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Assessing the Efficacy of *Moringa oleifera* Seed Powder in Ameliorating Spent Engine Oil Toxicity on Hydroponic *Acalypha wilkesiana* Stem Cuttings

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Abstract

Pollution is in flux due to pressures on machinery and oil exploration. The rapid study assessed the efficacy of *M. oleifera* seed powder (MOSP) and *A. wilkesiana* stem cuttings (AWSCs) in ameliorating toxicity of spent engine oil (SEO) using hydroponics, a completely randomized experimental design (CRD) and assigning phytotoxic scores. The study was carriedout under laboratory-contolled conditions and the ameliorative efficacy was assessed over a 14-day period by observing: root, shoot, weight, and observable phytotoxicity symptoms. AWSCs were exposed to various treatments: treatment 1-5 spiked with 10ml SEO per replicate and amended with MOSP (3, 6, 9, 12 and 15g), single amendment with 9g MOSP, negative control (distilled water), and a positive control (SEO-only). The positive control demonstrated severe phytotoxicity, characterized by no root growth, necrosis chlorosis, drying, and mortality. Tr1 and 2 demonstrated phytotoxicity symptoms as the positive control after seven days, Tr3 remained resilient with reduced toxicity for 12 days and showed signs of atrophy, while Tr4 and 5 demonstrated resilience and no phytotoxicity symptoms till day 14. Tr6 and the negative control demonstrated shoot growth and root development. MOSP significantly reduced toxicity in most contaminated groups. Treatment with SEO and respective MOSP concentrations resulted in root and shoot length(s) growth compared to the positive control, negative control, and MOSP biosorbent amendment at a high significance difference of $p \leq 0.0001$. The study showed MOSP biosorbed concentrations of SEO, neutralized toxicity, augmented the phytoremediative capability of *A. wilkesiana* and stabilized hydroponic solutions for optimal root growth and plant stability.

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Introduction

Environmental pollution has fluxed in the past few decades due to increased anthropogenic activities (Sharma, 2021). Spent, used or waste engine oil like anyother pollutant(s) has been implicated in a broad array of negative impacts (especially due to leakage and poor disposal) on environmental matrices (air, water soil and biota) and it constitutes an important concept in pollution studies (Armioni et al., 2024). Spent engine oil (SEO) poses a significant environmental threat, particularly in areas characterized by extensive industrial operations and inadequate waste management practices. The presence of SEO, which contain hydrocarbons and heavy metals, detrimentally impacts soil health, microbial functionality, and plant productivity (Ossai et al., 2020). These contaminants can alter soil structure, diminish nutrient bioavailability, and elevate toxicity levels, thereby hindering plant and animal development in affected ecosystems (Rodriguez-Rodriguez et al., 2016). Engine oil pollution also causes changes in chemical and physical characteristics of soil or water, subsequently giving way to edaphic accumulation (of heavy metals and nutrients) which ends up being bioaccumulated by plants (Mohsen and Hazim, 2023). Furthermore, the persistence of SEO in soil can facilitate the biomagnification of toxic substances in the food chain, posing substantial risks to human health and ecosystem integrity (Singh and Haritash, 2019). Prolonged exposure to these contaminants can lead to persistent degradation of soil quality and loss of biodiversity, underscoring the need for effective remediation strategies (Khan et al., 2011).

Contamination by spent engine oil poses a significant environmental threat, particularly in areas characterized by extensive industrial operations and inadequate waste management practices. The presence of spent oil, which contains diverse hydrocarbons and heavy metals, detrimentally impacts soil health, microbial functionality, and plant productivity (Ossai et al., 2020). These contaminants can alter soil structure, diminish nutrient bioavailability, and elevate toxicity levels, thereby hindering plant growth and development in affected ecosystems (Rodriguez-Rodriguez et al., 2016). Engine oil pollution also causes changes in chemical and physical characteristics of soil or water, subsequently it gives way to edaphic accumulation (of heavy metals and nutrients) which ends up being bioaccumulated by

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plants (Mohsen and Hazim, 2023). Furthermore, the persistence of spent oil in soil can facilitate the bioaccumulation of toxic substances in the food chain, posing substantial risks to human and ecosystem health (Singh and Haritash, 2019). Prolonged exposure to these contaminants can lead to persistent degradation of soil or water quality and loss of biodiversity, underscoring the need for effective remediation strategies (Khan *et al.,* 2011).

Phytoremediation or plant remediation is a biotechnology involving the use of plant products and plants in the accumulation, extraction and degradation of contaminants (Oseni *et al.,* 2020). This type of remediation involves processes like phytoextraction, phytostabilization, phytomining and rhizofiltration (Subhashini et al. 2017). Remediation of contaminants in polluted environments also involves absorption, translocation (from the root to the shoots), and accumulation (Miguel *et al.,* 2013; Sharma, 2021). Most endemic plants that survive or grow in polluted environments are good phytoremediators, and the success of using these plants could require the improvement of their remediative potentials by bioaugmentation (Sharma, 2021) or by chemoaugmentation (Oseni *et al.,* 2020). Furthermore, phytoremediation holds many advantages, some prominent merits include cost effectiveness, broader applications, reduced management and processing costs, soil profile conservation, metal leaching prevention and erosion control (Ali *et al.,* 2013). The introduction of plants or relative products into polluted environments may degrade, remove, volatilize, or immobilize pollutants (Oseni *et al.,* 2020). One importance which can not be looked away, is the effectiveness of bioremediaton in the improvement of soil fertility due to the introduction of organic biomass and matter (Mench *et al.,* 2009).

Many foreign and indigenous workers carried out research and reviews on phytoremediation or bioremediaton involving plants. Based on their findings, they concluded on the positive or potential outcomes of remediation and drawbacks in their respective research or review findings (Chen *et al.,* 2023). Certain plants are used as phytoremediators in environmental clean up, some plant species have been reported to be reliable in heavy metal remediation and some in organic pollutant remediation. A summary of some notable endemic and exotic phytoremediative plants used in remediation research include: *Abutilon indicum, Acalypha indica, Physalis minima* (Subhashini *et al.,* 2017), *Acalypha wilkesiana* (Dokania *et al.,* 2024; Iya and Assim, 2025), *Moringa oleifera* (Nwagbara *et al.,* 2022; Adhikari and Neupane, 2025), *Chromolaena odorata, Axonopus compressus* (Azorji *et al.,* 2024), *Aspilia africana* (Beckley and Chijioke-Osuji, 2015; Azorji *et al.,* 2024), *Abelmoschus esculentus* (Ajie and Adelegan, 2025), *Helianthus annuus*, *Cannabis sativa, Nicotiana tabacum, Zea mays* (Yan, 2020), *Phrgmites australis, Chenopodium marale, Chenopodium album, Lactuca serviola* (Aloud, 2023), *Calendula officinalis, Celosia cristata pyramidalis, Melastoma malabathricum, Iris lactea var. chinensis, Euphorbia milii, and Tagetes patula* (Liu, 2018) e.t.c.

Materials and Methods

Study area: The study was conducted in a controlled environment (laboratory) within the Federal University Otuoke ecosystem, Ogbia L.G.A., Bayelsa State, Nigeria. The coordinates for the study area lies within Latitude 4°42’23 ″N and Longitude 6°19’44 "E. This study was carried out to investigate the efficacy of both *A. wilkesiana* (Copper leaf plant) and *M. oleifera* (Moringa or Drum stick plant) in remediation.

Materials and equipment Sampling materials and equipments used included: 10 centimeter *A. wilkesiana* stem cuttings (AWSCs) sourced from the University Botanical garden, dried *M. oleifera* seeds, 15 litres of distilled water, one litre of SEO (contaminants utilized in spiking), 10 litres of Hoagland's hydroponic solution. Laboratory wares including: beakers, flasks, petri dish, test tubes, burettes, pipettes, measuring cylinders, digital thermometer, syringe, pH meters, funnels, filter paper, labcoat, disposable gloves, nose masks, mortar and pestle, mixing wares (spatula and ceramic bowls), measuring wares, (vernier caliper, meter rule, white thread, and tape rule) and electronic weigh. The model plants employed in the novel study were *A. wilkesiana*, also known as the copper leaf plant, or red variegated plant, the remediative-biosorbent used was *M. oleifera* seed powder (MOSP), and the contaminant administered was spent engine oil (SEO). The laboratory control-site followed a completely randomized design (CRD) or randomized complete block design (RCBD) pattern in the organization of hydroponic samples (Table 1.). Eight treatments (with two replicates each) were utilized in the study (i.e. 16 samples overall). Sample hydroponic plants were handled, organized, and labelled appropriately.

Procedure and Protocol: Precise protocol, procedures and precautions were taken to achieve desired results. *A. wilkesiana* stem cuttings: n= 16 (8×2) were first cut with a blade to allow for root growth then introduced into Hoagland's solution (10 ml) in 1 litre of water to acclimatize and accumulate nutrients, mimicking a typical soil environment for two days prior to spiking and amendment(s). Varying numbers (10, 20, 30, 40 and 50 seeds) of *M. oleifera* seeds were grinded with a ceramic mortar and pestle to powder. Hydroponic contaminanted solutions were prepared by placing 750 ml distilled water in glass wares, and then spiking with SEO in respective treatments prior to sample plant introduction. 10 ml of SEO was used to spike hydroponic solutions and varying concentrations of MOSP biosorbent were administered (3g, 6g, 9g, 12g, and 15g) to replicates (Table 1.). The amended and spiked hydroponic solutions were allowed to acclimatize for 24 hours and then the hydroponic stem cuttings were introduced (from the Hoagland's mixture) into the various contaminated hydroponic solutions (this was done to avoid any additive, antagonistic, or synergistic reactions from mixing, amendments and treatments). The treatments varied: 10 ml concentrations of SEO were administered for treatments 1-5 and for the positive control (PC) with varying amendments, the negative control (Clean control) was not spiked or administered any amendment (MOSP), and treatment 6 was administered 9g (amendment control only). The hydroponic

samples were contaminated to simulate pollution in the the laboratory-controlled environment. Spiked treatments, unspiked treatments, and amended controls were used in the study respectively (Table 1.).

Table 1. Experimental design for treatments and amendments

Treatment (TR)	Replicates (R)	Spiking (SEO in ml)	Amendment (MOSP in grams)
Positive control (Oil spike only)	2	10ml	No amendment
TR 1 (Oil spike and amendment)	2	10ml	3.0g (10 seeds)
TR 2 (Oil spike and amendment)	2	10ml	6.0g (20 seeds)
TR 3 (Oil spike and amendment)	2	10ml	9.0g (30 seeds)
TR 4 (Oil spike and amendment)	2	10ml	12.0g (40 seeds)
TR 5 (Oil spike and amendment)	2	10ml	15.0g (50 seeds)
TR 6 (Amendment only)	2	No spike	9.0g (30 seeds)
Negative (clean) Control	2	No spike	No amendment

The table above gives a subtle narrative from the novel methodology for treatments and amendments per replicate in 750 ml of contaminated and uncontaminated hydroponic solutions in the 14-day control study.

Morphometrics: The initial (Day 0) and final (Day 14) lengths and weights of the plants were estimated in centimeters and in grams using measuring rule, white thread, venier caliper and an electronic weighing scale to determine the hydroponic plant development and growth.

Phytotoxicity score: The phytotoxicity scores of spiked hydroponic stem cuttings (HSC) samples were estimated following the novel procedures adopted by Diogo et al. (2025), who formulated the VipTox approach (a novel observatory phytotoxicity scoring analysis). The developmental, morphoanatomical and phytotoxic observations of the hydroponic plant samples were organized in a way to ascertain the intensity of toxicity on each replicate using an arrayed toxicity profiling. These observed changes were organized by giving an array of scores from 0-10 (no morphoanatomical changes to morphoanatomical severity). The non-phytotoxic scores for un-spiked samples and unaffected samples were designated with capital letters. Every stage in the array potrays a sequential key with a score. Based on the procedures of Diogo et al. (2025), this formula was utilized:

$$\mathrm{PMD} = \frac{\sum(\mathrm{IS})}{n}$$

Here: PMD: means Phyto-Morphoanatomical Disruption, IS: means Individual Score per replicate, and n: means Complete number of replicates (all hydroponic samples). The PMD sequence key is expressed in the series below:

A.1) With chlorosis and necrosis.....**10**

2) With chlorosis and necrosis.....**B**

B. 1) Weak HSC...….....**9**

2) Underdeveloped leaves and/or roots.....**8**

3) With Abscission.....**7**

4) Without root growth.....**6**

5) Without atrophy.....**C**

C. 1) With intense epinasty.....**5**

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2) With mid-epinasty.....**4**

3) Without epinasty.....**D**

D. 1) No length or growth increase.....**3**

2) With length or growth increase.....**E**

E. 1) Reduced weight above negative control.....**2**

2) Reduced weight above negative control.....**1**

F. 1) Healthy and with no morphological changes..**0**

Software's and statistical analysis: Grammar checker and plagiarism softwares were used to check grammar errors, literal errors and for copyright appropriation. Data were organized as mean ± standard deviation in 8 replicates (n= 16). Visual PhytoToxicity assessment (ViPTox; Diogo *et al.,* 2025) was used in scoring morphoanatomical phytotoxic changes. One way ANOVA was also used to test the relationships between means at a $p \leq 0.05$ level of significance (later estimated as $p \leq 0.0001$).

Results and Discussion

Amongst bioindicators, plants remain important group of sentinels used in determining stress in ecosystems and change in environmental dynamics giving room for environmental legislation, mitigation and studying pollution (Ceschin *et al.,* 2023). Phytoremediative specimens used in environmental pollution must have bioaccumulating capacity, must be resilient, must possess appropriate rooting system, must have large mass and must have quick growth (Durumin Iya *et al.,* 2022). A number of researchers utilized *Acalypha spp*: *Acalypha indica* (Subhashini *et al.,* 2017), *A. wilkesiana* (Durumin Iya *et al.,* 2022; Dokania *et al.,* 2024; Durumin Iya and Assim, 2025), and *M. oleifera* (Nwagbara *et al.,* 2022; Adhikari and Neupane, 2025) in remediation of toxic heavy metals, inorganic and organic pollutants. These plant species have been utilized extensively in research, are endemic, environmental friendly, and also easily accessible for remediation. *A. wilkesiana is* of the Euphorbiaceae plant family, endemic to Sub-saharan Africa, Asia and the Americas (Durumin Iya *et al.,* 2018; 2023). Subhashini et al. (2017), Durumin Iya et al. (2018), Durumin Iya et al. (2022), Durumin Iya et al. (2023), and Durumin Iya and Assim (2025), posited in their findings that *A. indica* and *A. wilkesiana* have the capability to accumulate toxic heavy metals and hydrocarbons in varied parts (leaves, stems and roots). It possesses a good rooting system, and is resistant to environmental pollution (Durumin Iya *et al.,* 2022; Durumin Iya and Assim, 2025). It has the capability to remediate water pollution (Dokania *et al.,* 2023), soil and landfill pollution (Durumin Iya *et al.,* 2018; 2022; 2023; 2025).

M. oleifera is of the Moringaceae plant family, widely distributed, and endemic to Sub-saharan Africa, Asia, and the Americas (Lea, 2014). This plant species is known to be characterized with filtering, flocculating, sedimenting, coagulating and water cleansing characteristics. The seeds of this plant contain polypeptides, 15.5% lipids, 34.1% protein, and 15% carbohydrates (Lea, 2014). Proximate characteristics of one seed constitutes: 1.06% natural carbon, 10.55% Nitrogen, 1.95% water, 18.33 ppm of Phosphorus and a pH of 7.06 (Agboun *et al.,* 2016). It has been scientifically proven that *M. oleifera* seeds possess water purifying effects due to their cationic (poly-electrolytes) or organic polymerization capability. The polypeptides found in the seeds of this plant possess an isoelectric pH of 10-11, act as flocculating agents (molecular weight of 6-16 kDa) biosorbing suspended concentrations and flocs or forms sediments (Lea, 2014). Agboun et al. (2016) in their study posited that *M. oleifera* seeds enhanced biodegradation of crude oil at a 64.3% efficacy, also boosting microbial availability that enhanced oil degradation as well. Results from their study postulated that the presence of *M. oleifera* amendment in soil increased the numbers of oil destroying bacteria. Further postulations demonstrated that *M. oleifera* seed extracts had a buffering effect during remediative treatments as the pH of amended samples were between 7.24 to 7.70, and that of samples without amendment were between 7.24 to 7.44. The pH for the amended samples created an enabling environment for oil destroying bacteria.

Most research use phytoremediative specimens or bioremediative approach respectively in remediation research but there is paucity of the utilization of phytoremediation and bioremediation as a combined approach. Building on this, the combination of these two methods (phytoremediation by AWSC and biological remediaton by MOSP) in achieving remediation is an approach of necessity for ameliorative efficacy. We argue that the introduction of MOSP (biochemical and biological remediation) into *A. wilkesiana* hydroponic systems (phytoremediation) demonstrates a novel low-cost biotechnology and methodology for efficient remediation. MOSP neutralizes toxicity through biosorption while simultaneously stabilizing the hydroponic solution for optimal root and shoot growth of *A. wilkesiana*. From the study, it was observed that MOSP and *A. wilkesiana,* biosorbed and bioaccumulated concentrations of engine oil respectively. Smaller aliquots of engine oil were observed floating on the surface of hydroponic solutions compared to the original concentrations (10ml) administered at the beginning of the research. The observation of reduced floating oil concentrations suggested efficacy, indicating that the combined treatments (AWSCs and MOSP) likely flocculated and sequestered significant concentrations of SEO. The MOSP could have buffered and prevented some uptake by the sample plant(s) beyond their respective resilient levels. This underscores the cationic capability of MOSP proteins in binding hydrophobic pollutants, the accumulation and translocation capability of *A. wilkesiana.*

Furthermore, the pollution control was without amendment and the initial SEO concentration remained the same, while the negative control (distilled water) and treatment 6 were not spiked. The spiked samples (treatment 1-5) showed varying changes in suspended SEO concentrations: ≤3% (Tr1), ≤10% (Tr2), 35%

(Tr3), 45-50% (Tr4), 55-65% (Tr5), while the aliquot for the pollution control remained unchanged (100%). The higher percentage range values show a positive dose dependent concentration reduction in SEO due to MOSP amendment (Fig 1.). This meant that MOSP amendment sorbed considerable concentrations of SEO from the surface of hydroponic solutions. It was observed that higher amendment concentration(s) demonstrated no toxicity and yielded better results in hydroponic stem stability. Higher concentrations of MOSP biosorbent amendments yielded better ameliorative and remediative effects, showing that dosage might not always create poisons (Table 1.). This novel low cost biotechnology can be utilized in the remediation of waste water, polluted surface water and soils especially in developing countries.

Morphoanatomical characteristics of plants and animals (biota) serve as reliable indicators of instability in the environment, ecological stress, and fluctuating asymmetry (FA), all signs of developmental instability in biota (Abdulsalam *et al.,* 2025). Bioaccumulation, biomagnification, contamination and pollution are all implicated in their respective connections to developmental instability. In plants, the value of phytotoxicity in environmental research can not be underestimated as its' capabilities are underscored in reflecting environmental stressors and pollution (Lee *et al.,* 2010). Phytomorphoanatomical changes disrupt developmental stability in plants, affecting their environmental and functional dynamics which also depends on the intensity of these changes. Plant stress can be observed through developmental instabilities like stunting, stem cankering, deformations or weaknesses in the leaf, roots or stem (organ atrophy), phytotoxic and morphoanatomical observations like epinasty (curling or flagging), abscission (leaf drop), chlorosis and necrosis (Diogo *et al.,* 2025). The leaf, root, and stem deformities are signs of SEO burning from bioaccumulation and translocation. Assigning scores to developmental instability is important in estimating severity. The Phyto-Morphoanatomical Disruption (PMD) sequence key chart gives a profound clarification on associated and observed developmental instability and phytotoxicity in spiked samples (Fig 2.). Morphoanatomical atrophies (score 9 and 8) like alterations in the leaves, stems and root organs of plant are reliable indicators of physiological and metabolism dynamics or changes, important for plant development (Diogo *et al.,* 2023). Other atrophies (score 7, 6, and 5) and developmental instabilities (score 4 and 3) could cause plants to have weak immune systems giving way to pathogens, infections and parasites (Diogo *et al.,* 2025). Morphoanatomical changes can have deleterious effects on plant developmental stability, and processes (hormone stabilization, photosynthesis, translocation of plant nutrients, and osmosis) which are important for growth (Diogo *et al.,* 2023).

The measurements and observational records of biotic development is important in tracking their responses to anthropogenic and natural stressors in ecosystems. Morphological estimations and observations were recorded for *Acalypha wilkesiana* hydroponic stem cuttings samples (HSCs). The HSCs total length, (initial lengths), growth length (final length), initial weight, final weight, root growth, leaf growth and phytotoxic observations were all recorded to ascertain experimental efficacy of treatments and amendments. The initial lengths of all HSCs were measured at 10cm to precision, but individual HSC weight varied. The mean length and weight of spiked and un-spiked samples also varied with: pollution control, Tr1, and Tr2 having no mean increase in length (no growth), while Tr3, Tr4, Tr5, Tr6 and the clean control had considerable mean increases in length respectively (minimal to moderate to maximum growth). This length relationships show growth inhibition and no observed effect from amendment in spiked samples (PC, 1, and 2), which is a symptom of phytotoxicity (Table 3.). The initial weight of all HSCs were also measured with precision. The overall weight(s) and mean weight(s) of spiked and un-spiked samples varied respectively demonstrating that: pollution control, Tr1, Tr2, and Tr3 have no mean increase in weight, while Tr4, Tr5, Tr6 and the clean control had considerable mean increase in weight respectively (Table 3.). All developmental observations of the HSCs in the study reflect variations in inhibition, stimulation and phytotoxicity.

According to Barrow and Hartemink (2023), pH is important for plant development but is a core characteristic of soluble states of matter like liquids and not an edaphic characteristic except for some exemptions. The pH of the initial solution (distilled water prior to spiking) and all HSCs were estimated using a digital pH meter. Subsequently, on the 14th day (final day of experimentation), the pH and dissolved oxygen (DO) of all HSCs was also estimated. The pH and dissolved oxygen of all samples varied and demonstrated effects on the development and on phytotoxic observations. From the study, a pH of 4.3 to 5.4 demonstrated unfavourable conditions for HSC development while an ambiguous pH of 5.7 through to a pH of 7.6 showed slightly favourable to highly favourable conditions for HSC development. In hydroponic experimentation, DO is quite important for plant development (Qin and Ferrarezi, 2025). DO for positive control, Tr1 and Tr2 (3-4mg/L) demonstrated effects on development. Oil on the surface film of water could affect the concentration of DO in water. DO for Tr3, Tr4, Tr5, Tr6, and the clean control reflected favourable hydroponic conditions for HSC development.

Consequently, Tr6 (amendment only) reflected favorable optimal conditions and nutrient availability for development with a length increase of 49% in comparison with it's initial length. This could mean *Moringa oleifera* is suitable for nutrient enrichment or as a fertilizer for plant development. Tr5 reflected a dose dependent (amendment) and positive response to MOSP. In contrast, the pollution control, Tr1, and Tr2 reflected growth inhibition and weight decrease as a result of phytotoxicity and physiological stress (plant utilizing stored carbohydrates with no development). The clean control (Distilled water only), showed better development than the PC, Tr1, Tr2, Tr3 and Tr4. Although Tr5 and Tr6 showed enhanced growth rate due to MOSP amendment(s), PC, Tr1 through to Tr3 showed phytotoxicity and developmental instability compared to Tr6 and the clean control. The study posits that the development of *Acalypha wilkesiana* HSCs despite being a phytoremediative specimen is reactive and sensitive to

concentrations of SEO and MOSP amendment at varying levels. It has also demonstrated that this "green on green" biotechnology could standout as a reliable remediative prowess if realized and further finalized. The results of the study showed a significantly different pattern at $P \leq 0.0001$. The One-Way ANOVA confirmed the high significance of these results, proving that the variations in growth were a direct result of the treatment compositions rather than just random error (Table 4.).

Table 2. Physiochemical Observations of hydroponic solutions

Treatment (TR)	Initial pH (Distilled water)	Final pH	Final DO
Positive control	6.9	4.3	< 3mg/L
TR 1		5.2	<4mg/L
TR 2		5.4	
TR 3		5.7	6mg/L
TR 4		6.0	< 7-8mg/L
TR 5		6.2	
TR 6		7.6	8.5mg/L
Clean Control		6.8	> 9mg/L

Table 3. Developmental and Phytotoxic Observations

Developmental Observations							Phytotoxic scores	Phytotoxic symptoms
Treatment	Initial Length (cm)	Mean of Final Length (cm)	Mean Initial Weight (g)	Mean of Final Weight (g)	Shoot Growth	Root Growth		
Positive control	10 cm	10 + 0.000	12.50 + 0.300	12.355 + 0.305	No growth	No growth	10,9,7,6,5,3,1	Stunting, Epinasty (Curling/Flagging), Abscission (Leaf Drop), Chlorosis, Necrosis, Stem canker
TR 1	10 cm	10 + 0.000	12.00 + 0.300	11.765 + 0.355	No growth	No growth	10,9,7,6,5,3,1	Stunting, Epinasty (Curling/Flagging), Abscission (Leaf Drop), Chlorosis, Necrosis, Stem canker
TR 2	10 cm	10 + 0.000	11.25 + 0.550	10.975 + 0.455	No growth	No growth	10,9,7,6,5,3,1	Stunting, Epinasty (Curling/Flagging), Abscission (Leaf Drop),

								Chlorosis, Necrosis, Stem canker
TR 3	10 cm	10.35 + 0.150	13.55 + 0.250	13.375 + 0.255	No growth	Minimal Growth, Few roots growth (2 underdeveloped tap roots)	9,8,7,4,3,2	As at 12 days: Epinasty (Curling/Flagging), Abscission (Leaf drop), Chlorosis, Necrosis, Stem canker
TR 4	10 cm	10.80 + 0.200	12.80 + 0.300	12.925 + 0.305	No new leaves, slight shoot growth	Moderate Growth (5 tap roots)	BCDEF(0)	As at 14 days: No symptoms
TR 5	10 cm	11.90 + 0.100	11.90 + 0.700	12.150 + 0.750	1 new small leaf, shoot growth	Moderate Growth (7 tap roots)	BCDEF(0)	As at 14 days: No symptoms
TR 6	10 cm	14.90 + 0.100	12.95 + 0.350	13.270 + 0.340	3 new small leaves, shoot growth	Maximum Growth (9 tap roots)	BCDEF(0)	As at 14 days: No symptoms
Clean Control	10 cm	12.85 + 0.150	12.55 + 0.150	12.95 + 0.250	3 new small leaves, shoot growth	Maximum Growth (8 tap roots)	BCDEF(0)	As at 14 days: No symptoms

The table above gives a subtle narrative of the initial lengths and weights of replicates, treatment lengths and weights of replicates, shoot and root growth, and phytotoxic observations.

Table 4. One way ANOVA of Developmental Observations (length and weight)

SOURCE OF VARIATION	SS (SUM OF SQUARES)	DF (DEGREES OF FREEDOM)	MS (MEAN SQUARE)	F-STATISTIC	P-VALUE
Between Groups	43.85	7	6.264	238.64	≤ 0.0001
Within Groups (Error)	0.21	8	0.026		
Total	44.06	15			

The table above gives a summary of the one way analysis of variance (ANOVA) showing the results from the study reflects high significance difference

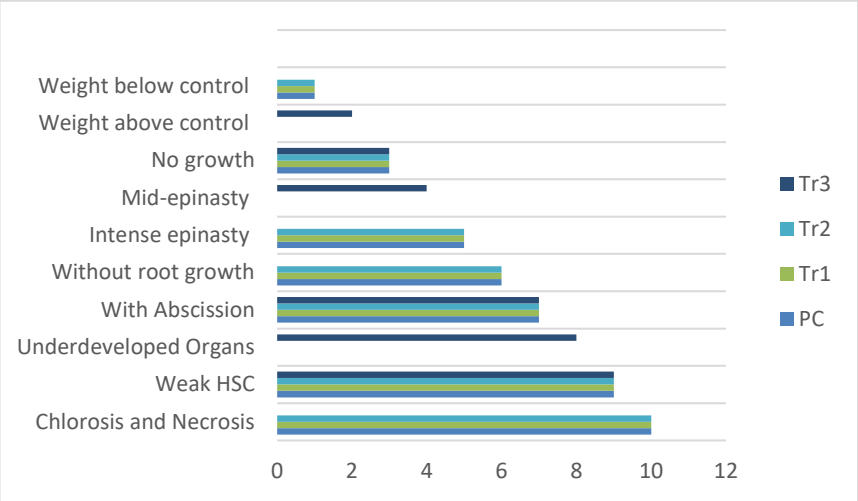


Fig 1. Phyto-Mor phoanatomical Disruption (PMD) sequence key charts adopted from Diogo *et al.* (202

Conclusion

This study successfully evaluated the efficacy of *Moringa oleifera* seed powder (MOSP) as a remediating biosorbent against the phytotoxic stressors of spent engine oil (SEO) on hydroponic *Acalypha wilkesiana* stem cuttings (AWSC). The experimental data confirms that SEO contamination significantly suppresses plant development, however, the application of organic phytoamendments can effectively mitigate these adverse effects. Observations of growth inhibition, atrophy, and morphoanatomical phytotoxicity confirm that SEO is highly toxic to *A. wilkesiana* in specific treatments, particularly the Positive Control and TR 1 through TR 3. These developmental instabilities correlate directly with recorded physiochemical data, where depleted Dissolved Oxygen (DO) and reduced pH values resulted in hypoxic and acidic hydroponic solutions.

The introduction of MOSP facilitated the buffering, flocculation, and sequestration of contaminants within the hydroponic solutions. This positively shifted the pH and DO levels, thereby enhancing the developmental stability and growth of the AWSCs. Conclusively, MOSP functions as a potent bioremediator that neutralizes acidity and aids oxygenation through its inherent cationic capabilities. These findings suggest that MOSP represents a low-cost, sustainable, and eco-friendly solution for the remediation and amelioration of petroleum hydrocarbon-contaminated water systems and soils. Further research into this novel "green-on-green" remediation method is necessary. Expanding the scope of assays (bioassays, molecular assays, concentration assays) to include higher contaminant concentrations and diverse plant species will further contribute to the body of knowledge in environmental biotechnology and remediation.

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