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Effect of neonicotinoide insecticide acetamiprid on super oxide dismutase activity in earthworm *Eisenia fetida*

Parveen Gill, Dommapati Sudhakara Rao, RK Gupta, Dharambir Singh, Anshu and Naresh Kumar

Abstract

Neonicotinoides are the most prominent class of pesticides in all over world and it is registered in approximately 120 countries globally and they are recognized as nicotine derivatives. Acetamiprid is the majorly used neonicotinoide insecticides and it has the detrimental effect on antioxidant enzymes of earthworms of *Eisenia fetida*. The SOD activities were significantly increased with an increase in the doses of acetamiprid at 48 hours of exposure time. Acetamiprid concentrations of 0.165 and 0.188 µg showed SOD activities of 6.903 U/mg protein and 7.330 U/mg protein at 48 hours of exposure respectively, while a concentration of 0.188 µg of acetamiprid showed SOD activity of 6.913 U/mg protein at 24 hours of exposure time. The study revealed that SOD activities are affected by acetamiprid.

Keywords: Earthworm, *Eisenia fetida*, neonicotinoide insecticide, acetamiprid, super oxide dismutase

Introduction

Neonicotinoide insecticides are among the most important insecticides applied to crops which have been the world's largest selling insecticide for many years (Jeschke *et al.*, 2011)^[7] and they are account for one-fourth of the entire insecticide market because of their excellent insecticidal activity (Kagabu, 2011)^[9], the neonicotinoides are particularly suited for controlling many insects with biting and sucking mouth parts especially if swallowed (Laurino *et al.*, 2011)^[10]. Acetamiprid is a second-generation neonicotinoide insecticide which was initially commercialized in Japan in 1995 by Nippon Soda mainly for foliar applications and direct soil uses are restricted (Elbert *et al.* 2008)^[3]. It has been proven that acetamiprid is more effective against pests if it used as foliar sprays as compared with applied directly to the soil (Palumbo *et al.*, 2001)^[11]. Earthworms are prominent invertebrates belonging to family lumbricidae and are dominant in temperate and tropical soils. They are hermaphrodite but self-fertilization does not usually take place. During laying eggs, the sexually mature worms exhibit distinctive epidermal ring shaped area called the clitellum, which has gland cells that secrete material to form a viscid, girdle like structure known as cocoon. Though the number of fertilized ova in each cocoon ranges from one to twenty for lumbricid worms yet only one or two survive and hatch. Growth rate is very fast during initial 70 days of life span. The mature worm can grow up to 1500 mg of body weight (Gill P. *et al.*, 2019)^[6].

Agricultural pesticide applications characterize severe hazards to wildlife and ecosystem resources which have led to a growing concern worldwide over the indiscriminate use of such chemicals (Rao, 2006)^[13]. Due to the presence of thin and moist body wall, earthworms are more susceptible to the pesticide contamination, thus they are the potential biomarkers of soil pollution. Upon exposure to pesticides the reactive oxygen species are formed and the antioxidant enzymes play an important role in protecting the earthworm cells against reactive oxygen species (ROS) (Feng *et al.*, 2015)^[4]. Acetamiprid is one of the potential threats to *Eisenia fetida* that effects the earthworm population in sub soil; therefore it is imperative to observe the effect of acetamiprid on earth worms.

The present study was carried out to see the effect of neonicotinoide insecticide acetamiprid on earth worm's antioxidant system with regard to super oxide dismutase activity and its response as a defence mechanism again the toxic effect acetamiprid.

Materials and Methods

Superoxide dismutase was assayed by measuring its ability to inhibit the photochemical reduction of nitro blue tetrazolium (NBT) adopting the method of Giannopolitis and Ries (1977)^[5].

Statistical Analysis

The standard statistical tools were used for analysis of data recorded in experiments. The experimental design for lab

study was completely randomized block with four replicates. A critical difference (CD) was calculated between the treatments by CRD (*in vitro*), accordingly, using software "OPSTAT", developed at the Computer Center, College of Basic Science and Humanities, CCS Haryana Agriculture University, Hisar, Haryana.

Results

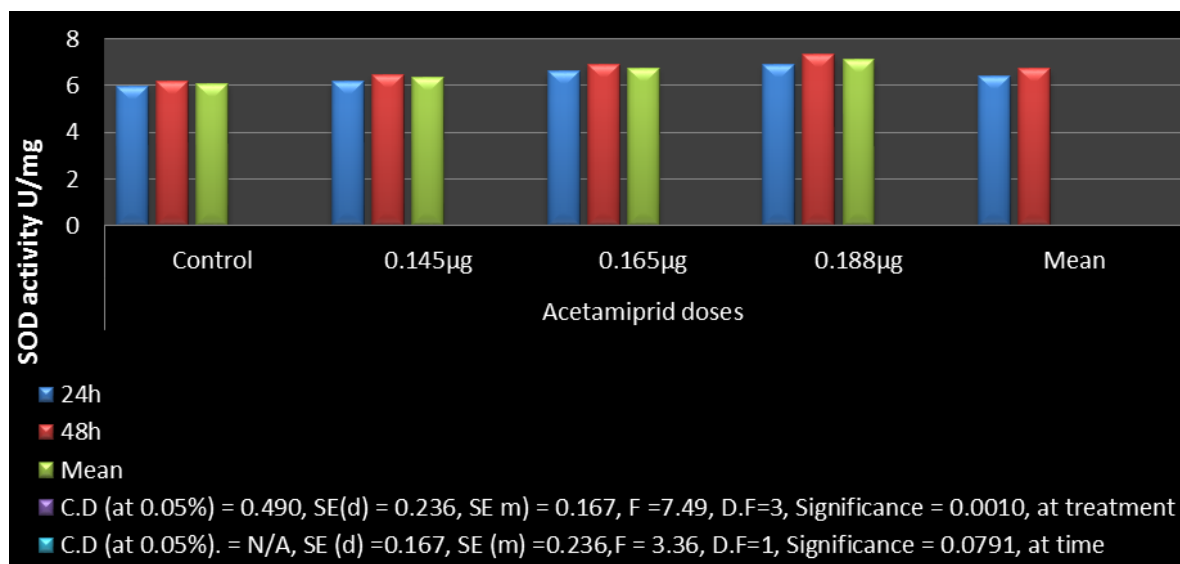


Fig 1: Effect of acetamiprid on SOD activity in *Eisenia fetida*

The results presented in Fig. 1 indicated that SOD activities were increased from 24 to 48 hr of exposure to all three applied doses against earthworms. The low dose of acetamiprid that is 0.145µg gave SOD activities of 6.213 U/mg at 24 hr and 6.443 U/mg at 48 hr of exposure. The medium insecticide dose used in the study was 0.165 µg resulted SOD activities of 6.615 and 6.903 U/mg for 24 and 48 hr of exposure time respectively. The highest dose chosen and given against earth worms resulted 6.913 and 7.330 U/mg of SOD activities for 24 and 48 hr of exposure period respectively. The enzyme activities were higher in all three doses given to earthworms compared to control. Low SOD activities were recorded in control group of earthworms compared to treatments. The SOD activities were increased by 15.44 per cent at 24 hr and 18.83 per cent at 48 hr of exposure period.

Discussion

Superoxide dismutase (SOD) activity has been studied with regard to acetamiprid at varied concentrations. The results of SOD activity revealed that it has completely depended on exposure time and concentrations of pesticide. Three doses of acetamiprid have been used during the experiment for determination of enzymatic activity. The SOD activity of 5.988 U/mg protein and 6.168 U/mg protein in control and treatment has been found at 24 and 48 hour of exposure time respectively.

The results of effect of acetamiprid on SOD activity are given in figure 1. It was observed that the activities of the enzyme have increased with an increase of acetamiprid concentration at 48 hour of exposure. Acetamiprid concentrations of 0.165 and 0.188µg had shown SOD activities of 6.903 U/mg protein and 7.330 U/mg protein at 48 hour of exposure

respectively, while a concentration of 0.188µg of acetamiprid had shown SOD activity of 6.913U/mg protein at 24 hr of exposure. Oxidative stress played an important role in the toxicity of various pesticides and can induce destructive reactive oxygen species (ROS). Accumulation of ROS such as hydrogen peroxide (H₂O₂) and superoxide radical (O²⁻) damages cellular components including DNA, proteins and lipids (Cooke *et al.*, 2003)^[2]. According Singh *et al.*, (2006)^[12] the level of SOD activity increase may be attributed to the increased biosynthesis of it to combat oxidative stress. On par to the results were also revealed by Juan *et al.*, (2016)^[8] where they have showed the significant increase in SOD activity with length of exposure time in treatments with insecticides compared to control.

Effect of acetamiprid on SOD activity in *E. fetida* by artificial soil method was done by Bing Li *et al.*, (2018)^[11] in which SOD activities were assayed at 7th, 14th, 21st and 28th days after treatment and found that the enzyme activity was increased from 7th to 14th day and later reached to a level on par to the SOD level in control at 21st day, later slightly inhibited at 28th day after treatment. These results are on par to the result of our study.

Conclusion

Acetamiprid is one of the most used neonicotinoide insecticides all over worldwide to control the insect for enhancing the production and productivity of crops yields, but ultimately they effect the beneficial soil fauna and other important soil organisms. Earthworm's farmer's friends are so affected due to indiscriminate use of this pesticide, causing harmful effects by reducing the antioxidant defence system. Earthworms contaminated with acetamiprid have showed increased activities of super oxide dismutase.

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