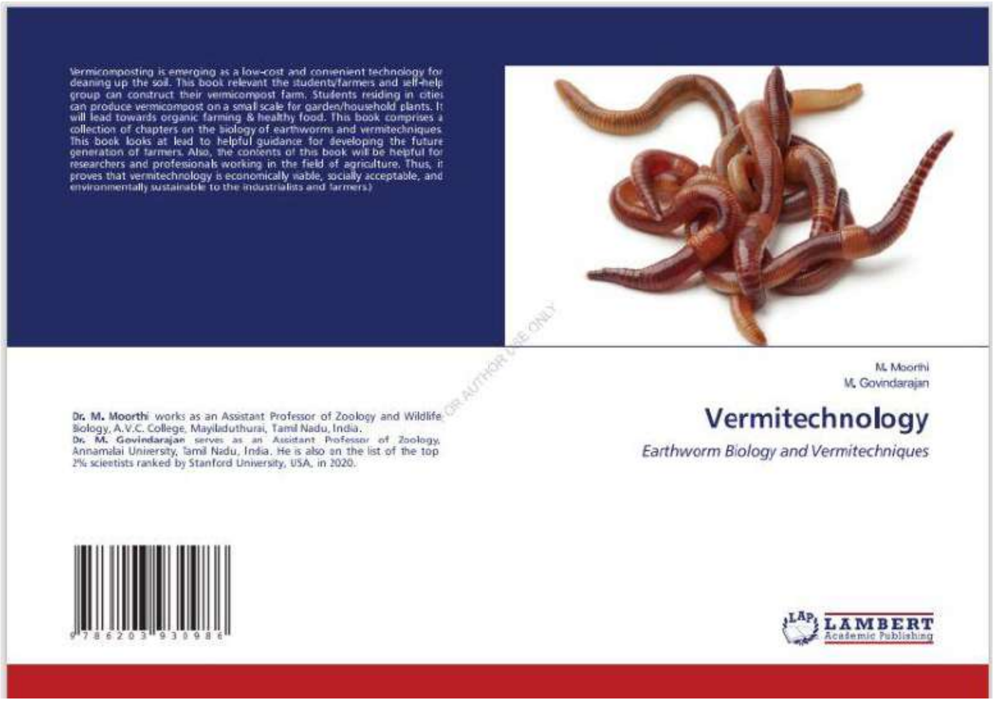
## Books Published




## Book Chapters

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CHAPTER

13

# Pesticides and Earthworms

**V. Kavitha and R. Anandhan**

Earthworms are the major terrestrial macrofauna, constitute more than 80% of the soil invertebrate biomass (Senapati and Dash, 1981; Sorour and Larink, 2001). Reynolds (1994) reported worldwide occurrence of 3,627 terrestrial earthworm species. So far, 402 species (357 native and 45 exotic peregrine species) of earthworms belonging to 66 genera and 10 families are known from India (Julka, 2001). Earthworm is an important soil organism in development and maintenance of nutrient value of soil by converting biodegradable material and organic waste into nutrient rich vermicast (Jansirani *et al*. 2012). They are also known as ecological engineers (Jones *et al*. 1994). Distribution and abundance of earthworms are governed by several ecological parameters viz. soil status, nutrients, temperature, moisture, season, adequate dissolved oxygen, pH and the presence of fertilizers and pesticides (Kale and Krishnamoorthy, 1981; Lee, 1985; Bhaskaran, 1986; Morgan, 1993; Vishwanathan, 1997; Curry, 1998; Bhattacharjee, 2002).

## IMPORTANCE OF EARTHWORMS

Earthworms can consume a wide range of unstable organic matter such as animal waste, industrial waste, sewage sludge, etc. The burrowing activity of earthworms enhances decomposition, formation of humus, development of soil structure, and cycling of nutrients. The product obtained by the modulation of organic waste in the earthworm gut is quite different from its parent waste material and is also known as black gold or vermicast (Lim and Wu 2015). Vermicompost increases the water holding capacity, porosity, and softness of soil thus requiring less tillage and irrigation. It is also rich in microbial diversity, nutrients, plant growth regulators (PGRs) and has properties of inhibiting pathogenic microbes (Mosa *et al*. 2015). Addition of earthworms and vermicompost to soil also maintains an optimum level of soil media in terms of metal concentration, soil porosity and aeration, pH, and electrical

conductivity (Pathma and Sakthivel 2012). Earthworms are important elements of soil biology because they contribute to mineralization and humification of the organic matter and help to change organic wastes into compost on an industrial scale (Reinecke and Reinecke, 1996). Earthworms play important roles in agriculture. They are considered not only as composting agents but also nature's ploughs, aerators, moisture retainers, crushers, and biological agents (Eguchi *et al*., 1995). Hence Aristotle and Darwin have referred earthworms respectively as the **"intestine of the earth"** and **"nature's plough to man"**. Earthworms are used as food for man (by Maoris in New Zealand, Japanese, natives in New Guinea, Africa) and also food for animals because of their high protein content. Earthworms are also used to cure human illness or diseases such as piles, fever, small pox, jaundice and removal of stones in bladder. Earthworm paste and its extract prevent the oxidative damage and also act as anti-ulceral, anti-inflammatory, anti-canceral, anti-coagulant and anti-bacterial agents.

The use of earthworms for toxicity testing is highly recommended by the European Communities (EC, 2004) and are considered as preferred bioindicators of chemical pollution (Reinecke and Reinecke, 2007). As with other organisms, biochemical biomarkers determined in earthworms exposed to contaminated soils provide a measure of pollutant bioavailability and toxic effects (Gambi *et al*., 2007). Earthworms are superb **'barometers'** or **'sentinels'** providing an early warning of deterioration in soil quality. This is important for protecting the health of natural environment and of increasing interest in the context of protecting human health (Beeby, 2001).

## MORPHOECOLOGICAL CLASSIFICATION OF EARTHWORMS

Earthworm species can be classified according to behavioral, morphological or physiological adaptations that enable them to partition available resources in the soil. The three main morphoecological categories are termed as epigeic, anecic, and endogeic (Bouche, 1977; Lee, 1985; Edwards and Bohlen, 1996).

**Epigeic** (soil surface dwelling) worms feed on plant litter, dwell on the soil surface or within the litter layer, tend to be heavily pigmented, and are ranging in size from small to large (*e.g. : Eisenia fetida, Eisenia andrei, Eudrilus eugeniae, Perionyx excavatus* and *Drawida modesta*).

**Anecic** (top soil dwelling) worms feed on plant litter and soil, live in nearly vertical permanent burrows, are dorsally pigmented, and large (*e.g.: Lampito mauritii* and *Octochaetona serrata*).

**Endogeic** (deep burrowing) worms are humus and soil feeders, form extensive horizontal burrow systems, are not heavily pigmented, and range in size from small to large (*e.g.: Octochaetona thurstoni, Allolobophora longa*). Endogeic worms have been further divided into polyhumic, mesohumic and oligohumic groups, which are separated, respectively, by the descending importance of organic rich mineral soil in their diet and increasing size. Earthworm species do not always fall clearly into these three main categories and may even exhibit traits of different groups at different life stages or under different environmental conditions.

Among the three groups of earthworm species, epigeic worms have greater potentiality for degrading organic wastes and endogeic worms have better capacity of protein conservation and soil turnover, whereas anecic worms remain in between these two categories (Dash and Senapati, 1980). Indian earthworms that could possibly be utilized for vermicomposting of organic wastes are *Perionyx excavatus, Lampito mauritii, Dendrobeena repaensis,* and *Metaphis hovlleti* (Kaushal *et al*., 1999).

## SOIL

Soil is a complicated heterogeneous system with the predominance of a solid phase constituted of soil organic matter, minerals, microbes, plants and animals (Nannipieri and Badalucco, 2003). The dynamic nature of soil is due to the activity of macro and microorganisms supported by availability of organic matter (Juma, 1993). Soil physico-chemical properties are the main factors affecting the availability and toxicity of some nutrients and heavy metals (Ge *et al*., 2000). These properties depend on the interactions between soils, microbes, plants, and animals. Some disturbances significantly impact soil biology and can be minimized to reduce their negative effects. These disturbances include compaction, erosion, soil displacement, tillage, catastrophic fires, certain pesticide applications, and excessive pesticide usage.

Earthworms are important in building soil, soil turnover, increasing the soil aeration, water infiltration and retention, maintain the soil fertility and formation of soil aggregates etc. Earthworms participate the soil forming process in five ways such as by influencing soil pH, by causing physical decomposition, by promoting humus formation, by improving soil texture and by enriching the soil with nutrients. Earthworms helps in changing the physical, chemical, and biological properties of soil.

## PESTICIDES

A pesticide is a chemical intended to kill, or destruct the population of a pest organism. Pests are unwanted insects, mites, plants, disease causing organisms, and other organisms that interfere with health or commerce. Insecticides target insects, herbicides target plants, fungicides target disease-causing fungi, and so on. Pesticides have assumed great importance in today's high yielding and intensive agricultural system as well as public health programme, the world over.

## INSECTICIDES

Based on the chemical nature, insecticides are classified into **inorganic** and **organic** insecticides. Several mineral compounds of arsenic, fluorine, copper, mercury, selenium, antimony, zinc, thalium etc., which do not contain carbon in the composition, constitute inorganic insecticides. These materials formed the earliest known insecticides and happened to be widely used before the advent of the synthetic organic insecticides. Organic insecticides are characterized by organic carbon bonding (*i.e.* C-C; C=C) and are classified into two groups : **botanicals** and **synthetic insecticides**.

## BOTANICALS PESTICIDES

The toxic principles are extracted from plants. It has remarkable low toxicity to mammals but toxic to insects eg. Nicotine (Tobacco), Rotenone (*Derris chinensis*), Pyrethrum (*Chrysanthemum coccineum, C.carneum* etc.) and Nimbecidine (Neem, *Azadirachta indica*). Nimbecidine is the only neem oil based phytopesticide with azadirachtin as the labelled active ingredient registered under 9(3) category (permanent registration) of Central Insecticide Board, India [It is also registered by EPA, USA (70387-1)].

## SYNTHETIC INSECTICIDES

Synthetic insecticides are synthesized in the laboratory and are classified into three main groups: **the organochlorines, organophosphorus and carbamates**. Organochlorines are chlorine containing compounds *eg.* endosulfan, aldrin, dieldrin, chlordane, heptachlor etc. Organophosphorus are phosphorus containing compounds which can be an ester of phosphoric acid (P = O) or phosphorothioate acid (P = S) *eg*. monocrotophos, dichlorvos, fenitrothion, parathion, diazinon etc. Carbamates are esters of carbamic acid, HOC (O) $NH_2$ *eg*. carbaryl, carbofuran, aldicarb, bendicarb etc.

The production of pesticides commenced in India in the year 1952 with production of 200 MT and it is increased to 66,470 MT in 1989. In recent years the production of pesticides has been increased to 1,02,740 MT. India also exports the pesticides to various countries. The export per year has been estimated around ` 680 crores. The Indian pesticide market for one year has been estimated to be ` 3,159 crores (Vasantharaj David, 2002). For the present study, widely used organochlorine insecticide endosulfan, organophosphorus insecticide monocrotophos, carbamate insecticide carbaryl and botanical insecticide nimbecidine that are affecting non-target organisms like earthworm have been selected. (Note: detailed information about these insecticides are given in materials and methods – section 3.1.4).

## PESTICIDES CONSUMPTION AND THEIR EFFECT ON NON-TARGET ORGANISM - *LAMPITO MAURITII*

Indian economy largely depends on agriculture. In the agricultural areas of many countries worldwide there is an increasing concern about soil contamination due to the widespread use of agropesticides (Bustos-obreg *et al*., 1998). In developing countries like India where the economy depends largely on agricultural products, one cannot afford to lose the harvest to pests. So the use of pesticides in present day agriculture is imminent for controlling pests and to achieve higher yield. In India, 15-20 % of the total harvest is destroyed by pests resulting in uncontrolled use of pesticides by the Indian farmers. Now a days pesticides become major input in crop production. India is the largest consumer of pesticides in South Asian countries where maximum (44.5%) consumption of the pesticides is by cotton crop. According to the statement of Vasantharaj David (2002), in India the per cent total consumption of agrochemicals (pesticides) are indicated below:

| | | |
|---|---|---|
| (i) Organophosphorus | : | 50% |
| (ii) Synthetic pyrethroids | : | 19% |
| (iii) Organochlorines | : | 16% |
| (iv) Carbamates | : | 4% |
| (v) Biopesticides | : | 1% |
| (vi) Others | : | 10 % |

Now a days earthworms have been shown to be affected by modern agricultural practices such as tillage and applications of pesticides and fertilizers (Edwards and Thompson, 1973; Edwards *et al*., 1995). Pesticides used to kill insect pests are applied in different ways, more commonly as a foliar spray or as a soil dressing (Kidd and James, 1992). Irrespective of place or mode of application, pesticides find their way into soil. Earthworms ingest the soil along with food materials (plant materials) and pass it out as fecal pellets. In doing so, they come in contact with pesticides externally at the body surface and internally through the alimentary canal and become unwanted victims of pesticides used on agricultural land (Lee, 1985; Brunninger *et al*., 1994). So, earthworms have become non-target recipients of many pesticides (Potter and Crenshaw, 1991). Some of the most effective pesticides are broad spectrum in action, and they may inadvently harm earthworms and other beneficial soil organisms and could affect neurological, haematological, biochemical, metabolic and reproductive functions as well as affect animal behavior, physiology, cellular structure etc.

Zhou *et al*. (2011) reported a decrease in growth and reproduction in earthworm *E. fetida andrei* when exposed to concentration of 5 mg/kg of mixture of cypermethrin and chlorpyrifos. Glyphosate and 2,4-D cause severe effects on the development and reproduction of *E. fetida* as reported by Correia and Moreira (2010). Shi-ping *et al*. (2007) reported that chlorpyrifos has an adverse impact on growth and reproduction in earthworms, but this is largely dependent on pesticide concentration and exposure period. Gobi and Gunasekaran (2010) concluded that intoxication of herbicide butachlor consumes the reserve energy from the chloragogen tissue which leads to reduced production of biomass and cocoon production.

Leena *et al*. (2012) reported a profound change in the testes of *E. kinneari* after 20-day exposure of dimethoate, suggesting a disturbance in the cellular enzyme system which in turn interferes with the process of normal gametogenesis. Jordaan *et al*. (2012) reported the effect of pesticide azinphos-methyl on maturation, growth, reproduction, burrowing activity, and ChE inhibition in *E. andrei*. Inhibition in superoxide dismutase (SOD) levels, induction of oxidative stress, and olive tail moments of single-cell gel electrophoresis of coelomocytes indicate DNA damage. Sorour and Larink (2001) reported that benomyl caused noticeable effect in E. fetida on the development and appearance of many spermatozoa; spermatids showing various degrees of hypo development that included some without acrosomes and some with abnormal acrosome development. According to Wang *et al*. (2015), neonicotinoid insecticides such as imidacloprid, acetamiprid, nitenpyram, clothianidin, and thiacloprid are toxic to earthworms and can significantly inhibit fecundity and cellulase activity in *E. fetida* and also damage the epidermal and midgut cells of earthworm. Benomyl is also known for its teratogenic properties, the most common being the disruption of microtubules and thus it is also known as a microtubule poison (Hess and Nakai 2000). In case of carbaryl, cholinesterase (ChE) inhibition was observed even at the lowest dose and the shortest duration of exposure to E. fetida. In addition, the biotransformation enzyme activities are also inhibited by this pesticide, but to a lesser extent. The biochemical responses investigated were sensitive, with evident effects observed even at the lowest concentration (Ribera *et al*. 2001). Capowiez *et al*. (2010) observed that pesticide application causes variation in cast production and nutrient content with variation in concentration of pesticides.

The earthworms have been used as model animals for studying the effects of agrochemicals on soil fauna. Herbicides have been reported to have adverse effect on feeding behaviour, which was reflected in the weight loss and reproduction capacity and survival of earthworms. The herbicide acetochlor caused adverse effect on the sperm number and DNA of Eisenia fetida (Xiao *et al*. 2006). Several studies have demonstrated the lethal activity of herbicides and pesticides on earthworms and histopathological effects (Sorour and Larink 2001; Lydy and Linck 2003; Gobi *et al.* 2004; Rombke *et al.* 2007; Mosieh 2009). Indiscriminate use of pesticides may affect non-target organisms in the soil and can cause serious damage to ecosystem. Smith *et al*. (1992) reported that soil animals, especially earthworms, are one of the best bioindicators of pesticide contamination. The agrochemical concentration is higher in surface layers. Earthworm activity is very much reduced in the soil surface layer (Cock *et al*. 1980).

Certain pesticides inhibit bacterial growth. A study reported the reduction of fungi upon organophosphorus pesticide treatment (Ambrogioni *et al*., 1987). The use of six insecticides in a cotton field reduced the soil fungal population by 75%. The bacterial population in this treated field returned to the levels as seen in the control field after 20 days (Vig *et al*., 2002). Soil treated with triazophos and lindane decreases and subsequently recovers the number of bacteria (Tu, 1978). Kavitha *et al*. (2020) reported that monocrotophos affected the bacterial and fungal population and also damaged intestine of Indian earthworm *L.mauritii* upto 15 days thereafter recovery was observed. Muthukaruppan *et al.* (2005) have reported the glandular cell enlargement in the intestine of the earthworm exposed to sublethal toxicity of herbicide butachlor and they have further observed that changes in the intestinal region may massively affect food intake and which inturn may indirectly inhibit earthworm reproductive capacity. An extreme (2–fold) nuclear swelling has been reported in *E. fetida* exposed to herbicides under different experimental conditions (Molnar, 1992). Gupta and Sundararaman, 1991 have reported the swollen nuclei and loss of chromatin material in carbaryl intoxicated *Pheretima posthuma.* Morowati, 2000 has reported that *Pheretima elongata* exposed to a field dose of herbicide glyphosate showed loss of epithelial cell structure in intestine, lacking regeneration of the cells and total loss of chromatin from first week to the third week of exposure and a marked regeneration of the cells in the fourth week of exposure. Bansiwal and Rai, (2010) observed that sublethal dose of organophosphate insecticide malathion has induced marked pathological changes in the body wall such as ruptured cuticle, with distortion of the shape of longitudinal muscle cells. Oluah *et al*. (2016) have been stated that after exposure to atrazine in the earthworm *Nsukkadeilus mbae*, damages were observed in chloragogenous layer, epithelial tissues, glandular enlargement of the epithelial tissues, prominent vacuolations and pycnotic cells. Kavitha and Anandhan (2018) observed that herbicide atrazine severely damaged intestine of Indian earthworm *L.mauritii* upto 15 days than 30$^{th}$ day of experiment due to microbial degradation of atrazine. Some of bacterial and fungal species in earthworm gut have the capacity to degrade pesticides. It was proved by Kavitha *et al*. (2011). They were observed that *L. mauritii's* gut bacterial and fungal species such as *K. pneumonia*, *E. aerogens*, *E. cloacae, B. subtilus*, *A. fumigatus*, A. *nigar* and *A. flavus* were able to survive and degrade endosulfan after 30 days of exposure, so the earthworms gut microbes might have played a major role in the biodegradation of pesticides. It is also strongly supported the results for recovery of intestinal epithelial lining in the 30th day of atrazine exposure.

## CONCLUSION

Increasing population growth and urbanization, especially in a developing country like India, necessitates producing more food. Food crops require fertile soil to grow. For terrestrial ecosystems, soil serves as a medium of entry to the nutrients. Continuous agricultural activities tend to decrease the soil fertility. However, the increasing application of herbicides and pesticides has also threatened the human environment and the ecosystems with deleterious consequences. The application of pesticides in the agricultural field is slowly eradicated the earthworm population in the soil. In future, the world economy is reduced by gradually decreasing agricultural yield due to infertile soil. Hence, we should avoid the inorganic pesticide application by using organic based pesticides. In future, the application of pesticides should be eliminated the earthworm populations. It was proved by many findings of researchers. The application of organic pesticides is the best remedy to protect our soil fertility.

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