HISTOLOGICAL ALTERATIONS IN THE BODY WALL OF EARTHWORM *ALLOLOBOPHORA CALIGINOSA* EXPOSED TO DIMETHOATE INSECTICIDES

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Received: 25 July 2023 / Accepted: 12 August 2023 / Published online: 30 September 2023

Abstract

Dimethoate is an organophosphorus insecticide with contact and systemic action used against a broad range of insects in agriculture. Dimethoate is highly mobile in soil. Its residues have been detected in soils, sediments, and water due to their non-regulated usage practice. Therefore, it affects the beneficial organisms that live in the soil, such as the earthworm. Earthworms are considered as soil engineers because of their ability to modify soils and plant communities, as earthworms ingest large amounts of soil and organic matter, they are continuously exposed to contaminants through their alimentary surfaces. The earthworm *Allolobophora caliginosa* was exposed to Dimethoate and the concentrations were range from 0.05 to 2.5 ppm. The changes in the body wall were represented by the appearance of vacuolated in the epidermis layer and muscle layers, the epidermis separating from the muscle layer then the cells were destroyed, and the effect increased by increasing the concentration of the pesticides.

Keywords: Earthworm, Dimethoate, Histology, Body wall.

Introduction

Dimethoate–DM is, an important organophosphorus (OP) pesticide, is widely used in controlling pests of different cereals, vegetables, fruit, and flies in animal houses in agricultural and sanitary hygiene [1].

It was introduced in 1956 and it is produced and used in many countries against a wide range of insect pests in agriculture, as well as against houseflies [2]. Dimethoate gains entry into the body through ingestion, inhalation, and skin contact [2]. Dimethoate is an organophosphate insecticide used to kill mites and insects systemically and on contact. Its mode of action is through inhibition of acetylcholinesterase. [3] Pesticides are known to produce morphological, anatomical and physiological changes in the vital organs such as reproductive, nervous, respiratory and osmoregulatory of different nontarget animals, such as earthworms and other beneficial organisms like arthropoda organisms. [4]. Like other pesticides, dimethoate also affects many non-target organisms, including soil organisms. In study of [5] Earthworms are terrestrial invertebrates belonging to the Phylum Annelida, Class Chaetopoda, Order Oligochaeta.

They have originated about 600 million years ago during the pre-cambrian era [6].

Earthworms are considered as soil engineers because of their ability to modify soils and plant communities [7, 8].

Earth worms are one of the most important organisms responsible for mechanical mixing of soil and play a major role in maintaining physical soil characteristics and processes such as aeration, water permeability and mineral turnover [9]. Furthermore, earthworms play an important role in the recycling and distribution of nutrients within the soil. They make nitrogen available for plant growth by feeding on organic matter within the soil then voiding casts with a low carbon: nitrogen ratio, the latter often containing fragmented litter which in turn is readily broken down by micro-organisms [10, 11]. organic matter stabilization and soil structure preservation [12, 13].

Earthworms often enhance plant growth [14], and the main mechanism is probably the release of N from organic matter [15]. Litter-feeding species in particular facilitate the transfer [16, 17].

Earthworms usually boost microbial activity, either through ingestion and gut passage, or by mixing soil with
organic matter and providing favorable microhabitats [18, 19, 20].
Earthworm is also used in traditional medical system [21]. The mucous and earthworm paste of Lampito mauritii contain anti-ulceral, anti-oxidative [22], and anti-inflammation properties.
Earthworm fibrinolytic enzyme in Eisenia foetida demonstrates anti-tumor activity on hepatoma cell [23] patterns.

Materials and Methods

Study Site:
The earthworm (Allolobophora caliginosa) was collected from an agricultural area free of pesticides, where the people of the region implant simple crops without pesticides. This region called Reman/ Badan/Ibb

Soil:
The soil was brought from an agricultural area free of pesticides and is totally free from pesticide contamination. Soils were sampled from the surface layers (0-20 cm) of corn fields and it sieved (opening 2 mm). The moisture content of the soil was determined by drying it at 105 °c for 24 h. Water was added to bring the moisture level to 45 percent (45 ml per 100 g of dry soil). also, the pH content of the soil was between 5-6.

Earthworm:
The earthworms A. caliginosa were obtained from an agricultural area free of pesticides. They were carefully brought to the Farm along with the moist soil. was in different age groups therefor maintained in our house with cow dung as a substrate and food. The adult earthworms were removed from culture which had well-developed clitellums (one group) and the juvenile earthworms which hadn’t developed clitellum were removed from culture (two groups) they were used in experiments.

The Experimental animals were divided into two groups (adult group, juvenile group); in order to study the impact of the sub-lethal concentrations (LC 50) and up-lethal concentrations (LC 90) of the dimethoate on the tissues of earthworm (Allolobophora caliginosa).

Fifty of adult earthworm were taken and divided into five groups, each group consisting of ten earthworms, and each group was placed in a different concentration of the dimethoate, and six group without dimethoate (it was control).

Dimethoate was mixed into the natural soil as an aqueous solution to give the desired concentration five concentrations was done by mixing dimethoate insecticides with natural soil.

Each concentration was prepared in kilo grams of the soil and divided into five quantities in plastic glasses. ten worms were added to each glass and covered with cover netting secured with Adhesive to prevent worms from escaping.

The glasses were maintained at room temperature and natural light. The same steps with the juvenile earthworms. It was concentrations (0.05, 0.1, 0.5, 1.0, 2.5) of Dimethoate.

Histological Studies:
The following steps were used in preparation of tissues:
Earthworms were removed from culture after 28 days, rinsed with tap water, and stored in plastic glasses of 5 litter capacity which contain damp cotton for 24 h to avoid the gut contents.
Dissection of earthworms were carried on in the laboratory of biology department, Ibb university, the earthworms were first anesthesia by using chloroform after that was cut ring.
Before histological preparations, the organs (rings) were immersed in saline solution (%0.75) for a few minutes to avoid contractions.
All these rings were then put in fixing solution, which was carny’s fluid for 24 hours When fixation period was completed the rings were put in10% formalin after that the rings tacks of Al- rafa laboratory.

Al- rafa laboratory accoutered slides, sections were prepared and mounted on slides according to the method of [24].

Results

Histological changes of the body wall:
Histological Structure of the body wall of A. caliginosa
The microscopic anatomy of the body wall illustrates that it is built up of outer layer of skin is the cuticle. the cuticle protects the earthworm from dehydration for short periods of time, and provides a tough layer of protection from abrasives contained in the soil through which the earthworm passes. Under the cuticle is the epidermis, this contains some fluids and the outer fastener for a complex of glands and nerves that carry fluid back and forth. Under the epidermis are two layers of muscle – a circular layer and a longitudinal layer, respectively. The circular muscle tissue is a vary thin, weak fiber and the longitudinal muscle is a thick, well-developed layer of actives muscle. These muscles provide the earthworm with its means of locomotion, contraction, and circular movement. The next layer of the body wall is the coelomic Lininger, or peritoneum, which forms the outer wall of the coelom [25].
Fig. 1: Tissue of the body wall (control group) of *Allolobophora caliginosa* with compact and distinct cells and normal nuclei in the Epidermis (Ep) and intact muscle layer circular (Cm) and longitudinal muscle layer (Lm) and goblet cell (GL) (H&E,X160).

The histological changes observed in body wall exposed to 0.05ppm of Dimethoate for 28 days showing vacuolated muscle fiber; vacuolated epidermis , lost epidermis cell in some regions (fig.2) , cuticle layer separating from epidermis and destroyed epidermis (fig.2), rupture longitudinal muscle (fig.3) in adult and the damage was intense with the entire muscle layer being disintegrated (fig.4) and exit muscle nuclei from circular muscle fibers and rippling circular muscle fibers (fig.5) in juvenile.

Fig. 2: Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (0.05 ppm) of Dimethoate to adult (VM= vacuolated muscle fiber, VE = vacuolated Epidermis. Ls = Lost Epidermis.) (H&E, X160).

Fig. 3: Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (0.05 ppm) of Dimethoate to adult (VM= vacuolated muscle fiber, VE = vacuolated Epidermis. Ls = Lost Epidermis, CS= Cuticle layer separating) (H&E,X160).

Fig. 4: Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (0.05 ppm) of Dimethoate to adult (RM= Rupture muscle) (H&E,X160).

Fig. 5: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.05 ppm) of Dimethoate to juvenile (VM= vacuolated muscle fiber.) (H&E,X160).
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When body wall exposed to 0.1 ppm of Dimethoate for 28 days showing vacuolated muscle fiber, vacuolated epidermis and destroyed epidermis in adult. In contrast the nuclei gathered have found in vacuolated which in circular muscles (fig.7), also found vacuolated in longitudinal muscle fibers and peritoneum rupture (fig.8) of adult and was vacuolated muscle fiber, vacuolated epidermis too the epidermis cell layer and longitudinal muscle were completely lost in certain regions in juvenile (fig.9), also found vacuolated in longitudinal muscle and fibers aggregation, circular muscle layer separating from longitudinal muscle layer, rupture in longitudinal muscle (fig.10,11), also found vacuolated in longitudinal muscle and circular muscle and exit muscle nuclei nil (fig.12).

**Fig. 6**: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.05 ppm) of Dimethoate (ExNM= Exit muscle Nuclei, RiM= Rippling circular muscle fibers) (H&E,X160).

**Fig. 7**: Histological changes in transverse sections of epidermis and body wall in adult *Allolobophora caliginosa* exposed to (0.1 ppm) of Dimethoate (VM= vacuolated muscle fiber, VE = vacuolated Epidermis, DE= destroyed Epidermis, N= nuclei) (H&E,X160).

**Fig. 8**: Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (0.1 ppm) of Dimethoate to adult (RP = Peritoneum Rupture, VM = Vacuolated Muscle) (H&E,X160).

**Fig. 9**: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.1 ppm) of Dimethoate (VM= vacuolated muscle fiber, VE = vacuolated Epidermis, LE= Lost Epidermis, LM = lost muscle fiber) (H&E,X160).

**Fig. 10**: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.1 ppm) of Dimethoate (VM= Vacuolated Muscle, RM= Rupture muscle, MS= Muscle layer circular separating from longitudinal muscle layer, Am= Aggregation muscle fiber) (H&E,X64).
Fig. 11: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.1 ppm) of Dimethoate (VM= Vacuolated Muscle, RM= Rupture muscle, MS= Muscle layer circular separating from longitudinal muscle layer, Am= Aggregation muscle fiber) (H&E,X160).

Fig. 12: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.1 ppm) of Dimethoate (VM= Vacuolated Muscle, VE= Vacuolated Epidermis, ExNM= Exit muscle Nuclei) (H&E,X160).

In body wall exposed to 0.5 ppm of Dimethoate for 28 days showing epidermis separating from the muscle layer and vacuolated muscle fiber(fig.13), Epidermis cells Elongate, Epidermis cells shrinkage, Rippling circular muscle fibers and Vacuolated Muscle (fig.14), Vacuolated Epidermis, Vacuolated Muscle, Rupture muscle and Nuclei Enlarge (fig.15) in adult and also found epidermis separating from the muscle layer and vacuolated muscle fiber(fig.16), Epidermis Inducting and Circular muscle overlap Epidermis (fig.17), Epidermis Inducting, Epidermis Mergers (fig.18) and Epidermis Separating from the muscle layer, Destroyed Epidermis, Circular muscle overlap Epidermis, Epidermis Inducting, Epidermis Mergers and Exit muscle Nuclei(fig.19) in juvenile earthworm.

Fig. 13: Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate (VM= vacuolated muscle fibre, ES = Epidermis separating from the muscle layer) (H&E,X160).

Fig. 14: Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate to adult (EE = Epidermis Elongate, ESh = Epidermis shrinkage, RiM= Rippling circular muscle fibers,VM= Vacuolated Muscle) (H&E,X160).

Fig. 15: Histological changes in transverse sections of epidermis and body wall of *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate to adult (VE= Vacuolated Epidermis, VM= Vacuolated Muscle, RM= Rupture muscle, NEn= Nuclei Enlarge) (H&E,X160).
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In body wall exposed to 1.0 ppm of Dimethoate for 28 days the histological Changes are very different compared with control; showing Muscle layer circular separating from longitudinal muscle layer and Vacuolated in Muscle, Exit muscle Nuclei (fig 20) and Vacuolated Muscle, epidermis Inducting, Pyrolyse muscle and Peritoneum Rupture(fig 21) in adult also found epidermis cells Elongate and shrinkage and Exit muscle Nuclei (fig 22), epidermis cells Elongate and shrinkage and Destroyed muscle (fig 23) epidermis cells Elongate and shrinkage, Vacuolated epidermis and Muscle, epidermis Mergers, Exit muscle Nuclei, epidermis Inducting(fig 24) in juvenile earthworm.

**Fig. 16:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate (ES = Epidermis separating from the muscle layer, VM= vacuolated muscle fiber) (H&E,X160).

**Fig. 17:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate (EI= Epidermis Inducting, MO= Circular muscle overlap Epidermis) (H&E,X160).

**Fig. 18:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate (EI= Epidermis Inducting, EMe= Epidermis Mergers) (H&E,X160).

**Fig. 19:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (0.5 ppm) of Dimethoate (ES = Epithelium Separating from the muscle layer, DE= Destroyed Epidermis, MO= Circular muscle overlap Epidermis, EI= Epidermis Inducting, EMe= Epidermis Mergers, ExNM= Exit muscle Nuclei) (H&E,X640).

**Fig. 20:** Histological changes in transverse sections of epidermis and body wall of *Allolobophora caliginosa* exposed to (1 ppm) of Dimethoate to adult (MS= Muscle layer circular separating from longitudinal muscle layer, VM= Vacuolated Muscle, ExNM= Exit muscle Nuclei) (H&E,X160).
**Fig. 21:** Histological changes in transverse sections of epidermis and body wall of *Allolobophora caliginosa* exposed to (1 ppm) of Dimethoate to adult (VM= Vacuolated Muscle, EI= Epidermis Inducting, MP = Pyrolyse muscle, RP= Peritoneum Rupture) (H&E.X160).

**Fig. 22:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (1 ppm) of Dimethoate (EE= Epidermis Elongate, ESh= Epidermis shrinkage, ExNM= Exit muscle Nuclei) (H&E.X160).

**Fig. 23:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (1 ppm) of Dimethoate (EE= Epidermis Elongate, ESh = Epidermis shrinkage, DM= Destroyed Muscle) (H&E.X160).

**Fig. 24:** Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (1 ppm) of Dimethoate (EE= Epidermis Elongate, ESh= Epidermis shrinkage= Vacuolated Epidermis, VM= Vacuolated Muscle, EMe = Epidermis Mergers, ExNM = Exit muscle Nuclei, EI= Epidermis Inducting) (H&E.X640).

In body wall exposed to 2.5ppm of Dimethoate for 28 days the histological Changes are very different compared with control; showing epidermis destroyed and vacuolated muscle fiber (fig 25), Vacuolated and Rupture in muscle , Destroyed epidermis and Inducting epidermis (fig 26), in adult earthworm, also found in juvenile earthworms vacuolated muscle fiber and destroyed epidermis(fig27) and Vacuolated Muscle , Muscle layer circular separating from longitudinal muscle layer, epidermis Inducting , , epidermis Mergers(fig28).

**Fig. 25:** Histological changes in transverse sections of epidermis and body wall of adult *Allolobophora caliginosa* exposed to (2.5 ppm) of Dimethoate (VM= Vacuolated Muscle fiber, DE= Destroyed Epidermis, N = nuclei.) (H&E.X160).
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Fig. 26: Histological changes in transverse sections of epidermis and body wall of *Allolobophora caliginosa* exposed to (2.5ppm) of Dimethoate to adult (VM= Vacuolated Muscle, RM = Rupture muscle, DE= Destroyed Epidermis, EI= Epidermis Inducting) (H&E, X160).

Fig. 27: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (2.5 ppm) of Dimethoate (VM= vacuolated muscle fiber, DE= destroyed Epidermis, N = nuclei) (H&E, X160).

Fig. 28: Histological changes in transverse sections of epidermis and body wall of juvenile *Allolobophora caliginosa* exposed to (2.5ppm) of Dimethoate (VM= Vacuolated Muscle, MS= Muscle layer circular separating from longitudinal muscle layer, EI= Epidermis Inducting, EMe= Epidermis Mergers) (H&E, X160).

Discussion

Typically the body wall of the earthworm consists mainly of the outer most cuticle secreted by the epithelium underlying it and the circular and longitudinal muscle layers. The epithelium is composed of a single layer including columnar cells, basal cells and mucous secreting cells [10]. The body wall of the control group in the present study was in line with this typical structure (fig.1). Histological examination of the body wall of *A. caliginosa* exposed to Dithemoate showed At 0.05ppm there was vacuolated muscle fiber; vacuolated epidermis and lost epidermis cell some regions (fig.2), Cuticle layer separating(fig.3), Rupture longitudinal muscle (fig.4) in adult and the damage was intense with the entire muscle layer being disintegrated (fig.5), Exit muscle Nuclei, Rippling circular muscle fibers (fig.6) in juvenile [26]. reported that the Neighboring cells in circular and longitudinal muscles appeared discontinuous (separated by narrow to large gap junctions) of the earthworm *Eisenia fetida* (Savigny) due to the exposure to pesticide, profenofos, also [27] were found. The circular and longitudinal muscle layer of the body wall of the Cr (VI) (Hexavalent chromium) treated *Eudrilus eugeniae* were also damaged as indicated by the loss of structural integrity and increased intercellular spaces the damage was more severe in those exposed to 893 mg kg-1. [28]. Found that the neighboring cells in circular and longitudinal muscles to be discontinuous, separated by narrow to large gap junctions when Earthworms (*Pheretima pegauna*) exposed to 80% LC50 for 48 h (Aza 3.13 μg cm−2) from commercial neem extract

At 0.1ppm there was vacuolated muscle fiber, vacuolated epidermis and destroyed epidermis, in contrast the nuclei gathered have found in vacuolated which in circular muscles of adult(fig.7), Similar Effect of nuclei gathered was reported due to the exposure of Butachlor Herbicide on Earthworm *Eisenia fetida* by [29]. Peritoneum Rupture (fig.8) in adult. The vacuolated muscle fiber, vacuolated epidermis too the epidermis cell layer and longitudinal muscle were completely lost in certain regions in juvenile (fig.9), similar effect of 893 mg kg-1 Hexavalent Chromium (Cr) on epidermis and muscle layers by reported [27]. Muscle layer circular separating from longitudinal muscle layer, Aggregation muscle fiber (fig.10,11), Exit muscle nuclei(fig.12). At 0.5 ppm there was epidermis separating from the muscle layer and vacuolated muscle fiber (fig.13), Rupture muscle and Nuclei Enlarge (fig.14) in adult and epidermis separating from the muscle layer and vacuolated muscle fiber(fig.15), Epidermis Inducting and Circular muscle overlap Epidermis (fig.16), Epidermis Inducting, Epidermis Mergers (fig.17) and Destroyed Epidermis, Circular muscle overlap Epidermis, Epidermis Inducting, Epidermis Mergers and Exit muscle nuclei(fig.18) in juvenile. similar [30] reported that the effect of 0.8mg/kg Atrazine on the earthworm (*N. mbae*) histology
showed that the epithelial tissue was vacuolated following cytolysis and the muscle layer has lost its compactness. Most of the epithelial cells have lost their nuclei.

At 1 ppm, of Dimethoate there were Muscle layer circular separating from longitudinal muscle layer and Vacuolated in Muscle, Exit muscle Nuclei (fig 20) and Vacuolated Muscle, epidermis Inducting, Pyrolyse muscle and Peritoneum Rupture (fig 21) in adult also found epidermis cells Elongate and shrinkage and Exit muscle Nuclei (fig 22), epidermis cells Elongate and shrinkage and Destroyed muscle (fig 23) epidermis cells Elongate and shrinkage Vacuolated epidermis and Muscle, epidermis Mergers, Exit muscle Nuclei, epidermis Inducting fig 24 in juvenile earthworm.

At 2.5 ppm there were epidermis destroyed and vacuolated muscle fiber (fig 25), Vacuolated and Rupture in muscle, destroyed epidermis and Inducting epidermis (fig 26), in adult earthworm, also found in juvenile earthworms vacuolated muscle fiber and destroyed epidermis (fig 27) and Vacuolated Muscle, Muscle layer circular separating from longitudinal muscle layer, epidermis Inducting, epidermis Mergers (fig 28).

All changes at 1.0. 2.5 ppm concentration of dimethoate similar changes which found by [27] were serious disintegration of cell margins and fusion of cells in the epidermis even at low doses of 0.24 mg kg-1 of Hexavalent Chromium, the entire epithelium being disintegrated, the epithelial cell layer was completely lost in certain regions at 893 mg kg-1 of cr. Epithelial sloughing was apparent by the space created between the epithelium and the circular muscle layer and [28] found some erosion of epithelium, fibrotic changes in the circular muscle and longitudinal muscle were noted in the earthworm exposed to the vermicomposting of paper cup. Cell necrosis was observed during this period. In temple waste vermicomposting, and [28] found the neighboring cells in circular and longitudinal muscles to be discontinuous when Earthworms exposed to ~80% LC50 for 48 h (Aza 3.13 μg cm−2), separated by narrow to large gap junctions also [31] reported that the epithelium separating from the muscle layer of the earthworm when the exposure to Normal Versus and Nano Cupper Enriched Engine Oil Previous studies suggested that earthworm skin has direct contact to the contaminated soils and is considered as a significant route to uptake of toxicants [32, 33]. Epidermis and cuticle represent a primary barrier that protects earthworm’s body from the environment and are also responsible for the transport of ions, thus allowing/blocking xenobiotics to enter the body [34].

**References**


التأثيرات النسيجية في جدار الجسم لدودة الأرض ALLOLOBOPHORA CALIGINOSA تعرضها لمبيد الحشرات الدايميثويت

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استلم في: 25 يوليو 2023 / قبل في: 12 أغسطس 2023 / نشر في: 30 سبتمبر 2023

الملخص

مبيد الدايميثويت ينتقل بشكل كبير في التربة ولقد اكتشفت بقاياه مترسبة في التربة والماء وذلك بسبب الممارسات غير المنظمة مما سبب أثر ضار على حياة الكائنات المفيدة بالترة والزراعة وذلك بسبب العوامل غير المنظمة مما سبب أثر ضار على حياة الكائنات المفيدة بالترة والزراعة.

تعتبر دودة الأرض مهندسة التربة لأنها قادرة على تحسين التربة والمجتمعات النباتية ولأنها تبتلع كميات كبيرة من التربة والمواد العضوية لذلك تتعرض لكميات كبيرة من الملوثات خلال استهلاكها الغذائي. تعرضت دودة الأرض ALLOLOBOPHORA CALIGINOSA لتركيزات من مبيد الدايميثويت (0.05ppm – 2.5 PPM) وظلت نحرة في طبقة البشرة ولم تظهر أي تأثيرات على الطبقات العضلية. وظلت نحرة في طبقة البشرة ولم تظهر أي تأثيرات على الطبقات العضلية. والخلايا العضلية كانت تتأثر وتتعرض للإصابة.

الكلمات المفتاحية: دودة الأرض، الدايميثويت، جدار الجسم

How to cite this article:


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