

Efficacy of *Eucalyptus camaldulensis* (Dehnh) in the Phytoremediation of Petroleum Hydrocarbon Polluted Soils

*Ayuba, D. K. (PhD)¹, Aliba N. V.², Oluwole, M. O.¹ and Nzamouhe, M.³

¹(Department of Forestry and Wildlife Management, FALS, Federal University Wukari – Nigeria.)

²(Department of Hospitality and Tourism Management, FALS,, Federal University Wukari – Nigeria.)

³(Department of Soil Sciences and Land Resources Mgt., FALS, Federal University Wukari – Nigeria.)

Abstract:

Background: In an attempt to assess the efficacy of *E. camaldulensis* in the phytoremediation procedure during decontamination of crude oil polluted soils, the following were determined: total dry biomass (DBM_T), phytoaccumulation of total petroleum hydrocarbon and its asphaltene fraction and the phytoextraction efficiency using bioaccumulation factor (BAF) and mass influx (M_{influx}) ratios.

Materials and Methods: The experiment was the split-split plot design where the crude oil contaminations (4 levels) were the main plot, the sub-plot factor were the soil amendments (4 levels) and the sub-sub plot factor were the tissues of the plant (3 levels).

Results: Results indicated that the plant can produce total dry biomass (DBM_T) of up to 6.17 g in heavily polluted soils (0.7 L/4.0 kg soil), accumulate both TPH and asphaltene in same contaminant's concentrations (0.16 g and 0.13 g respectively) with the roots accumulating more than other tissues, and it was able to uptake, translocate and accumulated the soluble TPH at a rate that was higher than it did for the soluble asphaltene.

Conclusion: It was concluded that the plant was efficient in the phytoremediation procedure as it tolerates contaminant's toxicity and extract and accumulate same in its tissues.

Key Word: Phytoaccumulation, Phytoextraction, Phytoremediation, Bioaccumulation factor, Mass influx

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I. Introduction

Plants are well known for their contributions to the wellbeing of man and the environment in various ways. They provide ecological support services such as nutrient cycling, reduction of carbon emission due to anthropogenic activities, erosion prevention, and remediation of contaminants. In addition, they contribute directly by producing timber and non-timber products; the non-timber products such as phytochemicals, among others, usually outweighs the former. Their ability to grow in contaminated environments because they adopt some avoidance mechanisms [1,2] in checkmating excessive contaminants by converting it to a less harmful form gives some plants the advantage of being effective natural remediation agent.

In addition, it is well-known fact that scientists have established that plants and its rhizosphere microbes could decontaminate polluted soils using different phytotechnologies such as *rhizofiltration*, *hydraulic barrier*[3], *phytostabilization*[4,5,6] and *phytoextraction*[7,8]. Others include *phytostimulation*[5, 9], *phytodegradation*[9, 10] and *phytovolatilization*[11, 12]. Additionally, *phytomining* that is described as the recovery of essential heavy metals from plants after phytoremediation of heavy metals contaminated sites as suggested by [13], is further extended to include recovery of organic contaminants such as crude oil accumulated in plants by [2]. However, it should be known that these various phytotechnologies are not mutually exclusive [5, 14].

In more recent times, scientists are becoming aware that despite the much stated roles of plants in ecosystems management, not all plants can be used effectively in the management of environmental pollution problems. However, several scientists have attested that phytoaccumulator plant species have the ability to remediate contaminated environments to an environmentally acceptable limit that enables other multiple land uses - depending on prevailing environmental factors. These phytoaccumulator plant species have been found to remediate organic and inorganic pollutants from both contaminated soil and groundwater aquifer. Some were found effective in the remediation of radioactive substances such as trinitoulene (TNT), heavy metals, polycyclic aromatic hydrocarbons, etc.

The choice of these phytoaccumulator plant species is very critical in the success of any phytoremediation method because petroleum hydrocarbon contaminations through leakages from storage tanks and pipelines, land disposal of petroleum wastes and accidental spills do create environmental problems associated with the inhibition of water infiltration capacity [15] and nutrient deficiency in soils that is necessary

to support plant growth [16]. It also affects microbial population that is crucial in the degradation of organic and inorganic contaminants in soil [17]. Phytoremediation is the process of applying plants using its various phytotechnologies for removing contaminants from the soil and ground water aquifer that has been developed over the past decades [18, 19, 20] and is considered to be an in-situ, cost-effective, less environmental destructive and more aesthetically pleasant solar powered treatment system [21, 22, 23].

According to [24], the choice of plant species for the phytoremediation procedure is based on the occurrence of plants under specific climatic conditions, its resistance to pollutant toxicity, presence of phenolic compounds in root exudates and their capability to reduce pollutant concentrations in soil. The problem is that not many plant species is studied for their efficacy as a phytoaccumulator plant, despite the upsurge of environmental degradation due to hydrocarbon pollution because of oil exploration as seen in the Niger Delta region of Nigeria. This conditions is further exacerbated as the Lake Chad Basin in Nigeria is earmarked for additional crude oil exploration in addition to the expansion of refining and petrochemical companies such as that of Dangote in Lagos, Nigeria.

Eucalyptus camaldulensis a tree native to Australia but widely cultivated successfully around the world. It belongs to the family Myrtaceae with about 800 species. They are evergreen, fast-growing, deep-rooting trees that is tolerant to poor soil conditions such as drought and moderate salinity [25]. To this extent, this paper assesses its ability to phytoextract, bioaccumulate and decontaminate total petroleum hydrocarbon (TPH) and its associated insoluble asphaltene content in its various tissues.

II. Material And Methods

2.1 The Study Area

The study was carried out at the nursery of the Federal University Dutse, Jigawa State in the North West geopolitical zone of Nigeria. Jigawa state shares an international border with Republic of Niger to the north and the Nigerian states of Yobe to the northeast, Katsina to the northwest, Bauchi to the southeast and Kano to the southwest. It covers an area of about 24, 516 square kilometers and lies between latitude 11° N to 13° N and longitudes 8°E to 10° 15' E. It has a population of 4,361,002 (NPC 2006 Census figure). It is underlain by granites, schists and gneisses of the basement complex that emanates from the Pre Cambrian rocks of the Chad Formation. The soils are generally sandy at the top and compact at depth with often hard pans having aeolian deposits from the sahara desert forming substantial part of the soils.

The mixing of the subsoil in these deposits has given rise to clayey subsoil, which dominates the northern parts of the state. The relief is generally undulating, but rock outcrops are common in areas of basement complex rocks. The climate is semi-arid characterized by a long dry season (June – September) and a short wet season (October – May). The climate varies considerably and is erratic. The mean annual temperature is about 25°C but the mean monthly values range between 21°C in the coolest month and 31°C in the hottest month. Total annual rainfall ranges from 600 mm in the north to 1000 mm in the south with variations leading to severe and prolonged droughts that causes crop failure, death of livestock and overall human sorrow. The vegetation falls within the Sudan Savannah vegetation belt, but traces of Guinea savannah vegetation are found in parts of the southern districts characterized by extensive open grasslands with few scattered stunted trees[26].

2.2 Materials

2.2.1 Plant material

Plants used as materials for the study were the seedlings of *Eucalyptus camaldulensis* obtained from a farm in Kiyawa, a town about 70 km away from Dutse, the Jigawa state capital. It was selected for its fast-growing, deep-rooting and abilities to tolerate poor soil conditions such as drought and moderate salinity in addition to its possible use in agroforestry systems in the Niger Delta prone contaminated area and the Lake Chad Basin where oil exploration is targeted in Nigeria.

2.2.2 Crude oil

Crude oil (Bonny light) was obtained from the Kaduna Refining and Petroleum Limited (A subsidiary company of the NNPC).

2.3 Experimental Technique

Seedlings of *E. camaldulensis* was cultivated in an uncontaminated pot soil at the nursery for two (2) months (November - December) to acclimatize prior to transplanting into crude oil contaminated soil. Experimental plots were plastic basins of known capacity (5 L). The soil medium was contaminated using four levels of crude oil (Control, 0.3 L/4.0 kg soil, 0.5 L/4.0 kg soil and 0.7 L/4 kg soil) as modified from the study of [27]. Seedlings of the plant species were transplanted early morning into the contaminated plastic medium for three months (January - March) and replicated three times.

2.4 Experimental Design and Treatments

The design for the study was the Split - Split Plot (3 x 4 x 3) experiment. This design was selected to ensure more precision to tissues of the plant species. The main plots were the Crude oil contamination (3 levels),

Sub plot were the soil amendments (4 levels) while the sub-sub plot factor were tissues of the plant species (3 levels).

The following were the treatments (soil amendments):

T₁=Control

T₂=NPK (g kg⁻¹soil)

T₃= Cow-dung (3:1 v/v)

T₄= NPK (g kg⁻¹soil) + Cow-dung (3:1 v/v)

Note: All experimental units were tilled and watered daily to ensure aeration.

2.5 Samples and Sampling Technique

2.5.1 Plant samples

Plant samples from each experimental unit were collected at the end of the three months; these were then divided into three groups: roots, leaves and stem cuttings (in case of the woody species) or root and turf (for the grass). All sampled plants were oven dried to constant weight using YC/JY series Analytical Precision Balance, China (0.001 precision) at ~ 60°C for at least 48 hours and weighed individually to determine plant biomass.

2.5.2 Soil samples

Soil was sampled randomly at depth of 0 - 10 cm (the surface and at middle) using soil auger from each sampling unit. The sampled soil was then homogenized and composite soil sample obtained from the experimental pots every 21 days for a period of three months. All soil sampled for residual TPH quantification were collected as quickly as possible to prevent exposure to the environment and subsequent error in measurements. In addition, except for the tests mentioned, all soil was oven dried and passed through a 2 mm sieve prior to analysis.

2.6 Determination of Plant Biomass

Plant biomass was then determined using the following mathematical relationships:

$$Bm = W_r(g) + W_s(g) + W_l(g) \quad \text{i (E. camaldulensis)}$$

where: W_r = Dry weight of root; W_s = Dry weight of stem; W_l = Dry weight of leaf;

2.7 Determination of Phytoaccumulation of Total Petroleum Hydrocarbon and its insoluble Asphaltene in Plant tissues

The accumulation of TPH and its asphaltene in plant tissues were determined using the method adopted by [28]. Ten grammes (10 g) of the air dried plant samples was mixed with 10 grams anhydrous sodium sulphate to remove moisture. The hydrocarbon was Soxhlet extracted with chloroform for 8 hrs. The chloroform extract was then evaporated in a pre-weighed dish and the amount of total petroleum hydrocarbons (TPHs) was determined with the loss of TPH as shown in equation ii. The crude oil extracted was suspended in n-hexane and filtered through tared filter paper to remove and determine the insoluble fraction (Asphaltene) in the mathematical relationship in equation iii

$$TPH_p = (W_E(g) + E_t(g)) - E_t(g) \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \text{ii}$$

$$ASP_p = (W_f(g) + F_t(g)) - F_t(g) \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \dots \quad \text{iii}$$

where: TPH_p = Phytoaccumulated TPH, W_E = Weight of extract, E_t = Weight of tared evaporating dish, ASP_p = Phytoaccumulated asphaltene, W_f = Weight of filtrate, F_t = weight of tarred filter paper.

2.8 Determination of Phytoextraction efficiency of Total Petroleum Hydrocarbon and insoluble Asphaltene in Plant tissues

2.8.1 Determination of Bioaccumulation Factor (BAF)

The bioaccumulation factor (BAF) for each plant-soil pair was calculated to obtain useful information using the formula that was modified as obtained from [29] as follows:

$$BAF = \frac{C_{root} + C_{stem} + C_{leaf}}{C_{soil}} \quad \text{vi}$$

where:

C_{root} = Dry weight of accumulated petroleum hydrocarbon in root (g g⁻¹)

C_{stem} = Dry weight of accumulated petroleum hydrocarbon in stem (g g⁻¹)

C_{soil} = Dry weight of accumulated petroleum hydrocarbon in soil (g g⁻¹)

Note: Equations vi was used to obtain BAF for *E. camaldulensis*.

BAF is the ratio of accumulated petroleum hydrocarbon in plant tissues to the residual petroleum hydrocarbon in soil. This model assumes that a certain mass of the contaminant had been taken up into the plant from the beginning of its growth in the contaminated soil to the time of harvest.

2.8.2 Determination of Uptake Kinetics

Time dependent uptake kinetics was based on the BAF values and this was useful in estimating the effectiveness of the phytoextraction potentials of the tested species. Derivation of the uptake kinetics was based on uptake of contaminants from the contaminated soil through destructive sampling and not based on the kinetics of physiological uptake of contaminants into the plant.

Note that the uptake rate is a continuous process though it may vary during the life of the plant therefore, the total mass of the contaminant removed (influx mass) was determined using equation vii below:

$$M_{influx} = C_{plant} \times BM \quad \text{vii}$$

where:

C_{influx} = Mass of contaminant in plant species (g)

C_{plant} = Dry weight accumulated amount of contaminant in plant species (g)

BM = Dry biomass of plant species (g)

2.9 Data Analysis

Data collected were analyzed using Descriptive Statistics and Analysis of Variance (ANOVA), the split-split plot model, using GenStat Discovery Edition 4 software but due to limitation of ranking the Generalized Linear Model (GLM) procedure of SAS (Statistical Analysis System, 1999) was also used. The probability level of certainty in the research was at 95 % confidence limit or $\alpha = 0.05$ although, $\alpha = 0.01$ was also used. Statistical means were compared using the Fisher's Least Significant Difference (LSD) at $p \leq 0.05$ and $p \leq 0.01$. Means were also represented with bar charts and line graphs for easy comparison.

III. Result

3.1.1 Distribution of total dry biomass (DBM_T) in *E. camaldulensis*

Mean squares excerpts from the analysis of variance for the distribution of total dry Biomass DBM_T in the tissues of *E. camaldulensis* species were presented in Table 1. The analysis revealed that there were highly significant variations ($p < 0.05$ and $p < 0.01$) for the amount of DBM_T accumulated in tissues of *E. camaldulensis* among the levels of crude oil contamination, soil amendments and the plant tissues.

Results of Table 2 revealed that the highest DBM_T was observed in C1 (Soil with no contamination) with mean value of 6.27 g. This was followed by that of C4 with mean value of 6.17 g. The least DBM_T was observed in C3 with a mean value of 5.71 g. In terms of the soil amendments during the experiment, the control soil amendment (T1) that did not differ significantly with T3 yielded the highest amount of DBM_T in the tissues of *E. camaldulensis* with mean values of 6.47 g and 6.52 g respectively. The least amount of DBM_T was observed from T4 with mean of 5.40 g. It was also noted that while the stem of this species yielded the highest value of DBM_T with mean value of 7.20 g, the least amount of DBM_T was observed in its leaf with mean value of 4.71 g.

Table 1: Mean Squares from the ANOVA for Distribution of Dry Biomass in Tissues of *E. camaldulensis*

| Source of variation | Df | Distribution of Dry Biomass | |
|--------------------------------|-----|-----------------------------|------------|
| | | Dry BM (g) | |
| Crude Oil Contamination | | | |
| REP | 2 | | 0.19520 |
| Crude Oil Conc. (A) | 3 | | 2.16816** |
| Error | 6 | | 0.08724 |
| Soil Amendments | | | |
| Treatment (B) | 3 | | 10.46182** |
| A x B | 9 | | 10.43667** |
| Error | 24 | | 0.04916 |
| Plant Tissues | | | |
| Plant Parts (C) | 2 | | 75.97559** |
| A x C | 6 | | 9.10925** |
| B x C | 6 | | 3.62193** |
| A x B x C | 18 | | 2.55480** |
| Error | 64 | | 0.02422 |
| Total | 143 | | |

** = Highly Significant at $p < 0.01$

Table 2: Distribution of Dry Biomass in different Tissues of *E. camaldulensis*

| Treatments | Dry BM (g) |
|--------------------------------|-------------------|
| Crude Oil Contamination | |
| C1 (0 L) | 6.27 ^a |
| C2 (0.3 L) | 6.05 ^c |
| C3 (0.5 L) | 5.71 ^d |
| C4 (0.7 L) | 6.17 ^b |
| Mean | 6.05 |
| p of f | 0.001 |
| S.E.D | 0.0696 |
| Soil Amendments | |
| T1 | 6.47 ^a |
| T2 | 5.82 ^b |
| T3 | 6.52 ^a |
| T4 | 5.40 ^c |
| Mean | 6.05 |
| p of f | 0.001 |
| S.E.D | 0.0523 |
| Plant Tissues | |
| Leaf | 4.71 ^c |
| Stem | 7.20 ^a |
| Root | 6.24 ^b |
| Mean | 6.05 |
| p of f | 0.001 |
| S.E.D | 0.0318 |

T1 = Control; T2 = NPK (g kg⁻¹); T3 = Cow-dung (3:1 v/v); T4 = NPK (g kg⁻¹) + Cow-dung (3:1 v/v); Figures with same alphabets within columns do not differ significantly for Crude contamination, Soil amendments and Plant species respectively p of f = Probability value of F. S.E.D = Standard Error Deviation.

3.1.2 Phytoaccumulation and partitioning of petroleum hydrocarbon in tissues of *E. camaldulensis* species

Results extracted from ANOVA for both TPH and its associated asphaltene from *E. camaldulensis* were presented in Table 3. The analysis showed that there were highly significant variability) ($p < 0.05$ and $p < 0.01$) in the amount of phytoaccumulated TPH and associated asphaltene in plant tissues of *E. camaldulensis* among the different levels of crude oil contamination, soil amendments and plant parts of leaf, stem and root.

Table 3: Mean Squares from the Analysis of Variance for phytoaccumulated TPH and associated asphaltene partitioned in tissues of *E. camaldulensis*

| Source of variation | df | Accumulation in <i>E. camaldulensis</i> | |
|----------------------------|-----|---|----------------|
| | | TPH (g) | Asphaltene (g) |
| Crude Contamination | | | |
| REP | 2 | 0.0004083 | 0.0000111 |
| Crude Oil Conc. (A) | 2 | 0.0212583** | 0.0158028** |
| Error | 4 | 0.0000500 | 0.0002264 |
| Soil Amendments | | | |
| Treatment (B) | 3 | 0.0556481** | 0.0296716** |
| A x B | 6 | 0.0112954** | 0.0053262** |
| Error | 18 | 0.0002213 | 0.0000830 |
| Plant Parts | | | |
| Plant Parts (C) | 2 | 0.0266028** | 0.0133444** |
| A x C | 4 | 0.0050444** | 0.0007972** |
| B x C | 6 | 0.0216472** | 0.0073864** |
| A x B x C | 12 | 0.0075889** | 0.0031799** |
| Error | 48 | 0.0001569 | 0.0001093 |
| Total | 107 | | |

** = Highly Significant at $P < 0.01$

Table 4: Phytoaccumulation and Partitioning of TPH and Asphaltene in Tissues of *E. camaldulensis*

| Treatments | TPH (g) | Asphaltene(g) |
|--------------------------------|-------------------|-------------------|
| Crude Oil Contamination | | |
| C2 (0.3 L) | 0.12 ^b | 0.09 ^b |
| C3 (0.5 L) | 0.12 ^b | 0.09 ^b |
| C4 (0.7 L) | 0.16 ^a | 0.13 ^a |
| Mean | 0.13 | 0.10 |
| p of f | 0.001 | 0.001 |

| | | |
|-----------------|-------------------|-------------------|
| S.E.D | 0.00167 | 0.00355 |
| Soil Amendments | | |
| T1 | 0.20 ^a | 0.15 ^a |
| T2 | 0.10 ^d | 0.09 ^c |
| T3 | 0.12 ^b | 0.10 ^b |
| T4 | 0.11 ^c | 0.07 ^d |
| Mean | 0.13 | 0.10 |
| p of f | 0.001 | 0.001 |
| S.E.D | 0.00405 | 0.00248 |
| Plant Tissues | | |
| Leaf | 0.12 ^b | 0.09 ^b |
| Stem | 0.11 ^c | 0.09 ^b |
| Root | 0.16 ^a | 0.13 ^a |
| Mean | 0.13 | 0.10 |
| p of f | 0.001 | 0.001 |
| S.E.D | 0.00295 | 0.00246 |

T1 = Control; T2 = NPK (g kg⁻¹); T3 = Cow-dung (3:1 v/v); T4 = NPK (g kg⁻¹) + Cow-dung (3:1 v/v); Figures with same alphabets within columns do not differ significantly for Crude contamination, Soil amendments and Plant species respectively p of f = Probability value of F. S.E.D = Standard Error Deviation.

The TPH and insoluble asphaltene content as partitioned in the tissues of *E. camaldulensis* species were presented in Table 4. From the results, it was observed that the highest accumulated values for both TPH and asphaltene was in the highest contamination level (C4) with mean values of 0.16 g and 0.13 g respectively. The least accumulated values for both TPH and asphaltene were recorded in C2 (0.12 g and 0.09 g respectively) and C3 (0.12 g and 0.09 g respectively) that did not differ significantly.

The results further showed that the control soil amendment (T1) yielded the highest values for both accumulated TPH and asphaltene with mean values of 0.20 g and 0.15 g respectively. On the other hand, while the soil amendment with the least accumulated TPH value was T2 with 0.10 g that of asphaltene was observed in T4 with a mean value of 0.07 g.

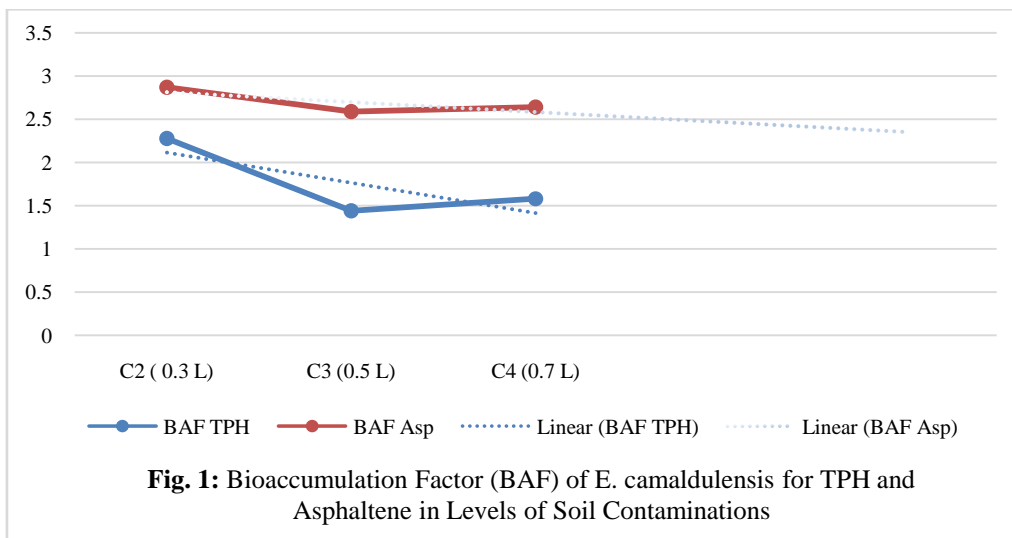
The roots of this species was observed to accumulate highest amount of both TPH and asphaltene with mean values of 0.16 g and 0.13 g respectively. It was further observed that while the stem had the least TPH accumulation with a mean of 0.11 g the leaf and stem that do not differ significantly for asphaltene accumulation was the least with 0.09 g and 0.09 g respectively.

3.2 Phytoextraction Potentials of *E. camaldulensis* in the Bioattenuation of Petroleum Hydrocarbon

The phytoextraction potentials for the tested plant species was determined using the bioaccumulation factor (BAF) and mass influx (M_{influx}) calculated from the results of accumulation of the petroleum hydrocarbon contaminants in the plant tissues.

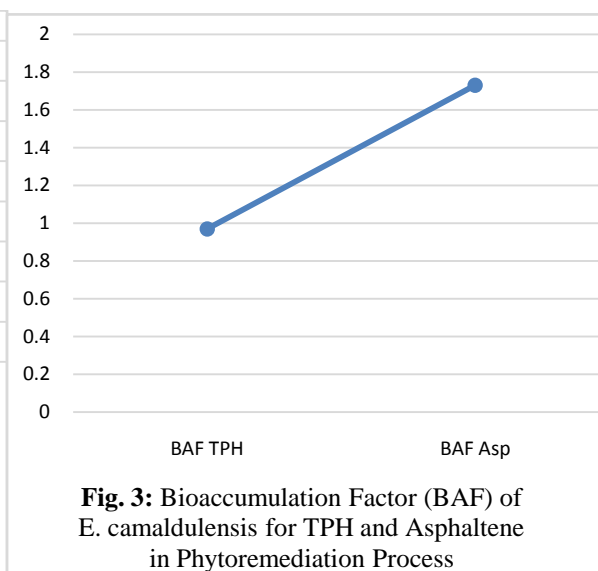
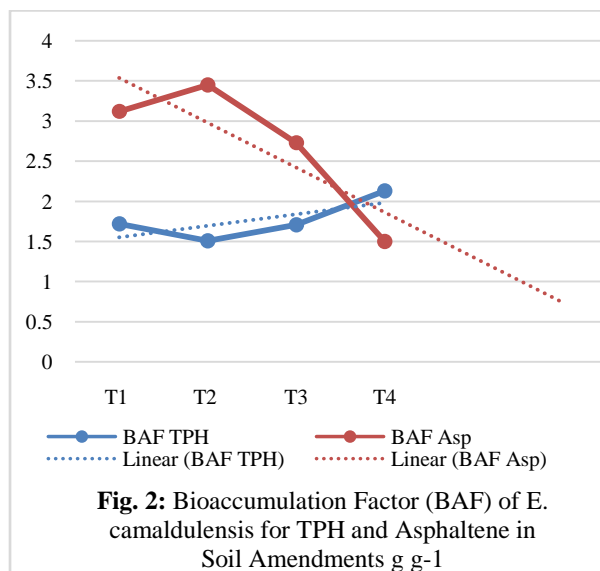
3.2.1 Bioaccumulation Factor (BAF) for petroleum hydrocarbon in plant species

Results of the mean values of BAF for both TPH and the insoluble asphaltene of the tested species were presented in figures 1 - 3. From the result, it showed that the BAF for the TPH was highest in C2 (2.28 g g⁻¹) and the least BAF value for TPH was recorded in C3 with mean value of 1.44 g g⁻¹. Similarly, the BAF for the insoluble asphaltene in C2 that did not differ significantly with that of C4 was the highest with a mean values of 2.87 g g⁻¹ and 2.64 g g⁻¹ respectively. Additionally, the higher the concentrations of contaminants in soil the lower the BAF for *E. camaldulensis*.



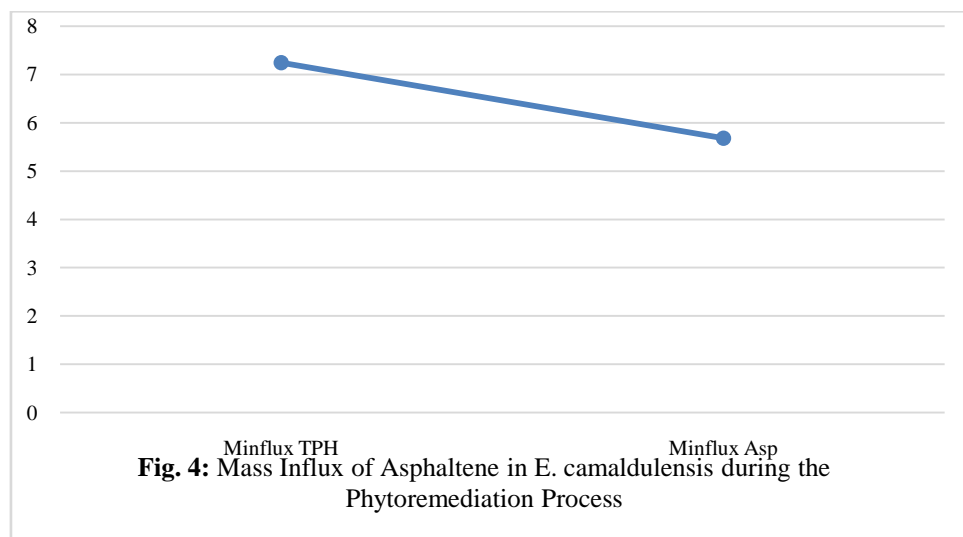
The results in Fig. 2 further revealed that T4 was the soil amendment with the highest BAF for TPH with mean value of 2.13 (g g^{-1}) and the least for TPH was T3 with mean value of 1.71 (g g^{-1}). In terms of the asphaltene, T2 yielded the highest BAF value of 3.45 (g g^{-1}) while the least was observed in T4 with a mean value of 1.50 (g g^{-1}).

In Fig. 3, it was observed that while the BAF for TPH in the tested species is less than 1 that of asphaltene is greater than 1. This implies that the accumulation capacity of the species for the insoluble asphaltene from soil is greater than that of the insoluble TPH with mean values 0.97 (g g^{-1}) and 1.73 (g g^{-1}) respectively.



3.2.2 Mass Influx (M_{influx}) of Petroleum Hydrocarbon in tissues of *E. camaldulensis*

The results of the mean M_{influx} values for TPH and the insoluble asphaltene of the tested species were also presented in Fig. 4. It was observed that while the M_{influx} for TPH in the tested plant species was above 7.24 g g^{-1} that of the asphaltene was 5.68 g g^{-1} . This indicated that the rate at which *E. camaldulensis* species uptake, accumulate and translocate TPH is higher than that of the insoluble asphaltene.



IV. Discussion

4.1 Total Dry Biomass (DBM_T) of *E. camaldulensis* during Bioattenuation of Petroleum Hydrocarbon

It was observed from the results that although biomass production by *E. camaldulensis* in uncontaminated soil (C1) yielded the highest value for DBM_T, the values observed in C4 the highest soil contamination level (0.7 L/4.0 kg soil) was closely following in DBM_T production. This means that the plant species can tolerate organic contaminant's toxicity and grow successfully as supported by [1]. More so, the result supported the fact that the plant did meet the toxicity criterion of plant selection for the phytoremediation process as reiterated by [2, 24].

The use of organic fertilizer of cow-dung (T3) as soil amendment in addition to daily tillage and watering at field capacity did encourage growth for the species. This could be possible because organic fertilizers are known to improve both physical and chemical characteristics of the soil. On the other hand, the stem of this plant was observed to yield the highest DBM_T that was closely followed by its leaves possibly due to some leaf senescence as a result of the escape of organic volatile substances through the stomata.

4.2 Phytoaccumulation and partitioning of petroleum hydrocarbon in tissues of *E. camaldulensis*

It was observed that phytoaccumulation of the soluble total petroleum hydrocarbon (TPH) and the insoluble asphaltene in the plant was highest in C4 (0.7 L kg⁻¹ soil). This implies that phytoaccumulation of the TPH and asphaltene in tissues of the tested species increases with increasing levels of hydrocarbon contamination in soils. In another vein, it means that the tested species did meet the criterion of being among the phytoaccumulator plants as supported by [24]. These plants are readily available and can grow fast hence, it will be effective in the clean-up of hydrocarbon polluted sites in Nigeria.

Aside the control treatment (T1) the soils amended with the cow-dung (T3: 3:1 v/v) did encouraged better phytoaccumulation of contaminants in tissues of the tested species. This could be because tillage and watering does improve permeability and availability of contaminants to plants as well as improve aeration in contaminated soil while both organic fertilizer did improve nitrogen and phosphorus thereby improving the activities of microorganisms in soil leading to reduced toxicity of contaminants and its subsequent accumulation in plants as supported by [30]. In addition, though the uptake and translocation of the contaminants were observed in all tissues of the plant, the root phytoaccumulate more than any other tissue.

4.3 Mass Influx (M_{influx}) of Petroleum Hydrocarbon in tissues of *E. camaldulensis*

The rate at which the tested plant species uptake, translocate and accumulate in its tissues is revealed in its M_{influx} values for both TPH and the insoluble asphaltene. From the results, it was obvious that the plant do uptake the soluble TPH contaminants faster and better than it did for the insoluble asphaltene fraction.

V. Conclusion

Based on the results of this study, it was concluded that *E. camaldulensis* species can tolerate organic contaminant's toxicity of up to 0.7 L/4.0 kg soil and uptake, translocate and accumulate same into all its tissues of root, stem and roots; though it did phytoaccumulate the contaminants better in its roots. Additionally, the

species could uptake, translocate and accumulate the soluble TPH much more than the insoluble fraction. Besides, the plant could achieve these when contaminated soil is treated with organic fertilizer (cow-dung at 3:1 v/v). To this end, the plant is recommended as good plant fit for use during the phytoremediation procedure but its performance could be less than other species if compared for such efficacy in remediation of petroleum contaminants.

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