

RESEARCH ARTICLE

Filter Paper Contact Test Method for Estimation of Toxic Effect of Chloropyriphose on Earthworm, *Eisenia foetida*.

Pawar SS and Ahmad Shahzad*

Department of Biology, Institute of Forensic Science, Nagpur, MS, India- 440001

*Research Students, Department of Zoology, Gov Vidarbha Institute of Science. and Humanities Amravati, MS, India- 444604

*Corresponding Author: ahmadshahzad0023@live.com

ABSTRACT	KEYWORDS
<p>Chloropyriphose (Organophosphate) is widely used by farmers for killing of insect. It is indirectly affects on non target organism like Earthworm which is very use full for maintain the fertility of soil. In the present study, we investigated the effect of chloropyriphose (Organophosphate) on mortality of earthworms. 1% of toxic test solution was prepared form selected pesticide. At the end of the experiment, the toxic concentration of 0.6ml/0.4 ml of distilled water, shown LC₅₀ value of mortality as compare to control set of experiment. The behavior of Earthworm has been changed like body folded and less active as per the increased the dose of the toxicants. We also observed higher toxic concentration chloropyriphose has shown melting and breaks down of earthworm body.</p>	<p>Earthworms, Organophosphate, LC₅₀, plastic box, filter paper etc.</p>

© 2013| Published by IRJSE

INTRODUCTION

Earthworm, *Eisenia foetida* is very useful for fertilizer. Earthworm is friend of farmer. Among the organisms with their living activity in soil, the earthworms are recognized for their important role regarding the improvement of physical and chemical characteristics of soil, and thus increasing its fertility (Abdul and bouche,1997; Aina, 1984; Edwards and Bohlen,1999). Knowing their dynamics in soil under the influence of different technological treatments, or as a result of fertilizers application, it is very important for soil fertility conservation (Aira *et al.*, 2006; Asawalam, 2006). As a good indicator of soil quality, earthworms were used as testing organisms by OECD in early 1980's for the registration of industrial fertilizers and pesticides before implementing them into the soil. More than forty of them are currently registered, although all run the risk of acute and sub acute toxicity. S is used in husbandry, in the habitation lawn, in gardens and in veterinary practice etc. All apparently share a common mechanism of cholinesterase inhibition and can cause similar symptoms of toxicity since they share this line of action, exposure to the same by multiple paths can run to serious additive toxicity or synergistic effect (Savage, *et al.*, 1988). Earthworms, called the "Intestine of Earth" by Aristotle,

are very important soil organisms that help in the disintegration of plant litter, such as the thatch layer, and in recycling of nutrients. (Waste) enrich the soil. Soil organisms can provide valuable indicators by which to study these issues. Recent developments in national and international legislation have sharpened the need for reliable, sensitive indicator organisms to use in research, monitoring, and regulatory testing.

MATERIALS AND METHODS

Experimental Animals- Earthworm (*Eisenia foetida*) brought from commercial supplier, Nursery Department of Forest, Amravati. This species was reared in the laboratory and adopted as the test species, recommended species by (OECD, 1984) guideline for testing of chemicals no. 207, earthworm, acute toxicity tests. We were used adult earthworm which was having above 250- 300mg of body weight.

2) **Pesticide-** We was selected chloropyriphose (Organophosphate) as test insecticide brought from agriculture pesticide market of Amravati. 1% solution of this test chemical was prepared in distilled water for the assessment of LC₅₀ mortality.

3) **Acute toxicity test** -Acute toxicity test was performed following the method described in the OECD (1984) guideline for testing of chemicals no. 207. This is a simple screening test to identify the toxic potential of the chemical to earthworm. The test petridish were a plastic round box which was transparent of 14cm diameter and 2cm height. Round filter papers (Whatman No. 1) were cut to the suitable size and placed in such a way that all sides were covered with filter paper. From the prepared test solution, experimental 10 boxes were arranged and in each box sprayed 0.1ml to 1ml test solution respectively. One blank test were prepared with 1ml of deionized water and used as a control. For each treatment, 3 replicates were used, each consisting of one earthworm per round box. Adult earthworms, which possessed clitellum and had an individual wet weight of 250–350mg, were selected for testing. Earthworms were washed briefly with deionized water, and were kept on moist filter paper for 3h to devoid the gut content, after which it was rinsed again with deionized water, blotted on the filter paper and placed in a test round box. An earthworm was introduced to each round box and the round boxes were covered with plastic film that had been punched with small holes by using needles. Tests were done in the dark at 28 ± 2 °C for 48 h. After 48 hours the earthworm was monitored for mortality by a gentle mechanical stimulus to the front part.

4) **Statistical analysis**- For the filter paper contact test method, based on the resulting 48h LC 50 values, the fertilizer will be classified as supertoxic (<1 ml/1ml), extremely toxic (0.8–0.9 ml/1ml), very toxic (0.6–0.7 ml/1ml), relatively nontoxic (>0.5 ml/1ml) (Roberts and Dorough, 1984).

RESULTS AND DISCUSSION

The earthworms are farmer's friend. Widely used for maintain the fertility of soil. The species *Eisenia foetida* is experimental animal recommended by (OECD, 1984). In the present experiment, we were used filter contact test method for the assessment of toxic effects of chloropyriphose (Organophosphate) on earthworm, *Eisenia foetida*. Experiment were set in three replicates, each replicate contain 10 boxes with earthworm species as a test animals and one box used as a control. 1% solution of chloropyriphose sprayed in 10 round boxes and each box contain 1 number of earthworm in one set shown (fig-1). 0.1ml – 1ml test solution compare with control which was having 1ml distil water (fig-2).

We were observed that the LC₅₀ mortality of chloropyriphose (Organophosphate) was found 0.6ml/0.4ml distilled water in contact filter paper test method shown. Therefore 0.6ml concentration of test solution showed very toxic to earthworm. In this

concentration body of earthworm was bending, shrinks and died in all three replicates.

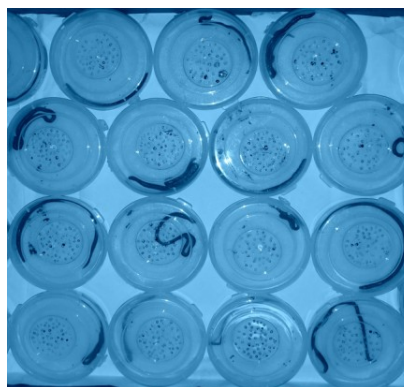


Fig-1

We were used filter paper contact test method for obtaining LC₅₀ for chloropyriphose (Organophosphate). This Organophosphate is very poisonous which affects on body of earthworm and Growth of Earthworm. We were seen in the test solution of 0.8ml, 0.9ml and 1ml of 1% chloropyriphose (Organophosphate), all earthworms were died and melt (fig-3). This concentration was shown supertoxic to earthworms. In the test solutions of 0.1ml and 0.2ml, the earthworms were very active and moving (fig-2). In test solutions of 0.3ml and 0.4ml the earthworm were slightly less active and moving, and in 0.5ml, concentration of test solution earthworm was live and less active was seen in this experiment.



Fig-2

We have used three replica test set for better resulting and calculated mortality. At the end of the experiment, we observed that 0.6 /ml concentration of chloropyriphose were shown LC₅₀ value of mortality. Kovilpathu 2013 was shown the LC₅₀ of superphosphate recorded as 210mg/5ml (i.e. 300 µg/cm²) (Kovilpathu, *et al.*, 2013). In some set like 1ml, 0.9ml was very highly affected on earthworm they have been break, melt and the body was destroying. It was shown directly effect on earthworm's body and growth.

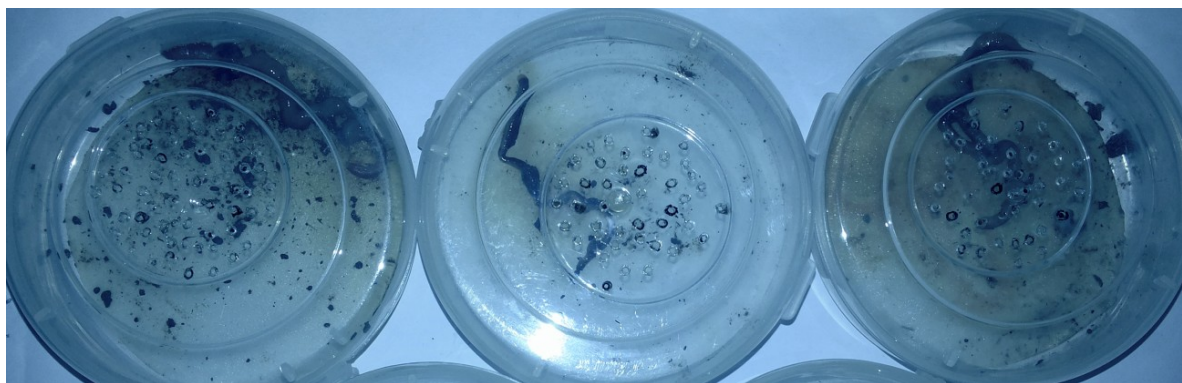


Fig-3

CONCLUSION

Filter paper contact test method is the best method for assessment of insecticide. By this method, we investigated LC₅₀ mortality of chloropyriphose (Organochlorine) on earthworm. Earthworm, *Eisenia foetida* is the test species recommended by OECD. We observed that 0.6ml concentration of test solution shown LC₅₀ mortality on earthworm *Eisenia foetida*. This species were highly affected from test solution concentration 0.6ml to 1ml. Therefore, it was statistically classified as supertoxic to earthworms.

REFERENCES

1. Abdul Rida AM, Bouché MB. Earthworm toxicology: from acute to chronic tests. *Soil Biology and Biochemistry*, 1997; 29, 699.
2. Aina PQ. 1984; *Pedobiologia*. 26, 131.
3. Aira M, Monroy F, Dominguez J.. *European Journal of Soil Biology*, 2006; 42, 127.
4. Anonymous: Annual report 2010-2011, Ministry of Chemicals and Fertilizers, Department of Fertilizers, Government of India, 2011 .
<http://www.fert.gov.in/annualreport/annualreport.asp> [5]
5. Asawalam D O,(2006) *Biology and Fertility of Soils*.42, 506.
6. Edwards CA, and Bohlen PJ. *Biology and Ecology of Earthworms*. 3 rd Edition. Chapman and Hall, London, 1996.
7. Kovilpathu Senthil, Kumar Abbiramy, Pankiras Ronald Ross and Jyothi Pillai Paramanandham. Assessment of acute toxicity of superphosphate to *Eisenia foetida* using paper contact method. *Asian Journal of Plant Science and Research*, 2013, 3(2):112-115
8. OECD, Organization for Economical and Cooperative Development Guideline for Testing of Chemicals, No. 207, Earthworm Acute Toxicity. OECD, Paris, France, 1984.
9. Roberts BL and Dorough HW. *Environ.Toxicol.Chem.*, 1984; 3, 67.
10. Savage E, Keefe T and Mounce K. Chronic neurological sequelae of acute organophosphate pesticide poisoning. *Arch. Environ Health*, 1988; 43: 38-45.

© 2013| Published by IRJSE

Cite this article as: Pawar SS and Ahmad Shahzad. Filter Paper Contact Test Method for Estimation of Toxic Effect of Chloropyriphose on Earthworm, *Eisenia foetida*, *Int. Res. J. of Sci. & Engg.*, 2014; 2(1): 23-25.

Source of Support: Nil,

Conflict of Interest: None declared