

## Original Research Article

### Toxic effect of Aluminium on reproduction and survival of *Eudrilus eugeniae* (Kinberg) on leaf litter

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#### A B S T R A C T

##### Keywords

*Eudrilus eugeniae*, cocoon, abnormal bulging, emancipation

Earthworms considered as farmers friend and an indicator of soil quality because they contribute enriched soil to agricultural field. These worms are major producers of natural manure without any factories and gift of farmers. Reproduction and survival of earthworms *Eudrilus eugeniae* were measured experimentally by exposing in moist leaf litter with Aluminium. The morphological changes observed during the exposure of animals were observed in laboratory conditions. The Reproduction and survival rate was higher in the control than Al treatments. The activity of Al concentrations is found to influence the size of the population and cocoon production and abnormal bulging of clitellar region and over all emancipation of the size of the body

## Introduction

Earthworms are well known for their ability to integrate the physical, chemical and biological domains of the soil ecosystem (Lee, 1985; Edwards, and Bohlen, 1996). Moreover, earthworms are present in many agro ecosystems where they influence nutrient cycling and hydrologic processes (Shipitalo and Edwards, 1993; Subler, 1997; Anitha, and Prema, 2003)

Vermicomposting, a novel technique of converting decomposable organic wastes into valuable vermicompost through earthworm activity is a faster and better

process when compared with the conventional methods of composting. Within a very short period, the nutrient rich good quality compost is prepared which is highly efficient, cost effective and ecologically sound input for agriculture (Tomati, 1985).

Earthworms have a great ability to consume all organic wastes, reducing their volume by about 50% and expelling the digested material as casting, which is a useful soil amendment and may be easily stored for agricultural use (Cikutovic, 1991).

Earthworms, on the other hand, have been used as biomarkers for assessing environmental pollution (Eduardo, 1998). There is an increasing concern about soil contamination due to the wide spread use of agro pesticides (Rodriguez and Eduardo, 2000). Among them, organo phosphoric compounds have been indicated to cause reproductive damage. Commercial parathion, a highly toxic organophosphate, is used in Latin America, and its application endangers the environment and the public health (Sobarzo and Eduardo, 2000; Rufus and Chaney, 1973)

There are numerous instance of metal toxicity in agricultural. Toxic amounts of Cu, Zn, or Ni have accumulated in soils from fungicides, unneeded fertilizers, and sewage sludge, or have occurred naturally. Most toxicity has occurred under intensive agricultural practices, in orchards, vineyards, and vegetable fields and can be quite expensive to alleviate (Morgan and Morgan, 1999).

The metal (Cd, Cu, Pb, Zn and Ca) concentrations in the tissues, ingesta (crop contents) and egesta (faeces) were investigated in two physiologically contrasting earthworm species (*Lumbricus rubellus* and *Aporrectodea caliginosa*) inhabiting soil exhibiting various levels of heavy metal contamination. In addition, a complementary soil layering experiment, conducted under laboratory conditions, was undertaken to investigate whether the distribution of Pb vertically within a soil profile influenced the relative metal accumulation patterns of these species (Ramalingam, and Thilagar, 2000) The accumulation of Zn and its effects on the growth, reproduction and life cycle of a dominant earthworm species *Drawida willisi* in Indian crop fields.

## Materials and Methods

### Selection and collection of test animals

A bulk sample of exotic, epigeic earthworm, *Eudrilus eugeniae* was obtained from M/S. Kamali farms, Kanuvai, Coimbatore, brought to the laboratory and maintained in glass tanks in vermicompost mixture supplied by the farm. The worms were acclimatized to the laboratory condition for 10 days.

### Preparation of pre-digested leaf litter-cowdung mixture

Leaf litter was periodically collected at different points in Coimbatore Racecourse and brought to laboratory. The leaves were sun dried and chopped into 4 to 5cm pieces. Organic mixture of leaf litter and dried cow dung was prepared at the ratio of 1:1 w/w sprinkled with minimum quantity of pollutant-free tap water and kept in large PVC tanks for days 21 predigestion (Stephenson, 2000). For better pre decomposition regular mixing and turning of the leaf litter-cowdung mixture were followed. After 21 days of pre digestion, required quantity of the predigestion, required quantity of the pre-digested organic mixture was used for mixing with the toxicant for further investigation.

### Selection of aluminium concentrations

For exposure of earthworms to different concentrations of aluminium (Al), aluminium sulphate  $[Al_2(SO_4)_3 \cdot 6H_2O]$  was used. The selection and the range of Al concentrations were based on the direction given in Dir 88/303/EEC (OIL 133, 1988). The Al concentrations selected for the study were 500ppm, 1000ppm, 1500ppm, 2000ppm, 2500ppm and 3000ppm,

respectively under T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>6</sub> test conditions.

### Exposure of worms to Al concentrations

Corresponding amounts of [Al<sub>2</sub> (SO<sub>4</sub>)<sub>3</sub> .6H<sub>2</sub>O] for each test Al concentration was weighed, dissolved in 100ml of toxicant free-tap water (uniformly for all test concentrations) and sprinkled with 2kg of pre-digested leaf litter- cow dung organic mixture. Aluminium added pre-digested mixtures of different concentrations were kept in individual trays (45cm × 30cm × 15cm) and were thoroughly mixed. Into each tray, 10 clitellate *Eudrilus eugeniae* were introduced. 2kg of pre-digested leaf litter- cow dung organic mixture (without aluminium as control) was also kept in similar tray and introduced with ten worms were maintained under laboratory conditions (28 ± 1°C) and at 60 to 70 percent of moisture by regular sprinkling with tap water every day. The trays were individually covered with thin cotton cloth in order to prevent the escape of the worms and also to provide darkness for the worms to work on the mixture. The worms were kept in aluminium- treated conditions for a maximum period of 90 days.

### Observation of worms during Al exposure period

At an interval of every 30 days, worms in each tray (control and treatments) were observed for the morphological changes (if any) due to aluminium treatment. Care was taken to observe the worms with minimum disturbance. At the end of the maximum period of 90 days, the compost mixture in all the trays were separately analysed for the number of adult worms surviving, number of cocoons and number of young ones. The obtained data were recorded and used for further analysis.

### Statistical analysis

Data on adult survival and reproductive parameters were analysed using non-linear regression models as described by Rajendra, 1990. The Karl Pearson's coefficient of correlation (r) surviving number of adults, Al concentration-number of young ones and Al concentration-number of cocoons (Maurya, and Chattoraj,1994)

### Results and Discussion

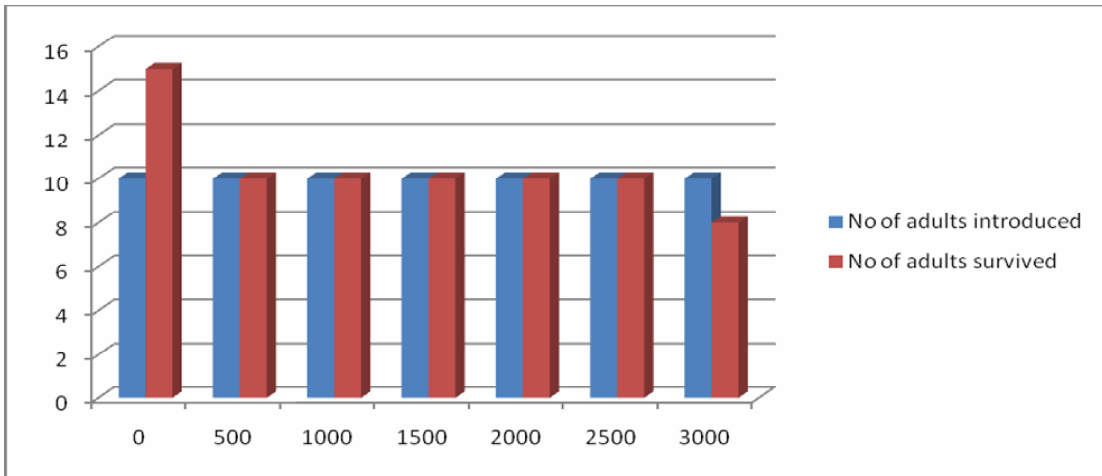
Plate 1 shows the bulk sample of *Eudrilus eugeniae* along with an adult clitellate worm. Plate 2 showed treatments with different concentrations of Al. Plate 3 provides the morphology of *Eudrilus eugeniae* under control and Al concentration treatments following 90days of exposure. Plate 3 shows cocoon from Al treated *Eudrilus eugeniae*. From Plate 1, it could be observed that the epidemic adult earthworm, *Eudrilus eugeniae* showed mass aggregation. The adult *Eudrilus eugeniae* measured about 15 ± 1.25cm.

From Plate 3, showing the texture of control and Al treated *Eudrilus eugeniae* for 90 days, it could be observed that the worm exhibited severe morphological abnormalities under Al – treatment when compared to normal worms. Abnormal bulging of clitellar region and over all emancipation of the size of the body could be observed to be more severe particularly in T<sub>4</sub> and T<sub>6</sub> treatments. Observation of cocoons of Al treated (for 90 days) of *Eudrilus eugeniae* also indicated that Al treated worms were more fragile and less pigmented compared to the cocoons of normal worms.

**Table.1** Data on adult survival, and production of cocoons and young ones of *Eudrilus eugeniae* exposed to aluminium for a prolonged period of 90 days

Aluminium Concentration (ppm)	Number of adults introduced	Number of adults Survived	Number of Cocoons	Number of young ones
0(control)	10	15	40	225
500	10	10	10	118
1000	10	10	2	4
1500	10	10	4	0
2000	10	10	1	0
2500	10	10	1	0
3000	10	8	0	0
'r' value		-0.75	-0.74	-0.79

**Figure.1** Survival of adult *Eudrilus eugeniae* in different concentrations of Al treatment



**Figure.2** Reproduction status of *Eudrilus eugeniae* in different concentrations of Al treatment

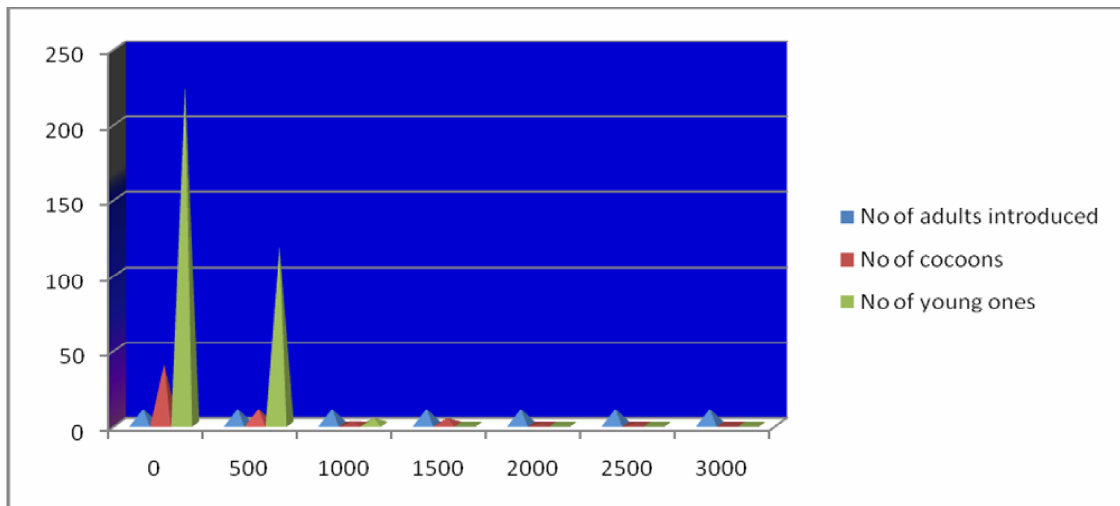


Table 1 and Figure 1 Provides the data on the adult survival and production of cocoons and young ones of control and Al treated *Eudrilus eugeniae* to different concentrations of aluminium for a prolonged period of 90 days. From Table 1, it could be observed that expect 500 ppm Al concentration, higher concentrations (from 1000ppm to 3000ppm) of aluminium appeared to be highly toxic to the worm.

At the maximum Al concentration of 3000 ppm, complete absence of cocoons and young one production besides the death of 2 adult worms were observed and showed in figure 2. The correlation coefficient (r) between Al concentration and adult survival, between Al concentration and cocoon production and between Al concentration and young ones production uniformly ranged between -0.74 to -0.79 (Table 1), there by indicating a negative impact of aluminium on the reproduction of the worms under toxic conditions.

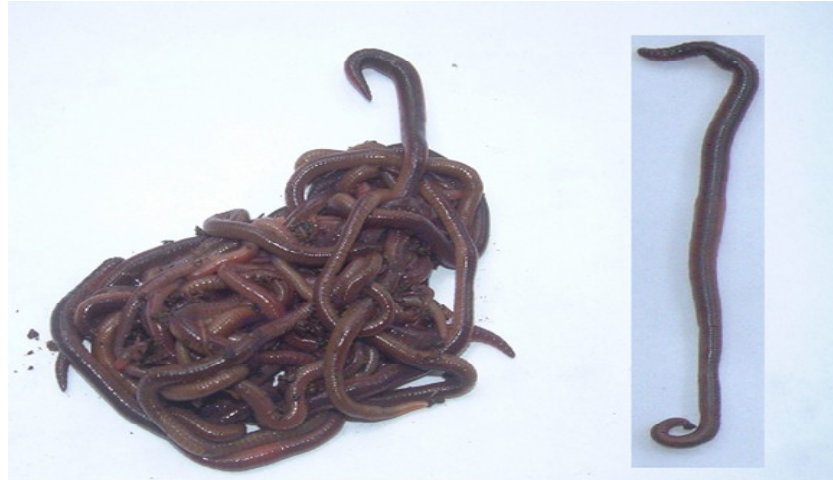
Pesticides and heavy metals are known to cause toxicological symptoms in the body of earthworms. (Venkateswara Rao, 2003) observed oozing of coelomic fluid from the body and swelling of clitellar region and posterior segments in *Lampito mauritii* exposed to different concentrations of fenitrothion. Similar morphological changes involving curling, excessive mucus secretion with bloody lesions and clitellar bulging in *Pheretima posthuma* exposed to aldrin, endosulfon, heptachlor and lindane were reported (Rajendra *et al.*, 1990). *Perionyx sansibaricus* and *Perionyx metaphire* exhibited abnormal symptoms such as body coiling, swelling of clitellar and

other body regions, reddening of dorsal blood vessels, oozing of coelomic fluids, and appearance of ring like constriction and damage of dermal skin when they were exposed to various doses of carbaryl, rogor and lindane (Maurya and Chatterraj, 1994).

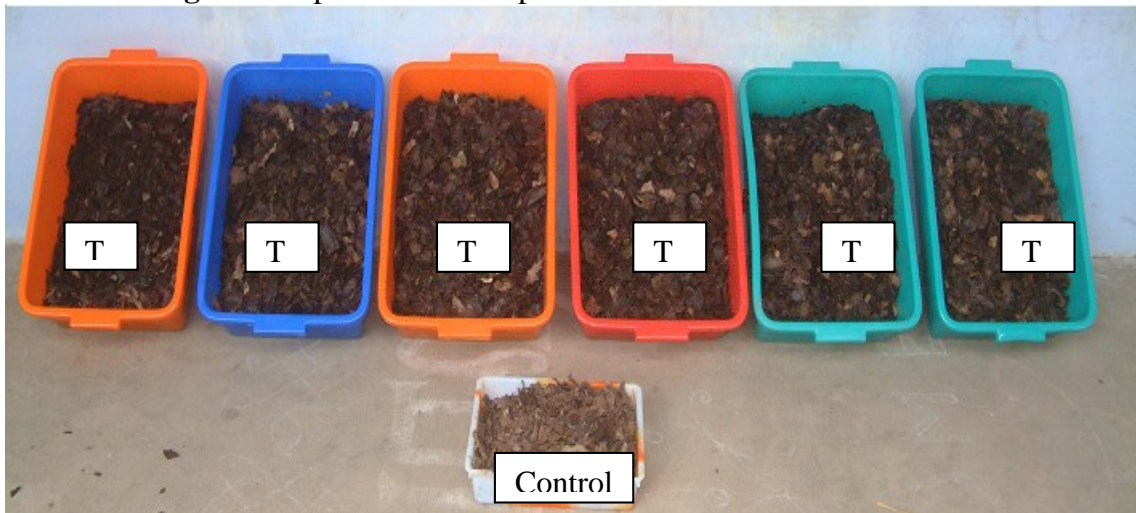
*Eisenia fetida* exposed to chlorpyrifos also showed similar abnormalities (Venkateswara Rao, 2003). In the present study, *Eudrilus eugeniae*, exposed to concentrations of aluminium ranging from 500ppm to 3000ppm (T1 to T6 treatments), exhibited severe morphological abnormalities compared to control (Plate 2). The abnormal bulging of clitellar region and severe emacipation of the size of the body could be observed in worms under higher concentration of aluminium treatments (T4 to T6).

The observed toxicological symptoms such as clitellar bulging and general emacipation of the worm body in *Eudrilus eugeniae* under Al treatment (in the present study) together with similar pathological symptoms reported in different species of earthworms exposed to various toxicants (Rajendra *et al.*, 1990; Maurya and Chatterraj, 1994; Venkateswara Rao, 2003). The reproductive ability of *Darwida willisi* at elevated concentrations of Zn resulting in a delay in completion of the life cycle and a decline in the total population (Ramalingam and Thilagar, 2000). Similar observations like unequivocal disturbance in the reproductive ability of the worms made in the present study.

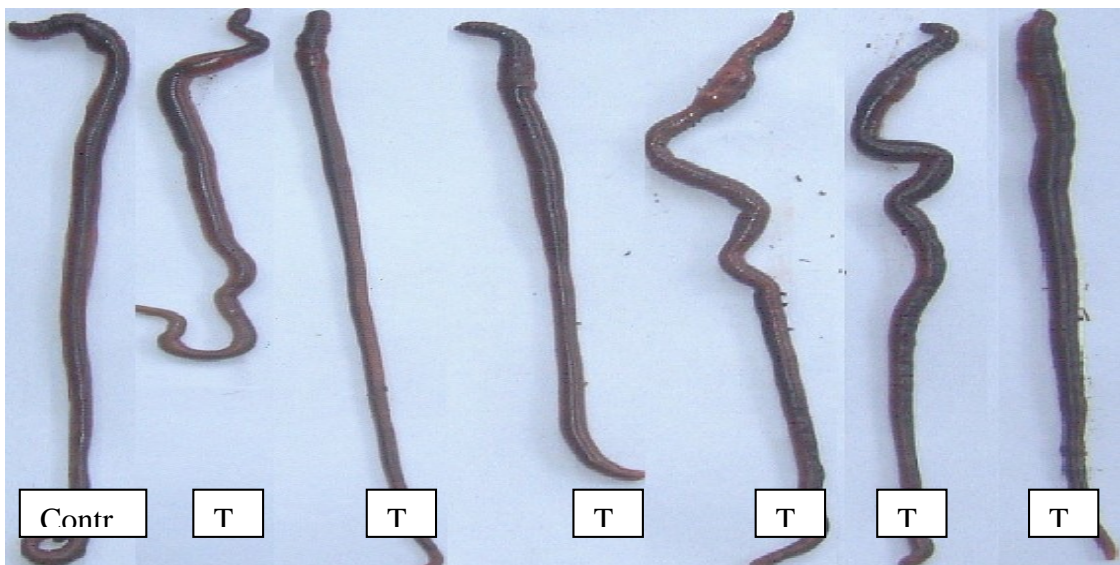
**Figure.1** Bulk sample of *Eudrilus eugeniae* and an adult clitellated worm



**Figure.2** Experimental set up of Al treated earth worm with control



**Figure.3** Morphology of *Eudrilus eugeniae* under control and Al treatments



Environmental degradation is a major threat in the world, and the enormous use of chemical fertilizers and effluent of industrial waste leads to the soil pollution through depletion of fossil fuels, generation of carbon dioxide (CO<sub>2</sub>) and contamination of water resources and effluents from many industries. It leads to loss of soil fertility due to regular use of fertilizers that has adversely impacted agricultural productivity and causes soil degradation and affects non target organisms. Concentration of toxicants affect the fauna of soil ecosystem particularly that of earth worm. The heavy metal, aluminium appears to impair the different stages of reproductive cycle of *Eudrilus eugeniae* under Al concentrations.

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