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Performance of *Mucuna pruriens* var. Veracruz following pollution with petroleum products

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Abstract

Petroleum products are derivatives of crude oil with long chain hydrocarbon. The fate of an event of spilling of these derivatives in an ecosystem dominated by a natural colonizing nitrogen fixing leguminous plant: *Mucuna pruriens* is the emphasis of this study. Petroleum products (Petrol: Premium Motor Spirit – PMS; Diesel: Automobile Gasoline Oil – AGO and Kerosene: Dual Purpose Kerosene – DPK) on the performance of velvet bean (*Mucuna pruriens* var. Veracruz) was experimented in the Botanical Garden of the department of Plant Science and Biotechnology, University of Port Harcourt. The petroleum products used as treatments which were Treatment 1: control – no pollutant was introduced, treatment 2: 1000ml of PMS, treatment 3: 1000ml of DPK; treatment 4: 1000ml of AGO; treatment 5: 500ml of PMS + 500ml of DPK; treatment 6: 500ml of PMS + 500ml of AGO; treatment 7: 500ml of DPK + 500ml of AGO and treatment 8: 333ml of PMS + 333ml of DPK + 333ml of AGO and. these treatments were replicated six times making it forty-eight potted bags containing 30kg of soil each. Reduction in the growth and development of plant was observed in the polluted treatments as against the control (non – polluted treatment) as a result of deleterious effect of the petroleum products on the morphological parameters (plant height, number of leaves and leaf area) tested for. The fresh and dry weights of plants in the polluted treatments were also lower than the control. Statistical ($p \leq 0.05$) evidence and visual rating reveals that treatment 5 (PMS and AGO) had the best growth amongst the polluted treatments while treatment 3 (diesel) affected the plants growth negatively. Conclusively, the presence of nodules in the roots of velvet bean made the soil to recover fast but the introduction of petroleum products on the growth and development of Velvet bean should be avoided.

Keywords: Growth, petroleum products, botanical garden, velvet bean, potted bags and morphological parameters.

Introduction

Human activities either directly or indirectly have resulted in the introduction of numerous chemicals into the environment. Pollution of the environment is no longer of local or regional concern; it has now reached a global proportion. Issues as climate change and ozone layer depletion serve to emphasize the global scope of the impact of environment for their proper growth, development and other requirements of life. Pollution as defined by Kinako and Awi-waadu, (2000) as the presence in the environment of materials or substance whose nature, location or quantity cause an undesirable environmental effect. According to Engelking (2005), pollution is the contamination of Earth's environment with materials that interferes with human, the quality of life or the natural functioning of living organisms and their physical surrounding. Most times, pollution is caused by human actions although some can be consequences of natural disasters like volcanic eruption, hurricane, flooding etc. There are 3 major types of pollution which are water, air and land / soil pollutions. Point – source pollution comes from a particular, distinguishable and limited source such as sewage pipelines and industrial smokestacks while non – point-pollution comes from uncontained, scattered or dispersed sources as contaminated water run-off from urban areas or automobiles emissions. Petroleum can be accidentally or deliberately introduced into the environment leading to serious pollution problems (Thousand, Bavda, Ovdot, kirsh, Sutton and Vadalie, 1999). These pollution problems often result in alteration of both the biotic and abiotic components of the ecosystem (Mueller, Resnick, Shelton and Pritchard, 1992). In Nigeria (where this study was conducted), the major cause of crude oil pollution is as a result of pipeline vandalisation by saboteurs (individuals and groups) seeking government attention to improve economic marginalization and ecological disaster occasioned by many years of unregulated crude oil exploration and exploitation by foreign companies. This has led to the loss of species diversity, loss of habitat, destruction of breeding grounds of aquatic organism and sometimes death of organisms including man (Ndimele, 2008; Ochekwu and Madagwa, 2013).

Petrol (Premium Motor Spirit – PMS), kerosene (Dual Purpose Kerosene –DPK) and diesel Automobile Gasoline Oil – (AGO) are essential petroleum products obtained through a fractional distillation process. Petrol also called gasoline is obtained at boiling temperature of about 30^0C - 200^0C (86^0F - 392^0F); kerosene is obtained at the boiling temperature of about 150^0C - 270^0C (302^0F - 518^0F) and diesel boils off beyond 275^0C (Dull, 1970). The components of crude oil have different sizes, weights and boiling temperatures and this can be separated by a process called fractional distillation. Petroleum gas (I to 4 carbon atoms), Gasoline or Petrol (5 to 12 carbon atoms), Gas oil or Diesel Distillate (12 or more carbon atoms), Kerosene (10 to 18 carbon atoms), Lubricating oil (20 to 50 carbon atoms), Heavy Gas or fuel oil (20 to 70 carbon atoms), Residue (70 or more carbon atoms)

Mucuna pruriens var. Veracruz belongs to the family Fabaceae and is the third largest among flowering plants with 650 genera and 20,000 species and about 100 varieties (Kay, 1979). The plant is a vigorous, annual, climbing legume. It is a shrub with long vines that can reach over 15m in length. The leaves are tripinnate; the sides of the leaves are often heavily grooved and the tips are pointy. The seeds are creamy - white, shimmy black or brown drift seeds. It is found in tropical African, India and Caribbean. Duke (1981) classifies velvet bean as a short – day plant. It is fairly tolerant of acid soils, the pH requirement for this plant is wide but for optimum yields, a pH above 5 is required [8]. Velvet bean can effectively absorb inorganic nitrogen from the soil and can respond to applied nitrogen just like any leguminous plant. Available soil nitrogen can reduce nodulation and effective nitrogen fixation in legumes. Nitrogen fertilization of velvet bean is not recommended because of the presence of Nitrifying bacteria (*Nitrosomonas* sp and *Nitrobacter* sp) in the nodules in the roots of velvet bean that can help convert atmospheric nitrogen into ammonium to nitrite and finally to nitrate which is the absorbable form (Anyanwu, Awi-waadu, Tanee and Ochekwu, 2014). Hence, the objectives of this study were to determine the effects of petroleum products (petrol, kerosene and diesel) on the germination and growth of velvet bean and to determine which of these products has the most deleterous effect on the soil.

Materials and Methods

Study area

The study was carried out at the Botanic garden of the department of Plant Science and Biotechnology, University of Port Harcourt from July 2018 to September 2018.

Collection of Seeds

Velvet bean seeds used for this study were collected from the International Institute of Tropical Agriculture (IITA) Ibadan, Oyo state, Nigeria while the petroleum products (PMS, DPK and AGO) were obtained from Nigerian National Petroleum Company (NNPC), mega filling station on East – West Road, Port Harcourt, Rivers State, Nigeria.

Collection of Soil, Experimental Design, Pollution Level and Layout

Thirty (30) kg of surface soil (0 – 15cm) were collected from the Botanical garden of the department of Plant science and Biotechnology, University of Port Harcourt and were added into the potted bags. A Completely Randomized Design with 8 treatments, 6 replicates giving rise to 48 observations was employed in the study. The treatments include

- Treatment 1: No pollutant was introduced into the soil (Control).
- Treatment 2: 1000ml of PMS (Petrol),
- Treatment 3: 1000ml of DPK (Kerosene);
- Treatment 4: 1000ml of AGO (Diesel);
- Treatment 5: 500ml of PMS + 500ml of DPK;
- Treatment 6: 500ml of PMS + 500ml of AGO;
- Treatment 7: 500ml of DPK + 500ml of AGO;
- Treatment 8: 333ml of PMS + 333ml of DPK + 333ml of AGO

Experimental method

The potted bags containing 30kg soil were polluted with the different pollutants except for the control which had no pollutant. The pollutants were left in the soil for 2weeks after which 5 velvet bean seeds were planted and thinned to 1 stand per bag after 2weeks after planting.

Parameters

Morphological and soil physico-chemical parameters were taken on the velvet bean plants and soil respectively. Morphological parameters which include plant height. number of leaves and leaf area were taken at 2, 4, 8 and 12 weeks after planting. Fresh and dry weights of the velvet bean plants were taken at harvest (12 weeks after planting). Soil physico-chemical parameters taken include soil pH, conductivity, soil Nitrogen, alkalinity and Total Organic matter (Table 1).

Table 1: Physico – chemical analysis of the pollutant before planting

Physico–Chemical Parameters/ Treatments	Nitrogen	Total Organic Matter (%)	pH	Conductivity (µS/cm)	Alkalinity (mg / kg)
Control	0.l5	5.8	6.48	0.11	4.0
PMS	0.084	4.2	7.54	005	10.6
AGO	0.098	3.7	6.18	007	8.2
DPK	0.098	7.2	6.65	012	0.208

Data Analysis

The data collected were analysed using (SAS, 2007) software and analysis of variance (ANOVA) was determined at 5 % level of probability. The means were separated using Least Significant Difference (LSD).

Result and Discussion

The deleterious effects of these pollutants on velvet bean plants were observed on one plant density of the velvet bean plant by evaluating the plant heights, leaf area, number of leaves, fresh and dry weights and comparism made with the control from planting to harvesting.

Effect of the pollutant on Plant Height, Number of Leaves and Leaf Area on velvet bean at Two Week After Planting

The velvet bean grown on polluted soil didn't perform well when compared with the control. By visual assessment plants grown in all the pollutants had poor growth but it was obviously observed with plant grown on treatment PMS + AGO + DPK polluted soil. Statistical evidence in the plant heights reveal that there were no significant ($P \leq 0.05$) differences recorded between the treatments control, PMS and DPK. These treatments however were significantly ($P \leq 0.05$) higher than other treatments. The other treatments were also not significantly ($P \leq 0.05$) different (Table 2). The number of leaves at two weeks after planting showed rather a different interpretation in which case treatment DPK was significantly ($P \leq 0.05$) higher than other treatments followed by treatment PMS + AGO and the rest treatments were not significantly ($P \leq 0.05$) different from each other (Table 2). Leaf area at two weeks after planting shows that the control was significantly ($P \leq 0.05$) higher than other treatments and treatments PMS, DPK, PMS + AGO, AGO + DPK and PMS + AGO + DPK were significantly ($P \leq 0.05$) similar but significantly ($P \leq 0.05$) higher than treatments AGO and PMS + DPK (Table 2).

The carbon to Nitrogen ratio (C: N) were altered when petroleum hydrocarbons contaminate the soil. The added carbon from the petroleum hydrocarbon stimulates the numbers of microbes but causes an imbalance in the carbon C: N ratio which may result in the immobilization of the soil nitrogen by the microbial biomass leaving none available for the growth of the plant (Adams and Duncan, 2003). Oil spillage on soil makes it unsatisfactory for plant growth (De Jong, 1980). This is due to insufficient aeration of the soil because of the displacement of air from the spaces between soil particles by petroleum. Udo and Fayemi (1975) reported that plants growing on polluted soil were generally retarded and showed chlorosis on leaves coupled with dehydration of plants indicating deficiency.

Table 2: Mean plant height (cm), number of leaves and leaf area (cm^3) on velvet bean at two weeks after planting

Treatment	Plant Height	Number of Leaves	Leaf Area
CONTROL	18.7^{a}	5.3^{ab}	25.7^{a}
PMS	15.2^{a}	5.0^{ab}	16.7^{b}
DPK	14.7^{a}	6.7^{a}	15.2^{b}
AGO	10.1^{b}	4.5^{ab}	13.0^{c}
PMS + AGO	11.9^{b}	4.7^{ab}	15.8^{b}
PMS + DPK	13.5^{b}	5.7^{ab}	14.2^{c}
AGO + DPK	11.5^{b}	5.5^{ab}	16.3^{b}
PMS + AGO + DPK	10.7^{b}	3.3^{b}	18.8^{b}
LSD	5.81	3.36	4.13

The values above are means of six replicates. Means with the same letters of the alphabet are not significantly ($P \leq 0.05$) different from each other

Effect of the pollutant on plant height, number of leaves and leaf area on velvet bean at Four Weeks After Planting

The control was seen to be significantly (P ≤ 0.05) higher than the other treatments. Treatments AGO, PMS + AGO, PMS + DPK, AGO + DPK and PMS, DPK and AGO were significantly (P ≤ 0.05) lower than other treatments (Table 3). At four weeks after planting number of leaves were negatively affected by the treatments in which case treatments PMS, DPK, PMS + AGO, PMS + DPK and AGO + DPK were significantly (P ≤ 0.05) lower than treatments control and PMS + DPK + AGO and significantly (P ≤ 0.05) higher than treatment diesel (Table 3). Leaf area at four weeks after planting showed that control was significantly (P ≤ 0.05) higher than other treatments (Table 3).

Table 3: Mean plant height (cm), number of leaves and leaf area on velvet bean (cm^2) at four weeks after planting

Treatment	Plant Height	Number of leaves	Leaf Area
CONTROL	37.2^{a}	23.2^{a}	37.3^{a}
PMS	23.5^{b}	14.8^{b}	17.8^{b}
DPK	28.5^{bc}	16.2^{b}	22.0^{b}
AGO	19.2^{c}	11.3^{c}	14.0^{c}
PMS + AGO	20.6^{c}	11.8^{bc}	22.0^{b}
PMS + DPK	19.7^{c}	12.2^{bc}	19.8^{b}
AGO + DPK	19.3^{c}	13.3^{b}	21.8^{b}
PMS + AGO + DPK	20.1^{c}	18.3^{a}	22.2^{b}
LSD	6.02	5.16	4.65

The values above are means of six replicates. Means with the same letters of the alphabet are not significantly (P ≤ 0.05) different from each other

Effect of the pollutant plant height, number of leaves and leaf area on velvet bean at eight weeks after planting

At eight weeks after planting, the effect of pollution had reduced as a result of the recovery process of the plant. The non – polluted treatment i.e. the control was significantly (P ≤ 0.05) higher than other treatments. Treatments AGO + DPK and PMS + AGO + DPK was significantly (P ≤ 0.05) lower than the other treatments (Table 4). Number of leaves at eight weeks after planting showed a different trend in which case treatments PMS, AGO, PMS + AGO and PMS + AGO + DPK was significantly (P ≤ 0.05) lower than the control but significantly (P ≤ 0.05) higher than treatments DPK, PMS + DPK and PMS + AGO + DPK (Table 4). The control was significantly (P ≤ 0.05) higher than other treatments for leaf area. Unlike other pollution levels after planting, treatments PMS, DPK and AGO had no significant (P ≤ 0.05) difference between them but were significantly (P ≤ 0.05) lower than treatments PMS + AGO + DPK and AGO + DPK and PMS + DPK (Table 4).

Table 4: Mean plant height (cm), number of leaves and leaf area (cm^2) on velvet bean at eight weeks after planting

Treatment	Plant Height	Number of Leaves	Leaf Area
CONTROL	50.9^{a}	40.5^{a}	42.5^{a}
PMS	38.7^{b}	22.7^{b}	23.8^{c}
DPK	41.9^{b}	23.0^{c}	23.5^{c}
AGO	42.9^{b}	21.5^{b}	24.8^{c}
PMS + AGO	37.3^{b}	26.0^{b}	27.5^{b}
PMS + DPK	30.1^{b}	19.2^{c}	26.0^{b}
AGO + DPK	29.6^{c}	18.8^{c}	23.8^{b}
PMS + AGO + DPK	28.2^{c}	25.0^{b}	26.2^{b}
LSD	8.50	6.73	4.35

The values above are means of six replicates. Means with the same letters of the alphabet are not significantly (P ≤ 0.05) different from each other

Effect of the pollutant plant height, number of leaves and leaf area on velvet bean at twelve weeks after planting (Harvest)

At harvest no significant (P ≤ 0.05) differences existed between the polluted treatments but they were all lower than the control for plant height. Number of leaves at harvest showed that the control was significantly (P ≤ 0.05) higher than other treatments and treatments PMS + AGO and AGO + DPK was next in the significant (P ≤ 0.05) rating as it was seen to be higher than treatments PMS, DPK, AGO and PMS + AGO + DPK. The control was significantly (P ≤ 0.05) higher than the rest treatments with treatments PMS, PMS + AGO and PMS + AGO + DPK being higher than treatments DPK, AGO and AGO + DPK (Table 5).

Okpokwasili and James (1995) reported that presence of petroleum – degrading bacteria has been demonstrated in Bonny light crude oil, kerosene and gasoline. These organisms are more abundant in crude oil (ca. 2.93 x 10^5 cfu/ml) than in kerosene (ca 3.22 x 10^4 cfu/ml) and gasoline (ca. 2.96 x 10^4cfu/ml). Growth of velvet bean would have been hindered because the relationship between the plant and the rhizophere can be affected by petroleum (Ochekwu, 2011).

Table 5: Mean plant height (cm), number of leaves and leaf area (cm^5) on velvet bean at twelve weeks after planting

Treatment	Plant Height	Number Of Leaves	Leaf Area
CONTROL	126.67^a	79.33^a	59.83^a
PMS	60.17^b	36.83^c	35.0^{bc}
DPK	66.37^b	36.50^c	27.5^c
AGO	60.53^b	31.83^c	26.33^c
PMS + AGO	67.68^b	53.0^b	29.83^b
PMS + DPK	55.62^b	35.33^c	33.33^{bc}
AGO + DPK	48.77^b	33.33^{bc}	29.67^c
PMS + AGO + DPK	49.25^b	29.17^c	39.5^b
LSD	15.19	6.73	4.35

The values above are means of six replicates. Means with the same letters of the alphabet are not significantly ($P \leq 0.05$) different from each other

Effect of fresh and dry weights on velvet bean at harvest

The fresh weight of velvet bean at harvest shows that the control was significantly ($P \leq 0.05$) higher than other treatments and no difference existed between among the other treatments. This was not the case for the more stable weight i.e. the dry weight that had the control as highly significant ($P \leq 0.05$) than treatments PMS + AGO and AGO and DPK next in the rating and these treatments in turn were significantly ($P \leq 0.05$) higher than treatments PMS, DPK, AGO and PMS + AGO and DPK (Table 6). AGO is a refined product of crude oil but more deleterious than crude oil due to its viscosity. Crude oil has a higher viscosity than diesel (Kinako, 1981).

The inhibitory effect on the growth of velvet bean treated with the pollutants could be a result of PAC (Polycyclic Aromatic compounds) and sulphur which are major components of these petroleum products hence causing the blockage of pores and resulting in low intake of nutrients. The yellowing of leaves observed might be due to deficiency of N as a result of reduction in Nitrates and inability to fix N by nitrogen fixing bacteria in the plant which is a leguminous plant. Oil pollutants in soil causes decrease in decomposer microbes' composition in soil (Atlas, Schofield, Moreli and Cameron, 1976) and is also known to inhibit nitrogen fixation by rhizobium (Munns, 1965).

Table 6: Mean fresh weight (g) and dry weight on velvet bean at harvest

Treatment	Fresh weight	Dry weight
CONTROL	180.8^a	50.45^a
PMS	44.37^b	15.39^c
DPK	34.57^b	15.45^c
AGO	22.43^b	7.35^c
PMS + AGO	22.17^b	8.41^b
PMS + DPK	28.11^b	10.20^c
AGO + DPK	29.91^b	11.22^{bc}
PMS + AGO + DPK	25.16^b	12.87^c
LSD	25.78	7.28

The values above are means of six replicates. Means with the same letters of the alphabet are not significantly ($P \leq 0.05$) different from each other.

Conclusion

The effects of petroleum products were visible on the leaves and vines of velvet bean on the different treatments concentrations of pollution. AGO affected the plants the most in all the morphological parameters tested for (plant height, number of leaves and leaf area). The overall results showed that plants treated with the pollutants were severely affected. Nodulations of velvet bean may have given rise to the various performances of the morphological parameters. This reduction in the growth of the plants may be as a result in the change of the physical and chemical properties of the soil which may directly or indirectly affect the growth of the plant by creating conditions impossible for water and nutrients to be available to the plants.

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