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Biodegradation kinetics of petroleum hydrocarbon in soil environment using *Mangnifera indica* seed biomass: A mathematical approach

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ABSTRACT

The biodegradation crude oil in polluted soil using *Mangnifera indica* seed biomass was investigated. The change in total petroleum hydrocarbon (TPH) concentration on the effect of the period of exposure with different dosage of *M. indica* seed mesh introduced in the different containers were examined and a decrease in TPH concentration was significant. The maximum specific rate and equilibrium constants were determined and the results obtained revealed that the values for room dried *M. indica* of various dosages at V_{max} 50–90 g, 500, 33,333.33, 50,000, 50,000, 27.78 (ppm/per day) and K_S 50–90 g, 0.5, 300, 2750, 1124.81 (ppm/per day) were observed. For sun-dried, *M. indica* of various dosages at V_{max} 50– 90 g, ∞, 1.6666.67, ∞, ∞, 100.00 (ppm/per day) and K_S 50–90 g, ∞, 16.67, ∞, ∞, 300 (ppm/per day) were recorded. The maximum specific rate and equilibrium constant was determined using the Lineweaver Burk plots Theory as well as excel plot of linear polynomial curve for comparison. Results revealed that the *M. indica* seed biomass ha promising effeminacy for the degradation of THP in the contaminated soil.

Capsule Summary: Lineweaver Burk theory was employed to evaluate the biodegradation kinetics of crude oil in a soil environment using *M. indica* seed biomass as a function of exposure duration and dosage of biomass and results revealed TPH concentration in soil was declined significantly.

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INTRODUCTION

Crude oil pollution in the environment has been a major concern to especially people living in the crude oil rich areas. Oil pollution due to spill could take place in water or on land. Crude oil polluted on land depends on the number of factors which include; the permeability of the soil, adsorption properties of the soil and the partition coefficient (Chaineau et al., 2001). Similarly, studies carried out by Iturbe et al. (2005) using sandy or loamy soil, and species of crude oil also confirmed the pronounced changes on the physiochemical properties and the increase on contaminated soil. And Brow et al. (1996) observed significant buildup of lead, iron and zinc in the contaminated crude oil in Niger Delta soil. In a related study, Mekl et al. (2005) showed that zinc distribution is more in an oil polluted sites than non-oil polluted areas and also observed that its distribution depends on certain soil properties as clay, organic matter and pH. Okolo et al. (2005) revealed zinc positive relationship with pH and effective cat ion exchange capacity whereas clay and organic matter did not affect zinc distribution in the soil polluted by crude oil. Other parameters including electrical conductivity, available phosphorus and total nitrogen in crude oil impacted soils were comparatively low, while the total organic carbon was high, compared with the reference site (Okpokwasili and Odokuma, 1990).

The assessment of the physiochemical values of leached polluted soil, Ifeadi and Nwankwo (1987), observed significantly the reduction in the porosity and organic matter content. Crude oil studied by (Andrade et al., 2004) showed a decrease in strength, stiffness and permeability of the clay attributed to formation of an open structure occasioned by the crude oil contamination. Certain mechanisms are involved in the fixation of crude oil soil. Such mechanisms were studied and represented by Iturbe et al., (2005) as adsorption, ion exchange capacity and chemical precipitation. Due to low absorption of certain soil types and the inability of the soil to accumulate on such soils, some component of the crude oil such as acryl-amide may migrate through the soil structures to pollute the ground water (Iturbe and Flores, 2004).

Crude oil also upsets the microbial biomass of the contaminated soil. Investigation of Chaineau et al. (2004) showed a decline of incepts and a slight increase. This study will examine the effects of crude oil on the selected physical properties of different soils of Niger Delta Nigeria which include: the percentage organic matter, porosity and bulk density, using different crude oil samples of different physiochemical properties. The applied volume variation effects of the crude oil samples on those soil properties will be studied. The experiment was carried out by Odukuma and Ibor, (2002), physiochemical properties of soil in relation to varying rates of crude oil pollution.

Crude oil extraction and transportation are often accompanied by environmental contamination which inevitably leads to pollution particularly in soil medium. Starting from the prospective stage down to the distribution of the end products degradation occurs in the environment in one way or the other (Odukuma and Ibor, 2002; Brow et al., 1996; Mekl et al., 2005). Petroleum hydrocarbons are toxic soil biotic and leads to the decrease in soil quality, making the latter useless for agriculture, therefore there is an obvious need in the technologies of hydrocarbon contamination removal from the soil. Presently, methods of biological remediation of oil contaminated soil are becoming more and more significant due to their ecological safety for the environment. Enzyme activity of hydrocarbon oxidizing microorganism is used in the biological method for petroleum hydrocarbon degradation.

MATERIAL AND METHODS

Sample collection

Soil sample (Loamy soil) was collected from Igbo Etche in Etche Local Government Area of Rivers State and then transported to the chemical/Petrochemical Department Laboratory for purpose of sampling (Analysis). The loamy soil sample was collected with the aid of shovel, hand trowel and plastic container. The plastic container was installed in a cooler to ensure that the physical parameters of the loamy soil sample do not change as a result of the environment effect. Part of the loamy soil sample was transport to the soil science and microbiology Department for analysis of important parameters and the remaining soil sample (loamy soil) was further subject into the experimental set-up as described in this research work.

Biodegradation procedure and analysis

Mangnifera indica was collected from Rivers State and transported to ADP office in Port Harcourt for the purpose of identification of the specie. After the identification the mango seed was subjected into two drying conditions, was thoroughly washed alongside was the room temperature specimen and further sun dried as well at room temperature. The drying period covered check 10 days. At this period of ten day a reasonable amount of moisture content was removed from the mango seed. After the experience the dried mango seed was introduced into a blender to be reduced to smaller size particles. This process was carried out for both sun dried and room temperature of the *Mangnifera Indica* (mango seed). Different reactors (containers) were set-up with equal quantity of loamy soil (10g) was introduced on each reactor and then the reactors were weighed to ascertain equal weight. The total react set-up for the investigation of crude oil degradation is twenty and each reactor contains 10 g of loamy soil with addition of 100ml of crude oil to all the reactors with variation in the concentration of the powder form of *Mangnifera indica* (mango seed) ranging from 50, 60, 70, 80 and 90 g. The experimental set-up was monitored for 2 weeks intervals before sampling, the concentration of each reactor for the purpose of crude oil concentration determination and other physiochemical parameters behavior in the system. This process was allowed to run for a period 84 days.

RESULTS AND DISCUSSION

The results obtained from this investigation are presented in Figures 1-20. From the above Figure 1 it is seen that the linear curve equation is expressed as -0.001x + 0.002 with the square root of the best feet R^2 = 0.014 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.002 with the slop of 0.006, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, $y = -0.001x + 0.002$
Slope = 0.001
Intercept $C = 1/V_{max} = 0.002$.
Therefore, V_{max} = 500 ppm/day., Slope = K_s/V_{max} = 0.001. Therefore $K_s = V_{max} \times$ Slope = 500 ppm/day × 0.001 = 0.5 ppm/day. Hence, the V_{max} = 500 and Ks = 0.5 which shows that the graph obeys the Lineweaver Burk plot.

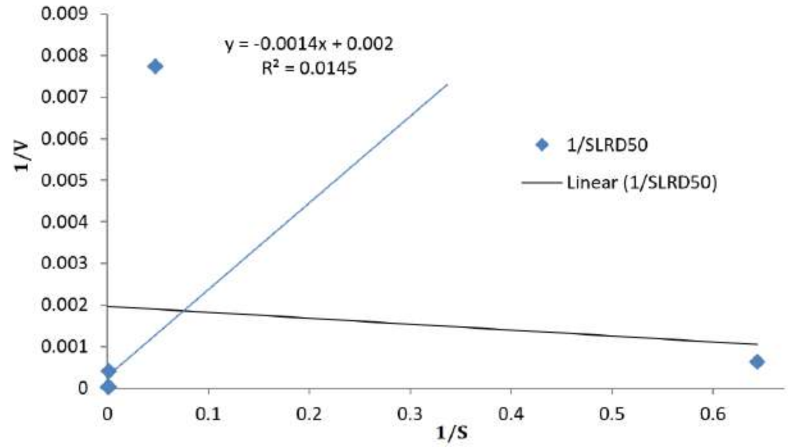

Fig. 1: Lineweaver Burk plot for $1/V_{max}$ verses 1/S for the degradation of TPH using 50 g of *M. indica* seed biomass

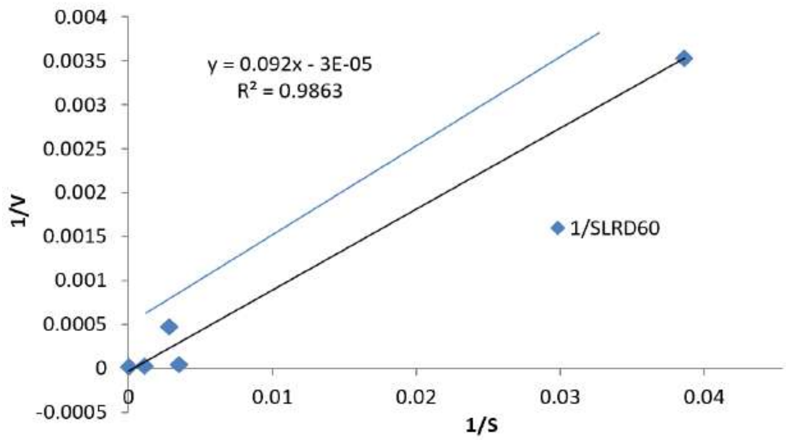

Fig. 2: Lineweaver Burk plot for $1/V_{max}$ verses 1/S for the degradation of TPH using 60 g of *M. indica* seed biomass

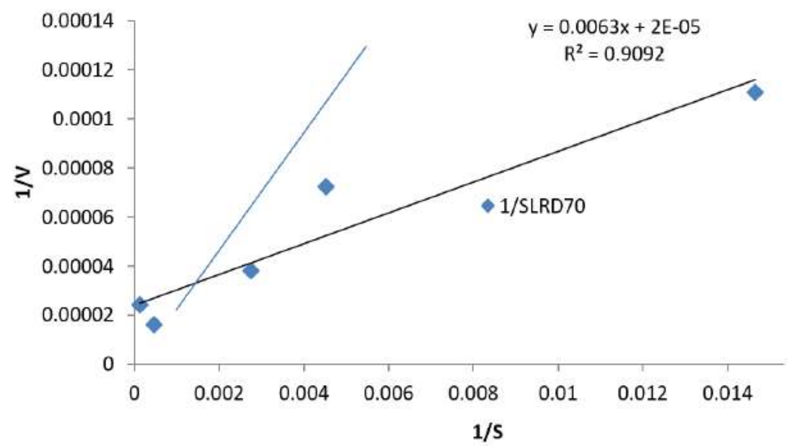

Fig. 3: Lineweaver Burk plot for $1/V_{max}$ verses 1/S for the degradation of TPH using 70 g of *M. indica* seed biomass

From the above Figure 2 it is seen that the linear curve equation is expressed as 0.092X-3E-5 with the square root of the best feet R^2 = 0.986 indicating the acceptability of the curve. Therefore, from the equation the intercept is -3E-5 with the slop of 0.092, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and K_s/V_{max} = slope).

Where, y = -0.092 - 3E – 05

Slope = 0.0092

Intercept C = $1/V_{max}$ = 0.0092

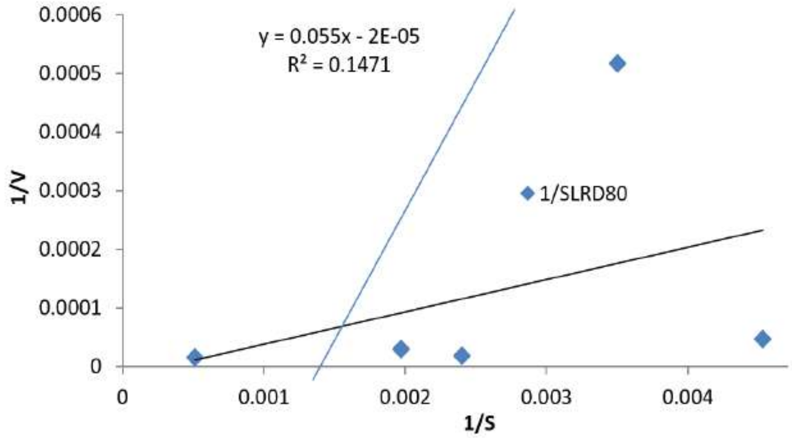

Fig. 4: Lineweaver Burk plot for $1/V_{max}$ verses 1/S for the degradation of TPH using 80 g of *M. indica* seed biomass

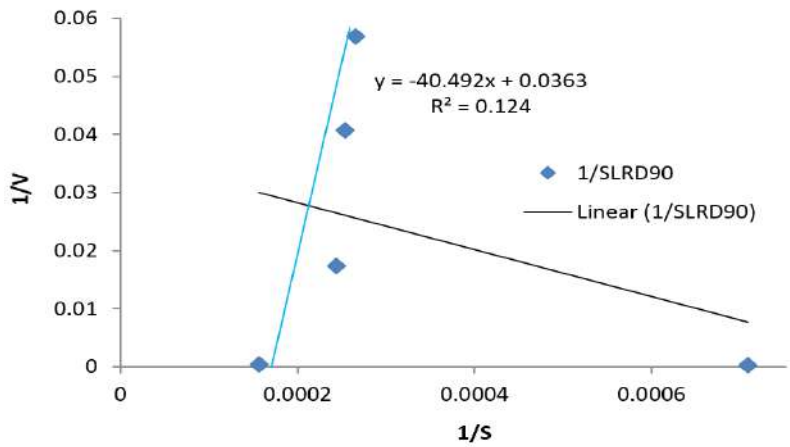

Fig. 5: Lineweaver Burk plot for $1/V_{max}$ verses 1/S for the degradation of TPH using 90 g of *M. indica* seed biomass

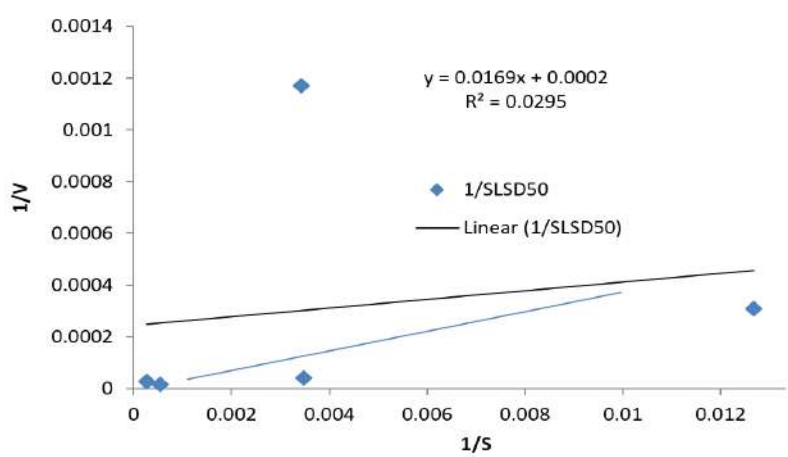

Fig. 6: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 50 g sun-dried *M. indica* seed biomass

Therefore, V_{max} = 33,333.3 ppm/day., Slope = K_s/V_{max} = 0.0092. Hence, the $K_s = V_{max} \times$ Slope= 33,333.3 ppm/day × 0.0092 = 3066.67 ppm/day. The V_{max} = 33333.33 and Ks = 3066.67 shows that the graph obeys the Lineweaver Burk plot.

From the Figure 3 it is seen that the linear curve equation is expressed as 0.006x+2E-5 with the square root of the best feet $R^2 = 0.909$ indicating the acceptability of the curve. Therefore, from the equation the intercept is = 2E-5 with the slop of 0.006, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, $y = -0.006x + 2E - 05$

Slope = 0.006

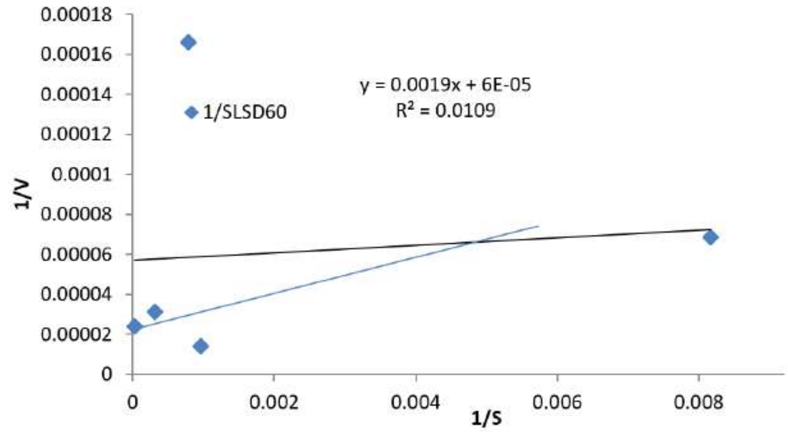

Fig. 7: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 60 g sun-dried M. indica seed biomass

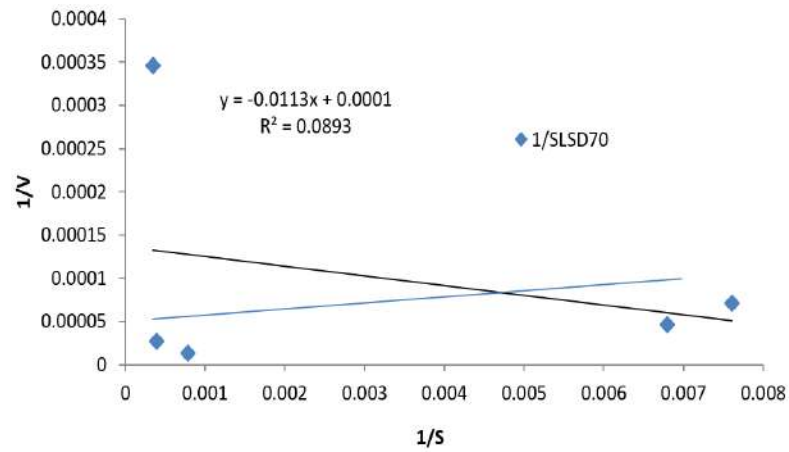

Fig. 8: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 70 g sun-dried M. indica seed biomass

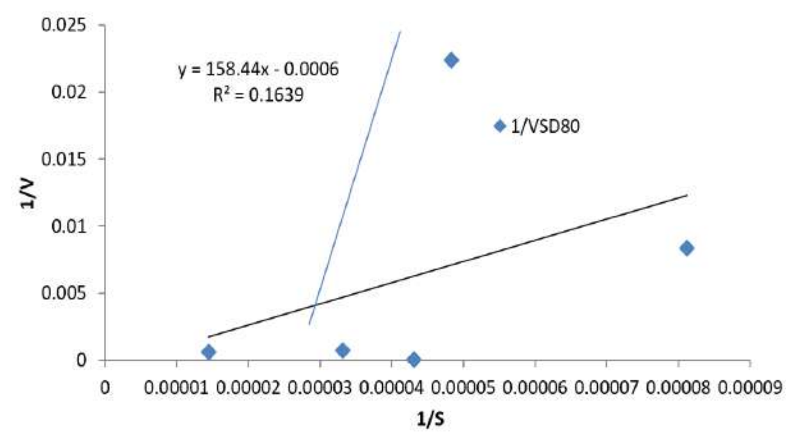

Fig. 9: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 80 g sun-dried M. indica seed biomass

Intercept C = $1/V_{max}$ = 0.006
Therefore, V_{max} = 50,000 ppm/day., Slope = K_s/V_{max} = 0.006.
$K_s = V_{max} \times$ Slope= 50,000 ppm/day × 0.006 = 300 ppm/day.
Hence, the V_{max} = 50,000 and Ks =300 shows that the graph obeys the Lineweaver Burk plot

From the Figure 4 it is seen that the linear curve equation is expressed as 0.055X - 2E - 5 with the square root of the best feet R2 = 0.147 indicating the acceptability of the curve. Therefore, from the equation the intercept is -2E-5 with the slop of 0.055, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = -0.055x – 2E – 05
Slope = 0.055

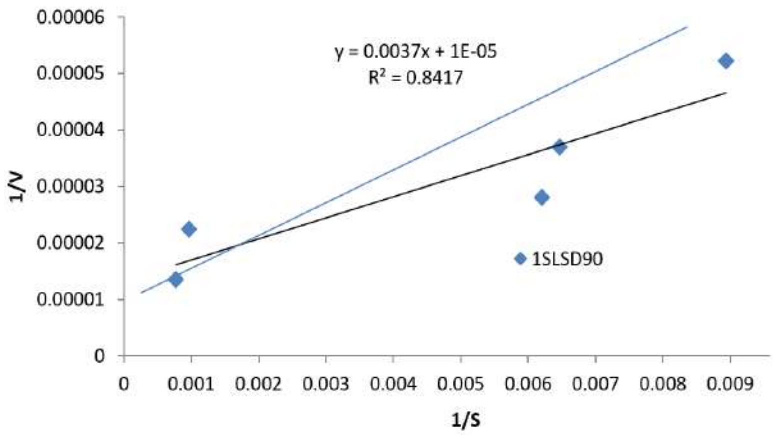

Fig. 10: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 90 g sun-dried M. indica seed biomass

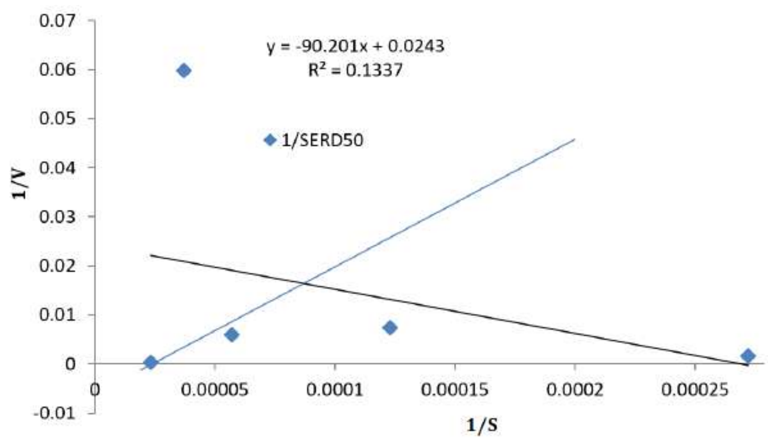

Fig. 11: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 50 g room-dried M. indica seed biomass

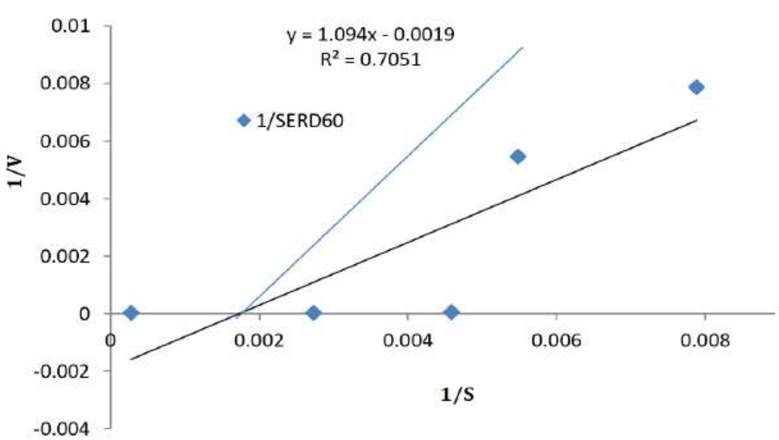

Fig. 12: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 60 g room-dried M. indica seed biomass

Intercept C = $1/V_{max}$ = 0.055
Therefore, V_{max} = -50,000 ppm/day., Slope = K_s/V_{max} = 0.055. K_s = V_{max} × Slope= -50,000 ppm/day × 0.055 = 2750 ppm/day. Hence, the V_{max} = 50,000 and Ks = 2750 shows that the graph obeys the Lineweaver Burk plot

From the above Figure 5 it is seen that the linear curve equation is expressed as -40.49x + 0.036 with the square root of the best feet R^2 = 0.124 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.036 with the slop of -40.49, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = -40.49*x* + 0.036
Slope = 40.49

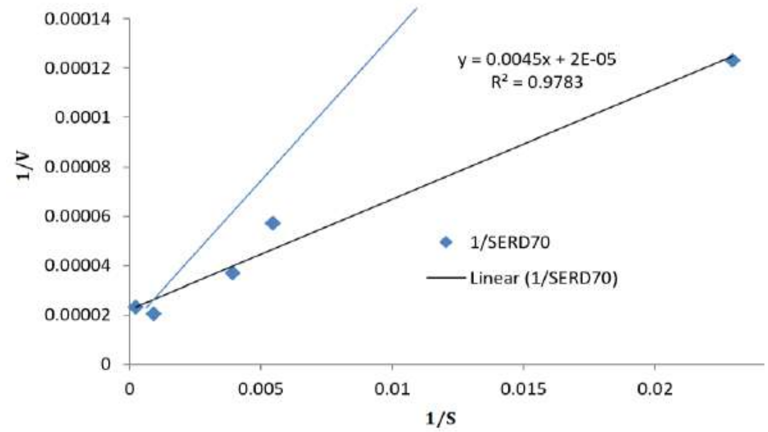

Fig. 13: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 70 g room-dried *M. indica* seed biomass

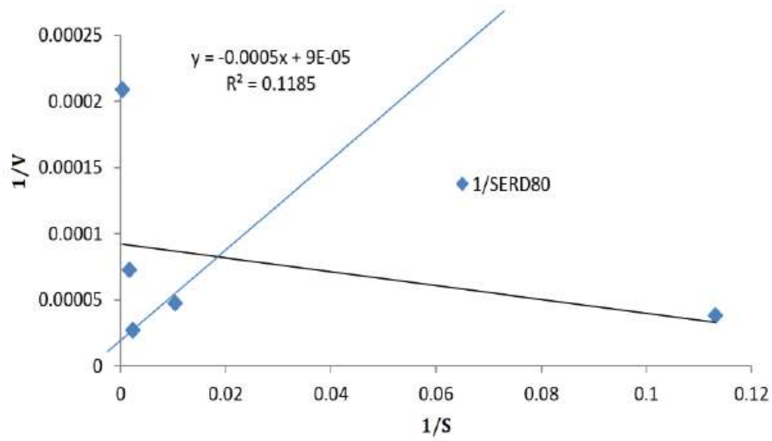

Fig. 14: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 80 g room-dried *M. indica* seed biomass

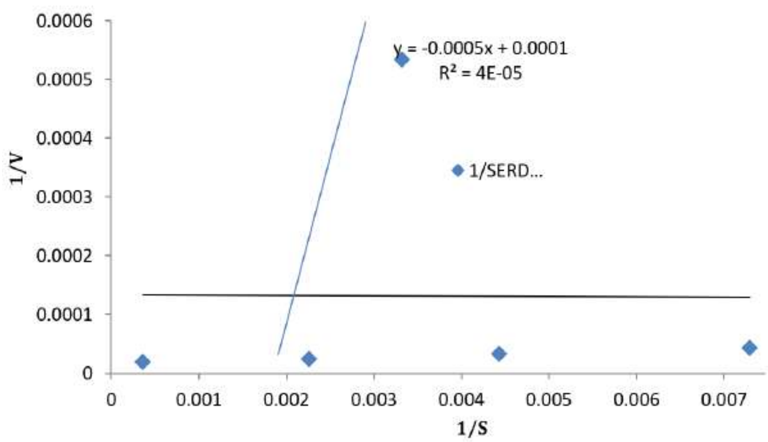

Fig. 15: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 90 g room-dried *M. indica* seed biomass

Intercept C = $1/V_{max}$ = 40.49
Therefore, V_{max} = 27.78 ppm/day
Slope = K_s/V_{max} = 40.49.
Therefore K_s = V_{max} × Slope= 27.78 ppm/day × 40.49 = -1124 ppm/day. Hence, the V_{max} = 27.78 and Ks = 1124.81 shows that the graph obeys the Lineweaver Burk plot.

From the Figure 6 it is seen that the linear curve equation is expressed as 0.016x + 0.000 with the square root of the best feet R^2 = 0.029 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.000 with the slop of 0.016, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = 0.0016*x* + 0.000., Slope = 0.0016
Intercept C = $1/V_{max}$ = 0.00016
Therefore, V_{max} = ∞ ppm/day

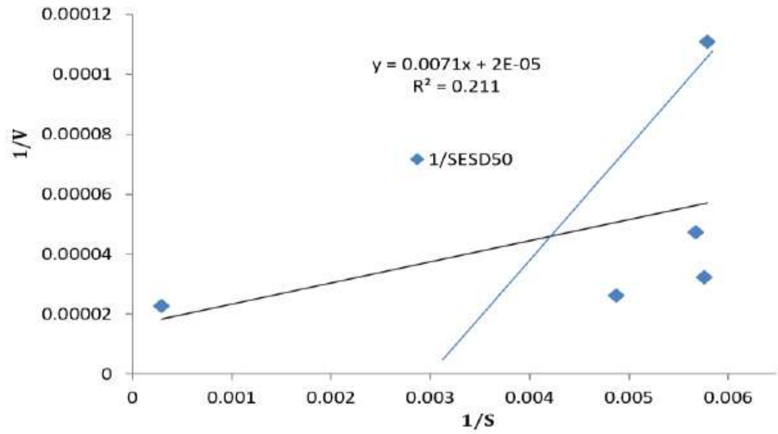

Fig. 16: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 50 g sun-dried *M. indica* seed biomass

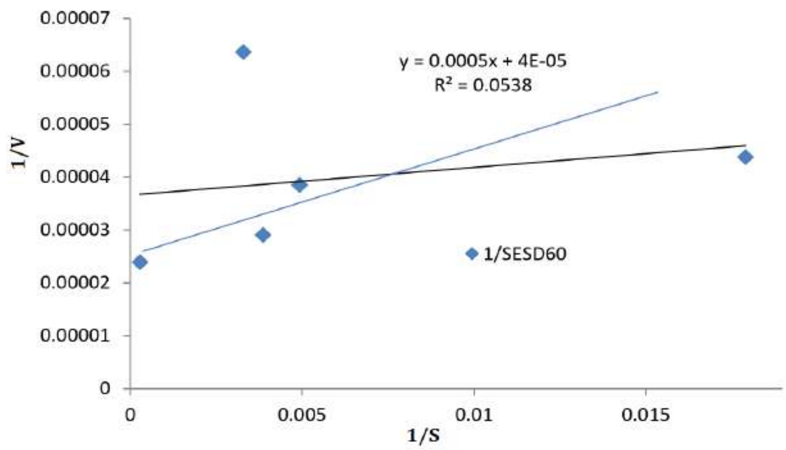

Fig. 17: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 60 g sun-dried *M. indica* seed biomass

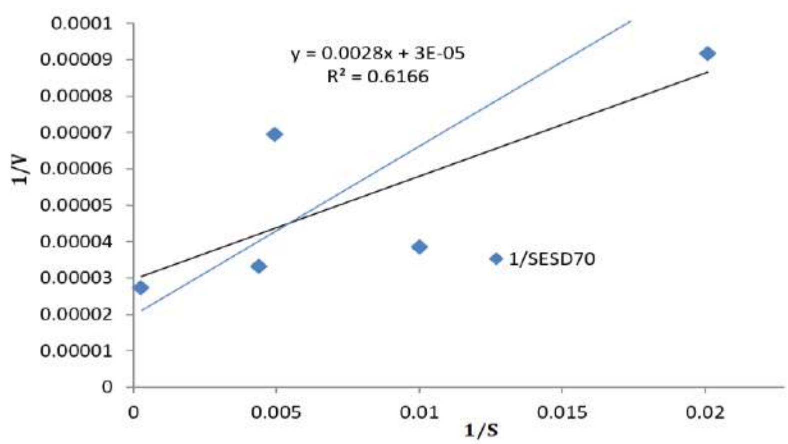

Fig. 18: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 70 g sun-dried *M. indica* seed biomass

Slope = K_s/V_{max} = 0.00016
K_s = V_{max} × Slope = ∞ ppm/day × 0.016 = ∞ ppm/day. Hence, the V_{max} and K_s did not obey the Lineweaver Burk plot.

From the above Figure 7 it is seen that the linear curve equation is expressed as 0.001 x + 6E-5 with the square root of the best feet R^2 = 0.010 indicating the acceptability of the curve. Therefore, from the equation the intercept is 6E-5 with the slop of 0.001, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = 0.001x + 6E – 05

Slope = 0.001
Intercept C = $1/V_{max}$ = 0.001
Therefore, V_{max} = 16666.67 ppm/day
Slope = K_s/V_{max} = 0.001
K_s = V_{max} × Slope= 16666.67 ppm/day × 0.001 = 16.67 ppm/day. Hence, the Vmax =16666.67 and Ks = 16.67 which shows that the graph obeys the Lineweaver Burk plot.

From the above Figure 8 it is seen that the linear curve equation is expressed as 0.011x + 0.000 with the square root of the best feet R^2 = 0.089 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.000 with the slop of 0.011, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = -0.011x + 0.000
Slope = - 0.011
Intercept C = $1/V_{max}$ = -0.011
Therefore, V_{max} = ∞ ppm/day
Slope = K_s/V_{max} = 0.011
K_s = V_{max} × Slope = 0.011 ppm/day × ∞ = ∞ ppm/day. Hence, the Vmax = 0.011 shows that it obeys the Lineweaver Burk plot, while Ks = ∞ is in a continuous process.

From the above Figure 9 it is seen that the linear curve equation is expressed as 158.4x-0.000 with the square root of the best feet R^2 = 0.163 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.000 with the slop of 158.4, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop) the lineweaver Burk plot.
Where, y = 158.4x - 0.000
Slope = 158.4
Intercept C = $1/V_{max}$ = 158.4
Therefore, V_{max} = ∞ ppm/day
Slope = K_s/V_{max} = 158.4
K_s = V_{max} × Slope= ∞ ppm/day × 158.4 = ∞ ppm/day. V_{max} = ∞ K_s = ∞ both of them are in a continuous process.

From the above Figure 10 it is seen that the linear curve equation is expressed as 0.003x + 1E-5 with the square root of the best feet R^2 = 0.841 indicating the acceptability of the curve. Therefore, from the equation the intercept is 1E-5 with the slop of 0.003, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = 0.003x + 1E – 05
Slope = 0.003
Intercept C = $1/V_{max}$ = 0.003
Therefore, V_{max} = 100.000 ppm/day
Slope = K_s/V_{max} = 0.003. Therefore K_s = V_{max} × Slope = 100.00 ppm/day × 0.003 = 300 ppm/day. Hence, the V_{max} = 100,000 and Ks = 300 67 shows that the graph obeys the Lineweaver Burk Plot.

From the above Figure 11 it is seen that the linear curve equation is expressed as 90.20x + 0.024 with the square root of the best feet R^2 = 0.133 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.024 with the slop of 90.20, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (Ks/V_{max} = slop).
Where, y = 90.20x + 0.025
Slope = 90.20
Intercept C = $1/V_{max}$ = 90.20
Therefore, V_{max} = 41.67 ppm/day
Slope = K_s/V_{max} = -90.20
K_s = V_{max} × Slope= 41.67 ppm/day × 90.20 = 3758.33 ppm/day. Hence, the V_{max} = 41.67 and Ks = 3758.33 67 shows that the graph obeys the Lineweaver Burk plot.

From the Figure 12 it is seen that the linear curve equation is expressed as 1.094x - 0.001 with the square root of the best feet R^2 = 0.705 indicating the acceptability of the curve. Therefore, from the equation the intercept is - 0.001 with the slop of 1.094, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (Ks/Vmax= slop).
Where, y = 1.094x - 0.001
Slope = 1.094
Intercept C = $1/V_{max}$ = 1.094
Therefore, V_{max} -1000 = ppm/day
Slope = K_s/V_{max} = 1.094
K_s = V_{max} × Slope= 1000 ppm/day × 1.094 = 1094 ppm/day. Hence, the V_{max} = 1000 and Ks = 1094 67 indicate that the graph obeys the Lineweaver Burk plot.

From Figure 13, it is seen that the linear curve equation is expressed as 0.004x + 2E - 5 with the square root of the best feet R^2 = 0.978 indicating the acceptability of the curve. Therefore, from the equation the intercept is 2E-05 with the slop of 0.004, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).
Where, y = 0.004x + 2E - 05
Slope = 0.004
Intercept C = $1/V_{max}$ = 0.004
Therefore, V_{max} = 50,000. ppm/day
Slope = K_s/V_{max} = 0.004
K_s = V_{max} × Slope= 50,000ppm/day × 0.004 = 200 ppm/day. Hence, the Vmax =-500,000 and Ks = 200. The V_{max} and Ks obeyed the Lineweaver Bulk plot.

From Figure14 it is seen that the linear curve equation is expressed as 0.000x + 9E - 05with the square root of the best feet R^2 = 0.118 indicating the acceptability of the curve. Therefore, from the equation the intercept is 9E-5 with the slop of 0.000, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (Ks/V_{max}= slop). Hence, the V_{max} =11111.11 and Ks indicate that the graph obeys the lineweaver Burk Plot.
Where, y = -0.000x + 9E - 05
Slope = 0.000
Intercept C = $1/V_{max}$ = 0.000
Therefore, V_{max} = 11111.11 ppm/day
Slope = K_s/V_{max} = 0.000

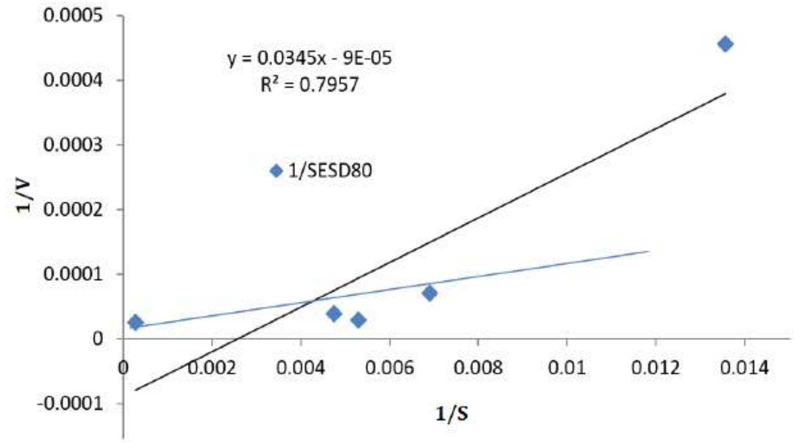

Fig. 19: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 80 g sun-dried *M. indica* seed biomass

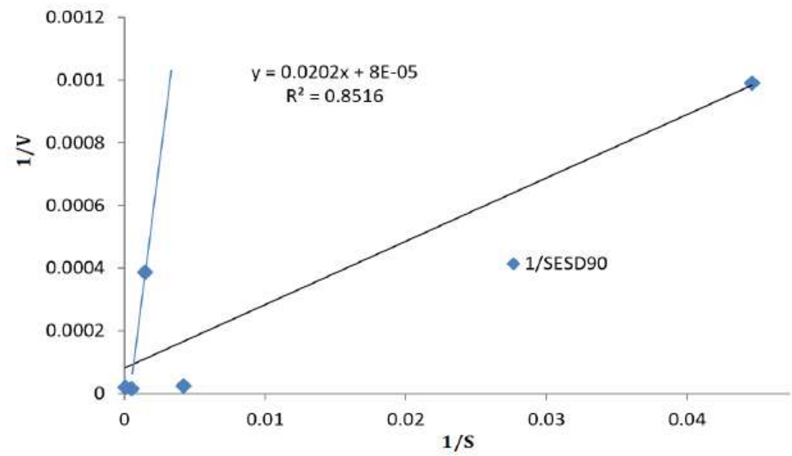

Fig. 20: Lineweaver Burk plot ($1/V_{max}$ Vs 1/S) for the degradation of TPH using 90 g sun-dried *M. indica* seed biomass

$K_s = V_{max} \times$ Slope= 11111.11ppm/day × 0.000 = 0 ppm/day. V_{max} = 11111.11 obeys the Lineweaver Burk plot while Ks = 0 which did not obey the Lineweaver Burk plot.

From the above Figure 15 it is seen that the linear curve equation is expressed as 0.000x + 0.000 with the square root of the best feet R^2 = 4E - 05 indicating the acceptability of the curve. Therefore, from the equation the intercept is 0.000 with the slop of -0.000, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, y = -0.000*x* + 0.000

Slope = 0.000

Intercept C = $1/V_{max}$ = 0.000

Therefore, V_{max} = ∞ ppm/day

Slope = K_s/V_{max} = 0.000

$K_s = V_{max} \times$ Slope= ∞ ppm/day × 0.000 = ∞ ppm/day. Hence, the V_{max} and K_s are both on a continuous process

From the above Figure 16 it is seen that the linear curve equation is expressed as 0.007x + 2E-05 with the square root of the best feet R^2 = 0.211 indicating the acceptability of the curve. Therefore, from the equation the intercept is 2E-05 with the slop of 0.007, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, y = 0.007*x* + 2E - 05

Slope = 0.007

Intercept C = $1/V_{max}$ = 0.007

Therefore, V_{max} = 50,000 ppm/day

Slope = K_s/V_{max} = 0.007

$K_s = V_{max} \times$ Slope= 50,000 ppm/day × 0.007 = 350 ppm/day

Hence, the Vmax = 50000 and Ks = 350 shows that the graph obeys the Lineweaver Burk plot.

From the Figure 17 it is seen that the linear curve equation is expressed as 0.000x + 4E-05 with the square root of the best feet R^2 = 0.053 indicating the acceptability of the curve. Therefore, from the equation the intercept is 4E-05 with the slop of 0.000, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, y = -0.000*x* + 4E - 05

Slope = 0.000

Intercept C = $1/V_{max}$ = 0.000

Therefore, V_{max} = 25,000 ppm/day

Slope = K_s/V_{max} = 0.000

$K_s = V_{max} \times$ Slope = 25,000 ppm/day × 0.000 = 0 ppm/day

V_{max} = 25000 obeys while Ks not possible because it did not obey Lineweaver Burk plot.

From the Figure18 it is seen that the linear curve equation is expressed as 0.003x+3E-05 with the square root of the best feet R^2 = 0.616 indicating the acceptability of the curve. Therefore, from the equation the intercept is 3E-05 with the slop of 0.003, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, y = 0.002*x* + 3E – 05

Slope = 0.002

Intercept C = $1/V_{max}$ = 0.002

Therefore, V_{max} = 33,333.33ppm/day

Slope = K_s/V_{max} = 0.002

K_s = V_{max} × Slope = 33,333.33 ppm/day × 0.002 = 66.67ppm/day. The Vmax = 33,333.33 and Ks = 66.67 indicates that the graph obeys the Lineweaver Burk Plot.

From the Figure 19 it is seen that the linear curve equation is expressed as 0.034x+9E-05 with the square root of the best feet R^2 = 0.795 indicating the acceptability of the curve. Therefore, from the equation the intercept is 9E-05 with the slop of 0.034, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, y = 0.034*x* - 9E – 05

Slope = 0.034

Intercept C = $1/V_{max}$ = 0.034

Therefore, V_{max} = 11111.11 ppm/day

Slope = K_s/V_{max} = 0.034

K_s = V_{max} × Slope= 11111.11 ppm/day × 0.034 = 377.78 ppm/day. Hence, the Vmax = 11,111.11 and Ks = 377.78, hence the graph obeys the Lineweaver Bulk Plot.

From the Figure 20 it is seen that the linear curve equation is expressed as 0.007x + 2E - 05 with the square root of the best feet R^2 = 0.211 indicating the acceptability of the curve. Therefore, from the equation the intercept is 2E-05 with the slop of 0.007, which is found useful in the determination of the maximum specific rate degradation ($1/V_{max}$ = intercept) and (K_s/V_{max} = slop).

Where, y = 0.020*x* + 8E – 05

Slope = 0.020

Intercept C = $1/V_{max}$ = 0.020

Therefore, V_{max} = 12,500 ppm/day

Slope = K_s/V_{max} = 0.020

K_s = V_{max} × Slope = 12,500 ppm/day × 0.020 = 250 ppm/day

Hence, the V_{max} = 12,500 and Ks = 250 indicates that the graph obeys the Lineweaver Bulk plot.

The degradation of the total petroleum hydrocarbon based on the factorial design mixture of the experimental set-up. Decrease in TPH was observed with increase in the period of exposure. The specific rate (velocity) of the reaction was determined including the reciprocal of the substrate (TPH) and the reciprocal of the specific rate (velocity) of the reaction. The data obtained from the process was used in the determination of the maximum specific rate of the reaction as well as the dissociation constant using Michael's Menten model equation. The detail explanation for each process is well resolved in each Figure as presented from Figure 1 to 20. It observed that most of the Figures do not obey the Lineweaver Burk Plot theory. Therefore, there is need to apply the polynomial theory to enable us obtained the equation of the line and the root of the best fit for the determination of the slope and the intercept. The intercept value is equated to $1/V_{max}$ whereas the slope value is equated to Ks/V_{max}. The findings revealed that the *M. Indica* has excellent efficiency for the degradation of hydrocarbon with very fast degradation rate, which can be used for the treatment soil contains petroleum hydrocarbons.

CONCLUSIONS

The following conclusion was drawn from the investigation. *M. Indica* biomass has promising crude oil bioremediation efficiency. Blended room dried *M. Indica* showed better crude oil remediation efficiency compared to the sun-dried. Crude oil remediation occurs faster in sandy soils than in loamy soils. Every 1 kg of soil polluted with 100 ml of crude oil requires 100 g of blended room dried *M. Indica* biomass to be completely remediated the TPH in 42 days. Oxygenation by addition of water enhances the rate of bioremediation significantly. Hence, this biomass could possibly be used for the treatment of soil contaminated with hydrocarbons.

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