ABSTRACT

Contamination of soil by organic compounds, especially hydrocarbon, is prevalent in industrialized and oil producing countries of the world. About 1.7 to 8.8 million metric tons of oil is released into the water and soils every year. This may pose a great threat to the environment and human being at large. This study evaluated the efficiency of organic wastes (biowastes) as supplementations for remediation of diesel fuel contaminated soil. Three organic wastes [tea leaf (TL), Soybean cake (SC) and Potato skin (PS)] and two economically viable plants (*Dracaena reflexa* and *Podocarpus polystachyus*) were utilized to evaluate the biodegradation of diesel fuel in soil contaminated with different concentrations of oil. For biodegradation studies, soils were treated with 20%, 15%, 10% and 5% (w/w) diesel fuel and amended with 10% and 5% TL, SC and PS. Completely randomized design was used for a period of 126 days under laboratory condition. At the end of 126 days, soil polluted with 20% diesel oil and amended with 5% TL recorded the lowest percentage of oil degradation (14.5%) and diesel utilizing bacteria at 30×10^5 colony-forming units (CFU) per gram of soil. The highest rate of biodegradation (95%) was recorded in soil polluted with 5% diesel oil and amended with 10% SC with the count of diesel utilizing bacteria at 210×10^5 CFU/g. First order kinetic showed that soil amended with SC had the highest rate of oil degradation and illustrates the least half-life for all the diesel fuel concentrations. Bioremediation of diesel fuel contaminated soil with biomass amendments was monitored for a period of one year under natural condition. Result indicates complete biodegradation of C_8 to C_{16} and remarkable biodegradation of C_{16} to C_{22} hydrocarbon fractions in contaminated soil amended with SC. In phytoremediation study, contamination of soil with 2.5% and 1% diesel fuel and amended with 5% of the three different organic residues was monitored for a period of 270 days under laboratory and natural conditions. About 98.8%, 90.3% and 19% oil loss

was recorded in soil amended with SC, polluted with 1%, 2.5% and 5% diesel oil planted with *D. reflexa*, respectively. However, diesel contaminated soil with *Dracaena* but without organic wastes recorded 62%, 52.4% and 8.5% for 1%, 2.5% and 5% contamination, respectively under laboratory condition. Also 91%, 84% and 13.8% oil loss was recorded in soil amended with SC, polluted with 1%, 2.5% and 5% diesel oil with *P. polystachyus*, respectively. The remediation process was influenced by oil concentration and organic biomass added. However, *D. reflexa and P. polystachyus* root did not accumulate hydrocarbons from the soil, thus indicating that the mechanism of the oil degradation was via phytovolatilization or rhizodegradation. Phytoremediation of co-contamination of soil with heavy metals (80 ppm Zn and 60 ppm Pb) and 2.5% diesel fuel was amended with 5% organic waste was studied for a period of 180 days. Significant bioaccumulation of Pb and Zn in the root and stem of *Dracaena* plant was observed. At the end of 180 days, 16.53 mg/kg and 12.2 mg/kg of Zn accumulation in root and stem while 16.7 mg/kg and 9.8 mg/kg of Pb in root and stem of *D. reflexa* was recorded, respectively. However, 11.8 mg/kg and 9.8 mg/kg bioaccumulation of Zn and Pb was observed in root of *P. polystachyus.* Potential of five diesel utilizing bacterial (DUB) isolates (*Stenotrophomonas acidaminiphila, Bacillus lichenifomis, Brevibacillus parabrevis, Ochrobactrum tritici, Pesedomonas citronellolis*) from oil-contaminated soil to degrade diesel fuel was studied in broth culture for 35 days at 32^0C. At the end of the incubation period higher percentage degradation was recorded for *Bacillus lichenifomis* (45.8%).

In conclusion, the results of these studies illustrated the potential of SC and the two plants (*D. reflexa and P. polystachyus*) as a good option for enhanced remediation of hydrocarbon-contaminated soil.

ABSTRAK

Pencemaran tarah oleh sebatian/bahanorganik terutamanya hidrokarbon adalah sangat lazim dinegara perindustrian yang menghasilhan minyak. Kira-kira 1.7 hingga 8.8 juta tan metrik minyak dilepaskan ke dalam air dan tanah setiap tahun. Pada umumnya, ini membawa ancaman kepada alam sekitar dan manusia. Kajian ini menilai kecekapan sisa organik sebagai pelengkap pada pemulihan tanah tercemar oleh bahan api diesel. Tiga sisa organik [teh daun (TL), kek soya (SC) dan kulit kentang (PS)] dan dua tumbuhan berdaya maju (*Dracaena reflexa* dan *Podocarpus polystachyus*) telah digunakan untuk menilai tahap biodegradasi bahan api diesel dalam tanah yang tercemar menggunakan kepekatan minyak berbeza. Tahap biodegradasi tanah telah dirawat dengan 20%, 15%, 10% dan 5% (w / w) bahan api diesel dan dipinda dengan 10% dan 5% TL, SC dan PS. Ini telah dikaji secara rawak di bawah reka bentuk lengkap bagi tempoh 126 hari di bawah keadaan makmal. Di penghujung hari yang ke-126, kadar peratusan terendah degradasi minyak adalah (14.5%) dalam tanah yang tercemar dengan minyak diesel sebanyak 20%, Seterusnya, dipinda menggunakan TL dan diesel pada 5% dengan kiraan bakteria pada 30×10^5 pembentukan unit koloni (CFU) bagi setiap gram tanah. Kadar tertinggi biodegradasi (95%) dalam tanah tercemar dengan 5% minyak diesel direkod dan dipinda dengan 10% SC dalam kiraan bakteria sebanyak 210×10^5 CFU / g. Keputusan hasil kinetik tertib pertama menunjukkan bahawa tanah yang dipinda menggunakan SC mempunyai kadar degradasi minyak paling tinggi serta menggambarkan separuh hayat yang berkurang bagi semua kepekatan bahan api diesel. Bioremediasi tanah yang tercemar dengan minyak diesel dan pindaan sisa organik dipantau untuk tempoh satu tahun di bawah keadaan semula jadi. Keputusan menunjukkan bahawa biodegradasi penuh C_8 hingga C_{16} dan biodegradasi luar biasa C_{16} ke C_{22} merupakan pecahan hidrokarbon di dalam tanah tercemar yang dipinda dengan SC.

Dalam kajian fitopemulihan, pencemaran tanah pada 2.5% dan 1%, dipinda dengan 5% daripada tiga bahan buangan organik yang berbeza dan dipantau bagi tempoh 270 hari di

bawah keadaan makmal dan semulajadi. Kadar kehilangan minyak direkod pada 98.8%, 90.3% dan 19% dalam tanah yang dipinda dengan SC. Tanah tersebut tercemar dengan 1%, 2.5% dan 5% minyak diesel dengan *D. reflexa.* Walaubagaimanapun, dalam tanah tercemar yang mengandungi *Dracaena* tanpa sisa organik, tahap pencemaran masing-masing dicatat pada 62%, 52.4% dan 8.5% 1%, 2.5% dan 5% di bawah keadaan makmal. Kadar kehilangan minyak direkod pada 91%, 84%, dan 13.8% dalam tanah yang dipinda dengan SC, tercemar pada 1%, 2.5% dan 5% minyak diesel dan *P. polystachyus*. Proses pemulihan dipengaruhi oleh faktor kepekatan minyak dan sisa organik biomas. Malah, akar *D. reflexa* dan *P. polystachyus*, tidak mengumpul sisa hidrokarbon dari tanah, sekali gus menunjukkan bahawa mekanisme degradasi minyak adalah melalui fitoremediasi. Proses fitoremediasi tanah yang tercemar dengan logam berat (80 ppm Zn dan 60 ppm Pb) dan tanah yang tercemar dengan 2.5% bahan api diesel, dipinda dengan 5% sisa organik bagi kajian selama 180 hari. Biaokumulasi logam Pb dan Zn dalam akar dan batang tumbuhan *Dracaena* diperhatikan. Pada akhir tempoh 180 hari, masing-masing sebanyak 16.53 mg / kg dan 12.2 mg / kg pengumpulan Zn diperhatikan dalam akar dan batang manakala 16.7 mg / kg dan 9.8 mg / kg Pb diperhatikan dalam akar dan batang *D. reflexa*. Walau bagaimanapun, hanya 11.8 mg / kg dan 9.8 mg / kg logam Zn dan Pb diperhatikan dalam akar *P. polystachyus.* Potensi lima bakteria menggunakan diesel pencilan (DUB) (*Stenotrophomonas acidaminiphila, Bacillus lichenifomis, Brevibacillus parabrevis, Ochrobactrum tritici, Pesedomonas citronellolis*) dari tanah yang tercemar dengan minyak diperhatikan selama 35 hari pada 320 °C bagi mengkaji pengurangan minyak diesel. Pada hari terakhir tempoh pengeraman, kadar peratusan degradasi yang tinggi direkod bagi *Bacillus lichenifomis* pada 45.8%.

Kesimpulannya, keputusan kajian ini menggambarkan potensi SC serta dua tumbuhan iaitu (*Dracaena reflexa dan Podocarpus polystachyus*) sebagai pilihan terbaik bagi meningkatkan daya pemulihan tanah tercemar dengan hidrokarbon.

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LIST OF SYMBOLS AND ABBREVIATIONS

AHB	Aerobic Heterotrophic Bacteria
BCF	Bioconcentartion Factor
CF–IRMS	Continuous Flow Isotope Ratio Mass Spectrometer
CFUs	Colony Forming Units
DUB	Diesel Fuel Utilizing Bacteria
EIA	Energy Information Administration
EPA	Environmental Protection Agency
GC-MSD	Gas Chromatography/ Mass Spectrometry Detector
HPCD	Hydroxypropyl[B]Cyclodextrin
ICP- OES	Inductively Coupled Plasma-Optical Emission Spectroscopy
INTF	Iodonitrotetrazolium Formazan
MSM	Mineral Salt Medium
NRC	National Research Council
NPL	National Priorities List
PS	Potato Skin
PAH	Polyaromatic Hydrocarbon
TL	Tea Leaf
TPH	Total Petroleum Hydrocarbon
TF	Translocation Factor
SC	Soybean Cake
USITC	United States International Trade Commission
VOCs	Volatile Organic Compounds