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COMPARATIVE STUDY OF EFFECT OF THE UNUSED PETROL AND USED (OIL MIXED) PETROL ON SOIL MACRONUTRIENTS, C\N RATIO OF SOIL AND CERTAIN ANTIOXIDANT LEVELS OF TWO LEGUMINOUS PLANTS

M. Arul Nangai
Department of Biochemistry, Dr. G. R. Damodaran College of Science, Avinashi Road, Civil Aerodrome Post, Coimbatore-641 014, T.N.

ABSTRACT

This study deals with the soil macronutrients and certain plant antioxidants, which can be used to assess the soil pollution in petrol mixed soil and garage soil. The study shows that unused petrol is responsible for greater plant stress in the two legumes studied compared to the used (oil mixed) petrol of garage soil.

INTRODUCTION

With the expanding automobile industry, there is an increase in fuel consumption, its production and spillage. The make shift garages contribute to soil pollution in a major way. These places are non-permanent utility areas and are abandoned when non-convenient for use. The study compares such garage soil samples with 5%, 15%, 25% petrol mixed soil. Petrol is representative petroleum product composed of various hydrocarbons. Petrol is volatile compared to other forms of fuel. The vapour is highly mobile within the soil void space and can accumulate in confined spaces. Many of the non-aliphatic hydrocarbons naturally present in petrol, as well as many anti-knocking additives are carcinogenic. The present study was done to compare the effects of unused petrol and used petrol on soil macronutrient levels, soil C\N ratio, percentage germination and levels of plant antioxidants like ascorbic acid, tocopherol, catalase, peroxidase, glutathione reductase and super oxide dismutase in two legumes *Cicer arietinum* (chick pea) and *Dolichos biflorus* (horsegram). The study may act as preliminary assessment for land reuse and plant growth potential.

MATERIALS AND METHODS

The red soil was obtained by taking soil and sand in the ratio of 3:1. Petrol was mixed with red soil in the ratio of 5%, 15% and 25% for the study. Mixed samples of the local garages were used for the present study. The soil pH values were measured using pH meter. Estimation of available nitrogen in soil samples was done by the alkaline permanganate method. The amount of phosphorus present in the soil samples was determined by Olsen's method. The potassium in soil was determined by neutral normal ammonium acetate method using flame photometry. The organic carbon content of the soil was estimated by method of Muthuvel et al. (1992). Germination studies were carried out as indicated by Zucconi et al. (1981). The legumes were grown for two-three weeks and leaf extracts were used to carry out the assays. Levels of vitamin C were determined by the method of Harris & Ray (1935) and vitamin E by Kivcak & Mert (2001) method. Protein was estimated by Lowry's method (Lowry et al. 1951). The catalase activity was determined by Sinha's method (Sinha 1972). Peroxidase activity of leaf extract was determined by method of Putter & Beker (1983). Glutathione

reductase activity was determined by Smith et al. (1988) method. The activity of superoxide dismutase was estimated by the method of Das & Chany (2000). All the assays were carried out in triplicate to obtain reliable results.

RESULTS AND DISCUSSION

The results of the study are given in Tables 1, 2, 3 and 4. The values of soil pH indicate alkaline nature of the garage soil samples so also is the pH of 25% petrol mixed soil sample in accordance with the earlier study (Diana et al. 2004). The nitrogen and phosphorus content is reduced in petrol mixed soil as compared to control soil sample (Diana et al. 2004). Both, soil disturbance and hydrocarbon addition increase soil pH (Udo & Fayemi 1975) and decreases available phosphorus (Naeth et al. 1987*)*. Low nitrogen is also due to soil disturbance, while hydrocarbon addition increases total nitrogen only slightly (Stahl & Williams1986, Stahl et al. 1988). The potassium content is found to be enhanced in all of the polluted soil samples compared to the control in this study. As expected, the C\N ratio is also altered to a large extent in the garage soil sample as compared to other polluted soil samples and the control sample (Diana et al. 2004). The percentage of germination of the legumes is altered to a lower extent (Smith et al. 2006). The antioxidants levels are reduced in both the legumes. The extent of reduction in both, non-enzymic (ascorbic acid, α-tocopherol) and enzymic antioxidants (catalase, peroxidase, glutathione reductase, superoxide

Table 1: pH of the control and polluted soil samples.

Samples	pH Values
Control	7.3
Garage sample	8.2
5% Petrol mixed soil	7.1
15% Petrol mixed soil	7.5
25% Petrol mixed soil	7.8

Table 2: Nirtogen, phosphorus, potassium, carbon and C/N ratios of control and polluted samples.

Samples Parameters	Control soil sample	Garage sample	5% petrol mixed sample	15% petrol mixed sample	25% petrol mixed sample
Nitrogen(mg/kg)	149.02	155.82	85.82	88.15	82.52
Phosphorus (mg/kg)	1192.69	985.37	666.5	696.6	686.47
Potassium (mg/kg)	128.8	176.68	164.70	160.14	158.51
Carbon (mg/kg)	230	1033	145	182	293
C/N ratio	14	144.5	24.26	22.3	22.2

Table 3: % Germination of the two legumes (Mean ± SD).

SampleSeed type	Control	Garage soil	5% petrol mixed soil	15% petrol mixed soil	25% petrol mixed soil
Chick Pea	8 ± 0.73	5 ± 0.66	5.6 ± 0.59	6.6 ±0.7	6 ± 0.9
Horse Gram	8 ± 0.75	6 ± 0.62	8 ± 0.62	7.3 ± 0.65	7.6 ± 0.58

Table 4: Antioxidant levels in chick pea and horse gram.

Soil Sample Type of Antioxidant	Control	Garage Soil	5% petrol mixed soil	15% petrol mixed soil	25% petrol mixed soil
Antioxidant levels of Chick Pea					
Ascorbic acid (mg/g tissue)	2.780 ± 0.01	2.05 ± 0.024	1.860 ± 0.01	1.72 ± 0.016	0.480 ± 0.033
α-tocopherol(mg/g tissue)	0.754 ± 0.003	0.560 ± 0.004	0.528 ± 0.01	0.425 ± 0.02	0.367 ± 0.01
Catalase(µg/mg protein)	0.576 ± 0.005	0.384 ± 0.01	0.295 ± 0.02	0.208 ± 0.00	0.192 ± 0.005
Peroxidase(µg/mg protein)	0.236 ± 0.04	0.222 ± 0.02	0.198 ± 0.01	0.165 ± 0.02	0.142 ± 0.002
Glutathione reductase(µg/mg protein)	0.168 ± 0.004	0.132 ± 0.003	0.106 ± 0.02	0.095 ± 0.00	0.074 ± 0.001
Superoxide dismutase(µg/mg protein)	0.582 ± 0.003	0.401 ± 0.02	0.324 ± 0.01	0.298 ± 0.03	0.172 ± 0.04
Antioxidant levels of Horse Gram					
Ascorbic acid(mg/g tissue)	1.95 ± 0.041	1.62 ± 0.016	1.46 ± 0.02	1.05 ± 0.016	0.98 ± 0.008
α-tocopherol(mg/g tissue)	1.007 ± 0.021	0.892 ± 0.019	0.732 ± 0.02	0.549 ± 0.01	0.458 ± 0.002
Catalase(µg/mg protein)	0.392 ± 0.002	0.318 ± 0.01	0.296 ± 0.02	0.205 ± 0.00	0.1 ± 0.005
Peroxidase(µg/mg protein)	0.172 ± 0.02	0.164 ± 0.01	0.151 ± 0.00	0.138 ± 0.01	0.096 ± 0.002
Glutathione reductase(µg/mg protein)	0.166 ± 0.002	0.129 ± 0.002	0.116 ± 0.02	0.099 ± 0.00	0.078 ± 0.001
Superoxide dismutase(µg/mg protein)	0.562 ± 0.01	0.411 ± 0.021	0.331 ± 0.01	0.289 ± 0.02	0.161 ± 0.01

dismutase) was similar.

Petrol may cause altered soil macronutrient levels of nitrogen, phosphorus, potassium and carbon due to soil disturbance (Diana et al. 2004). The C\N ratio of petrol mixed soil samples should allow plant growth. The percentage of legume germination shows that the germination of legumes is unaffected by the hydrocarbon based pollutants. The study also confirms that germination studies alone would not predict the subsequent growth of the species (Smith et al. 2006*)*. The antioxidant levels are indicative of plant stress. The present study reaffirms that there is significant toxicity of polycyclic aromatic hydrocarbons present in petrol to the plants (two legumes in this case) and the suitability of multiple biomarker assessment to characterize mechanism of oxidative stress and to serve as an early warning of phytotoxicity *in vivo* (Pakova et al. 2006*)*. It may be concluded from the present study that plants grown in unused petrol mixed soil samples show greater plant stress than the plants grown in garage soil sample with used (oil mixed) petrol. Fedorak and Westlake (1981) also reported a more rapid attack of aromatic hydrocarbons during the degradation of crude oil. This study would help in formulating strategies to recover some of the abandoned roadside garages, a necessary menace that springs up all around and in the outskirts of city limits.

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