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Role of nitrogen and phosphate dinamics to increase plant survival grown on oil contaminated soil

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Abstract. The petroleum mining industry plays an important role in the fulfillment of human needs but also has a negative impact on less productive soil due to oil contamination that causes soil nutrient degradation such as nitrogen and phosphate level. This research aims to study the dynamics of nitrogen and phosphate on oil-contaminated soil by utilizing multisymbiotic soil bacteria (*Pseudomonas sp*. and *Rhizobium sp*.) as an effort to improve the quality of nitrogen and phosphate availability that affect plant resistance to survive in the oil-contaminated soil. The research method was Randomized Complete Design with soil bacterial treatments (*Pseudomonas sp*. and *Rhizobium sp*.), consisting of 3 treatment i.e (1) *Pseudomonas sp*., (2) *Rhizobium sp*., and (3) combination of *Pseudomonas sp*. and *Rhizobium sp*. Each treatment at a concentration of 10^8 cells/gram with 4 repetitions. Data was obtained including Total Petroleum Hydrocarbon (TPH), P, C, N concentration in soil, plant growth and environmental parameters including temperature, pH, and soil moisture. Analysis of the data used one way variance analysis followed by LSD test. The results showed that plant survival on oil-contaminated soils is not only determined by the dynamics of nitrogen and phosphate availability but also by the availability of TPH content in the soil that indicated by optimal results when there is a multisymbiotic role of both bacteria that inserted into the soil.

1. Introduction

The main content in petroleum are paraffinhydrocarbons, saturated alicyclic hydrocarbons, and carcinogenic aromatic hydrocarbons and are organic pollutants [1]; [2]. Petroleum can be divided into four classes, namely: saturated, aromatic, asphaltene (phenol, fatty acids, ketones, esters, and porifrin), and resins (pyridine, quinolin, carbasols, sulphoxides, and amides). The type and class of hydrocarbons affect the biodegradation process of the petroleum [3].

Environmental pollution by petroleum and its derivatives is a very common problem found in petroleum mining areas. Oil spills resulting from petroleum mining activities lead to soil contamination, it effect in the very low nutrient content and very high hydrocarbon compounds [4], so that the soil needs to be processed first so that it can be used as a plant medium for plants [5]. One of them is through bioremediation which is currently considered an effective technology for transforming toxic components into less toxic products without any disruption to the surrounding environment [6]; [7].

Beside mycorrhiza, an organism that capable of increasing the availability of soil P is a solvent phosphate bacteria. These bacteria have the ability to dissolve the insoluble P and make available to the plant by dissolving it with organic acids [8]; [9]. Some of phosphate solubilizing bacterial have the ability to degrade hydrocarbon compounds. Research shows that some types of microbes are able to use their carbon sources in the process of hydrocarbon compounds degradation such as: Pseudomonas, Bacillus, Acinetobacter, Alcaligenes, Xanthomonas, Benecdea, Brevi bacterium, Methvlobacterium, Methylococcus, Mycobacterium [10]; [11]; [12]. Some certain bacteria have ability in increasing the availability of phosphate in the soil as well as the ability to degrade hydrocarbon compounds on polluted soil. This type of microbe is feasible to be involved in bioremidiating soil contaminated oil because the degraded hydrocarbon will produce mineral or nutrients that can be used for plant growth.

Legume plant is a unique plant because it is able to perform N-free fixation of air through the utilization of Rhizobium bacteria so as to form the root nodule [13]; [14]. In symbiosis between the legumes and bacteria that form the root nodule, each component of the symbion has a different role [15]. Plants play a role in photosynthate for Rhizobium bacteria and phosphate solubilizing bacteria that have the ability to degrade hydrocarbon compounds [16]; [17]. Rhizobium provides N elements for plants through N-fixation activities [15]; [18] and phosphate solubilizing bacteria provides P elements for plants and Rhizobium [16]; [17]; [19]. The interaction pattern between legumes and Rhizobium provides an information pattern of symbiotic relation mechanisms on legume crops so that plants can survive in their environmental conditions [15].

Analog with this multi-symbiotic pattern will be applied in this study which aim to examine how the dynamics of N and P level in the plant in playing a role against the disadvantage conditions of plants on oil contaminated soil using the legume plants.

Similar study was done by [20] about the influence of nitrogen and phosphorous on the growth and root morphology of plant. Based on these results study indicated that both N and P application significantly affected plant height, root collar diameter, chlorophyll content, and root morphology. On the other study, [21] did a study about the role of nitric oxide in plant responses to abiotic stress. Based on these study, it could be conclude that nitric oxide was a critical component in mediating hormone action, modulating gene expression, and protein activity. In addition, [22] showed that petroleum-derived substances continued to adversely affect the growth of plants even three years after soil contamination. Bioremediation supported by the use of microorganisms is an advantageous solution that permits the improving of the growth parameters of plants, as well as offsetting the harmful effects of petroleum-derived products upon the majority of the analyzed elements. [23] showed that concentration of N and P on oil contaminated soil enhance the biodegradation process by microbe.

2. Methods

The research method used Randomized Complete Design with soil bacterial treatments (*Pseudomonas sp*. and *Rhizobium sp*.), consisting of 3 treatments i.e (1) *Pseudomonas sp*., (2) *Rhizobium sp*., and (3) combination of *Pseudomonas sp*. and *Rhizobium sp*. Each treatment at a concentration of 10^8 cells/gram with 4 repetitions was performed on soybeans and mungbeans.

In the preparation stage, nutrient analysis activities was obtained on soil samples include N, P Content and Total Petroleum Hydrocarbon (TPH). Furthermore, the preparation of the oil-contaminated soil as follows.

a. The soil was dried for 7 days and then was sterilize using the oven at 160°C for 2 hours. After that the soil was left for 1-2 days to be cool and than move on to the 3 kg provided polybag.
b. The soil is added a basic fertilizer and moistened using a water sprayer, then incubated for 7 days. The basic fertilizer composition for 3 kg of soil consisted by 24 mg Urea, 24 mg KCL, 24 mg TSP.
c. The physical and chemical properties of soil were measured including pH, temperature, and soil moisture.

The bacterial culture was carried out at Unesa Microbiology Laboratory using Broth nutrient and nutrient agar. The concentration of bacteria in the solution was calculated by Haemocytometer. The culture of *Pseudomonas sp.* and *Rhizobium sp.* was transferred aseptically to several infusion bottles containing nutrient agar media, then incubated at room temperature (28-30◦C) for 24 hours. Three ose cultures of *Pseudomonas sp.* and *Rhizobium sp.* then inoculated into intravenous bottles containing aseptic nutrient brorth, and let them at room temperature for one week.

One week before the germinated seeds were planted, the concentration of bacteria 10^8 cells/ml for each gram of soil were incubated on the soil. The three germinated seeds were sown in each polybag, after 2 weeks there were two plants each polybag. Removal of weeds was regularly done and the soil moisture (80-85%) was maintained until soybeans or mungbeans reach the maximum vegetative period (aged 8 weeks after planting).

The harvest of the plants was obtained 8 weeks after planting. The data was obtained including total biomass of plants (roots, stems, and leaves), percentage of the effective root nodule (observing the root nodule visually by splitting the root nodule directly), level of TPH in soil by Gravimetri method, N and P content in the plant.

3. Results and discussion

The results obtained from this study include (1) the effect of *Rhizobium sp.* and *Pseudomonas sp.* to decrease of TPH level, content of N, P, effective root nodule, and soybeans biomass, (2) *Rhizobium sp.* and *Pseudomonas sp.* to decreased TPH, content of N, P, root nodules, and mungbean biomass.

3.1. Rhizobium sp. and Pseudomonas sp. affect on TPH decreasing, N, P Contents, root nodule, and biomass of soybean

The effect of *Rhizobium sp.* and *Pseudomonas sp.* to decrease the TPH, the contents of N, P, effective of root nodule, and biomass of soybean are shown in Table 1. The previous TPH concentration before treatment was 63.480 mg/k

Table 1. Effect of *Rhizobium sp. and Pseudomonas sp.* on TPH decreasing, N, P contents, effective of root nodule, and biomass of soybean.

Treatments	Decreasing of TPH (%)	P Content (%)	N Content (%)	Root nodule (%)	Biomass (g)
Controle	04.06±1.89	0.35±0.02	0.36±0.010	16.82±3.002	13.30±3.20
Rhizobium sp.	36.05±2.17	0.93±0.06	0.80±0.015	90.64±9.219	32.20±2.19
Pseudomonas sp.	36.00±1.90	1.01±0.02	0.86±0.040	98.13±2.304	33.75±2.04
Rhizobium sp. & Pseudomonas sp.	48.26±2.42	1.46±0.05	1.39±0.029	96.95±3.458	43.10±0.55

Table 1 indicate that the TPH decreasing in the soil is increased due to the combination of *Rhizobium sp.* and *Pseudomonas sp.* which affect also the soybean growth effectively. The combination of these two types of bacteria also showed the highest results for P and N level in the plant, effective root nodule, and plant biomass than single bacterial treatment of *Rhizobium sp.* or *Pseudomonas sp.* only.

3.2. Rhizobium sp. & Pseudomonas sp. affect on TPH decreasing, N, P contents and biomass of mungbean

Treatment with *Rhizobium sp.* and *Pseudomonas sp.* on the decrease of TPH, contents of N, P, and mungbeans biomass are shown in Table 2.

Table 2. Effect of *Rhizobium sp. and Pseudomonas sp.* on TPH decreasing, N, P contents, and biomass of mungbean.

Treatments	Decreasing of TPH (%)	P Content (%)	N Content (%)	Biomass (g)
Controle	18.59±7.10	0.15±0.017	0.46±0.21	25±2.33
Rhizobium sp.	45.42±7.17	0.56±0.075	1.05±0.21	66.02±3.62
Pseudomonas sp.	47.86±2.43	0.45±0.033	0.98±0.17	62.3±2.46
Rhizobium sp. & Pseudomonas sp.	59.12±9.12	0.65±0.074	2.86±0.14	75.68±2.13

Table 2 reflect that the TPH decreasing in the soil is increased due to the combination of *Rhizobium sp.* and *Pseudomonas sp.* which affect also the mungbean growth effectively. The combination of these two types of bacteria also showed the highest results for P and N level in the plant, and plant biomass than single bacterial treatment of *Rhizobium sp.* or *Pseudomonas sp.* only.

The decrease of TPH is caused by the degradation of hydrocarbon compounds by bacteria added to the oil-contaminated soil. It is known that *Pseudomonas sp.* has the ability to dissolve the phosphate and also degrades the hydrocarbon compound that resulting in mineralization which causes higher availability of nutrients due to hydrocarbon degrading bacteria, that capable of using hydrocarbons as a single carbon source [24]; [25]; [26].

The differences in TPH content in the soil can be affected by various factors. The ability of bacteria itself to degrade hydrocarbon compounds can also be affected by environmental factors that support the continuity of the process of degradation of hydrocarbon compounds by bacteria. *Pseudomonas sp.* degrades the carbon organic material and use it as a source of energy for the aerobic respiration process, so that the organic carbon content in the oil-contaminated soil was reduced, such as aromatic compounds, including benzene and toluene [27]; [28]; [29]; [30].

The mechanism of benzene biodegradation begins with the breaking of aromatic rings by the enzyme dioxygenase [31]. Microbes form dihydrodiol compounds in single ringed aromatic components. Furthermore, microbes perform metabolism and produce catechol or protocatechol compounds [31]; [32]; [33]. These compounds are then broken down with one of two mechanisms namely the ortho mechanism (Ortho pathway) or meta mechanism (Meta-cleavage pathway). Subsequent metabolic results are pyruvic acid, formic acid and acetaldehyde, then into the Krebs cycle which eventually results in H_2O, CO_2 and further compounds [31]; [34].

In addition, the biodegradation of hydrocarbons by microbial communities depends on community composition and adaptive response to the presence of hydrocarbons. Different adaptability causes not all types of bacteria are able to adapt to the presence of hydrocarbon compounds [3]; [4]. The bacteria used in this study were not isolated directly from the location of the oil-contaminated soil in Bojonegoro, which the soil come from, so that the adaptation period for these bacteria was required.

Biodegradation may occur in two or more steps, i.e biotransformation and/or mineralization [35]. The low percentage decrease in TPH levels due to the addition of hydrocarbon degradation type is probably caused by the degradation of hydrocarbons carried out by the bacteria still at the biotransformation stage [35]; [36]; [37]. This means that at that stage only subsequent compounds are produced which have not been degraded completely. These advanced compounds will be mineralized by other bacteria capable of degrading hydrocarbon compounds [38]; [39].

Tables 1 and 2 show that there had been an increase in P-content available in the soil due to the combination of treatment of bacteria than one bacteria solely. The highest increase in P-content occurs on soils with the addition of a combination of *Psedomonas sp.* and *Rhizobium sp.* The increase is greater than single-bacterial treatment. This suggests that *Pseudomonas sp.* bacteria have the ability to increase the availability of P-available in the soil [24]. That mean, the bacteria are able to dissolve the strongly absorbed phosphate in the oil-contaminated soil to be P-available on the soil. Increased P-available in the soil is caused by the presence of organic acids produced by bacteria such as citric acid, glutamate, succinate, lactate, oxalate, glioksalat, malate, fumarate, tartaric, and α-ketobutirat [3]. Microbes produce these organic acids through the process of glucose catabolism and the tricarboxylic acid cycle (TCA), which is a continuation of the glycolysis reaction [31]. These organic acids was link to the ions which bind P and decrease the pH, so P will change from P-bound to P-available. Therefore, soil microorganisms that can dissolve phosphate play a role in improving phosphorus deficiency. Soil microorganisms may also release the soluble inorganic phosphate (H_2PO_4) into the soil through the decomposition of phosphate-rich organic compounds [40]; [41].

Table 1 and Table 2 also indicate that the treatment of *Pseudomonas sp*. and *Rhizobium sp*. is able to increase the levels of N. The process of degradation of organic compounds by degrading bacteria of hydrocarbon compounds releases N which is bound to hydrocarbon compounds, so that N increases [16]; [17]. Beside that, the treatment of *Rhizobium sp*. serve the ability of nitrogenase enzyme to change the N2 to be avalilable for plant so that the acceleration of N metabolism will be increased [15]; [18]. This is indicated by the increasing of effective root nodule formed on the plant roots so that the N availability for the plants can be maintained which affect positively on the growth and development of the plants due to the supply of N and P as well as thus the survival of the plant can be maintained under adverse environmental conditions, for example on oil-contaminated soil [19]; [15].

It is known that oils could be absorb by plants that make oil to become phytotoxic compounds [42]; [43]; [44] because of the burning of plants or compression necrosis exposed by oil [45]. This effect is attributed to the death of cells due to the destruction of cell membranes by acidic compounds from oil [46]. This symptom is probably associated with a high concentration of oil up to more than 8% resulting in rapid leaf loss as a result of the influence of plant hormones [47]. This dramatic effect will occur when oil spraying is performed under extremely low environmental humidity conditions. This risk will increase with the presence of isoalkanes and cycloalkanes, leading to the exclusion of ethylene thereby encouraging the absorption process [48]. In general, the presence of oil with environmental stress conditions due to low humidity will make chronic symptoms, ie loss of vigour, so that leaves, flowers and fruits will easily fall and then affect the production of flowers and fruits. In addition, the presence of oil will disrupt gas exchange and water movement and metabolite compounds in plants [49].

Table 1 and 2 also show that generally there is a positive correlation between the percentage reduction of TPH, N contents, and P contents on plant biomass reflecting on growth and developmental processes of the plant grown on oil-contaminated soil. Optimal plant growth on oil-contaminated soil will be determined by low hydrocarbon compounds in plant medium due to increasing process of oil degradation and also determined by the availability of N and P contents in the plant. It is known that N is needed as a protein-forming material of both functional proteins and structural proteins of cells, while P is an element of the key nutrients involved in the transfer and storage of energy in cells. In generally it can be stated that the plant survival level in polluted soil depends on the availability of N and P elements in the soil which is reflected as a dynamic of N and P availability in the soil.

4. Conclusion

The combination of *Pseudomonas sp*. and *Rhizobium sp*. affected significantly on plant survival (indicated by plant biomass) on oil-contaminated soil. These results reveal that microorganism can improve the plant tolerance and survival in polluted soil mediated by improving plant nutrient status (N and P) in the soil. As a consequence, the multisimbiotic soil microorganism can be used as an alternative to increase the plant nutritional status in order to improve plant tolerance and plant survival in oil-contaminated soils.

References

[1] E M Ogbo and J A Okhouya 2011 Bioavailability of Some Heavy Metals in Crude Oil Contamined Soils Remediated with Pleurotus tuber-regium *Fr. Singer. Asian Journal of Biological Sciences* **4**(1) 53-61

[2] K Santhini, J Myla, S Sajani and G Usharani 2009 Screening of Micrococcus Sp from Oil Contaminated Soil with Reference to Bioremediation *Botany Research International* **2**(4) 248-252

[3] N Das and P Chandran 2011 Microbial Degradation of Petroleum Hydrocarbon Contaminants: An Overview *SAGE-Hindawi Access to Research Biotechnology Research International* **2011**

[4] P K Jain, V K Gupta, R K Gaur, M Lowry, D P Jaroly and U K Chauhan 2011 Bioremediation of Petroleum oil Contaminated Soil and Water *Research Journal of Environment Toxicology* **5**(1) 1-26

[5] S Hardjowigeno 2003 *Ilmu Tanah.* (Jakarta: Akademi Pressindo)

[6] V S Millioli, E-L C Servulo, L G S Sobral and D D Carvalho 2009 Bioremediation of Crude Oil-Bearing Soil: Evaluating the Effect of Rhamnolipid Addition to Soil Toxicity and to Crude Oil Biodegradation Efficiency *Global NEST Journal* **11**(2) 181-188

[7] N T Joutey, W Bahafid, H Sayel and N E Ghachtouli 2013 Biodegradation: Involved Microorganisms and Genetically Engineered Microorganisms *licensee InTech*

[8] J M Igual, A Valverde, E Cervantes and E Velázquez 2001 Phosphatesolubilizing bacteria as inoculants for agriculture: use of updated molecular techniques in their study *Agronomie* **21** 561–568

[9] H Rodriguez, R Fraga, T Gonzales and T Bashan 2006 Genetic of phosphate solubilizing and its potential applications for improving plant growth-promoting bacteria *Plant Soil* 287:15-21

[10] E Rosenber and E Z Ron 1996 *Bioremidiation of Petroleum Contamination In R. L. Crawford., & D. L. Crawford (eds)* Bioremidiation Principle and applications (Cambridge University Press. Cambridge) 100-124

[11] A Mrozik, Z Piotrowska-Seget and S Labuzek 2003 Bacterial degradation and bioremediation of Polycyclic Aromatic Hydrocarbons *Polish Journal of Environmental Studies* **12**(1) 15-25

[12] F Kafilzadeh, P Sahragard, H Jamali and Y Tahery 2011 Isolation and identification of hydrocarbons degrading bacteria in soil around Shiraz Refinery *African Journal of Microbiology Research* **4**(19) 3084-3089

[13] Qi Cheng 2008 Perspectives in Biological Nitrogen Fixation Research *Journal of Integrative Plant Biology* **50**(7) 784–796

[14] A G M Silva, A d C Agular, N Jorge, T de S A Costa and E G Moura 2017 Food quantity and quality of cassava affected by leguminous residues and inorganic nitrogen application in a soil of low natural fertility of the humid tropics *Bragantia, Campinas,* **76**(3) p.406-415

[15] J Clúa, C Roda, M E Zanetti and F A Blanco 2018 Compatibility between Legumes and Rhizobia for the Establishment of a Successful Nitrogen-Fixing Symbiosis *Genes* **9**(125)

[16] A Zaidi, M Ahemad, M Oves, E Ahmad and MS Khan 2010 *Role of Phosphate-Solubilizing Bacteria in Legume Improvement; In: Microbes for Legume Improvement* (Khan MS, Zaidi A, Musarrat J Editors *Springer*) pp 273-292

[17] A Afzal, A Bano, Rhizobium and Phosphate 2008 Solubilizing Bacteria Improve the Yield and Phosphorus Uptake in Wheat (Triticum aestivum) *Int. J. Agri. Biol.* **10**(1)

[18] T C de la Peña, E Fedorova, J J PueyO, M M Lucas 2018 The Symbiosome: Legume and Rhizobia Co-evolution toward a Nitrogen-Fixing Organelle? *Frontiers in Plant Science* **8** 2229

[19] G S Tagore, S L Namdeo, S K Sharma and N Kumar 2013 Effect of Rhizobium and Phosphate Solubilizing Bacterial Inoculants on Symbiotic Traits, Nodule Leghemoglobin, and Yield of Chickpea Genotypes *International Journal of Agronomy* **2013** 8 pages

[20] M Razaq, P Zhang, H Shen and Salahuddin 2017 Influence of nitrogen and phosphorous on the growth and root morphology of Acer mono *Plus One* **12**(2)

[21] M Simontacchi, A Galatro, F R Artuso and G E S 2015 María Plant Survival in a Changing Environment: The Role of Nitric Oxide in Plant Responses to Abiotic Stress *Frontiers in Plant Science* **6** (977)

[22] M Rusin, J Gospodarek and A Nadgórska-Socha 2015 The Effect of Petroleum-Derived Substances on the Growth and Chemical Composition of Vicia faba *L. Pol. J. Environ. Stud.* **24**(5)
[23] T K Vyas and B P Dave Effect of Addition of Nitrogen, Phosphorus, and Potassium Fertilizers on Biodegradation of Crude Oil by Marine Bacteria *Indian Journal of Marine Science* **39**(1)
[24] G Awasthi, A Chester, R Chaturvedi and J Prakash 2015 Study on Role of Pseudomonas aeruginosa on Heavy Metal Bioremediation *Int. J. Pure App. Biosci*. **3**(4) 92-100
[25] S Wasi, S Tabrez and M Ahmad 2013 Use of Pseudomonas spp. for the bioremediation of environmental pollutants: a review *Environ Monit Assess* DOI 10.1007/s10661-013-3163-x
[26] G Stamenov, S S Duric and T I H Jafari 2015 Bioremediation Potential Of Five Strains Of *Pseudomonas* Sp. *Matica Srpska J. Nat. Sci.* **128** 41-46
[27] X D Huang, Y El-Alawi, M D Penrose, B R Glick and B M Greenberg 2005 A multi-process phytoremediation system for removal of polycyclic aromatic hydrocarbons from contaminated soils *Environ Pollut* **81** 130-147
[28] X Liu, J Zou, Z Wang, X Hu, X Liang and J Wei 2012 Degradation of diesel pollutants in Huangpu-Yangtze River estuary wetland using a plant-microbes system *Procedia Environmental Sciences* **16** 656 – 660
[29] A Meliani 2015 *Bioremediation Strategies Employed by Pseudomonas Species In: Maheshwari D. (eds) Bacterial Metabolites in Sustainable Agroecosystem* Sustainable Development and Biodiversity (Springer : Cham) **12**
[30] D Halder and M Basu 2016 Role of Pseudomonas stutzeri MTCC101 in Cadmium Bioremediation *Int.J.Curr.Microbiol.App.Sci.* **5**(2) 139-148
[31] C Vogt, S Kleinsteuber and H H Richnow Anaerobic benzene degradation by bacteria *Microb. Biotechnol.* **4** 710–724
[32] S A B Weelink, M H A van Eekert and A J M Stams 2010 Degradation of BTEX by anaerobic bacteria: physiology and application *Rev. Environ. Sci. Biotechnol.* **9** 359–385
[33] Atashgahi S, Hornung B, Waals M J, Rocha U N, Hugenholtz F, Nijsse B & Smidt H 2018 A benzene-degrading nitrate-reducing microbial consortium displays aerobic and anaerobic benzene degradation pathways Scientific reports **8**(1) 4490
[34] K M Posman, C M DeRito and E L Madsen 2017 Benzene Degradation by a Variovorax Species within a Coal Tar-Contaminated Groundwater Microbial Community Applied and Environmental Microbiology **83**(4)
[35] M Y S Gonzalez, R Chandra, C C Zacarias, F R Padilla, M de J R Alanis and R P Saldivar 2018 Biotransformation and degradation of 2,4,6-trinitrotoluene by microbial metabolism and their interaction Defense Technology **14** 151-164
[36] Y Jiang, K J Brassington, G Prpich, G I Paton, K T Semple, S J T Pollard and F Coulon 2016 Insights into the biodegradation of weathered hydrocarbons in contaminated soils by bioaugmentation and nutrient stimulation Chemosphere **161** 300–307
[37] C U Emenike, B Jayanthi, P Agamuthu and S H Fauziah 2018 Biotransformation and removal of heavy metals: a review of phytoremediation and microbial remediation assessment on contaminated soil Environ. Rev. 1–13
[38] L Yerushalmi, S Rocheleau, R Cimpoia, M Sarrazin, G Sunahara, A Peisajovich, G Leclair and S R Guiot Enhanced 2003 Biodegradation of Petroleum Hydrocarbons in Contaminated Soil *Bioremediation Journal* **7**(1) 37–51
[39] H Moya, J Verdejo, C Yáñez, J E Álvaro, S Sauvé and A Neaman Nitrification and nitrogen mineralization in agricultural soils contaminated by copper mining activities in Central Chile *J. Soil Sci. Plant Nutr.* **17**(1)
[40] B K Yadav and A Verma 2012 *Phosphate Solubilization and Mobilization in Soil Through Microorganisms Under Arid Ecosystems*, *The Functioning of Ecosystems* (Prof. Mahamane Ali (Ed.)) ISBN:978-953-51-0573-2 *InTech*
[41] K P Ingle and D A Padole 2017 Phosphate Solubilizing Microbes: An Overview *Int. J. Curr.*

Microbiol. App. Sci. **6**(1) 844-852

[42] A M Agnello 2002 *Petroleum-derived spray oils: chemistry, history, refining and formulation* (Beattie GAC, Watson DM, Stevens ML, Rae DJ, Spooner-Hart RN, eds. Spray oils beyond 2000 Sydney University of Western Sydney)

[43] V Kocí, K Mocová, M Kulovaná and S Vosáhlová 2010 Phytotoxicity tests of solid wastes and contaminated soils in the Czech Republic *Environ Sci Pollut Res Int.* **17**(3) 611-23

[44] Azevedo R and Rodriguez E 2012 Phytotoxicity of Mercury in Plants: A Review. *Hindawi Publishing Corporation Journal of Botany* **2012**(6) pages

[45] B Ikhajiagbe, G O Anoliefo, M A Jolaoso and E O Oshomoh 2013 Phytoassessment of a Waste Engine Oil-polluted Soil Exposed to Two Different Intervals of Monitored Natural Attenuation Using African Yam Bean (*Sphenostylis stenocarpa*) *Pakistan Journal of Biological Sciences* Volume **16**(14) 680-685

[46] B O Okonokhua, B Ikhajiagbe, G O Anoliefo and T O Emede 2007 The Effects of Spent Engine Oil on Soil Properties and Growth of Maize (Zea mays L.) *J. Appl. Sci. Environ. Manage* **11**(3) 147 – 152

[47] M C Hodgkinson, D Johnson and G Smith 2002 *Causes of phytotoxicity induced by petroleum-derived spray oil In*: Beattie GAC, Watson DM, Stevens M, Rae DJ, Spooner-Hart RN, eds. Spray oils beyond 2000 (Sydney: University of Western Sydney) 170–178

[48] D Johnson, MC Hodgkinson and D Joyce 2002 *Potential effects of petroleum-derived spray oils on abscission, senescence and stress physiology of citrus* In: Beattie GAC, Watson DM, Steven ML, Rae DJ, Spooner-Hart RN, eds. Spray oils beyond 2000 (Sydney: University of Western Sydney) 185–192

[49] G Han, B X Cui, X X Zhang and K R Li 2016 The effects of petroleum-contaminated soil on photosynthesis of Amorpha fruticosa seedlings *Int. J. Environ. Sci. Technol.* **13** 2383–2392

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