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PHYTOTOXICITY OF PROFENOFOS 50% EC (CURACRON 50 EC) TO *VIGNA RADIATA* L. SEEDLINGS: I. STUDIES ON MORPHOLOGY AND PIGMENTS

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ABSTRACT
Profenofos 50% EC (curacron 50 EC) a hazardous pesticide commonly used in agriculture is an important contaminant of environment. Its presence in biological system has gained importance due to bioaccumulation in food chain. The phyto-toxic effect of profenofos was assessed based on the morphological traits of seedlings (Root length, Shoot length, Fresh weight and dry weights) and phyto-pigments (Chlorophyll a, Chlorophyll b, Total Chlorophyll, Caretonoid and Phaeophytin) of the test species, *Vigna radiate* L. The concentrations of pesticide chosen were based on EC50 and are in the range of 0.02, 0.05, 0.08, 0.1 and 0.2 % of profenofos. All morphological traits and pigments were significantly reduced with increase in pesticide concentration.

KEY WORDS: Profenofos, organophosphate, seedlings, morphology, pigments.

INTRODUCTION:

Pesticides of various categories are frequently used against a number of pests, in the field to increase the crop production, though these chemicals are highly toxic and represent a risk for the non-target organisms and finally finding their way to the food chain (Gupta, 2006). Profenofos (O-4-bromo-2-chlorophenyl-O-ethyl S-propyl phosphorothioate) is a broad-spectrum organophosphate pesticide used widely for agricultural and household purposes in India and other countries. Profenofos (PFF) had been investigated to be highly toxic to different organisms including mammal insects and fish. It also has been classified as moderately hazardous (toxicity class II) pesticide by WHO and it has a moderate order of acute toxicity following oral and dermal administration (WHO, Geneva, 1990, 2001) Organophosphates, including chlorpyrifos, are the most hazardous water pollutants, which have already led to biotype pollution in some regions. Since 1965, chlorpyrifos has been in use throughout the world. Pascoe (1993) had reported that, the methods of assessing have been improved regarding their sensitivity, precision and accuracy, but they are not sufficient to assess the effects on living organisms and bioavailability. Therefore, it is necessary to involve biological tests, namely bio-indicators, in risk assessment and water contamination detection. The effect of chlorpyrifos was assessed based on morphological traits (root and shoot length, fresh and dry weight of roots and shoots). Zhi-Yong *et al*. (2011). It has been observed that when the chlorpyrifos applied at the rates of 1.0 mg/l and 10.0 mg/l caused significant phytotoxic effects. (Gvozdenac *et al*., 2013). In contrast to the results of this study, Wang *et al.* (2007) showed that chlorpyrifos had not affected the growth of wheat and oilseed rape seedlings even at high rates, indicating that those species were not good indicators of the presence of that insecticide in water. The findings of Li *et al*. (2007) was that tolerance levels of crops are species-dependant and vary under different stress intensities (concentrations and types of pollutants) and growth stages (germination, emergence, vegetative growth *etc*.). Literature is rich in information referring to the phytotoxic and inhibitory effects of herbicides on germination, root and shoot growth (Boutin *et al*., 2004; White and Boutin, 2007), as well as the effects of seed-coating fungicides on germination (Klokocar *et al*., 1991, Stevanovic *et al*., 2009a, 2009b). However, very few reports can be found on the effects of insecticides, especially organophosphates, on these seed and plant traits.

Keeping a view on the above literature available on the phytotoxocity of organophosphates. Here an attempt has been made to find out the effects of an organophosphate, Profenofos on the seedlings of *Vigna radiata* L.(mung).

MATERIALS & METHODS

Experimental Protocol

Selection of profenofos concentration

The concentrations of pesticide, Profenofos chosen were, 0.02, 0.05, 0.08.0.1 and 0.2%. A control set was maintained with distilled water only for comparison purpose.

Selection of test seed

The prime pulse seed *Vigna radiata*, var. PDM 139 Samart popularly called as mung commonly used in eastern state of India, particularly Odisha State has been chosen for study. Healthy seeds of *Vigna radiata*, were obtained from OUAT extension Ratnapur, Ganjam for the experimentation. A standard filter paper method was used. Mung seeds (20 per replication) were placed in Petridishes (6’’) on filter paper moistened with 10 ml of test solution. The mung seeds surfaces sterilized with sodium hypochlorite for 10 minutes and were incubated in the dark at 25±2 °C for 7 days in Seed Germinator (REMI-6C). After seven days (168hours) the following assessments were made on germination (%), Phytopigments (Chlorophyll a, Chlorophyll b, Total Chlorophyll, Caretonoids and Phaeophytin) following the methods of Arnon (1949), length of seedling roots and shoots (cm) and fresh and dry weight (g) of roots and shoots. The experiment was set in three replicates.

Statistical Analysis

The data are expressed as mean values of (n=5) and were analyzed employing Correlation Analysis to determine whether the values were significantly different from control at 0.05P with 4 d.f. (Mishra and Mishra, 1980).

RESULTS

The observations made in relation to morphological traits and phytopigments are given in Fig. no.1-4, plate-1 and Table no.1 showing the correlation analysis of profenofos concentration (%) and parameters studied. In Fig. No.-1, the effect of profenofos on Mung seed germination (%) recorded after 96h of treatment under laboratory conditions were given. It was found that, there is a gradual decline in % seed germination with the increase in profenofos concentration and at 0.2% profenofos; the % germination was decreased up to 26.25%. Fig. No-2 showed the root and shoots length (in cm) and Root/Shoot Ratio (R/S) of 7days old seedlings after profenofos exposure. Both the parameters decreased with the increase in profenofos concentration. Fig.No-3 indicates the decline in fresh weight and Dry weight of Root and Shoot in gm with the profenofos exposure in comparison to control values. However, an Exception was found in case of 0.02% profenofos treatment where it elevated the parameters studied. The effects of profenofos on different phytopigments of 7days old Mung seedlings are depicted in Fig. no.-4. The pigments like Chlorophyll a, Chlorophyll b, Total Chlorophyll, Caretonoids and Phaeophytin decreased with the increase in profenofos concentration expect 0.02% of profenofos. The correlation analysis (r values) was carried out between each parameter observed with the Concentration of Profenofos (%) are given in Table No.-1.The morphological traits like % germination ,Shoot Length, Root fresh and Dry Weight and Shoot fresh weight showed a statistically significant result where as all pigments studied were not significant statistically.

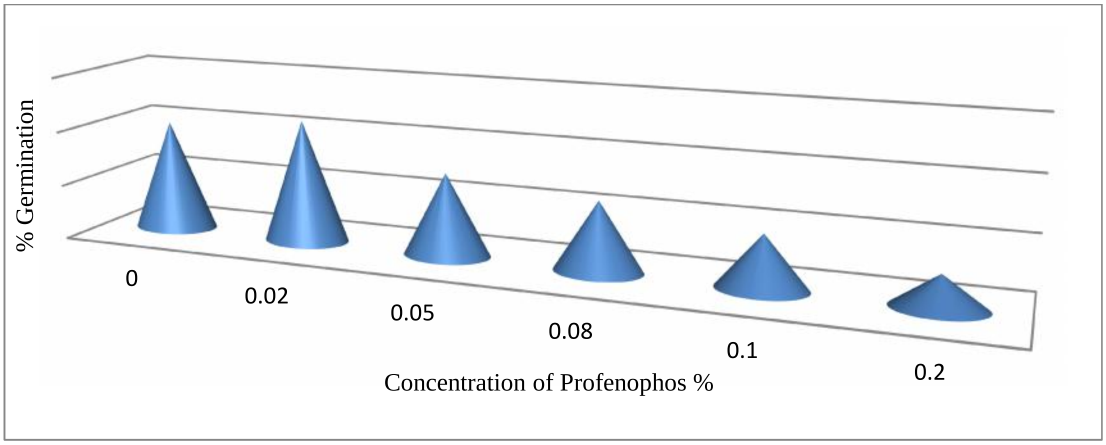

FIGURE 1: Effect of %Profenophos on Mung Seed Germination (recorded after 96h of treatment)

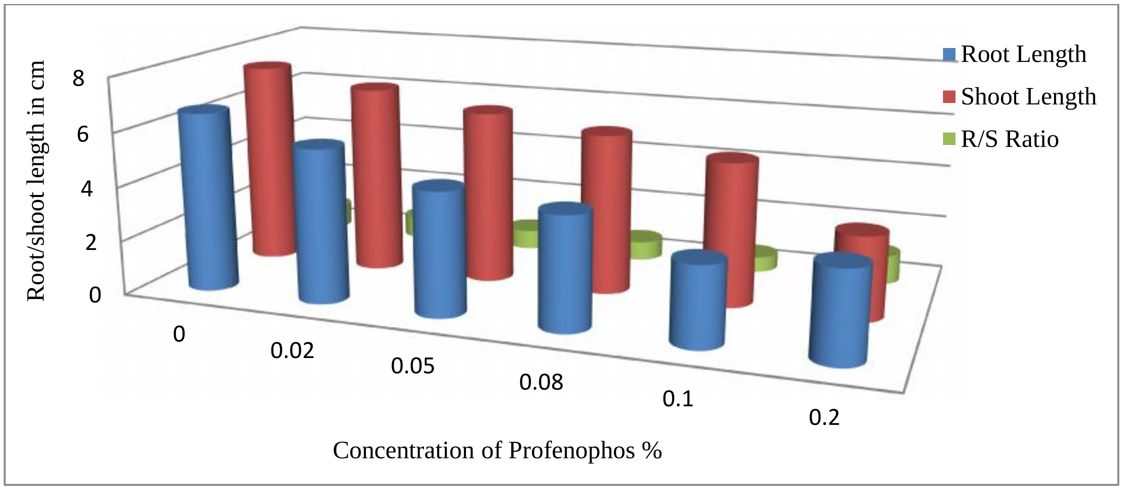

FIGURE 2: Phytotoxicity of profenophos on the 7days old morphology of Mung seedlings

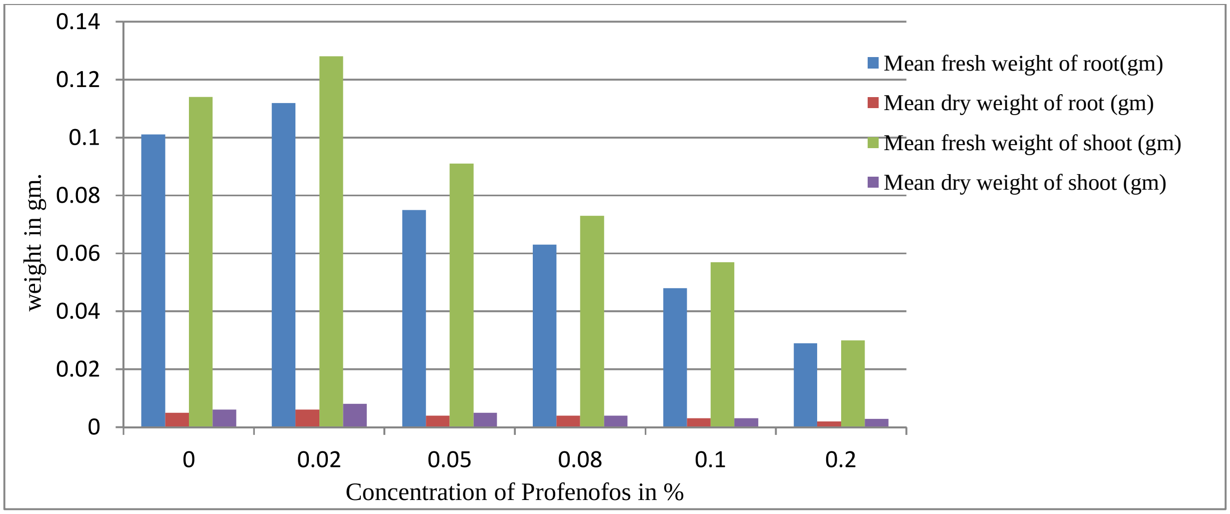

FIGURE 3: Changes in Fresh and Dry weight in Root and Shoot in 7days old seedlinds after profenofos treatment

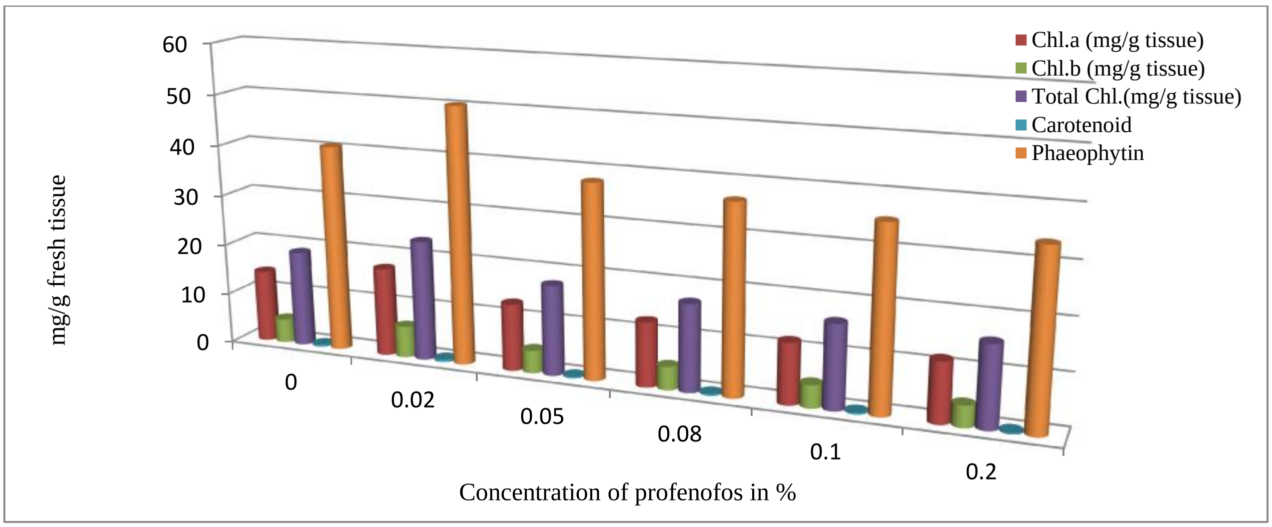

FIGURE 4: Changes in pigment content in 7days old mung seedlings after profenofos treatment

TABLE 1: The correlation analysis (r values) between each parameter observed in 7days old Mung (*Vigna radiata*, L.) seedlings with the Concentration of Profenofos (%)

Sl. No.	Correlation analysis between Parameters	Regression Equation	'r' value	d.f	Significance Level
1.	%Profenofos Vs % Germination	Y= -363.5x+90.67	-0.887	04	0.05P
2.	%Profenofos Vs Root length	Y= -16.21x +5.749	-0.684	04	NS
3.	%Profenofos Vs Shoot Length	Y=-21.86x+ 7.479	-0.997	04	0.001P
4.	%Profenofos Vs Root Fresh Weight	Y= -0.409x+0.102	-0.866	04	0.05P
5.	%Profenofos Vs Shoot Fresh Weight	Y= -0.482x +0.118	-0.896	04	0.05P
6.	%Profenofos Vs Root Dry Weight	Y= -0.018+0.005	-0.828	04	0.05P
7.	%Profenofos Vs Shoot Dry Weight	Y= -0.022x+0.006	-0.648	04	NS
8.	%Profenofos Vs Chlorophyll a	Y=-20.9x+15.01	-0.487	04	NS
9.	%Profenofos Vs Chlorophyll b	Y= - 6.258x+5.291	-0.368	04	NS
10.	%Profenofos Vs Total Chlorophyll	Y= -27.13x+20.22	-0.462	04	NS
11.	%Profenofos Vs Car0tenoid	Y= - 0.463x+0.448	-0.435	04	NS
12.	%Profenofos Vs Phaeophytin	Y= - 57.35x+43.47	-0. 474	04	NS

d.f = Degrees of freedom (n-2), Y= a+ bx, r values = correlation values , P = probability Level, NS= not significant

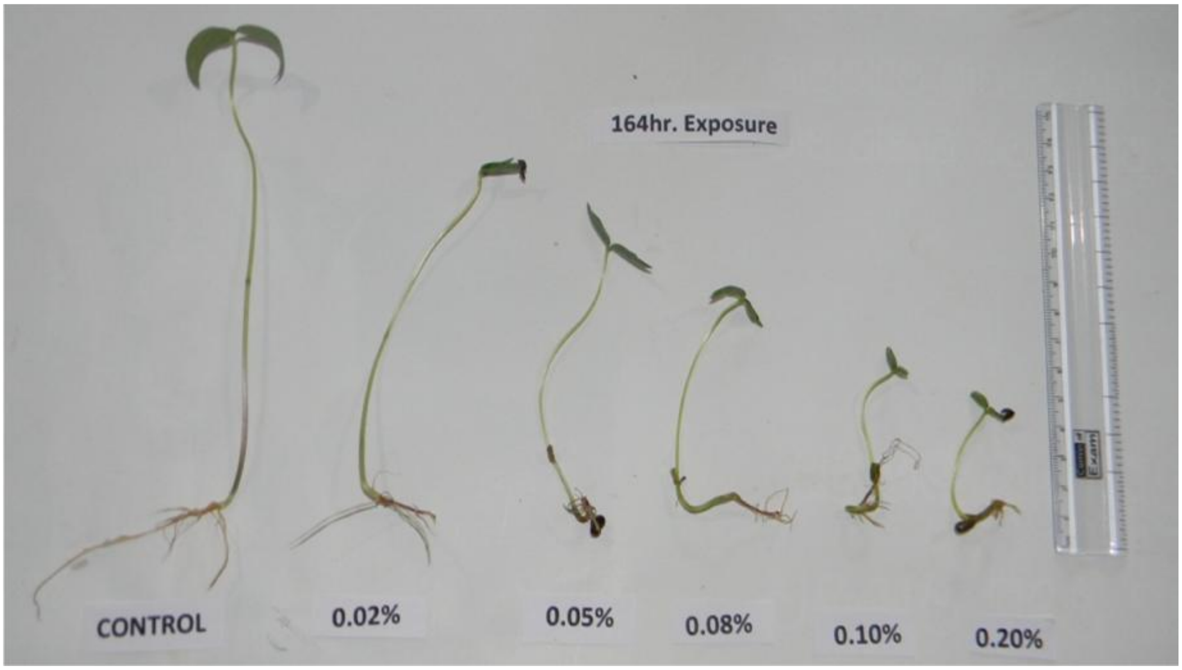

PLATE 1 : Changes in Morphological features in V.radiata (mung) 7 days old Seedlings after Profenofos (%) exposure (Control was grown with distilled water only)

DISCUSSION

Extensive use of organ phosphorus insecticide profenofos (PFF) for agricultural and house-hold purposes has led to serious environmental pollution, with potential risk to organisms in the ecosystem. The effects of chlorpyrifos on cultivated plants have been examined by several authors **(**Anonymous, 1989; Gvozdenac *et al*., 2013). In the present study, Profenofos caused phyto toxic effects on tested plant species, manifesting as root and shoot growth reduction or total inhibition. The growth and fruiting pattern of cotton plants were noticeably altered in association with the use of two systemic organophosphorus insecticides. Phorate treatments at planting time delayed seedling emergence and reduced the /lumber of plants per acre at one or more locations, and resulted in greater plant height and leaf size in all tests. Fruiting was delayed or reduced for up to 3 weeks in treated plots at two locations and was earlier and greater at two others. Insect and spider mite control obtained with these materials apparently was not a contributing cause for growth and fruiting differences (Leigh, 1963). The organophosphate compounds are taking the major share of insecticide consumption in India (Aditya *et al*., 1997). Chlorpyrifos is abroad-spectrum organophosphate insecticide being used for more than a decade to control foliar insects that affect agricultural crops, to reduce pod damage (Khan *et al*., 2009; Wu and Laird 2003; Rusyniak and Nanagas, 2004), and subterranean termites (Venkateswara Rao *et al*., 2005). Chlorpyrifos produces hazardous effects on the environment when it is applied directly on plants or mixed with soil (Howard, 1991). The absorption and translocation of chlorpyrifos residue by wheat and oil seed rape root has been studied by Wang *et al*. (2007) and it concluded that, the uptake rate of chlorpyrifos residue by these two plants increased with an increase in the amount of chlorpyrifos residue in soil. Parween *et al*. (2011, 2012) revealed that the exposure of an organophosphorous insecticide, chlorpyrifos proved depressing for nitrogen metabolism and plant growth in Vigna radiata L. Previous studies have demonstrated that dimethoate causes a reduction in plant growth, photosynthetic pigments and photosynthetic activity of Glycine max L. (Panduranga *et al.*, 2005) and *Vigna unguiculata* L. (Mishra *et al*., 2008).The adverse effects of mancozeb on morphological and anatomical traits of *Lens culinaris* L. at different developmental stages has been investigated by Bashir *et al*. (2007). The blocked growth might have resulted from the inhibition of normal cell division or elongation. Mishra *et al*. (2008), Mishra and Mohanty (2008) had studied the effect of Endosulfan on the seedling characteristics of *V. radiata* and *V. mungo* concluded that, even at very low concentration of the pesticide, there is overall inhibitions in the seedling parameters and morphological traits.

Disturbance of seed germination and mitosis could produce severe consequences for root growth and development. This was clearly demonstrated for the roots of *Pisum sativum* and *Zea mays* with different concentrations of the sulfonylurea herbicides chlorsulfuron and metsulfuronmethyl which caused severe injuries of the root growth (Fayez *et al.,* 1994).After the treatment with different concentrations of pesticides, we observed that the percentages of seed germination decreased while the concentrations increased for each concentration of profenofos. The root and shoot length, dry and fresh weight of both root and shoot also declined with profenofos exposure. Insecticide mixtures *viz*., Roket, colphos, Nimbicidine + Biolep, eucalyptus + lemon grass oil + neem oil and profenofos (organophosphate insecticide) generally used against *Helicoverpa* *armigera* significantly increased chlorophyll content 'a' in chickpea leaves while application of endosulfan + Biolep showed sharp decline in chlorophyll content 'a'. Endosulfan at 0.07% concentration with 0.5% Biolep proved more toxic to chlorophyll content 'a' than Endosulfan at 0.05%. (Srivastava *et al*., 2006).In the present case, the chlorophyll content and other pigments showed a decreasing trend but was not statistically significant.

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