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METABOLIC EFFECTS OF PESTICIDE EFFLUENTS ON *NICOTIANA TABACUM* AND *VIGNA RADIATA* PLANTS

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Abstract

Some important metabolic parameters were investigated in relation to various doses of pesticide effluents in two economically plants viz. *Nicotiana* and *Vigna* plants. Increasing doses of pesticide effluents caused decrease in both sugar and protein concentration almost similarly in both plants. However, activities of enzymes, catalase, peroxidase and MDA content were found to be stimulated by higher doses of pesticide factory effluents in both the plants. Protein and sugar content decreased at all doses in *Nicotiana tabacum* however there were increase in enzymatic activity like catalase and peroxidase. MDA content was also increased in same plant. On the other hand *Vigna radiata showed inducing nature of protein content at lower concentration (25%) and* thereafter inhibit at all doses while sugar content decreased at all concentrations. Enzymatic activity *viz.* catalase and peroxidase showed promoting nature at all concentrations. MDA content also increased at all concentrations. Thus it can be concluded that but for small differences in almost all the metabolic parameters towards their response towards pesticide industry effluent both the studied plants i.e. *Nicotiana* and *Vigna* were found to be quite susceptible towards pesticide effluents.

Key words: Pesticide effluent, *Nicotiana tabacum* L., *Vigna radiata* L., catalase, peroxidase, proteins, sugars, MDA content

Introduction

Pesticides and other organic pollutants are highly detrimental for the environment, plants and humans alike. Pesticides usually have direct compounds are toxic and carcinogenic in nature even at low concentration in liquid medium.

Along with municipal point source and industrial discharges, urban storm, water runoff has been identified as a primary source of pollution of surface water with pesticide residues. The process of pesticide removal from industrial wastewater is of great importance because of well-known pesticide resistance to microbial degradation and its ability of accumulation in the environment as well as possible carcinogenic and mutagenic properties. The main causes of surface water and ground water contamination are discharges from pesticide industries.

This aspect in view, the present investigation was carried out to study the effect of various doses of pesticide industry effluent on certain metabolic parameters in *Nicotiana* and *Vigna* plants.

Materials and Methods

In this study, assessment of certain metabolic parameters at different doses of pesticide industry effluents were carried out in two economically important plants viz. *Nicotiana* and *Vigna*. Pesticide effluent was obtained from Indian Pesticide Ltd. Located at Aishbagh, Lucknow (India). Liquid pesticide industry effluent was diluted to make its concentration in the % of 25, 50, 75 and the last dose of pure pesticide effluent i.e. of 100%.

Soil culture technique

Experiments were carried out in soil as pot culture under controlled glass house conditions. The soil samples for the experiment were collected from outskirts of Lucknow. These were allowed to dry in shade, then powdered in agate mortar, finally sieved through a 1 mm sieve before analyzing the different chemical characteristics of the soil.

Medium sized earthen clay pots were used to carry out the experiments. The central drainage hole provided in these pots for leaching purpose was covered with an inverted watch glass over a pod of clean glass wool. The soil was thoroughly washed with water filled into these pots. Repeated flushing with distilled water was carried out to maintain the soil pH.

Nutrient solution

The composition of the nutrient solution used was based on the Long Ashton formula (Hewitt, 1966). Analytical reagents (AR) were used to prepare both the macro and micronutrient solutions. A concentrated stock solution of each macronutrient was prepared separately by dissolving AR grade salts directly in distilled water, which on 500 times dilution gave the required concentration of the nutrients in the medium.

Following metabolic parameters were observed at 8 weeks growth of each group of plants. All parameters were carried out in the fresh leaf lamina which was finely chopped and 100 mg of the material for each parameter was taken and was ground in a clean pestle and mortar with 10 ml acetone.

Protein concentration was estimated by the method of Lowry *et al*. (1951) while sugar was estimated by the method of Dubias *et al*. (1956). Catalase activity was measured by the method of Euler and Josephson (1927). Peroxidase activity was measured by the method of Luck (1963). Level of lipid peroxidation was estimated in terms of malondialdehyde (MDA), a product of lipid peroxidation in plant samples by the method of Heath and Packer (1968).

Results and Discussion

A. Nicotiana tabacum

a. Protein content

It was found to be significantly decreased by 14.84, 24.35, 34.20 and 51.39% at 25, 50, 75 and 100% concentration respectively as compared to the control. It was found to be 126.003 μg/g fw for control plants, decreasing maximum to 61.260, for 100% effluent concentration (Table 1).

b. Sugar content

Sugar concentration decreased significantly. It was 65.933 μg/g fw for control plants and decreased by 50.19% at 32.840 μg/g fw for 100% effluent concentration (Table 1).

c. Enzyme activity

i. Catalase

Table 1: Effect of various concentrations of pesticide factory effluent on the concentrations of sugar and protein of *Nicotiana tabacum* L

Concentrations (%)	**Sugar concentration (µg/g fw)**	**Protein concentration (µg/g fw)**
Control	65.933± 3.059	126.003± 0.741
25	60.317±0.333 (-8.52%)	107.299*±2.735 (-14.84%)
50	54.400**±0.650 (-17.49%)	95.322**±7.265 (-24.35%)
75	44.556**±2.504 (-32.42%)	82.912**±3.371 (-34.20%)
100	32.840**±0.921 (-50.19%)	61.260**±1.320 (-51.39%)

All values are mean of triplicates ±□S.E. ANOVA ($p<0.01$)

**Significant ($p<0.001$) compare to control, *Significant ($p<0.01$) compare to control.

Table 2: Effect of various concentrations of pesticide factory effluent on different enzymes and MDA content of *Nicotiana tabacum* L

Concentrations (%)	**Catalase activity (µ moles H_2O_2 decomposed/mg fw)**	**Peroxidase activity (ΔOD/g protein)**	**MDA (µM/g fw)**
Control	32.33± 0.167	98.300± 0.900	48.757± 0.378
25	45.667**±0.667 (+41.23%)	154.867**±0.467 (+57.55%)	50.447*±0.336 (+4.20%)
50	60.667**±0.667 (+87.62%)	186.500**±1.500 (+89.73%)	53.823**±0.038 (+10.82%)
75	106.667**±1.667 (+229.89%)	194.733**±2.267 (+98.10%)	55.135**±0.365 (+13.10%)
100	166.667**±9.667 (+415.46%)	209.033**±0.167 (+112.65%)	65.515**±0.355 (+38.50%)

All values are mean of triplicates ±□S.E. ANOVA ($p<0.01$)

**Significant ($p<0.001$) compare to control, *Significant ($p<0.01$) compare to control.

The activity was drastically increased at increasing effluent concentrations. It increased by 415.46% of the control at 100% and 229.80% at 75%, respectively. Maximum increase is noted in the higher concentrations (Table 2).

ii. Peroxidase

Peroxidase activity increased significantly with increasing concentrations of pesticide effluent. The rate of increase is by 57.55, 89.73, 98.10 and 112.65% at 25, 50, 75 and 100% effluent concentrations, respectively (Table 2).

iii. Lipid peroxidation

MDA content in *Nicotiana tabacum* increased with progressive increase in effluent concentrations. It exhibits 4.20, 10.82, 13.10, 38.50% increase at 25, 50, 75 and 100% concentration, respectively (Table 2).

B. Vigna radiata

While in *Vigna radiata* plants following results were obtained with regard to same metabolic parameters.

a. Protein content

It was found to be decreased significantly (p<0.01) with increasing concentration of pesticide industry effluent except for 25% effluent concentration, where it was increased by 1.49% (Table 3).

Table 3: Effect of various concentrations of pesticide factory effluent on the concentrations of sugar and protein of *Vigna radiata* L

Concentrations (%)	**Sugar concentration (μg/g fw)**	**Protein concentration (μg/g fw)**
Control	76.567± 9.893	559.353± 4.870
25	48.667*±2.030 (-36.44%)	567.708±3.836 (+1.49%)
50	37.933**±1.033 (-50.46%)	529.684±0.684 (-5.03%)
100	35.150**±0.650 (-54.09%)	409.014±8.978 (-26.88%)

All values are mean of triplicates ±S.E. ANOVA (p<0.01)

**Significant (p<0.001) compare to control, *Significant (p<0.01) compare to control.

b. Sugar content

Sugar content was observed to decreased significantly ($p<0.01$) with increasing pesticide factory effluent concentrations by 36.44, 50.46 and 35.15% for 25, 50 and 100% effluent concentration (Table 3).

c. Enzyme activity

i. Catalase

Activity was increased on increasing effluent concentrations in dose dependent manner. It became 33.78% of the control at 100% and 18.12% at 75% effluent concentrations, respectively (Table 4).

Table 4: Effect of various concentrations of pesticide factory effluent on different enzymes and MDA content of *Vigna radiata* L

Concentrations (%)	**Catalase activity (μ moles H_2O_2 decomposed/mg fw)**	**Peroxidase activity (ΔOD/g protein)**	**MDA (μM/g fw)**
Control	149.000± 0.577	55.400± 2.600	63.550± 0.310
25	153.333±0.882 (+2.91%)	64.800*±1.000 (+16.97%)	67.668±0.378 (+6.48%)
50	176.000**±0.577 (+18.12%)	97.800**±1.400 (+76.53%)	73.548±2.248 (+15.73%)
100	199.333**±1.764 (+33.78%)	116.133**±0.267 (+109.63%)	83.970*±3.370 (+32.13%)

All values are mean of triplicates ±S.E. ANOVA ($p<0.01$)

**Significant ($p<0.001$) compare to control, *Significant ($p<0.01$) compare to control.

ii. Peroxidase

Peroxidase activity was increased significantly with increasing concentrations of pesticide effluent. Maximum increase was noted for higher effluent concentrations of 50 and 100%. The values were 64.8, 97.8 and 116.133 ΔOD/g protein for 25, 50 and 100% concentrations and 55.4 ΔOD/g protein for the control (Table 4).

iii. Lipid peroxidation

MDA content was measured in the plants for progressive increase in effluent concentrations. Increase observed was by 6.48, 15.73 and 32.13% for 25, 50 and 100% effluent concentrations, respectively (Table 4).

In the present study a decrease in protein concentration was found in higher concentrations of pesticide effluent. Protein and lipid content reduction could be due to disturbance in the nitrogen metabolism of the plants. Nitrogen cycle is responsible for the synthesis of proteins in plants. The results are in conformity with that obtained from a study conducted by Azmat *et al*. (2009) where considerable reduction in the level of protein and lipid content in the leaves of plant treated with various concentrations of effluent was observed. Similarly inhibition of protein content was also reported in synthetic pesticide dimethoate treated *V.radiata* plant.

A decrease in total sugar content was observed with increasing effluent concentrations. The reason for reduced sugar (carbohydrate) content could be retarded growth and reduced chlorophyll content in plants which in turn adversely effects photosynthesis and thus carbohydrate content was also reduced. A study conducted by Malla and Mohanty (2005) confirms the present study results.

Catalase activity was found to be increased with increasing effluent concentrations. This might be due to the fact that catalase is an iron enzyme. The analysis of the effluents showed that they contain excess amount of iron which might have increased catalase activity.

Activity of another antioxidant iron enzyme viz. peroxidase was found to be increased with increasing concentrations of effluents. Many chemicals including metals like iron which are present in the industrial effluent might be responsible for increasing the activity of peroxidase enzyme activity. Enhanced peroxidase activity might have caused due to stress conditions of the cell. Activities of peroxidase expression have been shown in several plant systems to be altered by stress chemicals and infection (Herbette *et al*., 2003).

The level of lipid peroxidation measured in terms of thiobarbituric acid reactive substances (TBARS) increased persistently with increase in effluent concentrations. MDA content is the decomposition product of polyunsaturated fatty acids of biomembranes and its increase shows the extent of membrane lipid peroxidation (Blokhina *et al*., 2003).

Thus it can be concluded that but for small differences in almost all the metabolic parameters towards their response towards pesticide industry effluent both the studied plants i.e. *Nicotiana* and *Vigna* were found to be quite susceptible towards pesticide effluents.

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