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Phytotoxicity of Salvinia *molesta* in Diesel Exposure

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Abstract

Phytoremediation is one of the methods to remove various types of pollutants from water and soil using plants. *Salvinia molesta*, an aquatic plant, is chosen in this study to determine its ability to degrade diesel as the pollutant in synthetic wastewater with different diesel concentrations (0, 8,700, 17,400, and 26,100 mg/L) for 14 days. Total petroleum hydrocarbon (TPH) has been used as an indicator to represent diesel concentration variation in wastewater. Degradation of TPH was 85.1% for diesel concentration of 8,700 mg/L, compared with only 53.9% in the corresponding control without plant. While, acute toxicity on *S. molesta* exposed in diesel concentrations of 17,400 and 26,100 mg/L was observed and eventually had caused the plants to die after 14 days of exposure. Additionally, throughout the phytotoxicity test, the biomass of *S. molesta* was found to fluctuate confirming inhibition on plant to survive with diesel contaminated water compared with the corresponding control without contaminant. Based on the results obtained it is suggested to decrease diesel concentration less than 8,700 mg/L in future study due to insolubility of diesel in water and the toxicity to the aquatic plants

Keywords: *Degradation, Salvinia molesta, Phytoremediation, Total petroleum hydrocarbon (TPH).*

1. Introduction

Diesel is the main fuel used in the world due to urbanization and industrial development causing insoluble oil-in-water emulsions among their basic contaminants. This problem is of considerable matter because it can cause trouble during conventional wastewater treatment [1]. Phytoremediation is a growing treatment approach that is fast gaining attention and offering efficient and economical clean-up of toxic wastewater contaminated with heavy metals [2], hydrocarbons [3], agricultural wastes [4], dyes [5,6], and chlorinated solvents [7]. Phytotoxicity has been used to test plants' ability to tolerate high concentration of diesel [8]. The approach is also applied to remove pollutants from wastewater with different capability. The use of plants in phytoremediation is cost-effective and ecologically sounds to remove, degrade, assimilate, and metabolize contaminants. Direct degradation, transpiration of volatile organic compounds via the leaves, biochemical transformations within rhizosphere by microbial, and mineralization process at the rhizosphere zone by associated mycorrhizal fungi and microbial consortia [9] are among the mechanisms in phytoremediation for organic contaminants are. According to Vieira et al. [10], microorganisms can degrade hydrocarbons with two conditions aerobic and anaerobic in contaminated soil or underground water [10]. Though, researcher prefers to use aerobic condition because of high efficiency removal in shorter time. However, the use of continuous aeration is considerably more expensive due to high biomass residues that need further treatment and increase energy consumption for aeration system.

In wastewater treatment, aquatic plants were applied to eliminate nutrients, heavy metals, suspended solids, toxic organics from agricultural landfill, urban storm water runoff and acid mine drainage [11]. In this study, an aquatic plant of Salvinia molesta was used. It can be easily found in hot and cold regions, and it is a Malaysian native plant. Besides having a rapid growth rate, this aquatic fern species also has the capability to accumulate pollutants. It is a free-floating fern; stems rootless, hairy, have horizontal stems that can be float with water surface, and grow at each node, a pair of floating or emergent leaves [12] with its growth is rapid and irregularly [13]. It can spread rapidly and prolifically into a monoculture which can shade out underwater natives, leaving large bare bottom areas [14]. Dhir et al. [15] demonstrated that natant Salvinia was able to accumulate high concentrations of chromium (Cr) from wastewater. The purpose of conducting this research is to identify the ability of S. molesta to survive in different concentrations of diesel-contaminated water and determine its efficiency to degrade diesel.

2. Materials and Method

2.1 Phytotoxicity Test Design

The investigation was performed under a greenhouse conditions in Universiti Kebangsaan Malaysia. In this study, 13 aquariums made of glasses were used for this phytotoxicity test to avoid oils sticking to the walls. The length, width and depth for aquarium were 30 cm each. It was filled with 15 L of synthetic wastewater containing diesel. Three replicates of aquarium (R1, R2, and R3) for each concentration were prepared and another three aquariums without plant as control contaminant (CC) as shown in Fig. 1. Only one aquarium is act as plant control (PC) (without diesel contaminant). Nine healthy aquatic plants of S. molesta (Fig. 2) at three weeks old were placed in each aquarium filled up with 15 L of water mixed with diesel available from a regional petrol station as synthetic wastewater with different concentrations (0, 8,700, 17,400, and 26,100 mg/L). The investigation was run in an open natural lab.

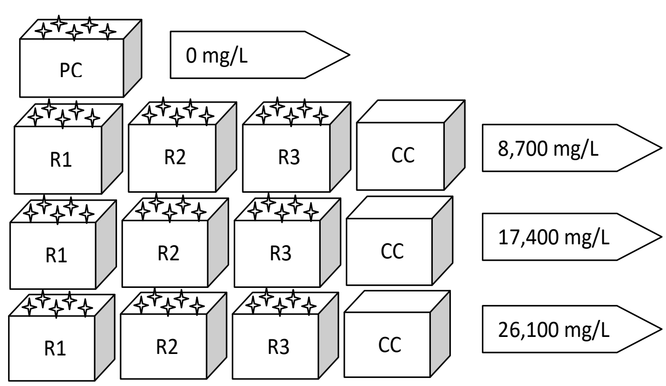

Fig. 1. Design of phytotoxicity test at different diesel concentrations.

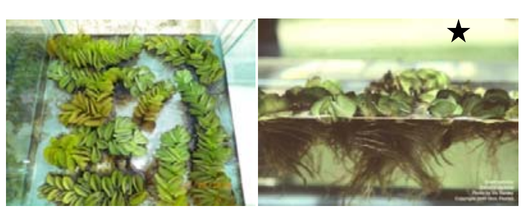

Fig. 2. The aquatic plant of Salvinia molesta [*13].

2.2 Properties of Water Quality

The phytotoxicity test was conducted throughout 14 days of lab work with samplings taken on day 0, 3, 7 and 14. The physicochemical variables of pH, temperature, T (˚C), dissolved oxygen, DO (mg/L), and Oxidation Reduction Potential, ORP (mV) in solution were monitored by a IQ150 multi-probe (I.Q Scientific Instruments, U.K.) for temperature, ORP and pH measurement, and GLI International dissolved oxygen sensor (Model 63, U.S.A) for DO.

2.3 TPH Detection in Water

Water samples of 100 mL each from growth medium were taken regularly in clean bottle from each aquarium on tasting time for all phyto-treatment to analyse diesel as total petroleum hydrocarbon (TPH). Liquid–liquid extraction method, and gas chromatography were used to determine TPH concentration in the water [16] according to procedure by the Environmental Protection Agency, EPA Method 8041 [17]. Dichloromethane (Merch, Germany) was utilized as a solvent. In 1 L separatory funnel, 100 mL (wastewater sample) and 25 mL dichloromethane (as solvent) were shaken for 2 min. The upper layer of water was removed and the bottom layer was taken whereby the water from the bottom layer was further removed using sodium sulphate. The residual water was evaporated for three to four days under an overhead fume hood. The remaining was then full up to 2 mL of GC vial. TPH concentration in samples were analyzed by GC–FID (Agilent Technologies, Model 7890A, GC System, U.K.). Capillary column gas chromatography was used for this purpose. The TPH degradation on each sampling day was calculated by Equation (1) [8]:

$$\text{TPH degradation}\,(\%) = \frac{TPH_0 - TPH_{SD}}{TPH_0} \times 100 \quad \ldots(1)$$

with, TPH0 is the total petroleum hydrocarbon on sampling day 0 and TPHSD is the total petroleum hydrocarbon on each sampling day.

2.4 Plant Sampling and Growth Measurement

The progress of S. molesta with various diesel concentrations (0, 8,700, 17,400, and 26,100 mg/L) was monitored during 14 days. On each sampling day (0, 3, 7, and 14), one plant was collected from each aquarium. S. molesta was washed wholly with water and then dried using cloth to determine its wet weight. Finally, the dry weight of plant was obtained by drying it at 70°C for 72 h in an oven (Memmert, Germany) [18].

2.5 Statistical Analysis

All results of removal efficiency and plant biomass were directed to the analysis of variance (ANOVA), using the GLM procedure in software SPSS version 16 (IBM, USA). Differences between means were tested using the Duncan method. All statistical analysis tests were performed using a significance level of 5% [19].

3. Results and Discussion

3.1 Observing Physical Parameters

As depicted in Fig. 3, physical parameters during the phytotoxicity test were determined for 0, 8,700, 17,400, and 26,100 mg/L diesel concentrations to determine the conditions of experimental work. In general, the results throughout the 14-day exposure show that the temperature and pH along the exposure period varied between 26.2-29.2°C and 5.3-7.2, respectively. In temperate climate, temperature (20–30°C) is suitable for biodegradation of mineral oil hydrocarbons [20]. PH in aquarium with diesel is acidic differ from plant control (0 mg/L) which in range of neutral among the treatments due to microbial utilisation of straight-chain hydrocarbons [8]. According to Ong et al. (2010), DO and ORP measurement throughout phytotoxicity test it is indicator of aerobic or anaerobic condition of treatment [21]. The DO range decreases from 6.22 to 1.58 mg/L, and ORP increased throughout 14 days from -105.4 to 105.5 mV. There were decreases in dissolved oxygen as shown in Fig. 3 but the environment of treatment was within aerobic and anoxic conditions. Faulwetter et al. [22] stated that aerobic processes were created at high ORP while lower ORP promote anaerobic processes. In addition ORP values were indicated the media of treatment in the aerobic and anoxic range.

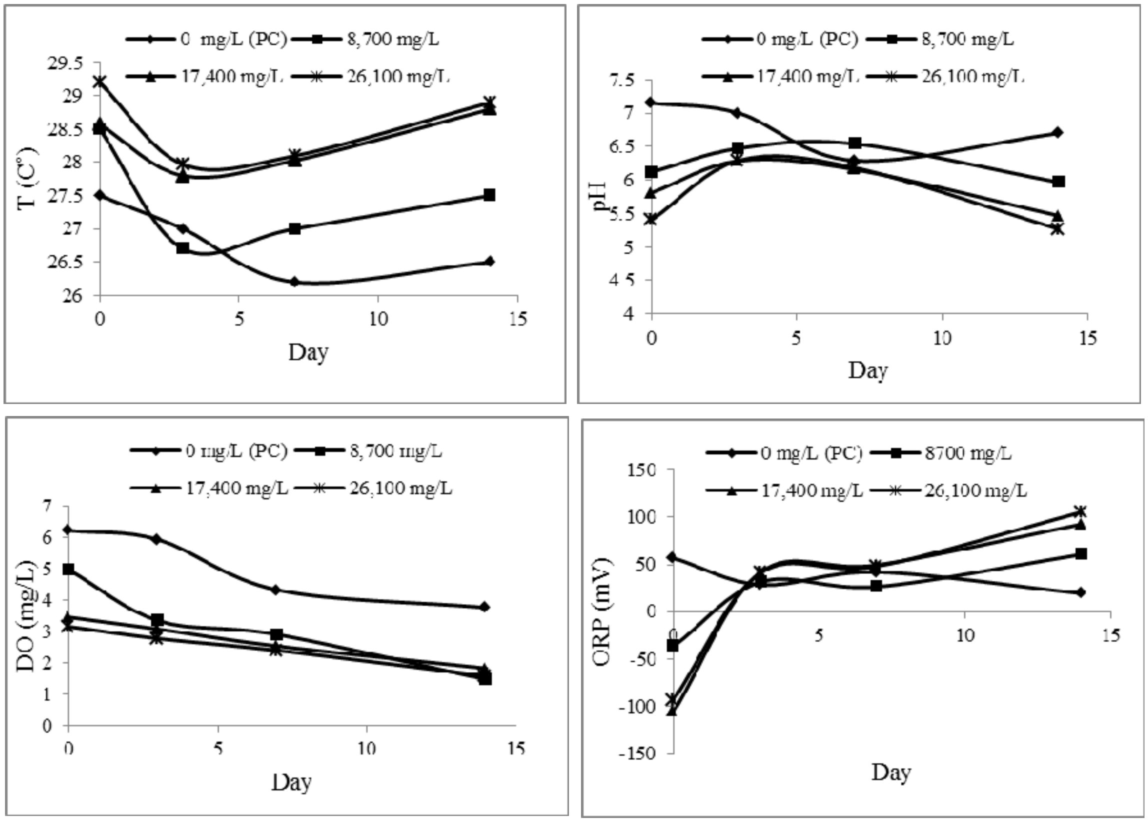

Fig. 3. Physical parameters throughout 14-day phytotoxicity test to diesel exposure

3.2 Plant Growth Throughout Phytotoxicity Study

Throughout the 14-day test, plant growth parameters were recorded as shown in Fig. 4. Generally, aquatic plant of S. molesta was inhibited when exposed to diesel contaminant compared to the corresponding control without contaminant. For diesel concentrations of 8,700 mg/L after 3 days of exposure, the plants were gradually withered and inhibited in their growth until day 14. The biomass of aquatic plant of S. molesta decreased until it died at the end of 14 days. The plants were inhibited directly after exposure where all plants died after 3 days in the higher diesel concentrations (17,400 and 26,100 mg/L). The inefficiency and death of the aquatic plant S. molesta was due to the high toxicity and water-diesel insolubility. Therefore, the floating layer of diesel was attached to S. molesta floating leaves and emergent roots and caused the death of plants.

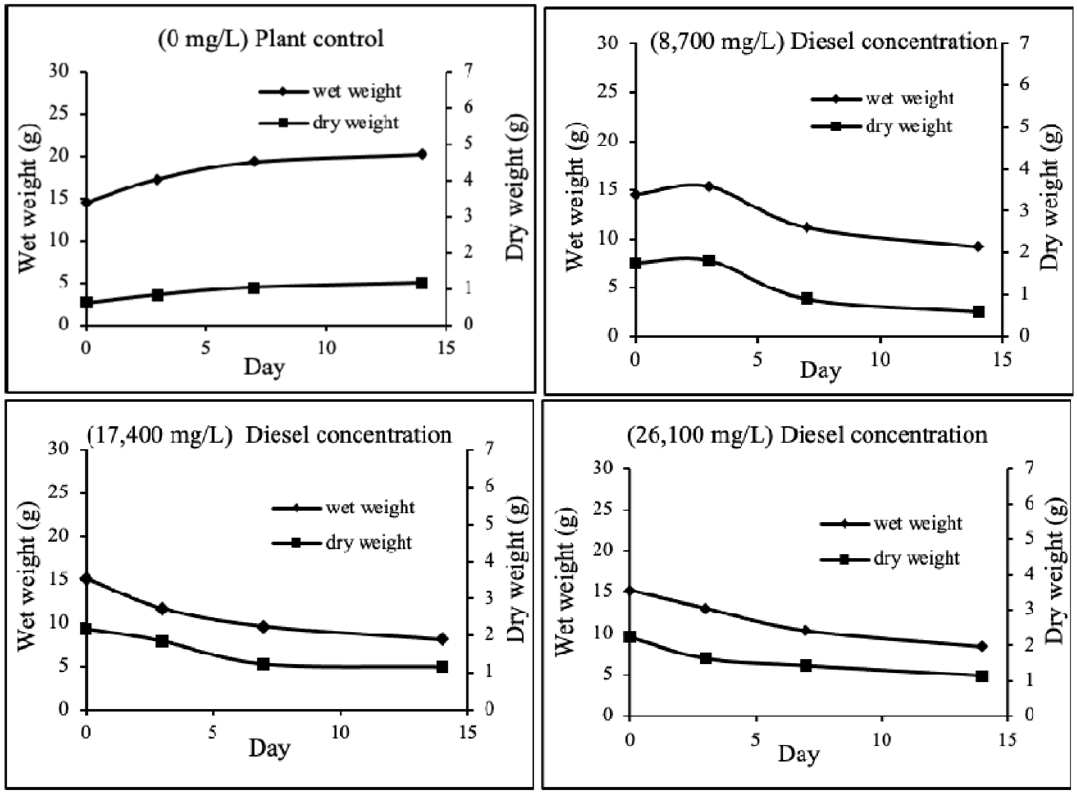

Fig. 4. Growth parameters of Salvinia molesta with different diesel contamination

3.3 TPH Removal

The GC results of TPH concentrations under different treatments of diesel contaminant (8,700, 17,400 and 26,100 mg/L) with plants and without plants (CC) throughout the 14-day treatment time are demonstrated in Fig. 5. For all concentrations, there were obvious decreases of TPH concentration, due to several reasons: 1) phytoremediation; 2) bioremediation; 3) absorption of diesel by plant leaves and roots; and 4) evaporation of hydrocarbons [20; 23; 24]. The percentage of TPH removal for different treatments of diesel contaminated water (8,700, 17,400 and 26,100 mg/L) with vegetated and the non-vegetated ones (CC) within 14-day treatment time are represented in Fig. 6.

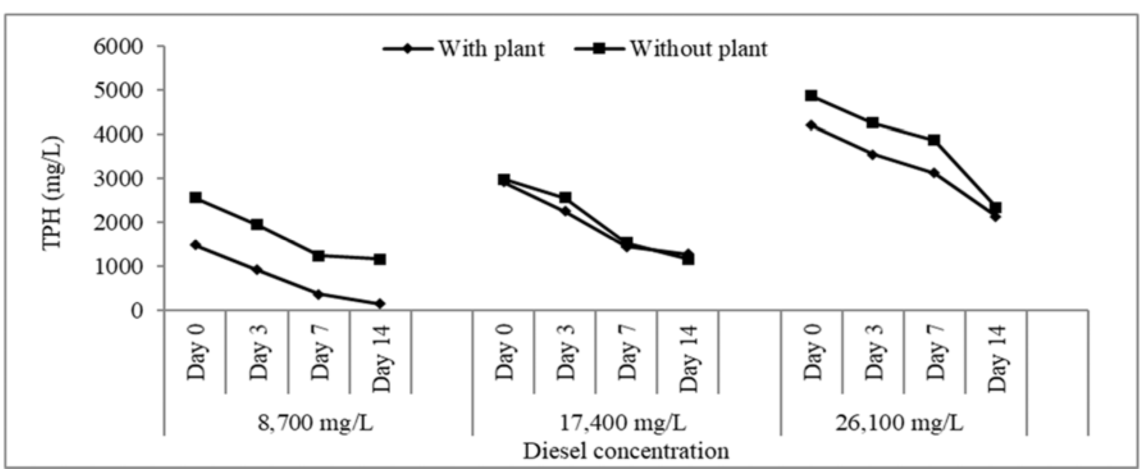

Fig. 5. TPH concentration throughout 14 day of diesel exposure.

The TPH degradation in different treatments was up to 85.1-48.1% while the degradation in the controls was only 49.7–57.8%. In this study, for concentration of 8,700 mg/L, at the end of 14 days the removal efficiency was 85.1% even though the S. molesta was inhibited after three days of exposure as illustrated in Fig. 4. Cohen et al. (2002), tested and found that S. molesta died with 1% (≈8,700 mg/L) diesel concentration similar results to our findings for 8,700 mg/L [25]. The results from the statistical analysis showed significant different between treatments with and without plants and also within days for 8,700 mg/L diesel concentration as visible in Fig. 6. S. molesta was inhibited directly after exposure in 17,400 and 26,100 mg/L and at the end of 14 days, all plants died. Cohen et al. [25], observed that when Azolla pinnata, Pistia stratiotes and S. molesta were floated in pots in site experiment containing soil contaminated at the surface with diesel fuel (2.4 Lm-2) and flooded with water, the plants were affected fast and led to plant die, thereafter bacterial flocks grow around the dead aquatic plants which enhanced the diesel degradation. According to Peng et al. (2009), the TPH can be degraded either by physical, chemical or biological action, while in control contaminant without plants; the degradation is usually due to volatilization, eluviation or photolysis [26]. So for this case, the action mentioned by Peng et al. (2009), worked together and we got the percentage of degradation of 54.9 and 48.1%, and 52.4 and 47.7% for treatment with diesel concentrations of 17,400 and 26,100 mg/L and its related control contaminant with the absence of plant respectively Fig. 6. Statistically analysis between treatments with and without plants for 17,400 and 26,100 mg/L diesel concentrations showed no significant difference between them due to the inhibition of the plant after direct exposure [27]. Vieira et al. [28] verified that the microorganisms were capable to remove TPH at 78 to 83% (initial concentration was 11,000 mg/L) from diesel oil and gasoline fuel polluted effluent after 15 days of treatment.

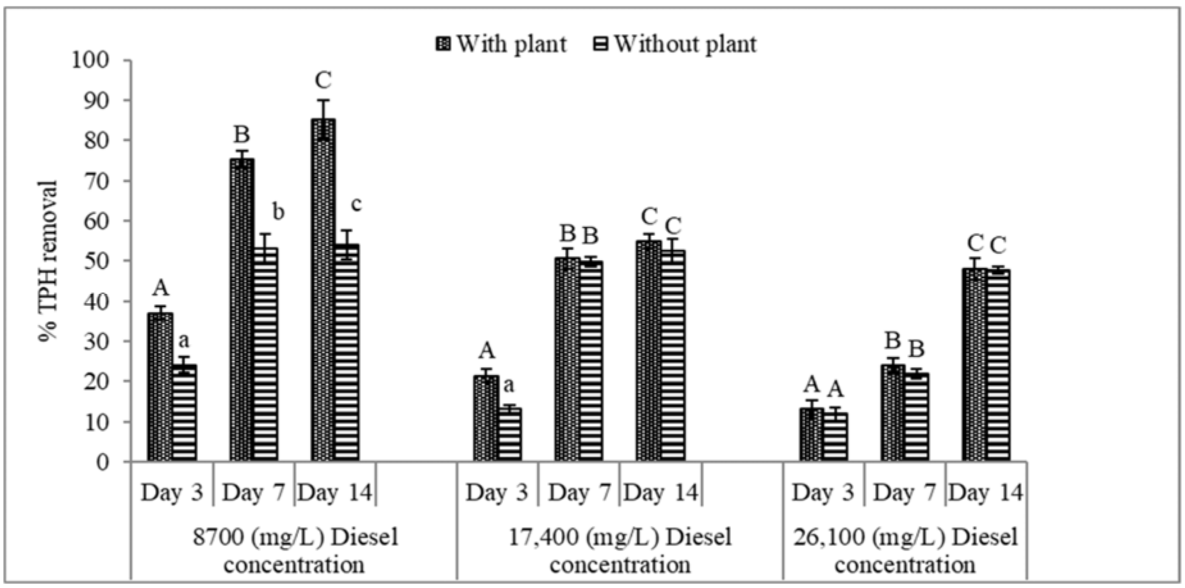

Fig. 6. TPH degradation by *Salvinia molesta* with: (a) 8,700 mg/L diesel (b) 17,400 mg/L diesel, and (c) 26,100 mg/L diesel. A, a: means significant different between treatments (with and without plants) at $p < 0.05$; AA: not significant different between treatments (with and without plants) $p > 0.05$

4. Conclusions

The results from phytotoxicity test with aquatic plant of Salvinia molesta has demonstrated the inhibition of S. molesta when exposed to water contaminated with diesel at concentrations of 8,700, 17,400, and 26,100 mg/L. The high removal efficacy was 85.1% for diesel concentration of 8,700 mg/L, due to physical, chemical, biological, volatilization, eluviations, and photolysis processes. Based on the results obtained it is suggested to decrease diesel concentration less than 8,700 mg/L in future study due to insolubility of diesel in water and the toxicity to the aquatic plants.

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5. References

[1] M. F., Imron, S. B., Kurniawan, H. S., Titah, “Potential of bacteria isolated from diesel-contaminated seawater in diesel biodegradation”, *Environ. Technol. Innov,* vol.14, pp. 100368, 2019.

[2] N.I., Ismail, S.R.S., Abdullah, M., Idris, H.A., Hasan, M.I.E., Halmi, N.H., AL Sbani, O.H., Jehawi, S.N.A., Sanusi, M.H. Hashim, “Accumulation of Fe-Al by *Scirpus grossus* grown in synthetic bauxite mining wastewater and identification of resistant rhizobacteria”, *Environ. Eng. Sci.*, vol. 34, pp. 367-375, 2017.

[3] N.H., AL Sbani, S.R.A., Abdullah, M., Idris, H.A., Hasan, I.A., Al-Baldawi, O.H., Jehawi, N.I. Ismail.,” Remediation of PAHs-contaminated water and sand by tropical plant (Eleocharis ochrostachys) through sub-surface flow system”, Environ. Technol. Inno., vol. 20, pp. 101044, 2020.

[4] L., Ivorra, P.G., Cardoso, S.K., Chan, C., Cruzeiro, K.A., Tagulao, “Can mangroves work as an effective phytoremediation tool for pesticide contamination? An interlinked analysis between surface water, sediments and biota”, J. Clean. Prod., vol. 295, pp. 126334, 2021.

[5] I.A., Al-Baldawi, S.R.S., Abdullah, A.F., Almansoory, H.A., Hasan, N.; Anuar, “Role of Salvinia molesta in biodecolorization of methyl orange dye from water”, Sci. Rep., vol. 10, pp. 1-9, 2020.

[6] E., Abdulqader, S.R.S., Abdullah, H.A., Hasan, N.I., Ismail, R.A.A., Rahim, M., Idris, “Plant-assisted remediation of wastewater contaminated with methyl orange using Scirpus grossus”, J. Environ. Biol., vol. 40, pp. 515-523, 2019.

[7] S. B., Kurniawan, A., Ahmad, N. S. M., Said, M. F., Imron, S. R. S., Abdullah, A. R., Othman, I. F., Purwanti, H. A., Hasan, “ Macrophytes as wastewater treatment agents: Nutrient uptake and potential of produced biomass utilization toward circular economy initiatives”, Sci. Total Environ., vol. 790, pp. 148219, 2021.

[8] I.A., Al-Baldawi, “Removal of 1,2-Dichloroethane from real industrial wastewater using a subsurface batch system with Typha angustifolia L”, Ecotoxicol. Environ. Saf., vol. 147, pp. 260–265, 2020.

[9] S.R.S., Abdullah, I.A., Al-Baldawi, A.F., Almansoory, I.F., Purwanti, N.H., Al-Sbani, S.S.N., haruddin, “Plant-assisted remediation of hydrocarbonsin water and soil: application, mechanisms, challenges and opportunities”, Chemosphere., vol. 247, pp. 125932, 2020.

[10] P.A., Vieira, R.B., Vieira, S., Faria, E.J., Ribeiro, V.L., Cardoso, “Biodegradation of diesel oil and gasoline contaminated effluent employing intermittent aeration”, J. Hazard. Mater., vol. 168, pp. 366-1372, 2009.

[11] A., Lohi, M., Cuenca, G., Anania, R., Upreti, L. Wan, “Biodegradation of diesel fuel-contaminated wastewater using a three-phase fluidized bed reactor”, J. Hazard. Mater., vol. 154, pp. 105-111, 2008.

[12] Prasad, M.N.V.: Aquatic Plants for Phytotechnology (2008), http://sumarsih07.files.wordpress.com/2008/09/aquatic-plant.pdf

[13] Jacono, C.C., Salvinia molesta (2003), http://salvinia.er.usgs.gov/html/identification.html

[14] Richard, A., Ramey, V.: University of Florida, UF/IFAS Center for Aquatic and Invasive Plants (2007). http://plants.ifas.ufl.edu/node/396

[15] B., Dhir, P., Sharmila, P., Saradhi, S., Abdul Nasim, “Physiological and antioxidant responsesof Salvinia natans exposed to chromium-rich wastewater”, Ecotoxicol. Environ. Saf., vol. 72, pp. 1790-1797, 2009.

[16] S.R.N., Alanbary, S.R., Sheikh Abdullah, I.A., Al-Baldawi, H., Abu Hassan, N., Anuar, A.R., Othman, F., Suja, “ Phytotoxicity of contaminated sand containing crude oil sludge on Ludwigia octovalvis”, J. Ecol. Eng., vol. 20, pp. 246-255, 2019.

[17] EPA Method 8041, http://www.caslab.com/EPA-Method-8041

[18] A.F., Almansoory, M, Idris, SR.S., Abdullah, N., Anuar, S.B., Kurniawan, “Response and capability of Scirpus mucronatus (L.) in

phytotreating petrol-contaminated soil", Chemosphere, vol. 265, pp. 128760, 2020.

[19] M.A., Norhan, S.R.S., Abdullah, H.A., Hasan, N.I., Ismail, "A constructed wetland system for bio-polishing palm oil mill effluent and its future research opportunities". J. Water Process Eng., vol. 41, pp. 102043, 2021.

[20] H., Chan, "Biodegradation of petroleum oil achieved by bacteria and nematodes in contaminated water", Sep. Purif. Technol., vol. 80, pp. 459-466, 2011.

[21] S., Ong, K., Uchiyama, D., Inadama, Y., Ishida, K., Yamagiwa, "Treatment of azo dye Acid Orange 7 containing wastewater using up-flow constructed wetland with and without supplementary aeration", Bioresour. Technol., vol. 101, pp. 9049-9057, 2010.

[22] J. L., Faulwetter, V., Gagnon, C., Sundberg, F., Chazarenc, M. D., Burr, J., Brisson, A. K., Camper, O. R., Stein, "Microbial processes influencing performance of treatment wetlands: A review", Ecol. Eng., vol. 35, pp. 987-1004, 2009.

[23] R.A., Wuana1, F.E., Okieimen, "Phytoremediation Potential of Maize (Zea mays L.) A Review", Afr J. Agr., vol. 6, pp. 275-287, 2010.

[24] E., Kaimi, T., Mukaidani, M., Tamaki, "Effect of Rhizodegradation in Diesel-contaminated soil under Different Soil Conditions", Plant Prod. Sci., vol. 10, pp. 105-111, 2007.

[25] MF., Cohen, J., Williams, H., Yamasaki, "Biodegradation of diesel fuel by an Azolla derived bacterial consortium", J. Environ. Sci. Health., vol. 37, pp. 593-606, 2002.

[26] S., Peng, Q., Zhou, Z., Cai, Z., Zhang, "Phytoremediation of petroleum contaminated soils by Mirabilis Jalapa L. in a greenhouse plot experiment", J. Hazard. Mater., vol. 168, pp. 1490-1496, 2009.

[27] I.A., Al-Baldawi, F., Suja, S.R., Sheikh Abdullah, M., Idris, "Preliminary Test of Hydrocarbon Exposure on Salvinia molesta in Phytoremediation Process", Revelation and Science, vol. 01, pp. 52-56, 2011.

[28] P.A., Vieira, R.B., Vieira, F.P., de Franca, V.L, Cardoso, "Biodegradation of effluent contaminated with diesel fuel and gasoline", J. Hazard. Mater., vol. 140, pp. 52-59, 2007.

السمية النباتية لسالفينيا موليستا في التعرض للديزل

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الخلاصة

تعد المعالجة النباتية إحدى طرق إزالة أنواع مختلفة من الملوثات من المياه والتربة باستخدام النباتات. تم اختيار سالفينيا مولستا ، وهو نبات مائي ، في هذه الدراسة لتحديد قدرته على تحلل الديزل باعتباره الملوث في مياه الصرف الصناعي بتركيزات مختلفة من الديزل (٠ ، ٨٧٠٠ ، ١٧٤٠٠ ، ٢٦١٠٠ مجم / لتر) لمدة ١٤ يومًا. تم استخدام إجمالي الهيدروكربونات البترولية (TPH) كمؤشر لتمثيل اختلاف تركيز الديزل في مياه الصرف الصحي. كان تدهور TPH 85.1٪ لتركيز الديزل البالغ ٨٧٠٠ ملجم / لتر ، مقارنة مع ٥٣,٩٪ فقط في الشاهد المقابل بدون نبات. بينما تسببت تركيزات الديزل البالغة ١٧,٤٠٠ و ٢٦,١٠٠ ملغم / لتر في حدوث سمية حادة في S. molesta وتسببت في النهاية في موت النباتات بعد ١٤ يومًا من التعرض. بالإضافة إلى ذلك ، خلال اختبار السمية النباتية ، وجد أن الكتلة الحيوية لـ S. molesta تتأرجح لتأكيد تثبيط النبات للبقاء على قيد الحياة مع المياه الملوثة بالديزل مقارنةً بالاحواض التي بدون ملوث فقط نبات مع الماء.