THE EFFECT OF SWINE WASTEWATER ON SEVERAL BIOLOGICAL PARAMETERS OF SPECIES EISENIA FETIDA: MORTALITY, BODY WEIGHT, PROLIFICACY

Mădălina IORDACHE, I. BORZA, Valentina ANDRIUCĂ, Daniela DUBIŢ

Banat`s University of Agricultural Sciences and Veterinary Medicine "King Michael I of Romania", Faculty of Agriculture, Timişoara, Romania
Calea Aradului 119, 300645 Timişoara, Romania,
E-mail: mada_iordache@yahoo.com

Abstract: *The objective of the study consisted of testing the effect of residual water resulted from swine breeding complexes on earthworms Eisenia fetida (Savigny, 1826) (Phyllum Annelida, Class Oligochaeta, Family Lumbricidae) under the following aspects: mortality, body weight, prolificacy. The experiment has been carried out in laboratory (microcosm experiment, according to the adjusted OECD methodology (Guideline for testing of chemicals – Earthworm, acute toxicity tests, no. 207/1984), using the test of artificial soil. The test lasted 14 days. All earthworms involved in the experiment were adults, with weight over 4000 mg. In each recipient were introduced 10 earthworms. Data recording was made in the 14th day: weight, mortality, prolificacy (number of cocoons). After 14 days of experiment, there was observed a decrease of earthworm number per recipients only for concentrations 12.5 and 25 ml swine wastewater per kg artificial soil, by 10% and 20%, respectively, related to control. Regarding the prolificacy, the cocoon number was the highest (17) at the concentration of 6.25 ml swine wastewater per kg artificial soil, followed by the concentration of 3.12 (15), which means with 30.76% and respectively with 15.38% higher than in the control recipient. These two concentrations were the most favourable. The highest concentrations of swine wastewater determined a decrease of cocoon number, with 15.39% as compared to the control recipient. In all concentrations of swine wastewater the earthworm biomass was higher than in the control recipient, the increase ranging between 0.17 – 6.73%. There were no notable changes at clitellum or locomotion levels after 14 days of exposure on swine wastewater.*

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Key words**:** *Eisenia fetida, earthworms, mortality, body weight, prolificacy, cocoons, swine wastewater*

INTRODUCTION

The aim of this research was to investigate the effect of swine wastewater resulted from the breeding complexes on earthworms *Eisenia fetida* (SAVIGNY, 1826), regarding the following aspects: weight, mortality, production of cocoons. Earthworms have been widely used to treat animal wastes [EDWARDS, 2004], as well as sewage sludge [KHWAIRAKPAM AND BHARGAVA, 2009; PRINCE ET AL., 1981; VIGUEROS AND CAMPEROS, 2002], and even human faeces [SHALABI, 2006]. This process is called vermicomposting, where earthworms enhance the microbial activity and the rates of decomposition of the material. This leads to composting or humification effect, in which the unstable organic matter is oxidized and stabilized [NATHASITH CHIARAWATCHAI, 2010]. Diluted swine manure applied in earthworms showed that earthworm population increased by 30% in 4 weeks, indicating the acclimation of the earthworms. A reduction in ammonia emission was observed of about 50% for the whole system. Higher water (+100%), carbon (+70%), and total nitrogen (+80%) gaseous losses were observed compared to conventional breeding on a slatted floor [LI, 2008].

Because the swine wastewaters exert an increasing environmental pressure, their management is important, especially by biological mechanisms.

MATERIAL AND METHODS

The experiments have been carried out in laboratory conditions according to the adjusted OECD methodology (Guideline for testing of chemicals – Earthworm, acute toxicity

tests, no. 207/1984), using the test of artificial soil. In order to test substances on earthworms, an essential importance is accorded to soil characteristics, because earthworms are organisms living into it. The differences occurred in the composition of soil influence the bio-availability and toxicity of pollutants; these might be less available in soils with high content of clay which can retain certain pollutants. The earthworms are chosen for this type of studies for several reasons. These organisms are ubiquitous and abundant in the soils of almost all terrestrial ecosystems, excepting deserts. They play an important role in soil formation, its aeration and in the circuit of the nutritive elements. Because these animals develop digging activity into the soil forming canals, consume detritus and other residues, they improve many parameters of soils, as aeration, a better infiltration of water, a higher microbial activity.

The OECD protocol used within this research uses the response of a single species, *Eisenia foetida* (SAVIGNY, 1826), commonly named red worm (Phyllum *Annelida*, Class *Oligochaeta*, Family *Lumbricidae*). Also, this species is known as compost worm or tiger worm because the segments of its body present transversal dark lines. Unlike other species of earthworms, this earthworm does not dig very deep into the soil, because it lives in soils rich in organic matter, which usually comes from decomposed plant debris or animal dejections, and which is located top of the soil, this organic matter serving as its food (fact which determined its common name). *Eisenia fetida* has a short life cycle, hatching from cocoons in 3-4 weeks and reach the sexual maturity in 7-8 weeks after hatching, at 20°C. This species is very prolific, each earthworm produces 2-5 cocoons/week, and from each cocoon hatched several worms.

The toxicity test used artificial soil (pH = 6) (adjusted by addition of $CaCO_3$). The composition of the artificial soil has been the following: 10% Sphagnum peat, 20% clay and 70% sand (fine sand, over 50% particles with sizes between 50-200 μ). Each recipient with artificial soil weighed 1000 g. The test lasted 14 days. All earthworms involved in the experiment were adults, with weight over 4000 mg. In each recipient were introduced 10 earthworms, covered by perforate plastic foil in order to avoid the evaporation of water, to maintain a good aeration and to avoid earthworm escape. At the beginning and at the end of experiment, the earthworms have been weighed, and before starting the test the worms have been washed with distillate water and incubated on wet paper for 24 hours, to empty their gut content.

Data recording was made in the 14th day: weight, mortality, prolificacy (number of cocoons). The chemical analysis showed the following composition of the swine wastewater (table 1):

Table 1

Chemical composition of the swine wastewater tested within experiment

Chemical parameter						
N total		P_2O_5		K_2O		pH
mg/l	%	mg/l	%	mg/l	%	pH unities
1665	0,17	179,35	0,018	1220	0,12	7,34

Into the testing recipients were added different quantities of swine wastewater (the dose of 12,5 ml has been established considering the field dose, the size of the recipient, and the amount of soil per recipient). The experiment consisted of four replicates. After 14 days, the alive earthworms have been counted, weighed, and the also the cocoons have been counted. At the end of the experiment, there were noticed the changes appeared at clitellum level, and the locomotion behaviour by observing the movement of each earthworm on a foil. The assessment of the mortality state was realised by touching the earthworm at the anterior part of the body (mechanical stimuli). The tested concentrations of swine wastewater were: 0 (control), 1.56, 3.12, 6.25, 12.5, 25 ml/kg artificial soil.

RESULTS AND DISCUSSION

The achieved results regarding the mortality and production of cocoons at the end of the test (after 14 days) are shown in tables 2 and 3.

Table 2

Earthworm mortality after exposure on swine wastewater

After 14 days	Dose of swine wastewater (ml/kg soil)					Control
	1.56	3.12	6.25	12.5	25	
Earthworms alive (mean values)	10	10	10	9	8	10

Table 3

Earthworm prolificacy after exposure on swine wastewater

After 14 days	Dose of swine wastewater (ml/kg soil)					Control
	1.56	3.12	6.25	12.5	25	
Number of cocoons (mean values)	13	15	17	12	11	13

After 14 days of experiment, there was observed a decrease of earthworm number per recipients only for concentrations 12.5 and 25 ml swine wastewater per kg artificial soil, by 10% and 20%, respectively, related to control. Regarding the prolificacy, the cocoon number was the highest (17) at the concentration of 6.25 ml swine wastewater per kg artificial soil, followed by the concentration of 3.12 (15), which means with 30.76% and respectively with 15.38% higher than in the control recipient. These two concentrations were the most favourable. The highest concentrations of swine wastewater determined a decrease of cocoon number, with 15.39% as compared to the control recipient (figure 1 and 2).

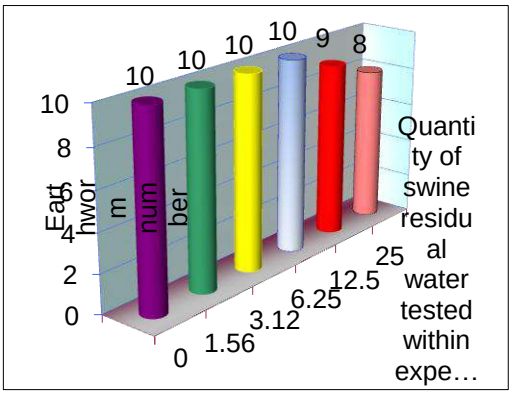

Figure 1. Earthworm mortality at the end of the test

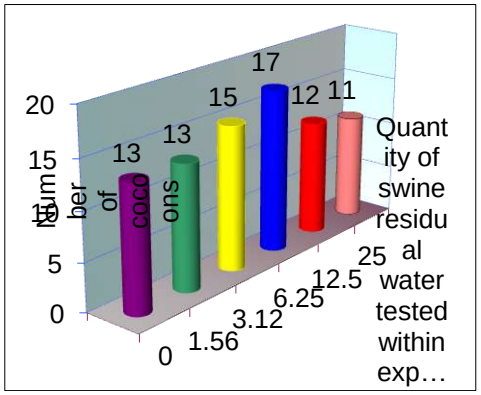

Figure 2. Earthworm prolificacy at the end of the test

The earthworm weight (biomass) at the end of the experiment is shown in table 4.

Table 4

Earthworm weight (biomass) at the end of the experiment

After 14 days	Dose of swine wastewater (ml/kg soil)					Control
	1,56	3,12	6,25	12,5	25	
Earthworm weight (mg)	4089	4228	4292	4281	4264	4021

In all concentrations of swine wastewater the earthworm biomass was higher than in the control recipient, the increase ranging between 0.17 – 6.73% (figure 3).

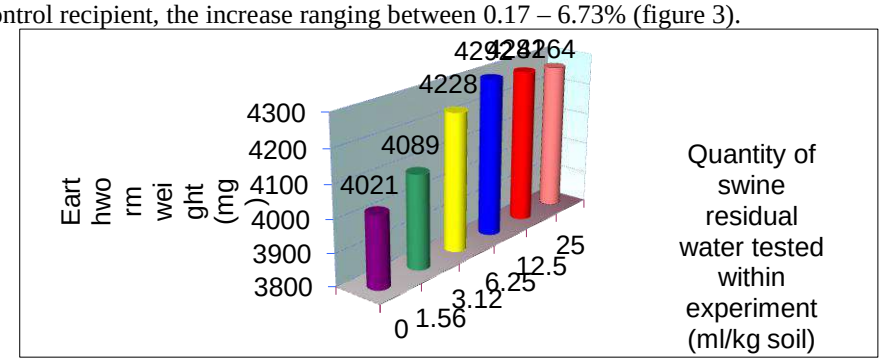

Figure 3. Earthworm weight (biomass) after 14 days of exposure on swine wastewater

There were no notable changes at clitellum or locomotion levels after 14 days of exposure on swine wastewater.

CONCLUSIONS

The performed study led to the following conclusions:

- After 14 days of experiment the earthworm number decreased at the concentrations of 12.5 and 25 ml swine wastewater per kg artificial soil (with 10% and 20%, respectively related to control.
- Regarding the prolificacy, the cocoon number was the highest (17) at the concentration of 6.25 ml swine wastewater per kg artificial soil, followed by the concentration of 3.12 (15), which means with 30.76% and respectively with 15.38% higher than control.
- The highest concentrations of swine wastewater determined a decrease of cocoon number, with 15.39% as compared to the control recipient.
- In all concentrations of swine wastewater the earthworm biomass was higher than in the control recipient, the increase ranging between 0.17 – 6.73%.
- There were no notable changes at clitellum or locomotion levels after 14 days of exposure on swine wastewater.

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