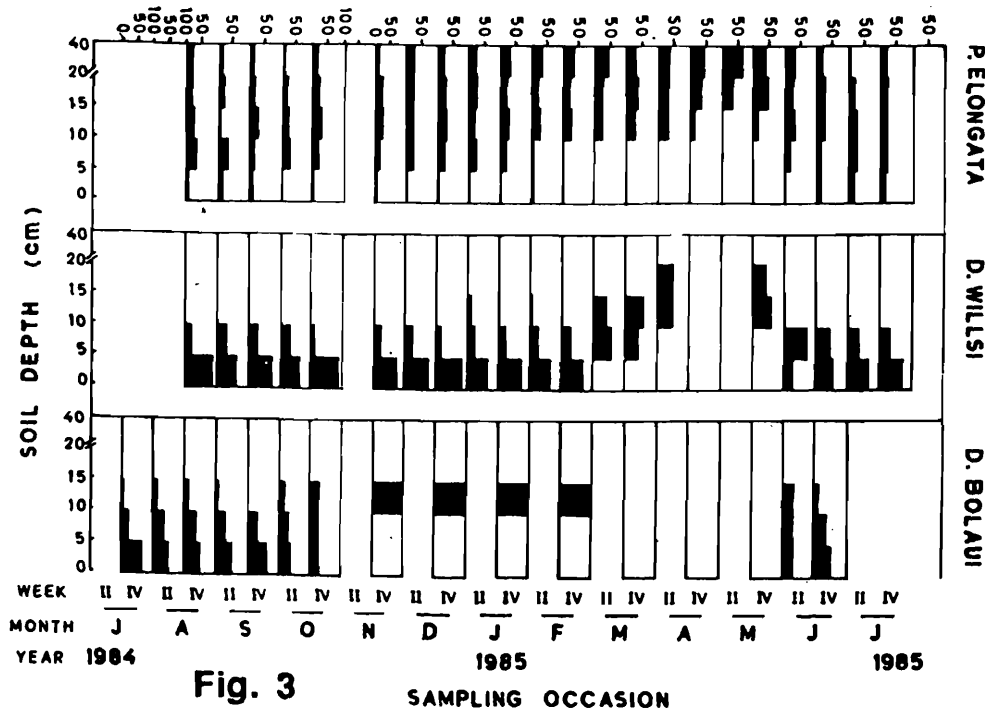


Fig. 3

Fig. 2. Fortinightly dynamics of total earthworm population of *D. bolauai*, *D. willsi* and *P. elongata* in pasture ecosystems of Orissa, India.

Fig. 3. Vertical distribution of *D. bolauai*, *D. willsi* and *P. elongata* earthworm population in pasture ecosystems of Orissa India.

This might be the reason of low density and biomass in comparison to the works reported by Senapati and associates (Table 4). Details of the population and biomass of earthworms in Indian ecosystems are dealt in respective papers. Dash and Patra (1977) in natural grassland, Senapati and Dash (1981) and Sahu *et al.* (1988) in organic waste deposit site, Mishra and Dash (1984) in natural forest ecosystem, Bhaduria and Ramakrishnan (1989) in shifting agriculture system have done pioneering work in the respective ecosystems (Table 4).

Vertical distribution of *D. bolau*, *D. willsi* and *P. elongata* earthworm population in pasture ecosystems of Orissa have been shown in Fig. 3. Vertical distribution is distinct in post rainy and summer than in comparison to rainy season. Studies on horizontal and vertical distribution have also been made by Dash and Patra (1977), Senapati and Dash (1981), Kale and Krishnamoorthy (1982), Krishnamoorthy (1985), Sahu *et al.* (1988) and Ismail *et al.* (1990). Worm body size (dry wt.) showed a significant positive correlation ( $r < 0.89$ ,  $p < 0.02$ ) with average depth of occurrence indicating greater probability of epigeic nature in small sized worm and endogeic nature in large size worm. There has been reports of earthworm migration upto 3 meter depth in search of suitable soil moisture (Edwards and Lofty, 1972). Survival of organism under adverse conditions through natural adaptation is one of the inherent mechanism for its successful continuation. Seasonal fluctuation of climatic and edaphic factor like rainfall, solar radiation, air temperature, soil moisture, evaporation from the soil surface and soil nutrients limit the activity of soil organisms. Too dry and too cold or too warm soil is unfavourable for soft bodied poikilotherms like earthworms which are devoid of any exoskeletal protective cover. Correlation coefficient values between different environmental parameters and population density of earthworms have been presented in Table 5. Total worm number shows high positive correlation with that of soil moisture, relative humidity and soil organic matter and negative correlation with that of solar radiation and actual evapotranspiration (Table 5). Similar correlation and regression models are also available from different agroecosystems (Dash and Patra, 1977); Dash and Senapati, 1980; Senapati and Dash, 1981; Kale and Krishnamoorthy, 1982; Krishnamoorthy and Ramachandra, 1988; Sahu *et al.* 1988; Bhaduria and Ramakrishnan, 1989). During hard summer and winter, earthworms undergo suspended metabolic stage for energy conservation. It may be formation of a diapause coiling of the worm in the soil cavity coated with colloidal mucus to prevent desiccation (Fig. 4) (Dash and Senapati, 1980) or quiescence where no coiling of the body is formed. Quiescence is a torper phase of non-diapausing and is considered comparatively as a less significant phenomenon of suspended metabolism than in comparison to diapause (Andrewartha, 1952).



Fig. 4. Diapausing of *D. willsi* earthworm.

Table 4. Population density (nos/m<sup>2</sup>) and biomass (g live wt/m<sup>2</sup>) of earthworms

Habitat	Population density	Population biomass
<b>(A) Grasslands and pastures</b>		
<b>(a) Natural grasslands</b>		
(i) Upland	12-387	20-78
(i) Lowland	64-800	6-60
(iii) Plain grassland	322	58.74
	53.5	7.5
	49-1364	34-122
<b>(b) Human interfered grasslands</b>		
(i) Grazed pasture (Upland)	75-272	12-70
(ii) Upland grazed pasture	0-140	0-26
(iii) Arable land		
Paddy-rice field (Upland)	32-1399	2-62
Paddy-rice field (Lowland)	0-246	0-25
(iv) Kitchen garden site	236-4964	1.02-14.87
(v) Countryard site	0-4496	0-31.80
<b>(B) Organic waste deposit sites</b>		
(i) Pasture receiving kitchen waste	0-8038	0-66.2
(ii) Farmyard manure garden	15-625	—
(iii) Dung deposit site	0-12617	0-51.4
(iv) Kitchen waste deposit site	600	—
(v) Wash basin waste receiving site	50-500	—
(vi) Straw thatched roof	800	—
<b>(C) Forests</b>		
<b>(a) Natural Forests</b>		
(i) Deciduous forest	24-131	7.0-28.5
(ii) Deciduous forest	73	9.8
(iii) Deciduous forest	83-247	—
(iv) Deciduous forest	273	63.94
(v) Deciduous forest	226-545	—
<b>(b) Human interfered forest</b>		
(i) Eucalyptus plantation	112-461	—
(ii) During fallow phases developed after shifting agriculture (Jhum)		
After 5 years	543	—
After 10 years	634	—
After 15 years	884	—
(iii) During cropping phases of shifting agriculture (Jhum)		
After 5 years	585	—
After 15 years	907	—

H = Handsorting. H and W.S. = Handsorting and Wet Sieving

## In different tropical ecosystems of India

Locality	Extraction method	Reference
Sambalpur, Orissa	H and W.S.	Senapati and Dash (1981)
Berhampur, Orissa	H	Dash and Patra (1977)
Bangalore, Karnataka	H	Krishnamoorthy (1985)
Madras, Tamilnadu	H	Ismail <i>et al.</i> (1990)
Sambalpur, Orissa	H and W.S.	Sahu and Senapati (1991)
Sambalpur, Orissa	H and W.S.	Senapati and Dash (1981)
Jyoti Vihar, Orissa	H	Pradhan and Mishra (1986)
Sambalpur, Orissa	H and W.S.	Pani and Senapati (1986), Pani (1987)
Sambalpur, Orissa	H and W.S.	Pani (1987)
Jyoti Vihar, Orissa	H and W.S.	Mohanty (1987)
Jyoti Vihar, Orissa	H and W.S.	Senapati (Per. Com.)
Sambalpur, Orissa	H and W.S.	Sahu and Senapati (1986)
Bangalore, Karnataka	H	Kale and Krishnamoorthy (1982)
Sambalpur, Orissa	H and W.S.	Sahu <i>et al.</i> (1988)
Rajgangpur, Orissa	H	Julka and Senapati (1987)
Jyoti Vihar, Orissa	H	Julka and Senapati (1987)
Ladukhai, Orissa	H	Julka and Senapati (1987)
Sambalpur, Orissa	H	Mishra and Dash (1984)
Madras, Tamilnadu	H	Ismail <i>et al.</i> (1990)
Bangalore, Karnataka	H	Krishnamoorthy and Ramchandra (1988)
Bangalore, Karnataka	H	Krishnamoorthy (1985)
Bargarh, Orissa	H and W.S.	Dash and Senapati (1991)
Bargarh, Orissa	H and W.S.	Dash and Senapati (1991)
Shillong, Meghalaya	H	Bhaduria and Ramakrishnan (1989)
Shillong, Meghalaya	H	Bhaduria and Ramakrishnan (1989)
Shillong, Meghalaya	H	Bhaduria and Ramakrishnan (1989)
Shillong, Meghalaya	H	Bhaduria and Ramakrishnan (1989)
Shillong, Meghalaya	H	Bhaduria and Ramakrishnan (1989)

H = Handsorting, H and W.S. = Handsorting and Wet Sieving

**Table 5. Correlation coefficient between environmental parameters and total number of tropical earthworms from pasture ecosystem of Orissa, India**

Parameter	<i>D. bolawi</i>	<i>D. willsi</i>	<i>P. elongata</i>
Solar radiation ( $10^3$ kj/m <sup>2</sup> /month)	-0.458 <sup>e</sup>	-0.662 <sup>c</sup>	-0.612 <sup>d</sup>
Rain fall (mm)	+0.177	-0.038	-0.014
Relative humidity(%)	+0.598 <sup>c</sup>	+0.547 <sup>b</sup>	+0.447 <sup>d</sup>
Actual evapotranspiration (mm)	-0.369 <sup>e</sup>	-0.559 <sup>b</sup>	-0.604 <sup>c</sup>
Air temperature (°C)	+0.091	-0.0336 <sup>e</sup>	-0.292 <sup>e</sup>
Soil moisture (g%)	+0.719 <sup>b</sup>	+0.782 <sup>a</sup>	+0.751 <sup>a</sup>
Soil temperature(°C)	+0.324	-0.103	-0.145
Soil organic matter (g%)	+0.643 <sup>c</sup>	+0.827 <sup>a</sup>	+0.887 <sup>a</sup>

a, b, c, d, e Significant at 0.001,0.01, 0.02, 0.05 and 0.2 respectively

### **Biomass and Secondary Production**

Biomass relates to the organismal tissue expressed in terms of unit of weight per unit area and time. Biomass could be represented as live weight, dry weight, ash free weight, carbon weight and as energy content. Organismal biomass is the media for production. But production indicates the role of an animal population as a continuing resource for its own population and for other members of the community. Production can indicate the strategy of the population in directing differentially mass and energy to other parts of the community and ecosystem.

Live biomass refers to live weight of the earthworm/worms available per unit area and time. Live biomass should be expressed as gut free live weight. It makes lot of difference to express live biomass with gut content and without it. Dry biomass of the worm could be quantified from gut cleaned and oven dried worm tissue kept at 85°C for 24 hours. Gut cleaning of the worm could be made by keeping them in wet filter paper and time period for gut evacuation depends on age group, species and other factors like temperature etc. However it is not always possible to calculate gut free live weight of the sampled worm. For this an alternative method is suggested for calculating dry worm tissue from preserved samples. Dry weight of the worm = [Formalin preserved weight + weight loss due to formalin preservation (Senapati and Dash, 1980) – gut content of the worm as calculated from the preserved worm – moisture content of the worm as calculated from the gut cleaned live worm tissue]. Moisture content of the live worm tissue has to be quantified by taking intact and live earthworms at each age group, species, season and habitat. This collection should be outside the sampling area. Secondary production (P) is the sum total of tissue utilised in growth (Pg) and tissues invested in reproduction (Pr) ( $P = Pg + Pr$ ) (Petrušewicz and Macfadyen, 1970). Tissues production (tissue utilised in growth) is estimated by Allens graphical method utilising the formula :  $Pg [V_i + V_{i+1}]/2 \times (W_{i+1} - W_i)$  where Pg = tissue production,  $V_i$  = number of individuals at a particular stage (attainig the successive size class at time  $t_i$ ),  $V_{i+1}$  = number of individuals of successive stages (attaining the successive size class in time  $t_i + 1$ ),  $W_i$  = mass of individuals at a particular stage and  $W_{i+1}$  = mass of individuals of the successive stages. Tissue

**Table 6. Dynamics of earthworm tissue (total production and tissue utilised in reproduction) and estimate of secondary production of *D. bolau*, *D. willsi* and *P. elongata* from pasture ecosystems of Orissa, India.**

Month (week) Year	<i>D. bolau</i>			<i>D. willsi</i>			<i>P. elongata</i>		
	Total live tissue	Tissue Production	Tissue utilised in reproduction	Total live tissue	Tissue production	Tissue utilised in reproduction	Total live tissue	Tissue production	Tissue utilised in reproduction
Jul. (IV) 1984	4.08		0.00						
		1.66							
Aug. (II)	13.23		0.04						
		0.00							
(IV)	5.06		0.32	2.47		0.01	12.43		0.11
		6.97			0.00			0.00	
Sep. (II)	12.05	0.36	1.92	1.60		0.01	11.02		0.12
(IV)	2.94		0.40	2.17		0.10	12.29		0.13
		2.23			0.59			4.29	
Oct. (II)	5.28		0.04	1.85		0.02	11.76		0.05
		0.00			0.03			2.41	
(IV)	0.36		0.00	1.63		0.01	13.47		0.05
		0.19			0.00			0.00	
Nov. (II)	—		—	—		—	—		—
		—			—			—	
(IV)	0.21		0.00	1.28		0.02	12.48		0.08
		0.00			0.00			0.00	
Dec. (II)	—		—	0.33		0.00	12.11		0.03
		—			0.09			0.00	
(IV)	0.07		0.00	0.69		0.01	9.19		0.05
		0.00			0.01			2.64	
Jan. (II) 1985	—		—	0.74		0.00	8.84		0.08
		—			0.00			0.00	
(IV)	0.05		0.00	0.66		0.01	7.12		0.04
		0.02			0.04			3.35	
Feb. (II)	—		—	1.76		0.06	7.66		0.06
		—			0.31			0.49	
(IV)	0.07		0.00	0.80		0.01	8.05		0.01
		0.00			—			—	
Mar. (II)	—		—	0.29		0.01	6.31		0.05
		—			0.15			0.00	

Month (week) Year	<i>D. bolau</i>			<i>D. willsi</i>			<i>P. elongata</i>		
	Total live tissue	Tissue Production	Tissue utilised in reproduction	Total live tissue	Tissue production	Tissue utilised in reproduction	Total live tissue	Tissue production	Tissue utilised in reproduction
(IV) 0.00	—	0.33 0.00		0.00	7.97 0.05		0.15	2.56	
Apr. (II)	—	—	0.16		0.00	7.15		0.03	
(IV)	0.00	—	0.00	0.00	0.00		4.46	0.52	0.00
May (II)	—	0.00	—	0.00	0.00		5.38	1.95	0.00
(IV)	0.00	—	0.00	0.39	0.20		8.99	0.53	0.03
Jun. (II)	0.37	0.18	0.00	0.57	0.00		8.20	0.00	0.03
(IV)	2.04	0.00	0.02	0.86	0.02		9.66	0.00	0.03
Jul. (II)	0.00	0.00	0.33	1.45	0.34		11.48	0.02	0.08
(IV)	3.03	0.00	0.32	1.10	0.00	0.01	13.74	1.58	0.08
Aug. (II)				—	—		—	0.00	—
(IV)				0.95	0.00	0.01	12.15	0.00	0.20
(a) *	Average biomass	2.87	0.96				9.62		
	B \$ SEM	0.98	0.15				0.57		
(b) **	Tissue production ( $P_g$ )	11.60 (224.39)		1.82 (35.19)			21.06 (407.24)		
(c) **	Tissue utilised in reproduction ( $P_r$ )		2.74 (53.05)		0.29 (5.67)			1.28 (24.83)	
(d) **	Total secondary Production (b + c)	14.35 (277.44)		2.11 (40.86)			22.34 (432.07)		

\* Values in g dry wt/m<sup>2</sup>/fortnight

\*\* Values in g dry wt/m<sup>2</sup>/yr (kJ/m<sup>2</sup>/yr)

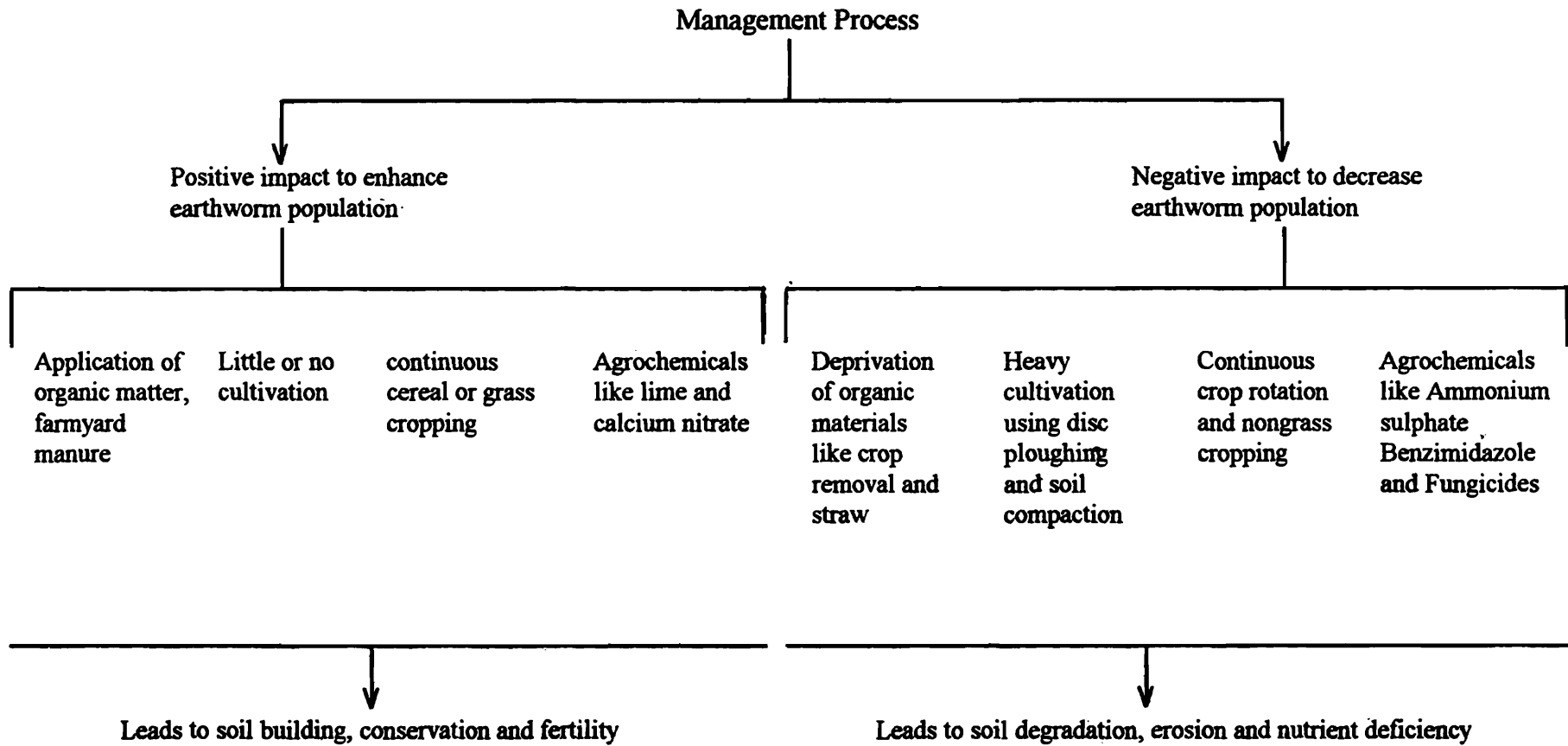
After Sahu and Senapati (1988), Sahu (1989), Senapati (1990), Senapati and Sahu (1991)

utilized in reproduction has to be calculated from sum total of cocoon produced per unit area and per year. Generally secondary production is quantified on annual basis. The energy value of earthworm tissue and cocoon could be quantified by Bomb Calorimetry or from previously reported value (19.34 kJ/g dry worm tissue by Golley, 1961), Biomass turnover value has been calculated from the rate of secondary production to average biomass (Senapati and Dash, 1981).

Table 6 gives the dynamics of earthworm tissue and estimate of secondary production of *D. bolau*, *D. willsi* and *P. elongata* from pasture ecosystems of Orissa. Average worm biomass (g dry wt/m<sup>2</sup>/fortnight) of about 2.87 + 1.10 for *D. bolau*, 0.96 + 0.16 for *D. willsi* and 9.6 + 0.58 for *P. elongata* have been reported during 1984 – 1985 period (Table. 6) (Sahu, 1989). Annual secondary production of 14.35 (277.44), 2.11 (40.86) and 23.04 (432.07) g dry wt/m<sup>2</sup>/yr (kJ/m<sup>2</sup>/yr) have been reported for *D. bolau*, *D. willsi* and *P. elongata* earthworm respectively during 1984-1985 study period. Analysis of individual components of secondary production shows that about 81%, 86% and 94% of total secondary production was contributed by tissue production and tissue utilised in reproduction contributed about 19%, 14% and 6% for *D. bolau*, *D. willsi* and *P. elongata*, respectively. This indicates more than three times energy investment in reproduction in *D. bolau* than in comparison to *P. elongata* which is expected in epigeic life strategy to enhance reproduction for compensation of mortality (Senapati and Dash, 1981, Sahu and Senapati, 1988; Senapati and Sahu, 1991). Highest biomass turnover value of about five has been reported for *D. bolau* earthworm. Lakhani and Satchell (1970), Satchell (1971) are the pioneering workers on secondary production of earthworm (*Lumbricus terrestris*) population from European temperate ecosystem. Dash *et al.* (1974), Dash and Patra (1979) and Senapati and Dash (1981) are the pioneer workers to report about secondary production in Indian earthworms and to signify their relationship to primary production. Secondary production in earthworms ranges from 50 to 250 kJ/m<sup>2</sup>/yr in temperate ecosystems whereas in tropical ecosystems it ranges from 70 to 700 kJ/m<sup>2</sup>/yr. thus tropical earthworms seem more productive than temperate worms. This inference very well supports the hypothesis by Pianka (1970) that : organisms inhabiting a variable (unpredictable) habitat are more productive than organisms occurring in fairly constant (predictable) environment. Intra habitat comparison reveals that epigeic species like *D. bolau*, epianecic species like *D. willsi* are more productive per unit biomass per day than the endogeic *P. elongata* worm, indicating their relative importance. *D. bolau* and *D. willsi* thus could very well thrive in man interfered environments. *D. bolau* abundance and role in cow dung deposit sites (Senapati *et al.* 1987; Sahu *et al.* 1988) and *D. willsi* abundance and role in paddy rice system (Pani and Senapati, 1986; Pani, 1987) support the above hypothesis. Pioneering work of Bhaduria and Ramakrishna (1989) on the population biology of earthworms under shifting agriculture indicated high resistance of *Tonoscolex horai* (Stephenson) and thus a hardy species suitable for application in higher elevations of North eastern India to promote biological soil fertility in low input shifting agricultural system. Management processes responsible to enhance earthworm population and to decrease it have been streamlined (modified after Edwards, 1981) for manipulation of proper population level so as to expect their positive role towards soil conservation, fertility and plant production (Fig. 5).

### Future Prospects and Problems

About 374 species of earthworms have been reported in India from different habitats. However population biology has been reported for about 40 species of earthworms and the details (population density, biomass and productivity) are known in about 10 species. It is important to know the organismal activity in its natural habitat before utilising it for any academic and economic interest.



**Fig. 5. Positive and negative impact of environmental management on earthworm population and soil conservation**

Autecological studies are thus very important to reveal the professional status of the organism. Since India has wide variety of habitat, species and climate it is never possible to certify a single species either for class room or for vermiculture based biotechnology. There is variation of the biological strategy of the organism at intra species and interspecies level which has already been reported by Senapati and Dash (1981), Pani (1987), Senapati *et al.* (1987), Sahu (1989), Sahu *et al.* (1988) and Bhaduria and Ramakrishnan (1989). Predictive models are always to be based upon large number of data. In India alongwith great variability in habitat condition, large number of species occurrence, with variety of biological but with few detail studies, it might not be possible to develop a really predictable model and thus it needs in depth study of different earthworm populations and communities. Because of extreme climates it is also equally important to know the biokinetic limit of different earthworm species particularly in relation to diapause and quiescence process to assess their impact and to understand the compensation mechanism. Alongwith natural stress, various physical, chemical and biological impacts of man's activity should also be looked into for their incorporation into the predictable models and for management.

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## **Reproductive Biology (Cocoon Morphology, Life Cycle Pattern and Life Table Analysis) in Earthworms**

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### **Synopsis**

Information on the reproductive biology of Indian earthworms is very much scanty and is absolute for the growth and development of vermiculture based biotechnology and for academic interest. Majority of the work done in this line relate to less than 5% of the total number of species so far reported. Authors and their associates have largely contributed in this aspect during the last decade. This paper summarizes the basic principles of reproductive biology in earthworms and has been dealt in eight sections including methodology, present state of information and future line of work to be done. The objective of this paper is mainly to introduce the subject to the students and research workers interested in this line.

### **Understanding the organisms through reproductive biology**

The process of producing young ones is the subject matter of reproduction. Reproductive process occupy the central position in the biomanagement programmes. Organisms could either be a pest (harmful) or a benefactor (useful) to man. Knowledge on the reproductive biology could help to curtail pest organisms and to enhance benefactors. Reproduction in earthworms is not only peculiar because of hermaphroditism but also there is a great paucity of information in this regard. More than 95% of zoologists have not seen earthworm cocoon. No standard text book describes details of reproductive biology in any single species of Indian earthworms although it serves as one of the basic biological specimen for introductory biology. Presently earthworm is not restricted as a basic biology material but is being established as a basic material for biotechnology (vermitechnology). Because of vast number of species available in India and because of their academic and economic value, understanding of reproductive biology of earthworms has become very important. This may help for : (i) understanding the organism, (ii) cocoon is most suitable for inoculation, storage and transport for vermiculture based biotechnology, (iii) bioindication and biocide tests and (iv) prediction of the population characteristics in captivity or in nature regarding impact of environmental variables. This paper deals with some aspects of reproductive biology in earthworms mostly available in Indian ecosystems.

### **Reproduction in earthworm**

Olive and Clark (1978) have distinguished three basic modes of reproduction in annelids (Fig. 1). Monotelic - in which a species breeds only once during whole life cycle. Polytelic - in which breeding occurs at several times during their life cycle. Both monotelic and polytelic species have discrete reproduction and are 'big bang' strategists. From physiological point of view one could

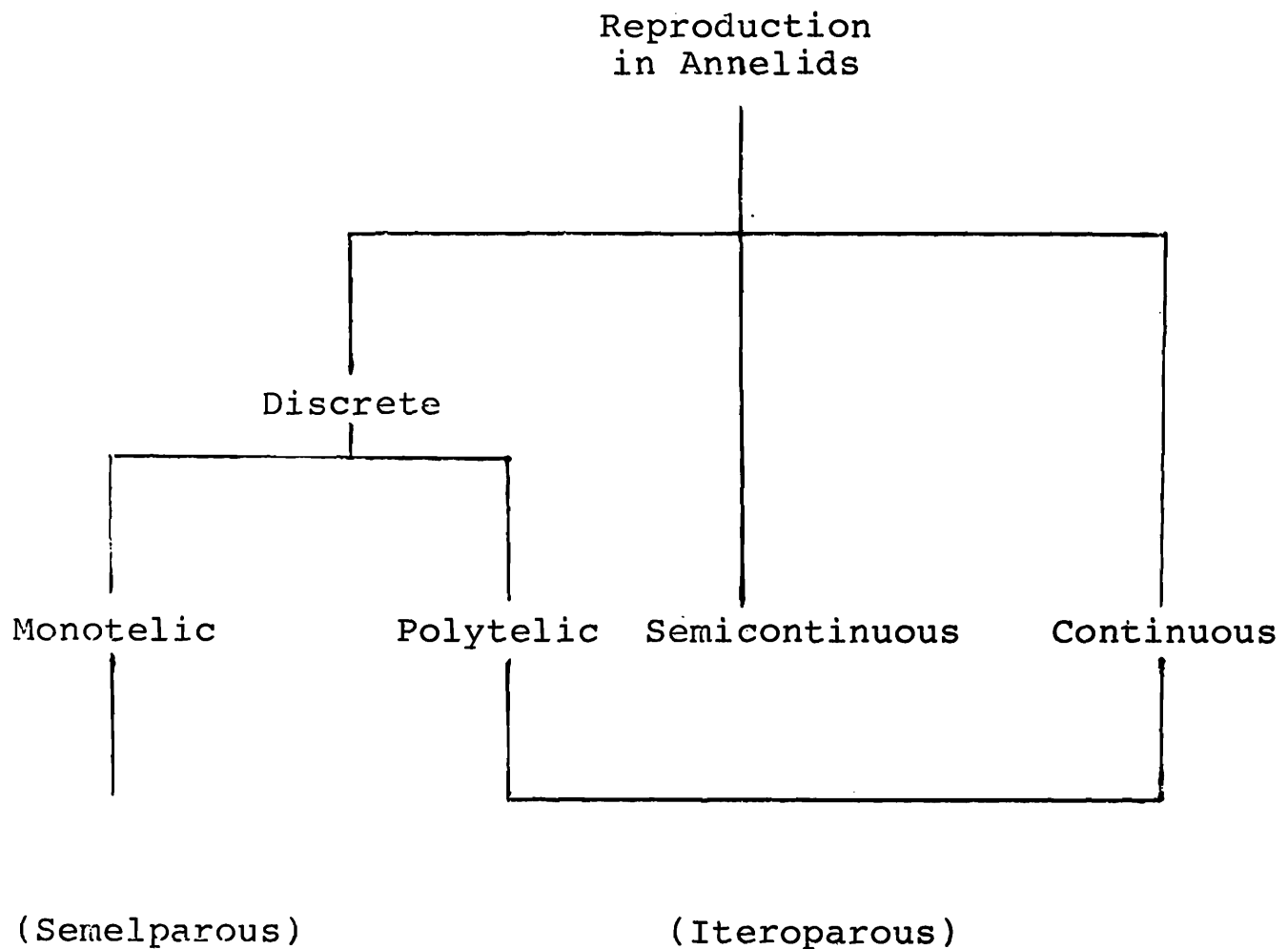


Fig. 1 Diagrammatic representation of reproduction in Annelids

distinguish discrete breeders which undergo reproductive crisis or crises during a year, from those who breed steadily over a period without such crisis. The third category is semi-continuous/continuous where the species breed several times during life and release gametes in a number of broods over an extended breeding season within one or more years. Earthworms were grouped among semi-continuous/continuous breeders (Lee, 1985). Cole (1954), Gadgil and Bossert (1970) have classified species that reproduce only once in their life time and die as 'semelparous' and those that reproduce repeatedly as 'iteroparous' (Fig. 1).

Earthworms are hermaphrodite and each worm produces both ova and spermatozoa. Exchange of sperms occur during copulation of two mature worms and results in the production of cocoon. Biparental mode of reproduction is more common method of cocoon production. Copulation has been observed in only few earthworms. Each species has a number of spermathecae in its anterior segments. A little behind the last pair of spermathecae is the female pore which opens to the ventral side. In *Metaphire posthuma* (previously *Pheretima posthuma*) clitellum remains in 14-16 segments. Genital papillae arise on each segment of 17 and 19 and the male generative apertures remain in 18th segment. The female pore is connected to the oviduct which brings the ova from the ovaries (situated on the posterior surface of septum 12/13) as well as other secretions to it. Each male pore is connected to a thicker tube which incorporates the tube from the testis (the vas, deferens) and from the prostate gland (Prostatic duct). Sequence of copulation has been given in Fig. 2 as observed in *Metaphire* sp. The copulating animals lie with anterior ventral surface facing

each other but each head pointing in opposite directions. The male pores of one come in contact with the spermathecal pores of the other. The sperms and the prostatic fluid are mutually discharged into the posteriormost pair of spermathecae, where they are nourished by some fluids (Tembe and Dubash, 1961). Each partner move backwards in sequential manner so as to discharge into the spermathecae in successive manner until all spermathecae are 'charged' The partners then separate after about an hour. The clitellum of each copulating pair gets enclosed in a colloidal secretion from the clitellum. The three layered wall of a cocoon is secreted by a type of clitellar gland cell containing large granules (Grove and Cowley, 1926). It contains protein and a chitinoid material which is probably chitin (Needham, 1969). Because of the presence of chitin, initially formed colourless cocoon darkens with exposure to air. Uniparental parthenogenesis with self fertilization is also known in some earthworm species where there is absence or retrogression of some secondary sexual organs like spermathecae, prostates etc. Methods of reproduction in some earthworms found in India is given in Table 1.

### Reproductive biology of Indian earthworm

Of the earthworms, the reproductive biology of the Lumbricidae is by far the best known from the reviews of Stephenson (1930), Satchell (1967), Edwards and Lofty (1972) and Lee (1985). But sufficient information was not available on Indian earthworms except in *Pheretima* (now *Metaphire*) a representative of the important tropical family Megascolecidae, to allow comparisons to be made with lumbricids. Pioneering works on the reproductive biology has been reported by Evans and Guild (1948 a & b), Gavrilov (1948), Michon (1954), Phillipson and Bolton (1977). First observation on the reproductive biology of Megascolecidae worm has been made by Bahl (1927, 1950), Oishai (1930) and Tembe and Dubash (1961). Ecology of reproductive biology of Indian worms was first reported by Senapati and Dash (1979), Senapati *et al.* (1979), Dash and Senapati (1980,1982). This was followed with a series of publications dealing with cocoon morphology, emergence patterns, growth, life cycle, life table, energy allocation to reproduction by Senapati and his associates (1980; Senapati and Dash, 1984; Sahu and Senapati, 1986, 1988, 1991; Sahu *et al.* 1988). Other works relating to the reproductive biology of Indian earthworms are by Kale *et al.* (1982) and Kale and Bano (1985).

**Table 1** Methods of reproduction in some earthworms found in India

Species	Type of reproduction	Reference
<i>Pontodrilus bermudensis</i>	S	Gates (1972)
<i>Eudrilus eugeniae</i>	S	Gates (1972)
<i>Pontoscolex corethrurus</i>	P	Gates (1972)
<i>Bimastos parvus</i>	P	Gates (1959)
<i>Eisenia fetida</i>	P, SP	Gates (1972)
<i>Lampito mauritii</i>	S	Gates (1972)
<i>Perionyx excavatus</i>	S	Gates (1972)
<i>Polypheretima elongata</i>	P	Gates (1972)
<i>Metaphire posthuma</i>	S	Gates (1972)
<i>Ocnodrilus occidentalis</i>	P	Gates (1959)
<i>Dichogaster bolau</i>	PP	Gates (1972)

#### Abbreviations :

S = amphimictic, reproduction sexual and biparental

SP = generally amphimictic with parthenogenesis in some individual

P = Parthenogenetic, reproduction uniparental

PP = Possibly parthenogenetic

### Cocoon collection and characteristics

Cocoons of earthworms are in general ovoid capsules, prolonged into short processes at both poles, when fresh they are whitish in colour and very soft jelly like and later become harder and colour varies from lemon yellow to olive green to pinkish red. Collection of the cocoon is a tedious affair of wet sieving and handsorting from the habitat soil. Selection of the sieve depends on the size of the cocoon and generally a sieve having pore size 500 $\mu$  is preferred (Senapati and Dash, 1979, 1984; Dash and Senapati, 1980). Cocoons may be stored at low or high temperature inside an incubator with and without soil medium (Dash and Senapati, 1980). Preservation of the cocoon is same as that of the worm and could be kept in 5% formalin. However for chromosomal study, live cocoons may be fixed in 1:3 acetoalcohol (Senapati per. cum.). From the field sampling generally three types of cocoons are collected. Live cocoons having variable colours and when examined under microscope or magnifying lens show live embryo at different stages of development. Dead cocoon which is generally black, contain putrefied material. Empty cocoon which is the cocoon case having a hollow space might be occupied by foreign organisms like nematodes, enchytraeids etc. or foreign materials like soil etc. (Senapati *et al.* 1979; Dash and Senapati 1980). Earthworm cocoons might possess different types of ornamentation at the tapering ends. The ornamentation might be short as in *Drawida willsi*, *Lampito mauritii* or may be long thread like structure as in *Drawida calebi*, *Glyphidrilus tuberosus* or may be flattened structure as in *Perionyx excavatus* (Senapati per. cum.). The cocoons of *Criodrilus* are spindle shaped with the ends drawn out to about 1.5 to 7 cm long corresponding to the length of the clitellum in this worm (about 32 segments) (Stephenson, 1939). Cocoon of *Megascolides australis*, the largest known earthworm is 75mm in length and 20mm in diameter and contains a single embryo which when hatched is as long as one foot. Cocoon of *Metaphire posthuma* is small and is about 2.0 mm in length and 1.5 mm in width. Comparative characteristic features of earthworms and their cocoon from temperate and tropical habitats of the world is given in Table 2.

Earthworm cocoon after undergoing incubation, produces young worms called juveniles which lack genital papillae and clitellum. Incubation period of cocoon vary in different species and also with environmental parameters. Incubation period ranges from 3 to 30 weeks in temperate worms whereas it is 1 to 8 weeks in case of tropical worms (Table 2.). Incubation period of *O. surensis* (4 weeks at about 25°C and 15 g% soil moisture) could be extended to more than 12 weeks at 20°C with 15 g% moisture level.

Low incubation period in tropical worms might be an adaptive strategy to cope with environmental drasticity (Senapati *et al.* 1979). Incubation period could be determined in laboratory condition by keeping fresh laid cocoons on moist filter paper spread over habitat soil. Young worm/worms hatch from the cocoon at the end of the incubation period. *Dichogaster bolawai* worm is reported to have the shortest incubation period of about one week duration at 20  $\pm$  2g% soil, moisture and 25  $\pm$  2°C of soil temperature (Sahu and Senapati, 1986). Incubation period of about four weeks has been reported for *Octochaetona surensis*, *Lampito mauritii*, *Perionyx excavatus*, *Ponotoscolex corethrurus*, *Metaphire posthuma* and *Polypheretima elongata* (Dash and Senapati, 1980; Kale *et al.* 1982; Senapati and Dash, 1982, 1984; Kale and Bano, 1985; Sahu and Senapati, 1986, 1988, 1991; Sahu *et al.* 1988). Usually a single worm emerges out of each cocoon. But in some cases two to three worms hatching from a single cocoon has also been reported (Dash and Senapati, 1980).

### Quantification of reproduction in earthworms

Rate of reproduction in earthworm is generally calculated from the ratio of total cocoons produced by the number of adults present within a particular time period and habitat/culture. Here it is presumed that the worm might be producing cocoon either sexually or parthenogenetically. To be more particular to consider sampling interval, the rate of reproduction may be calculated as : [(C x (SI/IP))/A. Second situation might be where information on cocoon production is not

available but incubation period of cocoon and number of juveniles hatched per cocoon are known from laboratory studies. So the formula for calculated rate of reproduction may be modified as :  $[(J/N \times (SI/IP))/A]$ . Third situation might be where no information is available about the cocoon but quantitative data are known for juvenile and adult worms. The rate of reproduction may be quantified from a simple formula :  $J/A$ . In a situation where the the investigators are unable to distinguish between juvenile and immatures the formula for quantification may be modified as :  $NA/A$ . Where A = average number of adults available per sampling occasion, C = average number of cocoon available in the samples per occasion, J = average number of juveniles available in the samples per occasion, IP = incubation period in days, N = average number of juveniles hatched per cocoons, NA = average number of non-adults available in the samples per occasion, SI = time interval between two successive sampling occasions in days. Utility of the above formulae have been verified on a peregrine earthworm, *Dichogaster bolau*. Dynamics of cocoons, different age class density and rate of reproduction in *D. bolau* from an upland grazed pasture has been presented in Table 3. Statistical analyses between (by 't' test) and among (by analysis of variance (Anova) different alternative formulae have been made in Table 4. Analysis of variance among four different ratios of reproductive rate at respective situations shows that they are not significant at 0.05 level ( $F = 1.73$ ) indicating the possibility of utilising any of these quotients for qualification of reproduction. Further details of these alternatives could be known from the publication of Sahu and Senapati (1988). Cocoon production (number/adult/year) varies with species and environmental condition (Satchell, 1967; Edwards and Lofty, 1972; Dash and Senapati, 1980; Lavelle, 1981; Krishnamoorthy, 1987; Sahu and Senapati, 1991). Considering both temperate and tropical earthworms it has been found that size of the worms bears a negative relationship with that of the number of cocoons produced per adult per year (Table 2). Worm diameter to cocoon diameter, worm biovolume to cocoon biovolume, worm dry weight to cocoon dry weight all bear significant positive correlation ( $r > 0.8$ ,  $p < 0.001$ ). It is possible to predict the characteristic features of the cocoon by utilising present regression models (Table 2).

### Seasonal dynamics of cocoons, emergence pattern and reproductive strategy in earthworms

Biological rhythmicity regulates the reproductive activity in earthworms. Accordingly availability of the cocoon in field condition is largely dependent on the species and the environment. Ecological category of earthworms very much depends on the species specificity and niche segregation (Senapati et al, 1979; Dash and Senapati, 1980; Sahu and Senapati 1991; Senapati and Sahu 1991). MacArthur and Wilson (1967) coined the term 'r'- selection and 'k'-selection to describe different ways in which the population might function to survive in non-competitive and competitive environments. Details of this aspect has been dealt in other papers of this volume. Here the discussion has been made with respect to three earthworms: *Dichogaster bolau*, *Drawida willsi* and *Polypheretima elongata* belonging to the three distinct ecological category. Most of the earthworms available in India belong to one of these categories. Fortnightly dynamics of total cocoons, juveniles and adult earthworms of *D. bolau*, *D. willsi* and *P. elongata* is given in fig. 3,4 and 5, respectively.

*D. bolau* is a small (Table 2), surface living, epigeic earthworm belonging to 'r' selected species restricted mostly to the litter horizon and is phytophagous (plant eating) in nature (Sahu and Senapati, 1986,1988,1991; Sahu et al 1988). Cocoons of *D. bolau* earthworm are available from end of July till October in upland grazed pasture. A peak number of about 5000 cocoons has been reported per square meter area during mid September when the adult worms were about 1150/m<sup>2</sup>. Rate of total cocoon to adult worms during peak cocoon production came to be 4.5. *D. bolau* worm showed single peak (discrete monotelic or semelparous) of cocoon production with unimodal emergence pattern of juvenile worms. Juvenile peak has been observed during mid August (Fig. 3). Thus exhibition of juvenile peak prior to cocoon peak justify the survival of *D. bolau* worm through cocoon during hard summer (soil temperature  $> 40^\circ$  and soil moisture

**Table 2 Comparative characteristic features of earthworms and their cocoon from temperate and tropical habitats of the world along with regression analysis**

A. Characteristic features of worm and cocoon

Climate	Ecological	Worm				Cocoon						
		Worm type	Dia- meter (mm)	Length (mm)	Bio- volume* (mm <sup>3</sup> )	Dry weight (mg)	Dia- meter (mm)	Length (mm)	Bio- volume* (mm <sup>3</sup> )	Dry weight (mg)	Incuba- tion period (week)	Number of cocoons/ adult/yr
1	2	3	4	5	6	7	8	9	10	11	12	13
Temperate												
<i>E. tetraedra</i>	Epigeic	3.0	45	318.09	-	1.3	2.0	2.66	-	-	-	(1)
<i>D. rubida</i>	Epigeic	3.5	55	529.16	-	1.7	3.7	8.40	-	-	-	(1)
<i>D. octaedra</i>	Epigeic	3.0	40	282.74	-	1.7	2.3	5.22	-	8.5	-	(1)
<i>D. subrubicunda</i>	Epigeic	4.0	80	1005.31	62.00	2.0	3.1	9.74	1.14	8.5	42.00	(1)
<i>A. chlorotica</i>	Endogeic	4.5	55	874.73	72.00	2.3	3.7	15.37	1.72	12.5	26.00	(1)
<i>A. rosea</i>	Endogeic	3.5	45	432.95	78.00	2.6	3.1	16.46	2.00	25-30	8.00	(1)
<i>A. caliginosa</i>	Endogeic	5.0	110	2159.85	126.00	2.3	4.6	19.11	2.58	25-30	26-42	(2)
<i>B. eiseni</i>	Epigeic	3.0	40	282.74	-	2.7	5.7	32.64	-	-	-	(1)
<i>L. rubellus</i>	Epigeic	5.0	110	2159.85	264.00	2.6	3.4	18.05	2.06	16.0	92.00	(1)
<i>O. cyaneum</i>	Endogeic	6.0	150	4241.15	662.00	3.4	6.0	54.48	7.34	-	13.00	(1)
<i>L. terrestris</i>	Epianecic	12.0	350	39584.07	1046.00	4.3	6.0	87.13	11.42	25-30	-	(1)
<i>E. foetida</i>	Epigeic	3.5	75	721.59	156.00	2.9	6.0	39.63	3.72	3-4	17.00	(1)
<i>A. longa</i>	Anecic	7.5	140	6185.01	430.00	3.7	6.9	74.19	12.00	10.0	8.00	(1)
<i>L. castaneus</i>	Epigeic	-	-	-	70.00	-	-	-	1.02	14.0	65.00	(1)
<i>D. mammalis</i>	Epigeic	-	-	-	18.00	-	-	-	0.86	-	17.00	(1)
Tropical												
<i>D. agilis</i>	Epigeic	5.0	70	1374.45	140.00	2.5	3.3	16.20	1.50	-	10.70	(3)
<i>D. terrae-nigrae</i>	Endogeic	18.0	700	178128.30	5600.00	9.0	12.5	795.21	63.00	-	1.90	(3)
<i>M. lamtoiana</i>	Epianecic	26.0	450	238918.12	6400.00	9.0	12.5	795.21	50.50	-	3.10	(3)

1	2	3	4	5	6	7	8	9	10	11	12	13
<i>C. zielae</i>	Endogeic	2.0	70	219.91	36.00	1.0	1.0	0.79	0.43	-	13.00	(3)
<i>S. perifera</i>	Endogeic	2.0	70	219.91	40.00	1.0	1.0	0.79	0.40	-	13.00	(3)
<i>M. anomala</i>	Endogeic	9.0	170	10814.93	610.00	5.0	5.0	98.17	20.00	3.0	6.20	(3)
<i>M. ghanensis</i>	Endogeic	9.0	300	19085.18	3200.00	6.0	10.0	282.74	26.50	-	1.30	(3)
<i>A. multivesiculatus</i>	Endogeic	6.0	330	9330.53	800.00	6.0	15.0	424.12	32.60	-	-	(3)
<i>A. opisthogynus</i>	Endogeic	4.0	200	2513.27	600.00	4.0	10.0	125.66	10.80	-	1.30	(3)
<i>D. bolau</i>	Epigeic	1.3	33	44.30	8.10	1.3	2.0	2.65	0.65	1.0	46.59	(6)
<i>D. willsi</i>	Epianecic	1.8	54	137.20	15.20	2.3	2.3	9.55	1.40	2.0	14.53	(6)
<i>D. calebi</i>	Epianecic	3.4	59	534.80	43.80	3.0	3.0	21.21	2.40	8.0	11.52	(4)
<i>O. surensis</i>	Anecic	3.8	92	1651.30	145.60	3.0	4.5	31.81	5.25	4.0	19.21	(5)
<i>L. mauritii</i>	Anecic	4.3	142	2057.80	185.30	3.5	5.0	48.10	6.10	4.0	14.25	(5)
<i>P. elongata</i>	Endogeic	4.5	192	5296.10	333.00	4.0	5.5	69.12	7.35	4.0	19.07	(6)

## B. Correlation and regression analysis

Parameter	Sample size (n)	Correlation coefficient (r)	Level of significance (p)	Regression equation $y = a + bx$
Worm diameter (mm)/cocoon diameter (mm)	28	+ 0.881	0.001	$y = 1.290 + 0.349x$
Worm biovolume (mm <sup>3</sup> )/cocoon biovolume* (mm <sup>3</sup> )	28	+ 0.910	0.001	$y = 32.610 + 0.004x$
Worm dry weight (mg)/cocoon dry weight (mg)	26	+ 0.920	0.001	$y = 3.323 + 0.009x$
Worm dry weight (mg)/incubation period (week)	17	+ 0.400	0.2	$y = 4.710 + 0.310x$
Worm dry weight (mg)/number of cocoons produced per adult per year	23	- 0.341	0.2	$y = 24.552 + 0.004x$

(1) Evans & Guild (1948, a), (2) Nowak (1975), (3) Lavelle (1978), (4) Senapati et al. (1979), (5) Dash & Senapati (1980), (6) Sahu (1989). \* Biovolume =  $\pi r^2 l$ ,  $r$  = diameter,  $l$  = length

**Table 3** Dynamics of cocoons (nos/m<sup>2</sup>/fortnight), different age class density (nos/m<sup>2</sup>/fortnight) and rate of reproduction of *D. bolau* earthworm in an upland grazed pasture

Month	(week)	Year	Cocoons (C)	Juvenile worms (J)	Non-adult worms (NA) (NA = J + I)	Adult worms (A)	Total worms (TW)	Rate of reproduction			
								$\frac{TC \times SI}{A \cdot IP}$	$\frac{J}{N} \times \frac{SI}{IP}$	$\frac{J}{A}$	$\frac{NA}{A}$
Jul.	(IV)	1984	0	1193	2659	372	3031	0.00	4.28	3.21	7.15
Aug.	(II)		76	1582	7167	871	8038	0.17	2.43	1.82	8.23
	(IV)		645	1208	3779	644	4423	2.00	2.51	1.88	5.87
Sep.	(II)		4956	78	3320	1132	4452	8.76	0.09	0.07	2.93
	(IV)		915	238	358	676	1034	2.71	0.47	0.35	0.53
Oct.	(II)		78	118	591	474	1065	0.33	0.33	0.25	1.25
	(IV)		0	0	40	80	120	0.00	0.00	0.00	0.50
Nov.	(II)		*	-	-	-	-	-	-	-	-
	(IV)		0	0	0	39	39	0.00	0.00	0.00	0.00
Dec.	(II)		-	-	-	-	-	-	-	-	-
	(IV)		0	0	0	39	39	0.00	0.00	0.00	0.00
Jan.	(II)	1985	-	-	-	-	-	-	-	-	-
	(IV)		0	0	0	39	39	0.00	0.00	0.00	0.00
Feb.	(II)		-	-	-	-	-	-	-	-	-
	(IV)		0	0	0	39	39	0.00	0.00	0.00	0.00
Mar.	(II)		-	-	-	-	-	-	-	-	-
	(IV)		0	0	0	0	0	0.00	0.00	0.00	0.00
Apr.	(II)		-	-	-	-	-	-	-	-	-
	(IV)		0	0	0	0	0	0.00	0.00	0.00	0.00
May	(II)		-	-	-	-	-	-	-	-	-
	(IV)		0	0	0	0	0	0.00	0.00	0.00	0.00
Jun.	(II)		0	0	40	80	120	0.00	0.00	0.00	0.50
	(IV)		39	118	591	119	710	0.66	1.32	0.99	4.97

\* Sampling not done

**Table 4 Anova and 't' values of rate of reproduction, calculated on the basis of proposed formulae considering whole year data**
**A.**

Sources of variation	Sum of square	Degree of freedom	Mean square	Calculated value of 'F'	Tabulated value of 'F' at 0.05 level	Significant (S)/Not significant (NS)
Between sample	20.63	3	6.88	1.73	2.8	NS
Within sample	238.67	60	3.98			

**B.**

Parameters considered	't' test between											
	$\frac{C \times SI}{A \times IP}$	and	$\frac{J \times SI}{N \times IP}$	$\frac{C \times SI}{A \times IP}$	and	$\frac{J}{A}$	$\frac{C \times SI}{A \times IP}$	and	$\frac{NA}{A}$	$\frac{J}{A}$	and	$\frac{NA}{A}$
Sampling occasion (n)	16		16	16		16	16		16	16		16
Mean value ( $\bar{X}$ )	0.91		0.71	0.91		0.54	0.91		2.00	0.54		2.00
Standard deviation (Sd)	± 2.24		± 1.28	± 2.24		± 0.96	± 2.24		± 2.89	± 0.96		± 2.89
Calculated value			0.31			0.35			0.46			1.92
Tabulated value at 0.05 level			2.04			2.04			2.04			2.04
Significant (S)/not-significant (NS)			NS			NS			NS			NS

Table 5 Demographic characteristics of some tropical earthworms from S. Africa and India in field condition

Worm type	Duration of the growth period (in months) (G)	Number of cocoons produced/adult/yr (F)	Expectation of life at hatching (in months) ( $e_x$ )	Demographic index (D.I.) (D.I. = $10^3 F/G.e_x$ )	Ecological index (E.I.) (W. $\bar{p}$ )	Reference
S. Africa						
<i>C. zielae</i>	18.0	13.0	3.3	219.0	3.2	Lavelle (1983)
<i>D. agilis</i>	15.0	10.7	3.4	210.0	3.0	Lavelle (1983)
<i>M. anomala</i>	20.0	6.2	6.2	50.0	32.0	Lavelle (1983)
<i>M. lamtoiana</i>	24.0	3.1	7.5	17.0	210.0	Lavelle (1983)
<i>D. terrae-nigrae</i>	36.0	1.9	11.6	4.5	575.0	Lavelle (1983)
<i>A. opisthogynus</i>	24.0	1.3	11.1	4.9	87.0	Lavelle (1983)
<i>M. ghanensis</i>	42.0	1.3	10.6	2.9	512.0	Lavelle (1983)
India						
<i>D. bolau</i>	2.8	46.6	1.7	9789.9	0.2	Sahu (1989)
<i>O. occidentalis</i>	2.5	25.0*	0.7	14285.7	0.4	Sahu (1989)
<i>D. willsi</i>	4.0	14.5	2.4	1510.4	2.0	Sahu (1989)
<i>O. surensis</i>	7.7	19.2	5.7	437.5	22.1	Senapati (1980)
<i>L. mauritii</i>	7.7	14.3	7.8	238.1	23.9	Senapati (1980)
<i>Melongata</i>	8.5	19.1	5.5	408.6	112.0	Sahu (1989)

\* Calculated on the basis of juveniles and assuming similar status as that of *D. willsi*

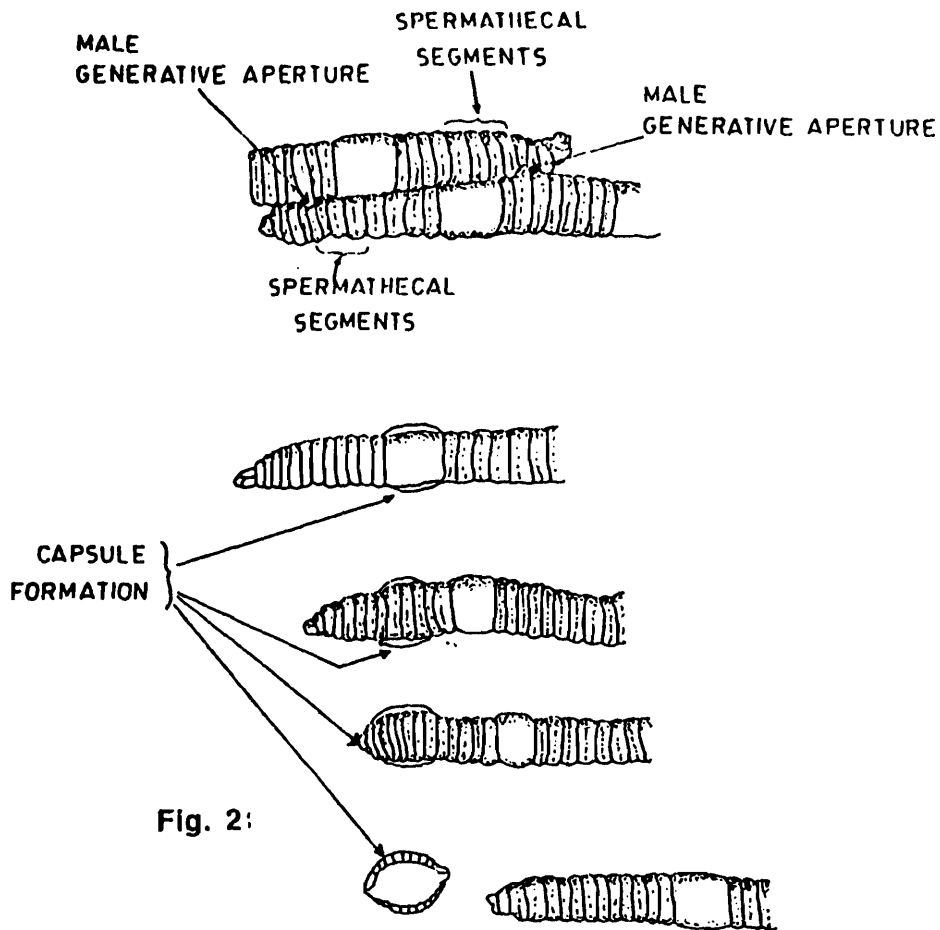


Fig. 2:

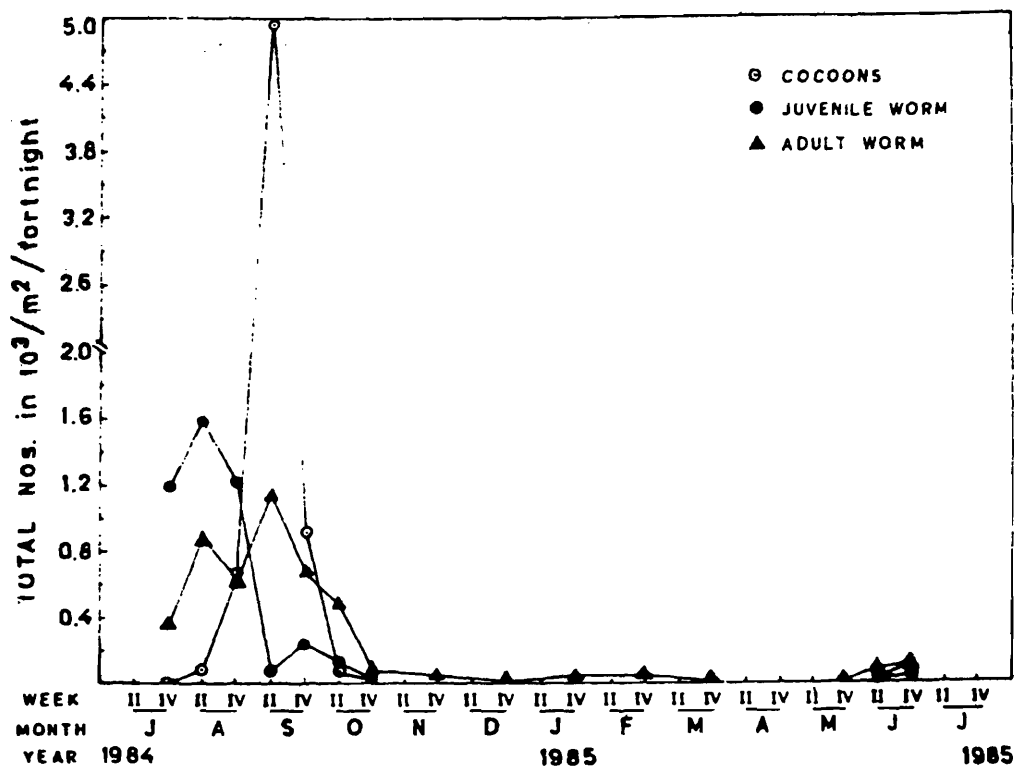


Fig. 3 SAMPLING OCCASION

Fig. 2. Sequence of copulation resulting cocoon formation in *Metaphire* sp.

Fig. 3. Fortnightly Dynamics of Total Cocoon, Juveniles and Adult Earthworms of *D. bolau*.

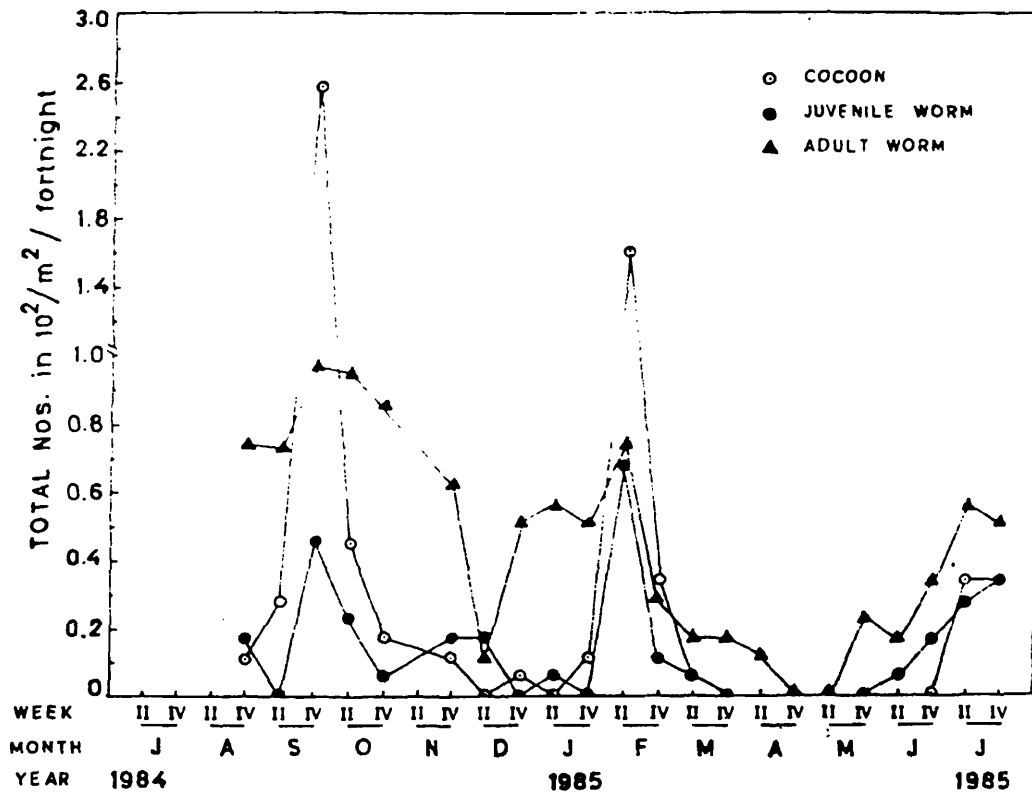


Fig. 4. SAMPLING OCCASION

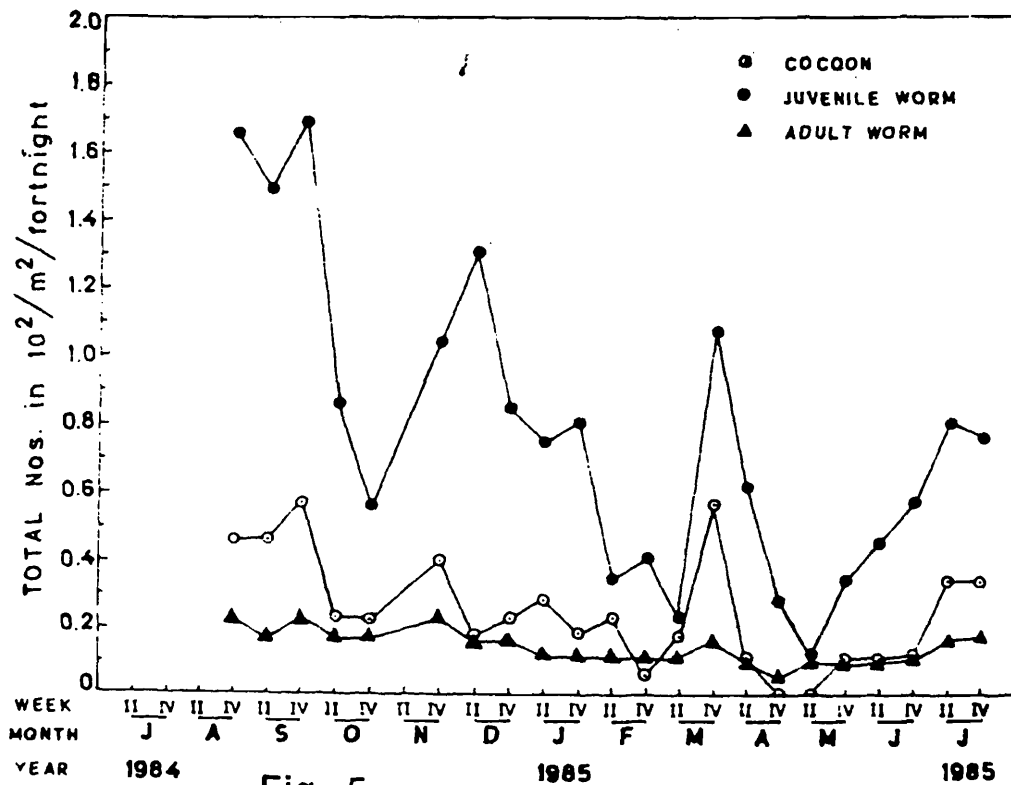


Fig. 5. SAMPLING OCCASION

Fig. 4. Fortnightly Dynamics of Total Cocoons, Juveniles and Adult Earthworms of *D. willsi*.  
 Fig. 5. Fortnightly Dynamics of Total Cocoons, Juveniles and Adult Earthworms of *P. elongata*.

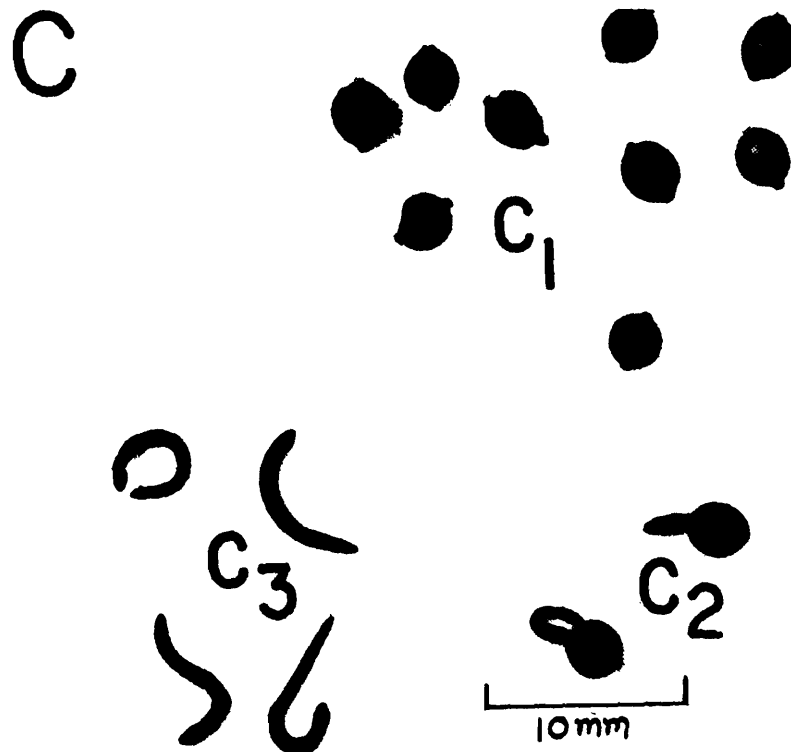


Fig. 6

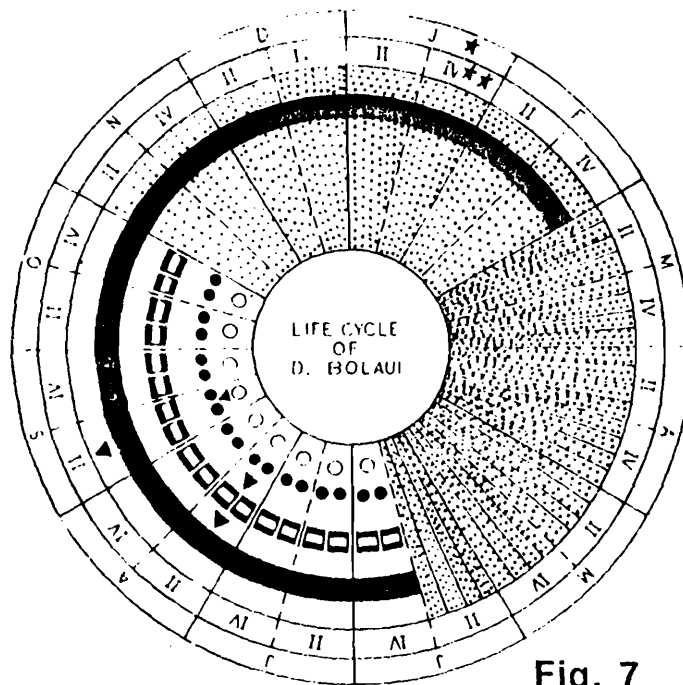


Fig. 7

DIAGRAMMATIC REPRESENTATION OF *D. BOLAU* LIFE CYCLE IN UPLAND GRAZED PASTURE AT BURLA, ORISSA INDIA.  
 ■ ADULT, ▨ IMMATURE, □ ACTIVE PERIOD, ▩ NO WORM,  
 ● JUVENILE, ○ COCOON, ▲ PEAK NUMBER, ★ MONTH, ★★ WEEK  
 ▭ INACTIVE PERIOD.

Fig. 6. Cocoon, just hatching and hatched juvenile of *D. willsi* earthworm C<sub>1</sub> Cocoons of *D. willsi*; C<sub>2</sub> Just hatching juvenile of *D. willsi*; C<sub>3</sub> Hatched juvenile of *D. willsi*.

Fig. 7. Diagrammatic Representation of *D. bolau* Life Cycle In Upland Grazed Pasture At Burla, Orissa, India.

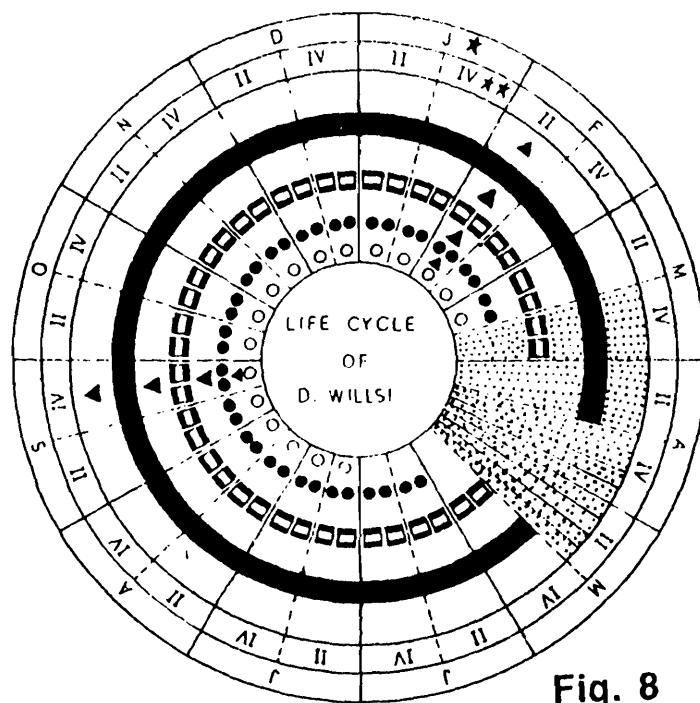


Fig. 8

DIAGRAMATIC REPRESENTATION OF *D. WILLSI* LIFE CYCLE IN PLAIN PROTECTED PASTURE AT SABALPUR, ORISSA, INDIA.  
 ■ ADULT, ▨ IMMATURE, ● JUVENILE, ○ COCOON,  
 □ ACTIVE PERIOD, ▤ INACTIVE PERIOD, ▩ NO WORM,  
 ▲ PEAK NUMBER, ★ MONTH, ★★ WEEK.

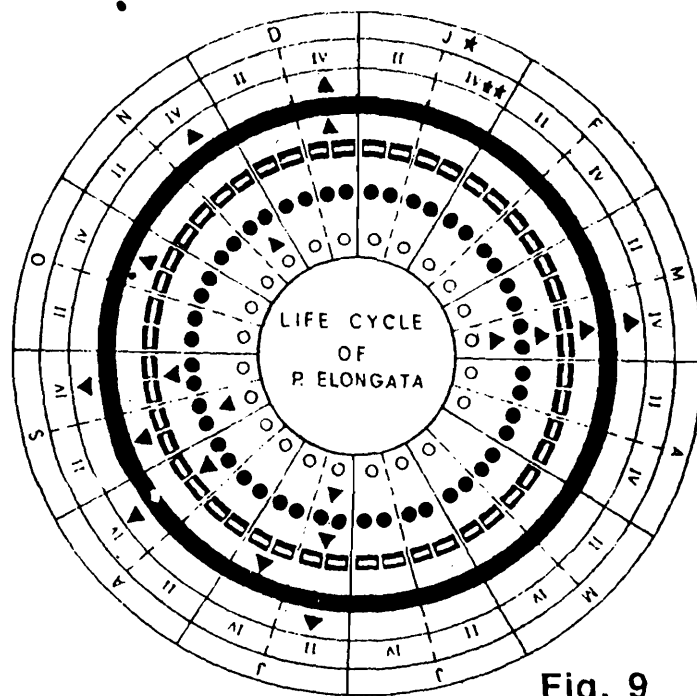


Fig. 9

DIAGRAMATIC REPRESENTATION OF *P. ELONGATA* LIFE CYCLE IN PLAIN PROTECTED PASTURE AT SAMBALPUR, ORISSA, INDIA.  
 ■ ADULT, ▨ IMMATURE, ● JUVENILE, ○ COCOON,  
 □ ACTIVE PERIOD, ▲ PEAK NUMBER, ★ MONTH, ★★ WEEK.

Fig. 8. Diagrammatic Representation of *D. willsi* Life Cycle in Plain Protected Pasture at Sabalpur, Orissa, India.

Fig. 9. Diagrammatic Representation of *P. elongata* Life Cycle in Plain Protected Pasture at Sambalpur, Orissa, India.

< 3 g%). *D. willsi* is a small medium sized (Table 2) epianecic earthworm mostly found at the top 10cm of soil depth and is phytogeophagous (plant and soil eating) in nature. Figure 6 exhibits cocoons and just hatched juveniles of *D. willsi* earthworm. Cocoons of *D. willsi* were available from end of August till mid of March. *D. willsi* showed two cocoon peaks one during September (rainy season) and another during February (post rainy season) (Fig.4). Total cocoon per square meter area was almost double during rainy season than in comparison to post rainy period. Rate of reproduction of *D. willsi* worm has been reported to be maximum of about 2.6 (Sahu, 1989). *D. willsi* showed double cocoon peak (discrete polytelic or interoparous) with bimodal emergence pattern of worms. *P. elongata* is a large (Table 2) deep dwelling endogeic earthworm belonging to 'K' selected species mostly found below 40cm of soil depth and is geophagous (soil eating) in nature. Cocoons of *P. elongata* were available throughout the year. It has exhibited five cocoon peaks (polytelic or interoparous) with multimodal emergence pattern of worm which breeds almost continuously (Fig.1 and 5). In lumbricids, single peak or prolonged emergence has been reported in surface living species whereas deep burrowing species showed either single peak or multiple peak of emergence (Satchell, 1967; Rundgren, 1977). Megascolecoid earthworms exhibit almost a similar manner as that of lumbricids. Interspecific variation and their functional importance with respect to respective strategy has been studied by Lavelle and his coworkers on South African earthworms (Lavelle 1978,1981,1983). Sahu and Senapati (1986,1988,1991) working on Indian earthworms have shown that variation in the reproductive strategy of 'r' selected *D. bolau* and 'K' selected *P. elongata* is an adaptation to cope with unpredictable and predictable environments respectively, indicating better adjustment of *D. bolau* to the changing environment of habitats disturbed by man such as compost pits, croplands, kitchen gardens etc.

### Life Cycle Study and Life Table Analysis in Earthworms

The probable duration for completion of life cycle could be quantified as per the method developed by Senapati *et al* (1979,1980), Dash and Senapati (1980). This is done by determining the time interval between two similar life stages, the latter derived from the former through the process of development. For example, cocoon produced during a particular time period will undergo incubation period, then juvenile will hatch and grow into adult worm through immature stage and finally resulting in cocoon production. The probable time period upto which the young hatched worm is expected to live is known as the expected life span. Life cycle of any earthworm to live is known as the expected life span. Life cycle of any earthworm could be determined in culture pots or from the field information. Diagrammatic representation of *D. bolau*, *D. willsi* and *P. elongata* life cycle has been given in figure 7,8 and 9, respectively. Life cycle of about 3 to 4 months in *D. bolau* at upland grazed pasture, 5 to 6 months in *D. willsi* and about more than a year in *P. elongata* at plain protected pasture has been reported by Sahu and Senapati (1986,1988,1991) (Fig. 7,8 & 9). Laboratory culture study has indicated life cycle period of around 1.5 months for *D. bolau*, 3 months for *D. willsi* and 7 months for *P. elongata* (Sahu, 1989). Life cycle period in field and laboratory condition has exhibited significant positive correlation ( $r = 0.98, p < 0.01$ ;  $r = 0.93, p < 0.01$ ) with worm body size.

Life table parameters could be quantified as per Deevey (1947) and Southwood (1966) considering the following columns:

$x$  = the pivotal age for the age class in units of time;  $l_x$  = the number surviving at the beginning of the age class  $x$  (out of a thousand originally born);  $dx$  = the number dying during the age interval  $x$ ;  $L_x$  = the number of animals alive between age  $x$  and  $x+1$  is calculated from the formula :  $l_x + l_{x+1} / 2$ ;  $T_x$  = total number of animal of  $x$  age units which survive beyond the age  $x$  and is quantified from:  $L_x + L_{x+1} + \dots + L_w$  where  $w$  = the last age, this is found by summing the  $L_x$  column from the bottom upwards;  $e_x$  = the expectation of life remaining for individuals of age  $x$  is calculated by using the formula:  $T_x/l_x$ . Demographic index (D.I.) could be calculated using the formula  $10^3 F/Ge_x$  where  $G$  = duration of growth period (interval between

juvenile to adult),  $F$  = number of cocoons produced per adult per year,  $e_x$ : expectation of life at hatching using above formula. Demographic characteristics of some tropical earthworms from S. Africa and India has been given in Table 5. Ecological category of these worms is mentioned in Table 2. Duration of growth period in some Indian earthworms varies minimally from 2.5 to 8.5 months whereas in South African worms it ranges from 15 to 42 months. Maximum number of about 47 cocoons could be produced by epigeic *D. bolawi* earthworm whereas it is about 15 to 19 cocoons in epianecic and endogeic earthworms (Table 5). Expectation of life at hatching is minimum for epigeic species. Demographic index is maximum in *D. bolawi* than in comparison to endogeic species like *P. elongata*. So it is possible to distinguish different ecological category of earthworm on the basis of the life table analysis and to predict the activity (Senapati and Dash, 1991; Lavelle, 1983).

### Future line of work

Out of about 374 species of earthworms reported in India, information on the reproductive biology of about 20 earthworms are only known. It is important to study the reproductive biology of each earthworm species available in different parts of the country. This will be helpful not only for biology teaching but also for application of biology for human welfare. Vermiculture based industries could manipulate reproductive activity for greater benefit. Some of the important tasks before the earthworm research workers in reproductive biology study are : (i) species wise characterisation of reproductive status (ii) possibility of utilising reproductive structure and cocoon for earthworm identification and classification (iii) selection of prolific breeders (iv) intraspecific variation of reproductive status basing on environmental factors, (v) biology of cocoon development and chromosomal analysis, (vi) identification of food additives and hormonal treatment for production of giant earthworm, (vii) development of methodology for utilization of earthworm cocoon for toxicity tests and cocoon production for stress impact, (viii) identification of acute, chronic and sublethal toxic agents for earthworm growth and reproduction, (ix) quantification of energy cost of reproduction and (x) process of cocoon storage and transport.

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## **Earthworm Gut Contents and Its Significance**

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Earthworms play an important role in the decomposition of organic matter and soil metabolism. Their contribution is mainly through : physical participation by feeding, fragmentation, aeration, turnover and dispersion; chemical participation by digesting organic substrate and contributing nutrients to soils; biological participation by grazing over microflora. An ecosystem functions through material cycling and energy flow. Trophic function of the heterotroph is important in regulating the ecosystem. On the basis of feeding habits, Evans and Guild (1947) were the first to distinguish earthworms into two distinct ecological categories of litter and soil feeding worms. Morphological information is available in support of ecological groupings of earthworms. Litter feeding group possesses active gizzard/s and soil feeding group has a well-developed active typhlosole. There is variation in the gut contents which could be utilised as tools for determining functional establishment of worms. Knowledge about different types of enzymes and microflora in the gut of earthworms is very essential to understand precise role of worms in the decomposition of organic matter and soil formation.

### **Earthworm Gut Morphology and Contents**

The earthworm gut is basically a straight tube extending from the mouth to the anus, its different regions are a muscular pharynx, oesophagus, intestine and associated digestive glands. The oesophagus may be further differentiated into two bulbous chambers, a muscular gizzard and thin-walled crop. There may be more than one gizzard depending upon the species. Various modifications in the digestive system in different worms depend upon the food taken by them. Gizzard is generally absent or rudimentary in earthworms which thrive on liquid or semi-liquid diet. Litter feeding species lack a typhlosole whereas it is well-developed in soil feeding worms. The gut contents usually comprise mucus, organic and mineral matter and organismal components (microflora and microfauna). A variety of digestive enzymes have also been reported from the alimentary canal of earthworms. These enzymes are usually correlated to the preferred diet of an organism (Wallwork, 1984). For example, many soil saprophages, such as oribatid mites, that feed on fungi possess trehalase. Earthworms, which feed on plant material other than fungi, do not have trehalase.

Analysis of gut contents in earthworms has revealed the occurrence of different kinds of symbionts like microfungi, bacteria, protozoans, etc. Maximum number of microfungi species are found in the foregut, gradually decreasing in number in the mid and hind gut with a minimum number in freshly laid casts (Dash *et al.*, 1985). The microfungi composition varies between species. It is well established that the earthworm gut provides suitable condition for the development of bacterial colonies since earthworm castings contain significantly higher counts of bacteria than in the surrounding soil. According to Atlavinyte and Lugauskas (1971), earthworms

increased the number of micro-organisms in soil as much as five times. Earthworms are, therefore, important in inoculating the soil with micro-organisms and their casts are foci for dissemination of soil microbes. They feed selectively on microflora by grazing over soil particles, thereby affecting the condition of soil (Senapati *et al.*, 1980). Different kinds of nematodes have also been recorded in their gut contents.

### Trophic Habits of Earthworms

Based on trophic functions, earthworms have been distinguished into three distinct ecological types by Bouche (1977), which represent different responses to soil constraints and possibly successive steps in earthworm evolution. Epigeic worms are small in size with limited period of activity and high mortality but balanced by high rate of reproduction. They subsist on high quality resources (litter and other organic matter). Endogeic worms are soil dwelling where they find buffered and predictable conditions with low quality of food. The third category of worms are aneciques and are intermediate between the litter and soil dwelling forms. They come to the surface for feeding and defecation but live in soil burrows. They have strong anterior muscles which help in burrowing, and maintain an extensive burrow system.

Earthworms in general are saprophages. They feed mainly on organic matter, usually the decomposed parts of plants, although micro-organisms of various kinds may also be ingested. Certain lumbricid species prefer food rich in nitrogen and sugar and low in polyphenols. Conifer needles and oak leaves with higher carbon/nitrogen ratios and polyphenol contents are not preferred. The intake of mineral soil alongwith organic matter may vary. Both epigeic and endogeic species ingest organic and mineral particles in an advanced state of decomposition (Wallwork, 1984). Deep burrowing aneciques emerge at night and draw down fragments of decaying leaves into their burrows where they are stored for sufficient decomposition before ingestion.

### Digestion

The ingested food in pharynx is subjected to acid mucus containing an amylase which is secreted by ductless pharyngeal glands. Muscular gizzard helps in fragmenting and grinding the food into small particles. In some species, calciferous glands discharge amorphous calcium carbonate particles coated with mucus into oesophagus. The function of this secretion is unknown but possibly it influences the pH of the intestinal fluid. Chemical changes in the degradation of organic material is carried through the enzymetic digestion, enrichment by nitrogen excrements, transport of organic and inorganic materials. Presence of digestive enzymes like amylase, cellulase, protease, lipase, chitinase and lichenase in the intestine signifies digestive ability of earthworms. These enzymes operate in a medium with a remarkable stable pH ranging between 6.3 and 7.3 throughout the length of the intestine which encourage the growth of bacterial colonies (Wallwork, 1984). Possibly, certain species of actinomycetes in the earthworm gut participate in the chemical transformation of organic materials, formation of clay/humus complexes, and production of cementing substances which improve the crumb structure of the soil. The gut communities of earthworms, therefore, play an important role in the decomposer system of the soil.

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## **Regeneration, Predators and Parasites of Earthworms**

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The architectural complexity achieved by an organism in structural organization in the evolutionary process has brought about limitation in the regeneration capacity of lost parts. The planarians, which are the simplest organisms with organ grade of organization have the maximum regenerative capacity and maintain an anteroposterior gradient during regeneration of lost parts.

Annelid worms, being the first group of coelomates with metameric segmentation and simple body plan possess a high degree of regeneration capacity. Various factors including intrinsic and extrinsic are involved in the regeneration process. Just as in planarians, an anteroposterior gradient may exist in these worms. The posterior segments when lost are replaced at a faster rate than the anterior ones. In the posterior region, newly regenerated segments to begin with are narrower than older segments. Only after addition of all the lost segments, they increase in diameter. During regeneration of the posterior end, the tip of the body remains open. Normally, anterior regeneration is a delayed process with the formation of wound tissue. The worm tolerates loss of only a few of anterior segments. Beyond this limit it fails to regenerate lost segments (Edwards and Lofty, 1977).

Earthworms that undergo obligatory or facultative diapause are termed “amphodynamic” These worms regenerate lost segments only during diapause. They have to enter into diapause state for regeneration of lost segments. Most of earthworms respond to dehydration and become quiescent. At this stage, they stop feeding, lose plenty of body water and regression of sexual characters takes place. Species that enter into quiescence state but not diapause are termed “homodynamic” In these species there is temporary sexual regression during the loss of caudal segments. The existence of inverse relationship between sexual reproduction and regeneration may be due to a single hormone regulating the phenomenon. Regeneration is mostly inhibited by secretions of supra-oesophageal ganglion. In case of homodynamic species, the release of this hormone depends on environmental conditions and the presence or absence of caudal injury. In amphodynamic group, the release of hormone determines the physiological state of worms (Mill, 1978).

The existence of an anteroposterior axis gradient for regeneration is an electrically controlled mechanism (Moment, 1953). Earthworms have voltage differences from one end to another and when a worm is cut, the voltage decreases sharply, which returns to original level in three weeks. The regeneration or growth stops after addition of a few segments because each segment contributes certain voltage and segment contributes certain voltage and as soon as voltage stabilises the growth ceases. Conclusive evidence to show the electrical stimulation or inhibition of growth is not available.

The capacity of regeneration of lost segments varies with the species. Normally, the épigéic forms have higher rate of regeneration than the endogéic species. This may again explain the environmental and physiological status of involved organisms. Épigéic worms, which are more prone to predatory attacks and do not enter diapause, may readily regenerate lost segments by altering levels of neurosecretions. Endogéic worms have to make major changes in their physiology to enter into diapause to regain lost segments.

The coelomocytes of earthworms are known to have a role in regeneration, immunity and wound healing. The greater number of cells in coelomic fluid rather than in blood has been attributed to the immune mechanism existing in the worms (Hostetter and Cooper, 1974). Depending upon the intensity of amputation, coelomocytes are forced through the wound by coelomic pressure and a plug appears by the production of pseudopodia, which are overgrown by epidermis and a scar tissue is formed. The scar tissue is later destroyed by phagocytosis of coelomocytes. These degraded tissues are called "blown bodies" (Mill, 1978). The epidermis is highly vascularised and like coelomocytes, haemocytes also contribute to plugging of walls of broken vessels. Thus, different types of free moving cells in the coelomic fluid and blood play significant role in wound healing mechanism.

### Predators of Earthworms

A large number of invertebrates and vertebrates are known to prey upon earthworms (Edwards and Lofty, 1977). Invertebrates include giant flat worms, carnivorous slugs, carabid and staphylinid beetles and centipedes. A carnivorous earthworm, *Agastrodrilus* sp., is reported to feed upon other earthworms. Frogs, toads, apodans, rodents, badgers, foxes, shrews, moles and birds are the major vertebrate predators. Though a long list of predators are known none of these live exclusively on worms. Quantitative data on the amount of earthworms eaten and their significance in the diet of predators are not available. Apodans, *Ichthyophrys* sp., are being maintained exclusively on worms for some other investigations at two laboratories in Karnataka. But no importance has been given to study food intake, assimilation and energetics of the predatory apodans. Fifty to sixty per cent of dry weight of a worm is protein and is found to contain all essential amino and fatty acids to meet the requirements of vertebrates. Observations on prey-predator associations and predation pressure on worm biomass under laboratory and field conditions are required.

Bengtson *et al.* (1976) recorded that predation of golden plover (*Pluvialis apricaria*) on worms in a hayfield in Iceland brought down the prey population to less than 50% when the grass cover was short. Similar observations on predation of gulls and starlings, on worms have also been conducted in New Zealand. Predatory pressure is more for surface living species and as such they have high productivity. Most of the carnivorous and omnivorous birds prey upon worms which constitute at least 5-7% of their diet.

Among mammals, some of insectivores have been recorded to prey upon worms. Most of the shrews feed on worms, though frequency of occurrence of worms in their gut contents may vary. Moles are the other group which feed largely on worms, the latter constituting 100% of their diet in winter and 50% in summer. Moles store worms in their burrows in paralysed condition by cutting off a few anterior segments (Macdonald, 1976). He has also dealt with detailed studies on the foraging behaviour of foxes on worms. They capture worms on small patches during the night with the help of their auditory senses in response to sound produced by worms while moving in soil crevices.

Earthworms are of secondary importance to the amphibians. Toads prey upon them as they surface at nights. Pieces of worms have been recorded from the gut contents of some toads collected from coastal areas of Karnataka (Mangalore). A few species of garter, grass and blind snakes are also known to feed on earthworms (Macdonald, 1983).

### Parasites and Pathogens

A large number of protozoans, nematodes, rotifers, flatworms, mites and dipteran larvae are found in the tissues and body fluids of worms (Edwards and Lofty, 1977). Among Diptera, a cluster fly (*Pollenia rudis*) parasitises the lumbricids. The other parasitic fly reported is *Onesia suvalpina*. *Histostoma murchiei*, a mite, is parasitic on the cocoons of *Allolobophora chlorotica*. Leeches are known to attack worms in vermeries. Some nematodes were found emerging from the

cocoons of *Eudrilus eugeniae* which affected their viability. Incidence of parasitism increased with the age of the host. Different species of worms have been reported to be susceptible to attacks of larvae of *Capillaria annulata*. A rotifer *Albertia vermicularis* was isolated from *Allobophora caliginosa* by Rees (1960). There are records of different species of parasitic nematodes from the coelomic cavity of worms.

Many ciliate and sporozoan parasites have been isolated from the body fluid and various tissues of earthworms (Edwards and Lofty 1977; Segun, 1978). Earthworms are also agents for the spread of parasites, acting as reservoirs or intermediate hosts for many parasites and pathogens. *Enterobacter aerogenes* bacterium isolated from the clitellar region of *Hoplochetella* sp. is found to be fatal to this octochaetid worm (Rao, *et al.*, 1983). *Bacillus thuringiensis*, which is used as a biocide, appears to effect the life of worms at a slightly higher dose than is recommended for insects (Smirnoff and Heimpel, 1961).

For a detailed understanding of the role of earthworms as carriers of diseases of plants and animals, it is very important to undertake an in depth study on soil borne pathogens and parasites of earthworms and other higher animals.

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## **Vermiculture : Scope for New Biotechnology**

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An indiscriminate growth of cities, increase in human population and vast industrialisation have led to an increased accumulation of waste materials. In turn these resulted in the vanishing of fertile lands which used to produce food and raw materials. Under such situations the main objectives of modern civilization is to make any available patch of land to be more resourceful. It is also essential to detoxify the environ which is exposed to hazards of chemical toxicants, and to hasten degradation of organic wastes for their recycling through biological agents. Alongwith the growth of industry, there is a demand for intensifying the animal husbandry, which again leads to a problem of disposal of animal effluents (Hartenstein and Bisesi, 1989).

### **Earthworms in Nutrient Fixation**

Soil improvement is very important to make use of even the less productive land to increase food production for ever increasing human population. Mineral aggregates are more stable in the presence of organic particles. Deficiency in organic carbon reduces storage capacity of soil for nitrogen, sulphur and phosphorus and leads to atmospheric acidity and reduction in soil fertility. The world's labile source of carbon has to be incorporated into soil before it escapes into atmosphere as methane and carbon dioxide. This requires economically and environmentally managed sound procedures. The worm cast in nature is found to contain the highest organic matter content than in those of other major soil fauna like ants and termites. Joshi and Kelkar (1952) have reported a higher electrical conductivity of casts that denotes an increase in the level of soluble salts in soil. They have also observed the casts to have greater nitrifying power than soils. The increased stability of casts and considerable amount of carbon and nitrogen than in the parent soil have been observed (Dutt, 1948; Bhaduria and Ramakrishna, 1989). The water stable aggregates make the soil more aerobic which is considered to be one of the main factors responsible for nitrogen fixation. Though the worm cast is considered to be a strong aggregate, its stability depends on concentration and type of organic matter, bacterial and fungal polysaccharides (Lee, 1985). These findings suggest that earthworms form one of the agents in organic matter breakdown and for rapid incorporation of detritus into the soil strata.

### **Vermicompost a Bio-organic Fertiliser**

Aristotle called earthworms, "The intestines of earth", and considered them as agents to restore soil fertility. Voracious feeding habits of selected species of earthworms on rich source of organic matter and their high reproductive potential are now being exploited in temperate countries. The nature of food and its availability and other physical parameters like temperature, light and moisture content, biological parameters like the density pressure, environmental conditioning created by their own activity influence their growth and fecundity (Evans and Guild, 1948; Mba, 1978, 1983; Neuhauser *et al.*, 1980). Evans and Guild (1948) and Kale and Bano (1985) also showed that the earthworms preferring nitrogen rich diet grow faster and produce more cocoons than those feeding on mineralised soil. Even litter feeding worms show an order of preference for certain leafy matter (Kale and Krishnamoorthy, 1981). The knowledge on the biology, food habits and habitat selections of worms are important factors for encouraging culturing of worms for degradation of animal waste, plant residues and wastes from the food processing units. Biomass

production of worms on one hand helps to enhance process of waste degradation and on the other hand residue free worms can be used as an alternate source of protein for poultry birds and fish.

Some earthworms such as *Eudrilus eugeniae*, *Eisenia fetida* and *Perionyx excavatus* are found to be very efficient and adoptable in cultures under semi-natural conditions in our country. Various agricultural wastes like post-harvest stubbles, sugar cane thrash, coir waste and paper pulp, and faecal matter of cow, sheep, horse, and biogas sludge of poultry droppings have been tried as food source for these worms. It was found that the breakdown of these materials was enhanced considerably in the presence of worms (Kale *et al.*, 1982, in press-1988; Bano and Kale, 1986; Kale and Bano, 1988). Disintegrable plant remains form a good source of manure on exposing them to earthworm activity.

The degraded organic matter by worm activity is called 'Vermicompost' which can be used as top soil or organic manure in fields to prevent organic carbon deficiency and soil erosion. The worm cast is a better source of organic manure over other anaerobically degraded compost because of the following facts. The worm cast is loosely packed granular aggregates of semi-digested matter that provides energy for establishment of various microorganisms. Some of microbes which are found in association with the cast are responsible for deodorising excrements derived from organic wastes with obnoxious odour (Watanabe, *et al.* 1982). The cast also forms suitable base for free living beneficial microbes whose activities are essential for releasing of nutrients to higher plants (Atlavinyte *et al.*, 1971; Atlavinyte and Vanagas, 1982). Thus an establishment of microenviron takes place in the presence of worms in a given media. Specially in tropical countries, earthworms cannot remain active throughout the year. The prevailing environmental conditions and type of soil bring about leaching of nutrients at a rapid rate. The existing microbial population fails to remain active due to lack of energy requirements for their activity. Under such circumstances, regular application of worm casts to fields improves the physico-chemical and biological properties of soil (Kale, *et al.*, 1990 - in press).

The worm activated soil or worm cast provides essential nutrients in available form to plants. Besides bio-chemical activities of established microbes and worm exudates have stimulatory effect on plant growth. Presence of earthworms in culture pots have positive effect on germination, growth and yield of crops. Atlavinyte *et al.*, (1971) have also shown an influence of worm activity on the density of microbes and in vit B12 level in soil. Springett and Syers (1979) showed increase in the herbage production on application of worm cast. The yield influencing substances are released into soil by earthworms, which is a specific character. Irrespective of the species of worms, growth and yield of crops show a definite increase in wormed worked soils than in controls. An increase in protein synthesis of *Agaricus bisporus* and radish *Raphanus sativus* has been reported when grown in the presence of worm cast (Galli *et al.* 1990; Tomati *et al.* 1990). Increase in uptake of nutrients in the level of available nutrients in symbiotic microbial association with cereals and some ornamental plants were observed in our studies (Kale *et al.*, 1987, 1990). These findings authenticate possibility of ameliorating soils by application of worm cast or worm worked soil wherever introduction of worms is not feasible. The culture maintenance and vermicompost production form an independent bio-technological unit for mass production of compost.

### Species Recommended for the Technology

Of many species of earthworms tested for mass cultivation all over the world, including the tropical and temperate regions, *Eisenia fetida*, *Eudrilus eugeniae* and *Perionyx excavatus* come in the order of preference for their ability to degrade the wastes. Their high biomass production may attain an increase of 40 to 90 times in a period of 3 to 6 months with adequate space and food (Neuhauser *et al.*, 1980; Kale *et al.*, 1988; Kale and Bano 1988; Viljeen and Reinecke, 1989; Mba, 1978, 1983; Reinecke and Hallatt, 1989). Frequent harvesting of worms brings down density pressure and enables continuous growth of worm population. An increased demand for fish meal in

livestock farms has resulted in a continuous escalation of the cost of fish meal. With proper management of vermiculture, the worm protein can supplement fish meal and the demand for animal protein can be subdued (Gurrero, 1983; Kale, 1986; Nandeeshha *et al.*, 1988).

Edwards and Thompson (1973) and Ireland (1977) reviewed the effect of various pesticides and heavy metals on earthworms. It is found that worms can act as "bioconcentrators" of heavy metals and other toxic organic compounds. This quality of worms can be made use of to minimise toxins from sewage sludge before applying the same as fertiliser to fields. This will bring down the risk of entry of these pollutants into plant system and then into sequential food chain. When worms are used for this purpose, they should be prevented from entering into food chain as they are found to concentrate very high levels of these toxins in their tissue.

Mass rearing and maintaining worm cultures and tapping of organic wastes for their maintenance has a good scope for developing it as a cottage industry in our country where there is no dearth for organic wastes, congenial climatic conditions and required man power. The tapping of resourceful technology is of utmost importance for the present day as "soil is the placenta of life."

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## **Vermitechnology in India**

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The availability of nutrients for sustained crop production has become a serious constraint in agriculture with the increased cost and shortage of fertilisers. The utilisation of organic wastes through the agency of earthworms is important for developing vermicomposting techniques. Earthworms could be made use of to meet needs for plant nutrients, recycling of biodegradable organic wastes and in solving problems of deteriorating soil conditions. Vermitechnology, is the method of converting wastes into useful products through the action of earthworms. It comprises three main processes :

1. Vermiculture-Rearing of earthworms.
2. Vermicomposting-Biodegradation of waste biomass in earthwormic way.
3. Vermiconservation-Mass maintenance of sustainability of waste lands through earthworms.

Utilisable products and benefits of vermitechnology are waste biomass management, animal protein production, organic pollution abatement, waste land conservation, and land reclamation, production of worm-worked manure, soil fertility and enhancement in plant production.

### **Prospects and Problems of Waste Management and Vermicomposting**

Environmental improvement is an accepted national goal. Because of low population and availability of inexpensive energy and enough raw materials, recycling of used material was not considered necessary in the past. Moreover, the amount of waste produced was within optimal limit and taken care of by nature. Under the present conditions of acute energy crisis and environmental degradation due to steep rise in population, it is very essential to develop suitable technology for recovery of energy from non-conventional sources like organic wastes which were once thought to be of no use. The concept of resource recycling is particularly relevant to agricultural production. The problem of organic recycling in soil improvement and crop production may be tackled by : (i) improvement in the process of composting by reduction in the processing period and enrichment in quality and (ii) utilisation of available organic residues and inorganic wastes in the natural plant production cycle. Waste biomass from domestic, agriculture, urban and industrial sources are the main cause of organic pollution in developing countries. A major portion of refuse, more than 60 per cent, constitutes decomposable materials. In fact, nothing is really waste and much of the waste is "vegetable matter in wrong place." Today's waste may be a raw material in the future and waste of one organism may be energy source for another. India produces about 3,000 million tonnes of organic wastes annually which could be utilised for recovering important resources like fertiliser, fuel, food and fodder. This huge amount of waste has also the potentiality to produce 400 million tonnes of plant nutrients besides biogas and alcohol (Dash and Senapati, 1986).

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\* Expanded by J.M. Julka

There are several way of organic waste biomass treatment and composting is one of the best, suitable and acceptable way for quality environment. Organic manure utilisation is useful in :

1. Narrowing the fertiliser gap upto about 25 per cent.
2. Improvement in water retention capacity and quality of soil by increasing humus content.
3. Besides N.P.K., supply of other essential nutrients,.
4. Reduction in leaching of nutrients.
5. Abatement of organic pollution.

Appropriate disposal of waste is most essential and beneficial from ecological and economical point of view. Decomposers like earthworms are also rate-regulators and bio-catalysts at organismal level. They stimulate composting both in enhancing manurial value and decreasing time. Application of vermicompost and earthworms increases the yield of paddy crops ranging upto 95% in grains and 128% in straw and root [Senapati *et al.*, (1985)]. Earthworm is physically an aerator, crusher and mixer, chemically a degrader and biologically a stimulator in the decomposition subsystem (Senapati and Dash, 1984). Epigeic (surface dwelling) worms, depending upon high quality nutrient, are good biodegraders. Vermicomposting, broadly speaking, involves three main phases :

1. Collection of wastes, shredding, mechanical separation of metal, glass, ceramics, etc. and storage of organic wastes.
2. Composting of organic wastes by earthworms. Organic wastes may be first utilised for production of biogas and residual slurry added to vermicomposting beds.
3. Sorting of large undecomposed wastes which can be used for land filling or reprocessing. Earthworms are harnessed for protein production and vermicompost is used as a biofertiliser.

### **Prospects and Problems of Waste Land Management and Vermiconservation**

There is no simple and short term answer to complex and long term problem of agriculture. Long term strategies and food security cannot only depend upon industrialised agriculture which is energy-wise inefficient and unable to preserve the quality of an ecosystem. Waste lands are increasing through lack of proper management and amount to about 20 per cent of land surface. If implications of high doses of fertilisers and pesticides, industrial wastes, irrigation and deforestation are considered, the proportions of waste lands will increase significantly. This needs biological ways of improving soil system. Endogeic earthworms are potential conservators of these systems through their high efficiency of harvesting energy in nutrient poor soil. On the contrary, epigeic worms do not play any role in soil formation and are of no use in reclamation of waste lands.

It is well known that earthworms enhance soil fertility, and attempts have been made to introduce worms in poor soil or to increase their population by adding organic matter or fertiliser. Earthworms have been used in reclaiming flooded areas that are subsequently drained and put into cultivation (Edwards and Lofty, 1977). These worms have been successfully introduced to newly established areas of artesian irrigation in U.S.S.R. for improving soil formation.

### **Vermitechnology Development at Global and National Levels**

The concept of vermitechnology was started from the middle of the twentieth century. First vermicomposting plant was set up at Hollands Landing, Ontario, Canada. Since then,

vermicomposting has been earnestly undertaken in the United States of America, Italy, Japan, and now being initiated in France, Israel, etc. Alongwith success stories, there are also instances like the Philippines where vermicomposting industries have collapsed because of lack of social acceptance and extension education. Japan procures about 3000 million tonnes of worms from U.S.A. to take care of huge paper and spinning mill wastes. Another 180,000 tonnes/year is required by thousands of eel farmers in that country. Vermiconservation of waste lands is of recent origin and there is a great scope for its development all over the world.

Worm farming does not involve a very skilled technology. Women, young, old, handicapped, literate and illiterate can easily take it up. In India, vermitechnology is still in the developing stage. Dash and Senapati (1988) compared the vermicultural characteristics of 7 Indian species (including two peregrine and commonly cultured species, *Eisenia fetida* and *Eudrilus eugeniae*). Kale and Bano (1986) employed an African worm, *Eudrilus eugeniae*, for degradation of organic wastes. The casts thus obtained were used as bio-organic fertiliser.

Vermitechnology has a bright future in India. Vast resources of unskilled labour and huge quantities of organic wastes could be tapped for this purpose. But important problems of vermitechnology are :

1. Proper species selection.
2. Development of procedure depending on local resources and needs.
3. Integrated agriculture programme alongwith vermitechnology.
4. Land use constrain and marketing, etc.

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## **Selection of Suitable Vermicomposting Species under Indian Conditions**

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Waste materials are increasing enormously with the growth of human population, agriculture and industrialisation. The disposal of these materials has become imperative for a healthy and quality environment. In this regard recycling of utilisable wastes is feasible. Preparation of organic manure from wastes can immensely help in rural based economy. It has been demonstrated that earthworms can process household garbage, city refuse, sewage sludge and waste from paper, wool, and food industries (Hartenstein *et al.*, 1979; Appelhof, 1980; Senapati and Dash, 1982). Earthworms dominate the soil invertebrate biomass (more than 80 per cent) in different ecosystems of the world. Various studies on energy channelisation through earthworms in tropical environs and effect of earthworm gut enzymes on the reduction of carbon/nitrogen ratio in soil show that earthworms can be utilised in the decomposition of waste organic biomass (Dash and Patra, 1977; Senapati and Dash 1982). These organism are able to process 10 to 20% of the net energy input into an ecosystem which is an indication of their importance in the decomposer subecosystem (Senapati and Dash 1984). Vermicomposting is earthwormic way of sanitation measure for waste biomass. More than 500 species of earthworms occur in India. Out of these, vermicomposting potentiality of only 3-4 species is known. It is therefore, very necessary to tap such potentialities of several other species for the development of indigenous vermiculture. It may be possible to select suitable vermicomposting species for different Indian regions with distinctive climates.

### **Biological Scaling Method of Selection**

Functional capability of an organism can be measured by biological scaling procedure. Analysis of interspecific variations in earthworms, determination of their life cycle strategy, selection pressure and niche segregation will provide useful information for vermiculture, vermicomposting and vermifeed preparation.

1. *Allometry* : This relates to relationship between the body size and various biological functions. Byzova (1965) reports an allometric relationship between the body size and rate of respiration among 6 species of earthworms. A high correlation between oxygen consumption and size of pigmented species of earthworms has been observed. On the contrary, this correlation for the unpigmented species is insignificant. Further, small sized pigmented worms with higher metabolic level show a horizontal migration in surface soil as compared to large sized unpigmented ones. Size relationship of earthworms with pH tolerance, sensitivity to ultraviolet rays, nitrogenous excretion and resistance to desiccation have also been demonstrated. Allometric scaling of poikilotherms like earthworms could be made use of in predicting their potentiality and application.

2. *Trophic study* : Decomposers have generally been classified as carnivores, microbivores, and saprovores. They are also subdivided into necrotrophs, biotrophs and saprotrophs. This classification refers to dynamic relationship between a decomposer and its food. Necrotrophs include some herbivores, plant parasitic microbes, predators and microtrophs, and have a strategy of short term exploitation of living organisms which results in rapid depletion of food resources. Biotrophs (root feeding nematodes, etc.) have a long term exploitation of their living food sources that is dependent on continuous existence of the host. Saprotrophs utilise dead food and majority of decomposers fall under this category. Trophic characteristics of different forms of earthworms as categorised by Bouche (1977) can be recognised. Epigeic worms are phytophagous, endogeic worms are geophagous and aneciques being phytogeophagous.

3. *Niche segregation* : Niche was first viewed as the ultimate distributional unit. Ecologically, it was defined as the functional role and position of the organism in its community. Recently, Pianka (1978) has defined the niche as 'all the various ways in which a given organismic unit conforms to its particular environment (an organismic unit is an individual, a population or a species).' Studies on niche segregation of phytophagous, geophagous and phytogeophagous earthworms are useful in identifying suitable species for vermiculture.

4. *Selection pressure* : Ecological studies show two types of distinct population growth equations. Firstly, the populations with unlimited resources and least or no competition, and restricted to a limited favourable time period having maximum population followed by zero or minimum population. Secondly, the populations relate to limited resources and severe competition, and have continuous occurrences. The evolution of earthworms and their present day continuation, recolonisation and establishment are associated with selection pressure of various environmental parameters. Selection pressure seems to have initially favoured the short life cycle of surface feeding worms, and must have swung as the climate ameliorated to favour the slow and resource conserving life style of the soil dwellers.

### 'r' - and 'K' - Selection

Several ecologists in recent years tend to determine the life styles of an organism as the product of one or two kinds of selection, termed 'r' - selection and 'K' - selection (Pianka, 1970). 'r' selection is associated with patchy, heterogeneous, and unpredictable environments with erratic changes in population size, high mortality counteracted by high reproductive rate, small body size and rapid development to maturity. 'K' - selection operates in predictable conditions, where population size is not much influenced by climatic changes, competition is intense and reproductive rates are low and development to maturity is long as individual body size tends to be large.

Epigeic or endogeic earthworms are expected to exhibit many of the attributes associated with 'r' selection, e.g. high productivity and fecundity, short life span and high metabolic rate with small body size. 'r' selected worms are usually early colonisers in newly-created environments. Deep burrowing anecique worms show characteristics of 'K' - selection, e.g. low fecundity, long life span and low metabolic rate with large body size. 'K' - selection worms rarely leave their burrows.

### Implications of Biological Scaling in Vermitechnology

The small body size of epigeic worms associated with high population density, turn over and reproductive rate, and exploitative bioenergetics with low duration of incubation and life cycle, and thriving on high energy substrate are most suitable for vermicomposting. These are also some of the characteristics of 'r' selection species.

An African species, *Eudrilus eugeniae*, and an European worm, *Eisenia fetida*, are being cultured in several parts of the world. These species have also been transported to India, and *Eudrilus eugeniae* is being cultured in South India for producing biofertiliser. Initial studies show that indigenous compost worms like *Perionyx excavatus*, *Perionyx sansibaricus* and *Dichogaster bolau* could also be taken up for vermicomposting. Some species of *Hoplochaetela*, *Drawida*, *Lampito* and *Moniligaster* may also be considered for utilisation in vermicomposting. These species are surface dwellers and deeply pigmented, and morphology of their alimentary canal (reduced or absence of typhlosole) indicated epigeic way of their life. Certain litter dwelling species of *Perionyx*, *Amyntas* and *Megascolex* may also be used in the degradation of organic wastes.

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## **Soil Organisms and Their Role in Humification of Organic Matter**

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Soil is the portion of loose materials which covers the earth's surface on which the plants can grow. The organic nutrients, essential for the growth of plants and animals, are derived from soil. These inorganic constituents occur in the parent rock and during formation of soil these are recombined into forms useable by large number of diverse organisms (Burgess and Raw, 1967; Kuhnelt, 1976).

The solid phase of soil has two main constituents, namely mineral material, derived from some parent material by weathering and organic material. Both of the components undergo a process of decomposition under the action of various physical, chemical and biological agents (Wallwork, 1970).

### **Soil Organisms – the Living Community**

The faunal community in soil is essentially formed of the following elements :

#### *Producers*

The proper soil does not have the producer which can build up organic material out of inorganic matters. The soil surface and the top layers contain autotrophic plants such as green algae, blue-green algae and other micro forms.

#### *Primary consumers*

**Herbivores** : They are poorly represented in the soil. However, some Protozoa (*Amoeba*, *thecamoebae*), snails of the genus *Carychium* and some mites are essentially recognised as algae feeders. The tardigrades feed on moss. Some hemipteran insects and nematodes are efficient root suckers. There are other insects like mole crickets, fly and beetles larvae, wire worms and myriapods which are considered as root nibblers.

**Fungus-feeders** : The mycelia are eaten by a variety of animals like nematodes, some snails and slugs, some myriapods, collembola, a few beetles and oribatid mites.

**Bacteria-feeders** : Many soil Protozoa and nematodes prefer definite species of bacteria.

#### *Detritivores*

**Primary decomposers** : The bacteria, fungi, Protozoa, some dipteran larvae, Isopoda, Mites, nematodes, etc. play very important role as detritivores.

**Secondary decomposers** : The droppings of all the digesters of fresh plant litter are further decomposed by the secondary decomposers. This group includes earthworms, enchytraeids, diplopods, Collembola and some Diptera larvae.

#### *Secondary consumers*

**Carnivores** : A number of soil organisms are carnivores. The most important of these are predaceous Protozoa, enchytraeids, nematodes, mesostigmatid mites, tabanid larvae, snails, beetles, pseudoscorpions, spiders, etc.

## Organic Matter in Soil

The organic matter, the plant and animal materials, ranges from bacteria, fungal hyphae, soft leaf tissues to tough woody substances, single celled Protozoa, nematodes, soft-bodied worms and insect larvae, arthropods with tough exoskeletons to large vertebrate animals and excreta. These complex substances are usually made up of carbohydrates, simple sugars, starch, cellulose, hemicellulose, pectins, gums, mucilage, proteins, fats, oils, waxes, resins, alcohols, aldehydes, ketones, organic acids, lignin, phenols, tannins, hydrocarbons, alkaloids, pigments and many other products.

## Decomposition of Organic Matter

Decomposition may be defined as the mechanical disintegration of dead plant or animal remains leading to the formation of humus, when the gross cell structure is no longer recognisable. It may otherwise be clarified as breaking down of complex organic molecules to carbohydrates, water and mineral components.

*Processes of Decomposition* : The decomposition of organic matter depends on the physico-chemical conditions of the sub-soil as well as on the nature of the organic matter (Dickinson and Pugh, 1974). In wet spongy ground where water prevents access of sufficient air, the process of decomposition is very slow and incomplete resulting in the formation of peat and undecomposed residues accumulate over parent rock. In other case where the condition is opposite to that of peat formation, the process is known as decay. It is carried out by various organisms like bacteria and beetles. The third category of decomposition is known as putrefaction and occurs in a condition where water is abundantly present and air has little or no access. The fourth and the most effective process of decomposition is humification where organic matter decomposes in the presence of adequate water and air. The end product of the transformation is a complex substance called humus.

*Phases of Decomposition* : The plants and small animals having soft tissues are usually decomposed by soil microflora. The tougher and chemically stable tissues are broken down by the action of both soil fauna and microflora (Seastedt, 1984). The organic matter may be decomposed at the soil surface or incorporated directly into the soil for decomposition.

*Decomposition of Plant Organic Matter* : The chemical constituents of plant litter are generally categorised into cellulose, hemicellulose, lignin, water soluble sugars, amino acids and aliphatic acids, ether and alcohol-soluble fats, oils, waxes, resins, pigments and proteins.

The phylloplane fungi attack easily decomposable sugars in the leaf surface. As a result the leaf becomes senescent, the fungi containing cutinase, pectinase and cellulase penetrate the cuticle and disintegrate the cell walls. Micro-organisms like bacteria (*Pseudomonas*, *Arthrobacter*, *Clestridium*, *Bacillus*, *Aerobacter*, etc.) and fungi (*Aspergillus*, *Mucor*, *Penicillium*, *Trichoderma*, *Cladosporium*, etc.) attack the accumulated leaf litter on the forest floor which darkens and becomes weathered. The water soluble substances, mainly sugars, organic acids and polyphenols are leached into the soil.

As the amount of water soluble polyphenols becomes less due to weathering, the litter becomes more palatable to other decomposers. The palatability varies greatly among the soil invertebrates. Increase in polyphenolic materials precipitate protein complexes in leaves and make them less digestible to the soil fauna. If the tannin content is minimum, the phytophagous insects and earthworms become more active. The microbial activity is followed by mechanical disintegration of organic matter through the action of dipteran larvae, Isopoda, mites, nematodes, tardigrades, etc. The carbohydrates like starch are broken down chemically to produce carbon dioxide or methane, alcohol and organic acids. The lignins are transformed into smaller substances by bacteria. The protein is split into amino acids which are either utilized by the organisms as food or are further acted upon by microorganisms giving rise to ammonia and carbon dioxide, alcohols and

organic acids. Other nitrogenous substances are also converted by microorganisms into ammonia and carbon dioxide.

The second phase of decomposition starts in the presence of earthworms, enchytraeids, millipedes, Collembola, some Diptera larvae, rotifers and oribatid mites. The major role performed by these soil invertebrates is in the form of fragmentation of litter. During the initial stages of litter fragmentation, these organisms provide a more suitable physical substrate for microbial growth. However, certain chemical changes occur during litter fragmentation. The enzymes secreted by the symbiotic gut flora in some invertebrates breakdown cellulose. Some animals synthesize humic substances in their digestive tract.

The fragmentation of litter further accelerates microbial invasion and tissue breakdown initiated by the microflora which in turn favours further attack by other soil animals. The microbes again disintegrate the remaining organic material and a complex substance, the humus, is formed and this process is known as humification (Edwards *et al.*, 1970).

The activities of the fauna and micro-flora are complementary and intricately interrelated. The number of microbes is high in soils with earthworms. The breakdown of organic matter into simpler forms and its incorporation into soil largely depends on the feeding by soil animals alternating with growth of microorganisms.

### Breakdown of Cellulose, Hemicellulose and Lignin

**Cellulose** : Nearly 20-40% of litter is cellulose (holocellulose), structurally organised in microfibrils made up of long polymer chains of glucose. Only a few cellulolytic saprophytes can decompose native cellulose. In the beginning an extra-cellular catalyst,  $C_1$ , converts native cellulose to shorter chains of glucose units, which are hydrolysed by a second extracellular enzyme,  $C_x$ , and changed to cellobiose, celotriose or celotetraose. These glucosides are then further hydrolysed by P-glucosidase to simple sugars which are metabolised to carbon dioxide by aerobes or to organic acids and alcohols by anaerobes.

**Hemicellulose** : There is no structural relationship between cellulose and hemicellulose. Hemicellulose is named from their occurrence as an amorphous mass around the cellulose strands. The hemicellulose is broken down by hemicellulose depolymerizing enzymes secreted by the microbes.

**Lignin** : It is considered as the third major constituent of plant litter forming about 5% of succulent plant material and about 15-35% of the woody parts of trees. In fresh plant litter, lignin occurs in the secondary layers of the cell wall and to some extent in the middle lamella. It is quite resistant to microbial attack, as it is surrounded by polysaccharides. In the decomposition of lignin, extensive demethylation occurs forming hydroxyl benzene derivatives which ultimately produce organic acids and can be utilized by the fungus as an energy substrate. The basidiomycetes, ascomycetes, actinomycetes and several groups of bacteria are the main decomposers of lignin.

### Decomposition of Woody Material

The fresh wood is attacked by the beetles (scolytids, buprestids, cerambycids and curculionids) and later by dipteran larvae. Some of the beetles carry fungi which proliferate in the wood. The fungal mycelia move through cracks and often tissues and then insect larvae, especially of the Scarabaeidae, millipedes and isopods attack the moist tissues. The faeces of these animals provide physical substrate for microbial growth. Gradually the wood becomes soft, fragmented and incorporated into the humus layer.

### Decomposition of Animal Material

The decomposition of animal material (carrion) is initiated by bacteria which is followed by the action of flies. The fly larvae bore into the interior and become abundant. They feed on the material liquified by the bacteria and the larval skin secretes a bactericidal chemical which inhibits the bacterial process. The smell emitted from the material attracts other organisms (Protozoa, nematodes, beetles, etc.) present in the soil. The fly larvae leave the material before pupation. The other animals also leave the decomposed material and distribute organic residues into the surrounding soil through their droppings.

### Contribution of Soil Organisms to the Breakdown of Organic Matter

Soil microorganisms and animals contribute to the breakdown of organic matter in various ways :

1. The microorganisms degrade the organic matter through the action of enzymes secreted by them.
2. Animals disintegrate plant and animal tissues and provide suitable substrate for invasion by microorganisms.
3. Animals selectively decompose and chemically change the organic residues.
4. Animals mix the organic matter thoroughly.
5. Animals transform plant residues into humid substances.
6. Animals form complex aggregate of organic matter with the mineral part of soil.

### Role of Earthworms in Humification of Organic Matter

Earthworms play an important role in the breakdown of plant litter and incorporate it into the soil.

Their contribution may be summarised by the following points :

1. *Removal of leaf litter* : The lumbricid earthworms (*Lumbricus terrestris*) are capable of removing upto 90% of leaf and other litter material through consumption from the soil surface in temperate regions.
2. *Litter burial* : The castings of many species of earthworms cover the litter, bringing it closer to litter decomposers.
3. *Fragmentation* : The earthworms and enchytraeids fragment the litter material, increasing the surface area on which others may feed.
4. *Incorporation* : The burrowing activities help in incorporating fragmented and decomposed plant material throughout the soil horizons.
5. *Effect on the C : N ratio* : While passing through the gut of earthworms, the ratio of carbon to nitrogen in the ingested plant material is lowered, so that it can be directly assimilated by plants.
6. *Effect on the soil microflora* : The faeces of earthworms contain more microflora than the surrounding soil.

### Major Factors Influencing Decomposition

*Temperature* : It greatly affects the growth and activity of soil organisms. It is apparently more important as an ecological factor for soil fauna as they can respond to temperature changes. Most of the soil microflora are mesophiles and require maximum and minimum temperature for growth and activity., The thermophilic organisms also need particular range of temperature for growth.

There are many observations which indicate that seasonal changes in temperature are related to decomposition rates.

**Moisture** : The decomposition process is influenced by the amount of moisture in the organic matter as well as in the air. During the drought, decomposition of mull and mor litter is retarded and the number of saprophagous animals are reduced. Several groups of organisms depend on moisture for maintenance of their activity and movement through the soil.

**Soil Atmosphere** : The aerobic organisms require oxygen for respiration and oxidative assimilation. Carbondioxide regulates the pH of microhabitats which influences the growth of many organisms.

**pH** : There are remarkable differences in the microflora and fauna of acid and alkaline soils. It is also observed that many potential decomposer organisms are less active or inactive when the pH is below 5.0.

**Light** : This factor is certainly important for heating the soil but it also directly affects the distribution of soil organisms. The negatively phototactic soil animals are generally absent from the surface layer of soil. The growth of some fungi is also affected by light.

**Nitrogen content** : The decomposers use carbon as an energy source while nitrogen is assimilated into cell proteins and other compounds. Thus in early stages of decomposition high nitrogen content in the organic matter prompts the process.

**Polyphenol content** : In plant litter decomposition, the polyphenols play a major role. The polyhydroxy phenols in plants comprise from 5-15% of their dry weight.

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## **Myriapods in Relation to Soil : Their Identification, Collection and Preservation**

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The myriapods form an important group of soil animals comprising mainly centipedes and millipedes, which occur in association with soil mites, termites, isopods, earthworms, pseudoscorpions, cockroaches, nematodes and molluscs. They have a tendency to remain near the moist surroundings, although – excessive moisture may lead to desiccation and death due to end-osmosis.

The millipedes cause damage to crops only under forced condition of drought and not by choice and are, therefore, known as secondary pests. Nevertheless, like earthworms, they have a definite role in upturning the soil by ingestion followed by defecation, as well as in the disintegration of humus and decomposition of leaf - litter in agricultural fields. In the absence of vegetable matter the millipedes consume soil which is rich in decaying organic matter.

### **Millipedes – As Humus Decomposers**

Diplopods are vegetarians and saprophagous. The litter feeding forms are pillmillipedes, Iulids and flat-back millipedes. The polydesmids or the flat-back millipedes feed on moist decaying leaves along with soil, while some species feed on dead worms, molluscs, insects and vertebrates. The humus and the rotting substances in soil have a texture which is preferred by the millipedes to that of living plant tissues and the animals are attracted to dilute concentration of sugars. Undoubtedly, the use of farmland manure in humid conditions or growing of some humus producing crops greatly increases the population of millipedes in soil, particularly if the soil is not disturbed by ploughing.

### **Chemical Basis for Consummation of Humus**

The preference of decomposing matter by millipedes is co-related with the nitrogen, carbohydrate and moisture contents of the humus. The micro-biota acts to release available food substances and decrease the polyphenol and tanning concentrations. The polyphenol and tanning contents of leaves counters the effect of sugars upon palatability (Sakwa, 1974). The release of simple sugars in the decomposing tissues serves to increase the palatability of leaf-litters and also act as an index for the nutritiveness of the food. The millipedes are, therefore, able to assess the nutritive value of their food in relation to its palatability.

### **Millipedes Attack Crops Under Forced Condition**

The millipedes, like centipedes, remain centered around moist places. The excessive moisture is, however, dangerous because it causes desiccation and death due to end-osmosis. Under conditions of drought these animals may attack the tender growing crops for their moisture requirement, and a return to their normal diet of decomposing matter and humus is most unlikely due to attraction of sugars in plant sap. This is the reason why the myriapods do not normally migrate but remain concentrated around moist place or in case they do not find vegetable crops of

their choice fulfilling their moisture requirements they burrow deep into the soil exhibiting positive geotaxy.

### Millipedes as Soil Turners

While some of the species of millipedes are surface roamers and subsist on top soil, the others are deep burrowers. Nevertheless, both the categories of species play a vital role in turning over the agricultural soil, thereby making it more fertile. The presence of organic matter in the soil greatly influences the surface or burrowing activity of the millipedes. The burrowing activity is extensive in poor soils, while higher surface activity has been reported in soils rich in organic matter. They assimilate most of the organic matter and defecate the soil along with nitrogenous excretory products. Like those of earthworms (Kale and Krisnamoorthy, 1976), the castings of millipedes are rich in humus, urea and ammonia salts. The excrements contain more humus than in the ingested soil. Defecation also dissociates the mineral organic matter complexes from unavailable form into available form (Bano and Krishnamoorthy, 1977).

### Classification

Chilopoda, Diplopoda, Pauropoda and Symphyla were formerly included under a single class Myriapoda (Myrias - thread like; pedes - feet) on the basis of their possessing many legs and the body presenting two major subdivisions, the head and the trunk. These are now recognised widely divergent phylogenetically so as to merit the status under separate classes, which along with the Hexapoda are placed in the subphylum Mandibulata with a pair of jaw - like trophic appendages, the mandibles.

#### *Class Diplopoda*

The name diplopoda means 'double footed' and most of the segments in these animals are provided with two pairs of legs, a condition arising from the confluence of two adjacent tergal segments. A majority of diplopods are included in the subclass Chilognatha and their integument is hard and horny.

#### Subclass 1. Chilognatha

##### Order Oniscomorpha

Family Glomeridae

Family Sphaerotheridae

##### Order Limacomorpha

Family Glomeridesmidae

Family Zephromiodesmidae

Family Polyzoniidae

##### Order Colobognatha

Family Pseudodesmidae

Family Platydesmidae

Family Siphonophoridae

##### Order AscospERMomorpha

Family Chodeumidae

Family Brachychaetomidae

Family Craspedosomidae

Family Heterochodeumidae

- Order Proterospermomorpha
  - Family Polydesmidae
  - Family Cryptodesmidae
  - Family Strongylosomidae
- Order Opisthospermophora
  - Family Stemmiulidae
  - Family Blaniulidae
  - Family Iulidae
  - Family Spirostreptidae
  - Family Spirobolidae
- Order Penicillata
  - Family Polyxenidae

### Subclass Pselognatha

### Class Chilopoda

The centipedes resemble insects more than any other group of land arthropods. They have their body demarcated into two sub sections - the head and the trunk. The head bears a pair of multisegmented antennae and three pairs of mouth parts. Behind the head is the first segment of the body called Basilar Segment - containing appendages called maxillipede or Toxicognaths - a name so given for bearing a pair of poisonous claws. The animal captures its prey with the mandibles, apparently injects the poison through the toxicognaths, immobilises the prey and kills it. At the tip of strong piercing terminal claws are the orifices of the paired venom glands, situated at the base of the maxillipedes.

On the basis of the number of body segments and paired legs, the centipedes are differentiated into four orders belonging to two subclasses. These are :

Subclass	Epimorpha	Body segments complete at hatching time.
Order	Geophilomorpha	Body long, thread-like, segments 31-171 with equal number of paired legs.
Order	Scolopendromorpha	Body long and strong, segments 21-23 with equal number of paired legs.
Subclass	Anamorpha	Body segments incomplete at hatching time.
Order	Lithobiomorpha	Body not very long, segments 15 with equal number of paired legs.
Order	Scutigromorpha	Body short, segments 15 with 12 pairs of fragile legs.

In the present communication the author discusses mainly about the centipedes belonging to the order Scolopendromorpha which is further divided into two families, the Scolopendridae and Cryptopidae. The members of the family Scolopendridae have four ocelli on each side of the cephalic plate below the base of antennae, while those of the family Cryptopidae are blind.

The centipedes are bisexual but copulation in these animals does not seem to occur in strict sense. The males deposit their spermatophores on the ground which later find attachment to the female orifice, accidentally passing over them. Since the seminal receptacles of the adult females are always found to contain spermatozoa, the females have repeatedly produced offspring in captivity.

It, therefore, appears that the centipedes once inseminated carries excessive quantity of sperms, which may probably last for the life time. Whereas millipedes are known to have internal breeding. All the centipedes are oviparous and the females exhibit a tremendous amount of parental care - in guarding their young ones. The eggs are laid in cluster held together by a sticky solution and female keeps the brood spore enclosed by her body and lifted up from the ground within the legs. If the brooding females are disturbed beyond a certain point the female reacts sharply by leaving the area immediately to a safer place or failing in attempt would devour the brood hurriedly but very seldom yielding to the captor. The cannibalism, therefore, is by force and not by choice.

### Methods of Collection and Preservation of Centipedes

Collecting centipedes and locating their possible hide-outs/niches requires a bit of skill coupled with experience. The centipedes though beautiful in looks and colour are repulsive in total appearance. They are found singly under stones, logs, leaf litter, cow dung, flower pots, bark of trees, sheath of banana stems, etc.

The most suitable habitat where one can expect to find them is an area with optimum moisture and temperature conditions near a forest patch, around ponds, lakes or river banks or on the hill slopes, inside termite mounds, etc. They are nocturnal and the most appropriate time for their collection is either early in the morning or in the evening hours. However, in hills it can be collected throughout the day specially on the hill slopes, the area which receives filtered sunlight through tall trees and the floor is covered with leaf litter and the soil is gravel. The stones or the other objects when upturned or removed disturb the animals and being photo-negative and nocturnal they retreat underground and escape in the loose damp soil or leaf litter. They run very fast in a zig-zag manner using pebbles, grass roots or humus as cover for their defence. *Cormocephalus dentipes* Pocock which is very common in Western Himalaya makes a network of fine galleries under stones for its activities and escape.

### *Preservation for the Taxonomic Studies*

While collecting centipedes, extreme care is required to be taken to hold the animal gently through its trunk but firmly with the help of a long forceps to prevent damage to its body. If not properly handled, its walking legs, antennae or anal legs may be damaged or broken; thereby rendering it unfit for taxonomic studies. The centipedes are preserved in 70% alcohol or 2% formalin. The size of the container should be big enough so that the captured animal can move freely within the killing reagent before it dies, and it should not get curled up or coiled. The disadvantage of preserving in small tubes is that it gets coiled, crumpled and when drawn out for study, the specimen may be damaged.

Specimens preserved in 2% formalin should be washed with running water immediately after it is brought to the field laboratory, if possible, within 4-5 hours after collection because the animal preserved in formalin for long becomes hard, brittle and difficult for study. These should be transferred to 70% alcohol for permanent preservation. It is also recommended that a few drops of glycerine should also be added in each collection tube so that the specimens remain soft and in perfect or in more or less fresh state at the time of study. To avoid jumbling of specimens due to shrinking or coiling not more than 2-3 centipedes are preserved in a tube or container.

### *Some Non-Conventional Methods of Preservation*

Besides the routine methods of preservation, it is recommended to preserve centipedes by some non-conventional methods, which though time consuming but are convenient for storage and economical in the use of preservatives and also cause least damage to the specimens. These methods are as follows :

Freshly killed animal is drawn out of the killing jar and placed singly on one side of the microscopic slide (if the animal is small in size), stretched straight and longitudinally placed, keeping the antennae anteriorly, walking legs laterally and anal legs posteriorly directed. On the other side of the slide the collection label is placed and the slide is gently tied with sewing thread. In this manner a number of specimens collected from the same or different locality, can be placed in one large jar filled with 70% alcohol in undamaged and stretched condition for a number of years. Addition of a few drops of glycerine to the preservative is recommended.

The second method for preservation of material collected in the field is more practical because large catches from different localities can be placed together but in a slightly different manner. The freshly killed specimens after being brought to the laboratory are taken out of the killing bottle as early as possible and straightened with the help of forceps (because it is easy to straighten a freshly killed one) and placed on a suitably cut square shaped butter paper sheet, which are then again placed on a thin layer of absorbent cotton also cut squared. It is advised to keep 3 or 4 or even more specimen on each sheet parallelly. The cotton is then rolled along with collection data, like finger rolls. Several such finger-like rolls, soaked in alcohol can then be conveniently put in any polythene bag or jars with more preservative in a routine manner. It is also recommended that such bags or containers be sealed till such time when they are required for study.

### *Collection for Study in Captivity*

For the study of behaviour, ecology or biology in captivity undamaged live material is required. They can be kept alive in a container having damp earth and humus also collected from the actual habitat. The animal can be kept alive for a long time in captivity in a large sized glass trough half filled with soil as above. A wire mesh or a perforated plastic lid, a sieve or a black linen cloth tied with thread round the rims of the glass trough may be used to prevent the animal to escape. The soil is then periodically moistened to its optimum requirement. It may be noted that excessive moisture may lead to fungal infection whereas the lack of it to desiccation. It is also advisable to replace the soil periodically. The best food that the centipedes easily take are the termites. However, earthworms can also be given. Cannibalism exhibited by the centipedes in captivity hampers studies on these animals. Due care is, therefore, to be taken in selecting the size of the glass trough as well as the number of specimens and proper amount of food.

### **Some Interesting Facts and Fallacies About Centipedes**

*The Name Centipede* – A misnomer : ‘Centum’ means hundred and ‘Pedes’ means feet i.e. hundred legs but the number of legs in centipedes varies from 12-171 pairs. All the centipedes do not have hundred pairs of legs and similarly all the millipedes do not have thousand pair of legs. The name centipede, therefore, is a misnomer.

*Centipede Enters The Human Ear – A Myth* : As it is evident from the various vernacular names that the most common myth about them is that the centipede enters a human ear or a nasal passage. This is not a fact. the fact is that the animal is nocturnal in habit and photo-negative in nature. It, therefore, tends to keep itself away from the light to retire in dark places. Then it may be any dark place, why not a human ear or a nasal passage.

*Economic Importance* : Their economic importance has not been appreciated despite the fact that these animals are poisonous and their bite is dangerous. Because of being carnivorous in habit the centipedes are predators on many species of pests of crops such as some millipedes, grubs of some harmful beetles and moths. Even small birds and snakes are reported to have been eaten by the centipedes. On account of retiring habits the centipede tends to escape notice using poison as an organ of defence. There are numerous scattered accounts of the ill effects of their bite. The centipede bite may cause oedaema, lymphangites with inflammation of skin and sub-cutaneous tissue, ulceration and in some cases a localised narcosis takes place. If no first aid is given a gangrenous sloughing may also form.

Since no vaccine or anti-serum against the venom is available application of Tincture Iodine or rubbing in of the crystals of Potassium permanganate or Liquid ammonia after making a nick between the two punctures, is recommended to serve as a first aid. Dressings of Hydrogen peroxide and fomentation or a hot compression, if necessary, is also advised. Anaesthetics used by dentists and Asprin and Cortison may be given (Jangi, 1984).

Since the local infection associated with bite is not uncommon, a routine treatment should include anti-tetanus serum and procaine penicillin (600,000 units). The centipedes otherwise are not naturally vicious. They will not harm the human being and will try to escape than bite. Even the largest among them seldom bite unless molested. The best prevention is, perhaps, to stay out of their way.

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