In Vivo Test of Dicofol on Cocoon Production and Viability of Earthworm *Pontoscolex corethrurus* Fr. Mull

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Abstract. The uncontrol using of pesticides, harmful to the environment, health, and it would have impact to non-target animal as earthworm. This study describes the effect of the Dicofol to cocoon production and viability of earthworm *Pontoscolex corethrurus* Fr. Mull., has been done in July - Augustus 2016 at the zoology laboratory of Biology Department of Universitas Negeri Padang. The experiment used the Completely Randomized Design (4 treatments 6 replications). The treatments are with 0 g / l (P1), 0.002 g / L (P2), 0.004 g / L (P3), and 0.006 g / L (P4) and 0.008 g / L of Dicofol that diluted to water. The Data of production and viability of earthworm cocoons *Pontoscolex corethrurus* Fr. Mull collected during 30 days in alternate day. Data analyzed by ANOVA and Duncan New Multiple Range Test at p <0.05. The results showed that the average number of cocoons production at P1 30 cocoons (the highest), P2 16 cocoons, P3 7 cocoons, and the P4 and P5 0 cocoons (the Lowest). The average percentage of cocoons viability were highest in P1, and P2 (100%); P3 (10%) and the cancel at P4 and P5 (0%). It can conclude that the pesticide Dicofol decreased the production and viability of the earthworm cocoons *Pontoscolex corethrurus* Fr. Mull.

1. Introduction
The use of Pesticides has been able to increase agricultural production in the world, to reach approximately US $ 25 billion per year [1]. Pesticides are often used by farmers exceed the recommended dose for farmers think with more and more pesticides are used then it will get better results.

Dicofol pesticides is one of the insecticides that are often used by farmers in Indonesia. Dicofol insecticides include but are not specific class of organophosphate certain kill insects, but can also cause deadly poisoning or other organisms or non-target animals. Various forms of organophosphorus compounds are often used to control insects and use more frequently and more than other groups of insecticides. The result is the market is dominated marketing of pesticide compounds [2].

This insecticide achieve the goals coming in through the mouth, skin or respiratory. Insecticides Dicofol is one pesticide to poison insects, including organophosphates group containing carbon and phosphorus, and can be neurotoxic in humans. Dicofol very effectively used to combat and eradicate plant pests such as aphids, rice weevils, flies, leafhoppers and so on [3].

Nonetheless, the unwitting use of insecticides Dicofol continuous negative impact on the environment both humans, animals and soil biota. The impact of these insecticides can be death,
obstacle metabolic activity, inhibition of behavior, reproduction and hatchability soil biota. One of them is the earthworm is the soil biota that are often found on agricultural land and have a beneficial role in the soil ecosystem. Earthworms and microbes on agricultural land have function to degradation substances in the process of decomposition and mineralization of organic matter. The decomposition process of organic matter causes changes in soil structure so that it can improve soil aeration and water holding ability of the soil \[3,4\].

Some research has been conducted regarding the effect of an insecticide on earthworms as practiced by \[5\] about the use of Insecticide Endosulfan Effect on Production and Viability of Cocoon Earthworm *Pontoscolex corethrurus* Fr. Mull and \[6\] about the use of Insecticide Carbofuran Effect on Production and Viability of Cocoon Earthworm *Pontoscolex corethrurus* Fr. Mull. In both of these studies found a decline in production and quality earthworm cocoons.

Based on this background, this research aim to investigate the effect of insecticides on production and viability Dicofol earthworm cocoons *Pontoscolex corethrurus* Fr. Mull.

2. **Methods**
This study was conducted in July-October 2016 in the Laboratory of Zoology Department of Biology, Universitas Negeri Padang. The study design used Completely Randomized Design (CRD), with 5 treatments each treatment consisted of 6 replicates. The concentration of treatment based on dose conversion from laboratory scale to field scale are:

- **P1.** 0 g / l (control) (Solvent / water)
- **P2.** 0.002 g / l Dicofol
- **P3.** 0.004 g / l Dicofol
- **P4.** 0.006 g / l Dicofol
- **P5.** 0.008 g / l Dicofol

**a. Dicofol solution**
Concentration is fluid density Dicofol insecticidal active ingredient. Dicofol insecticide concentration used was 60%. To make Dicofol insecticide concentration of 0.002 g / l is to enter by using a micro pipette Dicofol as 2,5μ then enter it into the measuring cup and add the solvent until the volume becomes 10 ml, this will continue to make further treatment.

**b. Observations earthworm Cocoon Production**
Production To detect earthworm cocoons *Pontoscolex corethrurus* Fr. Prepared Mull 25 Plastic jars the size of 5 liters, then filled with Growing Media Standard 1 kg of the next volume mencapurkan Dikofol solution according to treatment and stir until smooth and each jar was added polybags and labeled. Each jar Plastic included 5 earthworms and on the ground placed rice bran. Then plastic jars covered using gauze. 1 time every 5 days each plastic jars sorted and counted the number of earthworm cocoons produced.

**c. Observations earthworm cocoon Viability**
Cocoon produced by the worms of each treatment was calculated, separated, and placed on filter paper moistened with water that has been deposited for 3 days in a petri dish container. Then once placed on a hard strain and covered with filter paper that has been moistened. Every day spraying by using distilled water 2 times a day, as well as to see the cocoon hatch. Total cocoon hatch is calculated every day until the 15th day of incubation.

**d. Data Analysis**
Data are number of cocoons produced and cocoon hatch in each treatment group were tested by ANOVA, followed by DNMRT test (*Duncan’s New Multiple Range Test*).
3. Results and Discussion

Based on research conducted at the Laboratory of Zoology Department of Biology, Universitas Negeri Padang, found that Dicofol have an influence on production and viability earthworm cocoons *Pontoscolex corethrurus* Fr. Mull, as shown in Table 1.

| No. | Treatment | Total average cocoon |
|-----|-----------|----------------------|
| 1.  | P1        | 30<sup>a</sup>       |
| 2.  | P2        | 16<sup>b</sup>       |
| 3.  | P3        | 7<sup>c</sup>        |
| 4.  | P4        | 0<sup>d</sup>        |
| 5 . | P5        | 0<sup>d</sup>        |

Description: the numbers in the same column followed by the same Superscript letters, not significantly different (P <0.05) in DNMRT test

The results showed that the production and viability of earthworms *Pontoscolex corethrurus* Fr. Mull., Conducted over 30 days of observation with various concentrations showed a highly significant difference between treatments. The higher concentration of insecticide Dicofol given further reduces the number of cocoons produced. The highest number of cocoons found in P1 treatment (control) is 30 cocoon. While the number of cocoon lowest is 0 at P4 and P5 treatment because there are treatments applied to turn off all the test animals. Total cocoon at significantly different P1 with P2, and P3. Total cocoon on P3 is the lowest of all experimental units were still alive. It also shows that there are significant differences among treatment and decrease the number of cocoon in line with the increase in dose.

It most likely cause a decrease in the number of cocoon production between treatments is active insecticidal compounds of Dicofol can enter the body worms either through the mouth or skin and such compounds to also on the reproductive system. In addition the nature of systemic Dicofol also stored and circulated in the body of the worm.

The decline in fecundity due to disruption of the process of oogenesis mainly on the process for entrainment Dicofol to vitelo genesis cause poisoning when opened or can not be found will be dissected cocoon prepared for ovulation, while in treatment P1 (control), P2, and P3 will still find many cocoons or eggs.

According to [5] fecundity on earthworm cocoons *Pontoscolex corethrurus* Fr. Mull., A decline due to the influence insecticides are toxic to the body tissues of animals and systemic works as a contact poison, stomach poison, and the poison can breath in through digestion and respiration. Cocoon production is a subject that can adjust to some kinds of conditions, especially in response to food. Total cocoon issued a the connecting link between one generation to the next generation.

This result contrary to the result of research [7], that the earthworms *Pontoscolex corethrurus* Fr. Mull., which produces the highest number compared to other worm cocoons of 98 cocoons within one year under laboratory conditions. In his research explains that *P. corethrurus*, *Dichogaster affinis*, *Metaphire houleti* and *Octochaetona beatrix* has the ability to self-fertilization or parthenogenetic species can produce a cocoon without mating. Almost all exotic species of earthworms in the humid tropics that reported at least facultative parthenogenic, while local species only produce cocoons in case of marriage.

The existence or hormone production in the earthworm is strongly influenced by the feed given so that will affect to the amount of cocoons produced. According to the research result [5] that the earthworm population depends on the strongly type and concentration of insecticide using. The differences between the number of the cocoon of earthworms hatched caused by changes in
temperature and limited sources of food reserves in the cocoon and the ability detoxication or excrete toxicant associated with the concentration of insecticide.

**Table 2.** The average yield Viability earthworm cocoons

| No. | Treatment | Mean percentage (%) hatching cocoon |
|-----|-----------|-----------------------------------|
| 1.  | P1        | 100a                              |
| 2.  | P2        | 100a                              |
| 3.  | P3        | 10b                               |

Description: The numbers in the same column followed by the same Superscript letters, not significantly different (P < 0.05) in DNMRT test

Based on Table 2 are known percentage of the earthworm *Pontoscolex corethrurus* Fr.Mull. cocoon hatchability, in the treatment of P1, P2, and P3, there are differences in average percentage of hatching cocoon. In the treatment of P1 and P2, have an average percentage of the cocoon hatchability as same as namely 100%. While on treatment P3 cocoon hatching percentage as much as 10%.

The percentage hatchability earthworm cocoons *Pontoscolex corethrurus* Fr. Mull., which was seen among treatment P1 and P2 is 100%. While the lowest percentage of cocoon hatchability seen in treatment P3 is 10%. It is suspected in Dicofol P3 has been able to be embryotoxic, causing death. It could be Dicofol which also neurotoxic properties has led to the failure of the embryo develops for failing the formation of the central nervous system. The percentage difference in hatchability for Dicofol pesticides are systemic and embryos are also toxic.

Suspected poisoning has occurred against the ovum in the process of oogenesis. If indeed this is the case then there is no development of the embryo from the beginning [5]. While on treatment P1 and P2 do not affect the viability cocoon hatchability or suspected in the concentration Dikofol though present in the worm body tissue but still on his tolerance threshold.

The decreased amount due to insecticide Dicofol cocoon has the power to kill broad spectrum against insects and earthworms. These compounds can enter the body through physical contact, digestive system, and respiration [5]. Soil moisture and temperature also affect the hatching success earthworm cocoons and removal of the field and laboratory conditions [7].

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