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Comparative Assessment of Three Fungal Genus in Mycoremediation of Spent Engine Oil: A Brief Review

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ABSTRACT

Spent engine oil is composed of various aliphatic hydrocarbons, aromatic hydrocarbons, lubricative additives, and traces of heavy metal. Improper disposal of spent engine oil can lead to deleterious effects on humans due to spent engine oil properties, which can exert toxicity, mutagenicity, and carcinogenicity on cells and organs. The conventional method to remove hydrocarbon in the spent engine oil is not only expensive but unable to degrade the hydrocarbon completely. In comparison, the mycoremediation approach has been reported to be environmentally friendly, efficient, and cost-effective. The main objective of this review article is to identify the fungal isolate which is most efficient to degrade spent engine oil by assessing the biomass production and the percentage of spent engine oil degraded. Based on the comparative information obtained, *Mucor* sp. showed the highest biomass production in the presence of spent engine oil. *Trichoderma* sp. and *Aspergillus niger* were found to have average biomass production and it depending on the strain and incubation period. Both *A. flavus* and *A. nidulans* were found to have the lowest biomass production. In terms of spent engine oil degradation, *Mucor* sp, *Trichoderma* sp. and *A. niger* showed >55% degradation as compared to *A. flavus* and *A. nidulans* which have less than 50% degradation. Therefore, from the results of the study, it can be concluded that *Mucor* sp. has the best potential to degrade spent engine oil within a short period based on the high biomass production and percentage of degradation. The comparative data also suggest that by selecting the right strain and right incubation period, the percentage of spent engine oil degradation by using *Trichoderma* sp. and *A. niger* could also increase.

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1 Introduction

Engine oil or more correctly, 'engine lubricant' is widely applied in the engine to reduce the frictional force and keep the metals fresh. After the engine oil had been exposed to extremely high temperature and mechanical pressure, it will be referred to as waste oil or spent engine oil due to physical and chemical impurities being formed (Boichenko et al. 2021). Spent engine oil is a mixture of hydrocarbon molecules and organic compounds with some organometallic constituents and encompassed various aliphatic chains and aromatic hydrocarbons (Ossai et al. 2020). According to Singh and Haritash (2019), the hydrocarbon contaminants such as polycyclic aromatic hydrocarbon (PAHs) and metals are hazardous due to their deleterious effect including toxicity, mutagenicity, and carcinogenicity that concerns the public. Furthermore, spent engine oil products can also exert adverse effects on other living organisms (ATSDR 1995; Adeleye et al. 2018), and severely affect agriculture by decreasing the yield of crops (Ismail et al. 2021).

Previous findings by Al-Hawash et al. (2019) have shown that fungal species were capable of reducing a wide range of pollutants as the fungal mycelia structure was able to produce non-specific enzymes and acids to solubilize the insoluble substrates. A recent study showed fungal secreting extracellular enzymes and acids through their mycelia which can effectively degrade the hydrocarbon molecules (Adekunle et al. 2015). However, the main key to successful mycoremediation is to employ the right fungal species in the removal of specific pollutants (Sing, 2006) but this information is scarcely documented. Albeit various hydrocarbon degrading fungal species have been identified, the search for an ideal species for successful mycoremediation of spent engine oil remains challenging due to the complexity of hydrocarbon components found in the spent engine oil (Stanley and Immanuel 2015).

Environmental quality regulations in Malaysia set a maximum content of oil sewage and industrial effluent discharge limit to 10 mg/L (Environmental Quality (Industrial Effluents) Regulations, 2009). Despite the regulation, 121 oil spillage cases were reported from 2009 to 2016 in Malaysia (Fong 2016) and some cases were left unreported due to the occurrence in smaller areas. Industrial manufacturers, disregarding the negative environmental impact, are still disposing of spent engine oil in open sources as a shortcut option to reduce their downstream costs by not processing the hydrocarbon waste. Therefore, there is an urgent need for remediation to reduce the spent engine oil contaminant found in the environment. Hence, this study focuses on the potential of fungal species on hydrocarbon degradation by reviewing their tolerance towards spent engine oil based on the fungal biomass and the percentage reduction of spent engine oil in the growth medium.

Various previous studies have identified five potential fungal species namely, *Mucor* sp., *Aspergillus flavus, A. niger, A. nidulans*, and *Trichoderma* sp. which have a wider potential for mycoremediation. These fungal species were inoculated on the different growth selective mediums spiked with spent engine oil and the fungal biomass production and the percentage reduction of spent engine oil were recorded by previous researchers.

2 Biomass Production

Based on the previous studies' conclusion (Table 1), each of the tested fungal species possessed different tolerance toward spent engine oil, which influences the biomass production of the fungal species on the culture medium containing spent engine oil. Among the five fungal species, *Mucor* sp was observed to attain the highest biomass production within a comparatively short incubation period of 7 days. This is followed by the *Trichoderma* sp. and *A. niger* these were able to achieve moderate to high

Table 1 The growth of different fungal species cultivated on different growth mediums supplemented with spent engine oil

Species	Fungal biomass production	Composition of the growth medium	Incubation period at room temperature	Reference
Mucor sp.	High	2 mL of spent engine oil + 40 mL of BHB	7 days	Ong et al. 2018
A. niger	Moderate	10 mL of spent engine oil + 40 mL of BHB	7 days	Ong et al. 2018
	High 5^{th} day/OD: 8.000 40^{th}day/OD: 5.250	1 mL of spent engine oil + 100 mL of mineral salt	21 days	Ahmad et al. 2015
	High	2 mL spent engine oil + 10 mL of MSB	40 days	Adekunle and Adebambo 2007
A. nidulans	Moderate	10 mL of spent engine oil + 40 mL of BHB	7 days	Ong et al. 2018
A. flavus	Moderate	10 mL of spent engine oil + 40 mL of BHB	7 days	Ong et al. 2018
	Moderate	1 mL of spent engine oil + 100 mL of mineral salt	21 days	Ahmad et al. 2015
Trichoderma sp.	High	10 mL of spent engine oil + 40 mL of BHB	7 days	Ong et al. 2018
	Moderate- High (+26% to +75%)	BHA complemented with 1% (v/v) of used engine oil	7 days	Daccò et al. 2020

BHB: Bushnell Hass Broth; BHA: Bushnell Hass Agar; MSB: Minimal salt broth biomass production but needed a relatively long incubation period. Furthermore, both *A. nidulans* and *A. flavus* were able to achieve moderate biomass production but with a longer incubation period relatively. Hence, the sequential order of fungal species tolerance towards spent engine oil based on their observed biomass production is *A. flavus*≈*A. nidulans*<*A. niger*<*Trichoderma* sp. <*Mucor* sp.

Various *Mucor* species can achieve high biomass in BHB containing spent engine oil. According to Marchut-Mikolajczyk et al. (2015), *M. circinelloides* secretes various metabolites that emulsify hydrophobic hydrocarbons and increase hydrocarbon interaction with degradative enzymes such as lipase and alcohol dehydrogenase (ADH) (Durón-Castellanos et al. 2005) and metabolizes the hydrocarbons available in spent engine oil to be utilized for cell biomass production (Balaji et al., 2014; Chimezie Dirisu, 2015).

Biomass production by *Trichoderma* sp. ranged from moderate to high when cultured on the medium containing spent engine oil as a sole carbon source (Daccò et al. 2020). This suggested that *Trichoderma* sp. has moderate to high tolerance toward spent engine oil and can consume spent engine oil as a carbon source to grow (Thenmozhi et al. 2013). Various species in this genus including *T. harzianum*, *T. pseudokoningii*, and *T. viride* (Ravelet et al. 2000; Saraswathy and Hallberg 2002) were reported to degrade pyrene, a recalcitrant hydrocarbon found in spent engine oil, as a carbon source. *Trichoderma* sp. possesses an *N*-acetylation detoxification pathway that enables this fungal species to degrade recalcitrant aromatic hydrocarbons structures to support growth (Cocaign et al. 2013; Kupareva et al. 2013; Zafra et al. 2015). Because of this, *Trichoderma* sp. is the most common fungal species that are isolated from the soil contaminated by petroleum (Makut et al. 2014).

A. niger was observed to have moderate biomass production after 7 days of incubation (Ong et al. 2018), but was able to achieve higher biomass production if the incubation time is extended (Ahmad et al. 2015). Adekunle and Adebambo (2007), also reported a fluctuating growth pattern of *A. niger* for 40 days before reaching the maximum growth peak, which was attributed to the different ratios of spent engine oil added in the experiment. Despite the inconsistent growth pattern observed, *A. niger* can secrete lipase, catalase, and lignin peroxidase to degrade hydrocarbon to support growth (Vatsyayan and Goswami 2016).

At present, information on the biomass production by *A. nidulans* on spent engine oil supplemented media is in scantly. Ong et al. (2018) reported moderate *A. nidulans* biomass production after 7 days of incubation in BHB containing spent engine oil and this might be due to the shorter incubation period. Similarly, moderate biomass production was reported when *A. flavus* was inoculated on the medium spike with hydrocarbons (Ahmad et al. 2015; Ong et al. 2018).

2.1 Reduction of Spent Engine Oil

Table 2 showed all the reported fungal species which were capable to degrade and utilize spent engine oil as the sole carbon source of growth (Thenmozhi et al. 2013). The percentages of spent engine oil being degraded varied according to the present of fungal species, different lengths of the incubation time, type of medium, and the extracellular enzyme secreted by each of the fungal species. As per table 2, the sequential order of the fungal species' effectiveness in reducing spent engine oil in ascending order is *A. nidulans*< *A. flavus* <*Trichoderma* sp. ≈*A. niger*≈*Mucor* sp.

According to Szewczyk and Długoński (2009), *Mucor* sp. can achieve up to 55% reduction in spent engine oil. The species *M. circinelloides* was reported to produce an extracellular emulsifier to emulsify hydrocarbons to increase the bioavailability of hydrocarbons to fungal's degradative enzymes (Marchut-Mikolajczyk et al. 2015). Balaji et al. (2014) reported that *M. racemosus* expresses a relatively higher concentration of lipase enzyme in spent engine oil, compared to other fungal species. These species were also able to produce organic acids that metabolize spent engine oil effectively and then utilize the spent engine oil as a sole carbon source to support growth (Thenmozhi et al. 2013; Paper and Nwinyi 2019). These circumstantial shreds of evidence suggested that *Mucor* sp. is consistent in degrading a high percentage of spent engine oil.

In the case of *Trichoderma* sp, Elshafie et al. (2020) reported that *T. harzianum* strain T22 can degrade a high percentage of spent engine oil, while the finding of Ong et al. (2018) was contradictory and these researchers reported a relatively low percentage of spent engine oil being degraded by *T. harzianum*. This indicated the percentage of spent engine oil degraded by *Trichoderma* sp is highly dependent on the strain and the incubation period. Various species of this genus including *T. hamatum*, *T. harzianum*, *T. koningii*, *T. viride*, *T. virens*, and *T. asperellum* have shown to degrade low-molecular-weight PAHs such as naphthalene and phenanthrene, or more complex PAHs such as anthracenebenzo[a]anthracene, benzo[a]fluoranthene, benzo[a]pyrene, and chrysene (Cerniglia and Sutherland 2010, Lieckfeldt et al. 1999, Zafra et al. 2015). Probable mechanisms for PAHs degradation are hypothesized for *Trichoderma*, including the production of laccases (Cazares-Garcia et al. 2013), peroxidases (Per) (Cristica et al. 2011), and dioxygenases (Hadibarata et al. 2007). This was confirmed by Zafra et al. (2015) and Balcázar-López et al. (2016) for *T. atroviride* who reported that laccase production can cleave the aromatic ring of PAHs, which resulted in the degradation of PAHs compound present in spent engine oil. Even though Table 1 illustrates a moderate to high biomass

Table 2 *In vitro* potential of various fungal species in reducing spent engine oil

Species	Isolated	Reduction (%) of spent engine oil	Composition of medium	Incubation period	References
Mucor sp.	*M. ramosissimus* IM 6203 from Poland	55.5%	5 % of spent engine oil in synthetic medium	10 days	Szewczyk and Długoński 2009
	Ota, Ogun State, Nigeria	OD increased from 0.715 to 1.978	2 mL of spent engine oil + 30 mL of MS	12 days at room temperature	Paper and Nwinyi 2019
A. niger	Shah Alam, Selangor, Malaysia	15.85%	10 mL of spent engine oil + 40 mL of BHB	7 days at room temperature	Ong et al. 2018
	Sokoto Metropolis, Nigeria	61.80%	1 mL of spent engine oil + 100 mL of mineral salt	20 days at room temperature	Ahmad et al. 2015
	Ota, Ogun State, Nigeria	OD increased from 0.292 to 1.731.	2 mL of spent engine oil + 30 mL of MS	12 days at room temperature	Paper and Nwinyi 2019
	Pudukkottai, Tamilnadu, South India	40.5%	Czapeck dox broth with 1 % v/v used motor oil	30 days at room temperature	Thenmozhi et al. 2013
A. nidulans	Shah Alam, Selangor, Malaysia	17.75%	10 mL of spent engine oil + 40 mL of BHB	7 days at room temperature	Ong et al. 2018
A. flavus	Shah Alam, Selangor, Malaysia	11.80%	10 mL of spent engine oil + 40 mL of BHB	7 days at room temperature	Ong et al. 2018
	Sokoto Metropolis, Nigeria	44.60%	1 mL of spent engine oil + 100 mL of mineral salt	20 days at room temperature	Ahmad et al. 2015
	Ota, Ogun State, Nigeria	OD increased from 0.213 to 0.617	2 mL of spent engine oil + 30 mL of MS	12 days at room temperature	Paper and Nwinyi 2019
Trichoderma sp.	*T. harzianum* strain T22 (*Th*-T22)	70.16%	5 mL of BHB and 1% (v/v) of used engine oil	45 days	Elshafie et al. 2020
	Shah Alam, Selangor, Malaysia	15.02%	10 mL of spent engine oil + 40 mL of BHB	7 days at room temperature	Ong et al. 2018

BHB: Bushnell Hass Broth; MSB: Minimal salt broth

production by *Trichoderma* sp., but the percentage of spent engine oil degraded is not so consistent compared to *Mucor* sp, but relatively better than *A. nidulans* and *A. flavus*.

Results of the previous studies suggested that *A. niger* can degrade a mere 15.85% spent engine oil after 7 days of incubation (Ong et al. 2018) to as high as 61.80% after 20 days of incubation (Ahmad et al. 2015). *A. niger* isolated from India was reported to achieve an average of 40.5% spent engine oil degraded (Thenmozhi et al. 2013). The differences observed can be attributed to the different incubation periods and locations. The ability of *A. niger* in reducing spent engine oil was also attributed to the secretion of some specific enzymes such as lipase which hydrolyzes the triglycerides structures (Gupta 2016), manganese peroxidase, and lignin peroxidase which can degrade PAHs compound present in spent engine oil (Ameen et al. 2016). Overall, *A. niger* is a good species that can be used in the remediation of spent engine oil but it needed a longer incubation period to achieve similar results as *Mucor* sp.

Ong et al. (2018) and Paper & Nwinyi (2019) recorded a relatively low amount of spent engine oil degradation with *A. flavus* but the finding of Ahmad et al. (2015) was contradictory and these researchers reported higher percentage degradation (44.6%) by this fungi. Although the fungal strain used by both Paper and Nwinyi (2019) and Ahmad et al. (2015) was obtained from Nigeria but this difference in spent engine oil degradation might be associated with the difference in incubation time. Furthermore, Kota et al. (2014) also had a different opinion and suggested that *A. flavus* is ineffective in degrading crude oil as compared to *Trichoderma* sp., this might be because *A. flavus* could not degrade PAHs compound effectively as compared to other potential fungal species (Chaudhry et al. 2012; Haritash and Kaushik 2016). Moreover, due to poor spore formation *A. flavus* have a relatively slow growth compared to *A. niger* (Marín et al. 1998), which causes lower cell biomass production and enzyme secretion and this might be associated with the low spent engine oil degradation percentage.

There is limited information available regarding the use of *A. nidulans* in remediating spent engine oil which suggests that *A. nidulans* might not be as good as other fungal species. Furthermore, various previous studies showed that the genus *Aspergillus* has the potential of degrading various types of hydrocarbons such as aliphatic and aromatic, but most of the reported studies concentrated on the *A. niger* and *A. flavus* (Olajire and Essien 2014).

Conclusion

Results of the study can be concluded that among the tested various fungal species, based on both biomass production and effectiveness in reducing spent engine oil within a relatively short incubation period, *Mucor* sp. is the best species. Further, *Trichoderma* sp. and *A. niger* also have the potential of reducing spent engine oil if the right strain and suitable incubation time are selected. Due to limited reports on fungal species oil degradation mechanisms, extensive research is required to be conducted on mycoremediation of the spent engine oil contaminant.

References

Adekunle, A.A., & Adebambo, O.A. (2007). Petroleum Hydrocarbon Utilization by Fungi Isolated From Detarium Senegalense (J.F Gmelin) Seeds. *Journal of American Sceinces*, *3*(1), 69-76.

Adekunle, A.T., Ester, B.B., Peter, A. O., Bankole, O. S., et al. (2015). Characterization of new glycosophorolipid-surfactant produced by *Aspergillus niger* and *Aspergillus flavus*. *European Journal of Biotechnology and Bioscience*, *3*(4), 34-39.

Adeleye, A. O., Nkereuwem, M. E., Omokhudu, G. I., Amoo, A. O., et al. (2018). Effect of microorganisms in the bioremediation of spent engine oil and petroleum related environmental pollution. *Journal of Applied Sciences and Environmental Management*, *22*(2), 157-167.

Agency for Toxic Substances and Disease Registry (ATSDR) (1995). *Toxicological profile for fuel oils*. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

Ahmad, S.A., Sadiya, S., & Alhaji, S.I. (2015). Biodegradation of used engine oil by fungi isolated from mechanic workshop soils in Sokoto Metropolis, Nigeria. *Sky Journal of Soil Science and Environmental Management*, *4*(6), 64-69.

Al-Hawash, A. B., Zhang, X., & Ma, F. (2019). Removal and biodegradation of different petroleum hydrocarbons using the filamentous fungus *Aspergillus* sp. RFC-1. *Microbiology Open*, *8*(1), e00619.

Ameen, F., Moslem, M., Hadi, S., & Al-Sabri, A.E. (2016). Biodegradation of diesel fuel hydrocarbons by mangrove fungi from Red Sea Coast of Saudi Arabia. *Saudi Journal of Biological Sciences*, *23*(2), 211-218.

Balaji, V., Arulazhagan, P., & Ebeneze, P. (2014). Enzymatic bioremediation of polyaromatic hydrocarbons by fungal consortia enriched from petroleum contaminated soil and oil seeds. *Journal of Environmental Biology*, *35*(3), 521-529.

Balcázar-López, E., Méndez-Lorenzo, L.H., Batista-García, R.A., et al. (2016). Xenobiotic Compounds Degradation by Heterologous Expression of a *Trametes sanguineus* Laccase in *Trichoderma atroviride*. *PloS One*, *11*(2), e0147997.

Boichenko, S., Yakovlieva, A., Kale, U., & Nagy, A. (2021). Analysis of technological potential for utilization of waste aviation lubricating materials. *Technology audit and production reserves*, *2*(1), 58.

Cazares-Garcia, S.V., Vazquez-Garciduenas, M.S., &Vazquez-Marrufo, G. (2013). Structural and phylogenetic analysis of laccases from *Trichoderma*: A bioinformatic approach. *PLoS ONE*, *8*(1), e55295.

Cerniglia, C.E., & Sutherland, G.R., (2010). Degradation of polycyclic aromatic hydrocarbons by fungi. In Timmis, K.N. (Ed). *Handbook of Hydrocarbon and Lipid Microbiology*, Springer, Berlin, Germany.

Chaudhry, S., Luhach, J., Sharma, V., & Sharma, C. (2012). Assessment of diesel degrading potential of fungal isolates from sludge contaminated soil of petroleum refinery, Haryana. *Research Journal of Microbiology*, *7*(3), 182-190.

Chimezie Dirisu, N.G. (2015). Isolation and characterization of hydrocarbon–utilizing fungi from fresh water swampy soil. *Microbiology Research International*, *3*(2), 33-36.

Cocaign, A., Bui, L. C., Silar, P., Chan Ho Tong, L., et al. (2013). Biotransformation of Trichoderma spp. and their tolerance to aromatic amines, a major class of pollutants. *Applied and Environmental Microbiology*, *79* (15), 4719-4726.

Cristica, M., Manoliu, A., Barbaneagra, T., & Ciornea, E. (2011). Compared analysis of catalase and peroxidase activity in cellulolytic fungus *Trichoderma reesei* grown on medium with different concentrations of grinded wheat and barley straws. Alexandru Ioan Cuza" University of Iași New Series, Section IIA 12: 89.

Daccò, C., Nicola, L., Temporiti, M.E.E., et al. (2020). *Trichoderma*: evaluation of its degrading abilities for the bioremediation of hydrocarbon complex mixtures. *Applied Sciences*, *10*(9), 3152.

Durón-Castellanos, A., Zazueta-Novoa, V., Silva-Jiménez, H., et al. (2005). Detection of NAD^+-dependent alcohol dehydrogenase activities in YR-1 strain of *Mucor circinelloides*, a potential bioremediator of petroleum contaminated soils. *Applied Biochemistry and Biotechnology*, *5*, 279-288.

Elshafie, H. S., Camele, I., Sofo, A., Mazzone, G., et al. (2020). Mycoremediation effect of *Trichoderma harzianum* strain T22 combined with ozonation in diesel-contaminated sand. *Chemosphere*, *252*, 126597.

Environmental Quality (Industrial Effluents) Regulations. (2009). Malaysia Department of Environment, https://www.doe.gov.my/portalv1/wp-content/uploads/2015/01/Environmental_ Quality_ Industrial_Effluent_Regulations_2009_-_P.U.A_434-2009.pdf accessed on 1 August 2011.

Fong, F. (2016). 121 oil pollution cases reported between 2009 and 2015. [*Press release*]

Gupta, V.K. (2016). *New and future developments in microbial biotechnology and bioengineering*: *Aspergillus* system properties and applications. Elsevier publication.

Hadibarata, T., Tachibana, S., & Itoh, K. (2007). Biodegradation of phenanthrene by fungi screened from nature. *Pakistan Journal of Biological Sciences*, *10*, 2535-2543.

Haritash, A. K., & Kaushik, C. P. (2016). Degradation of low molecular weight polycyclic aromatic hydrocarbons by microorganisms isolated from contaminated soil. *International Journal of Environmental Sciences*, *6*, 472-482.

Ismail, H. Y., Farouq, A. A., Rabah, A. B., Muhammad, A. B., et al. (2021). Effect of Soil Contamination with Crude Petroleum on Cowpea: An Insight into the Prospects of Crop Production in Nigerian Frontier Basins. *Journal of Environmental and Agricultural Studies*, *2*(2), 50-62.

Kota, M. F., Hussaini, A. A. S. A., Zulkharnain, A., & Roslan, H. A. (2014). Bioremediation of Crude Oil by Different Fungal Genera. *Asian Journal of Plant Biology*, *2*(1), 16-23.

Kupareva, A., Mäki-Arvela, P., Grénman, H., Eränen, K., et al. (2013). Chemical characterization of lube oils. *Energy & fuels*, *27*(1), 27-34.

Lieckfeldt, E., Samuelsi, G.J., Nirenberg, H., & Petrin, O.A. (1999). Morphological and molecular perspective of *Trichoderma viride*: Is it one or two species? *Applied and Environmental Microbiology*, *65*, 2418-2428.

Makut, M. D., Ogbonna, A. I., Ogbonna, C. I. C., & Owuna, M. H. (2014). Utilization of petroleum products by fungi isolated from the soil environment of Keffi Metropolis, Nasarawa State, Nigeria. *International Journal of Science and Nature*, *5*(2), 222-225.

Marchut-Mikolajczyk, O., Kwapisz, E., Wieczorek, D., & Antczak, T. (2015). Biodegradation of diesel oil hydrocarbons enhanced with *Mucor circinelloides* enzyme preparation. *International Biodeterioration & Biodegradation*, *104*, 142-148.

Marín, S., Sanchis, V., Sáenz, R., et al. (1998). Ecological determinants for germination and growth of some *Aspergillus* and *Penicillium* spp. from maize grain. *Journal of Applied Microbiology*, *84*(1), 25-36.

Olajire, A.A., & Essien, J.P. (2014). Aerobic degradation of petroleum components by microbial consortia. *Journal of Petroleum & Environmental Biotechnology*, *5*(5), 1.

Ong, G.H., Ho. C.C., Lim, V.B.F., Wong, Y.Y., et al. (2018). Isolation and identification of potential fungal species for spent engine lubrication oil remediation in Peninsular Malaysia. *Remediation Journal*, *28*(3), 91-95.

Ossai, I. C., Ahmed, A., Hassan, A., & Hamid, F. S. (2020). Remediation of soil and water contaminated with petroleum hydrocarbon: A review. *Environmental Technology & Innovation*, *17*, 100526.

Paper, C., & Nwinyi, O. C. (2019). Earth and Environmental Science Effect of Saw-dust on soils contaminated with waste lubricating oil Effect of Saw-dust on soils contaminated with waste lubricating oil. *IOP Conference Series: Earth and Environmental Science, Volume 331, International Conference on Energy and Sustainable Environment 18–20 June 2019, Covenant University, Nigeria*.

Ravelet, C., Krivobok, S., Sage, L., & Steiman, R. (2000). Biodegradation of pyrene by sediment fungi. *Chemosphere*, *40*, 557-563.

Saraswathy, A., & Hallberg, R. (2002). Degradation of pyrene by indigenous fungi from a former gasworks site. *FEMS Microbiology Letters*, *210*, 227-232.

Singh, H. (2006) "Mycoremediation: fungal bioremediation". *Hoboken: Wiley-Interscience, 1-28.*

Singh, S. K., & Haritash, A. K. (2019). Polycyclic aromatic hydrocarbons: soil pollution and remediation. *International Journal of Environmental Science and Technology*, *16*(10), 6489-6512.

Stanley, H.O., & Immanuel, O.M. (2015). Bioremediation potential of *Lentinus subnudus* in decontaminating crude oil polluted soil. *Nigeria Journal of Biotechnology*, *29*, 21-26.

Szewczyk, R., & Długoński, J. (2009). Pentachlorophenol and spent engine oil degradation by Mucor ramosissimus. *International Biodeterioration & Biodegradation*, *63*(2), 123-129.

Thenmozhi, R., Arumugam, K., Nagasathya, A., Thajuddin, N., et al. (2013). Studies on mycoremediation of used engine oil contaminated soil samples. *Pelagia Research Library Advances in Applied Science Research*, *4*(2), 110-118.

Vatsyayan, P, & Goswami, P. (2016). Highly active and stable large catalase isolated from a hydrocarbon degrading *Aspergillus terreus* MTCC 6324. *Enzyme Research*, 4379403.

Zafra, G., Moreno-Montaño, A., Absalón, Á.E., & Cortés-Espinosa, D.V. (2015). Degradation of polycyclic aromatic hydrocarbons in soil by a tolerant strain of *Trichoderma asperellum*. *Environmental Science Pollution Research*, *22*(2), 1034-1042.