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Review

# Microbial communities in petroleum refinery effluents and their complex functions

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## ABSTRACT

Petroleum refinery effluents (PRE) are a significant cause of pollution. It contains toxic compounds such as total petroleum hydrocarbons (TPH), and polycyclic aromatic hydrocarbons (PAHs), as well as heavy metals. They show a huge threat facing the aquaculture habitats, human health, and the environment if they are not treated before discharging into the environment. Physical and chemical procedures are used to treat hydrocarbon pollution in PRE, but these techniques often result in the formation of hazardous by-products during the remediation process. However, PRE contains various microbial communities, including bacteria, yeast, microalgae, and fungi. The bioremediation and biodegradation of oil contaminants are the primary functions of these microbial communities. However, these microorganisms can perform various additional functions including but not limited to heavy metals removal, production of biosurfactants, and nitrogen fixation. This review contributes to the comprehension of natural microbial communities and their complex functions in petroleum refinery effluents. Understanding microbial communities would facilitate the advancement of innovative biotechnology aimed at treating PRE, improving bioremediation processes, and potentially transforming PRE into valuable by-products. Moreover, it assists in determining the most effective bioaugmentation strategy to enhance biodegradation and bioremediation in PRE. The review highlights the potential for sustainable green approaches using microbial communities to replace toxic chemical therapies and expensive physical treatments in the future.

## 1. Introduction

Petroleum-derived products, like petrochemicals and fuels, are critical for economic growth and play an important part in our daily lives. Petroleum is a fossilized form of fuel that is a vital form of energy. Oil refineries treat crude oil and transform it into useful petroleum-based goods such as gasoline, kerosene, and jet fuel which are significant in meeting the world's greedy need for energy and chemicals. Despite their advantages, it harms potentially the ecological systems and the health of living creatures. There are many main sources through which petroleum-based pollution can enter the environment and cause adverse effects including refinery pollutants, automotive discharge, and inadvertent leaks and spills during the investigation, shipping, refining, and preservation (Imam et al., 2019). Refinery discharge is the major source of soil and water contamination because it contains petroleum products such as hydrocarbons (polycyclic aromatic hydrocarbons (PAHs), and total petroleum hydrocarbons (TPH)), and additional organic chemicals for example toluene, benzene, phenols, ethylbenzene, and xylene. It is extremely important to treat refinery contaminants and biological

approaches are recognized as more useful in this regard rather conventional methods (Imam et al., 2021).

Microorganisms, with their extraordinary physiological adaptability, are now recognized as a significant factor in the alteration of contaminants in refinery effluents. The microbial populations that inhabit petroleum refinery effluents are diverse and highly receptive to the wastewater's particular chemical constitution. Microbial consortiums change and develop to successfully decay, eliminate, or trap contaminants ranging from hydrocarbon-rich fractions to heavy metals and an extensive variety of chemical compounds (Lukhele et al., 2021). This review aims to elucidate the complex function of microbial communities, including bacteria, microalgae, fungi, and yeast play in defining the fate and effect of petroleum refinery effluents on aquatic ecosystems and human health. Addressing the microbe community of refinery effluents is critical not only for optimizing wastewater treatment systems but also for reducing the environmental effects of the discharge of effluents.

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## 2. Petroleum refinery effluents (PRE)

The wastewater or fluid waste produced throughout the refinement procedure of crude oil into different petroleum-based goods is referred to as petroleum refinery effluents (PRE) or oil refinery effluents. Between 0.4 and 1.6 times as much PRE is created during the processing of crude oil products. Therefore, based on the current output of 85 million barrels of crude oil per day, the global production of effluent is 33.5 MBpd (Yan et al., 2014). The primary pollutants found in petroleum refinery effluents and sludge are hydrocarbons, polycyclic aromatic hydrocarbons (PAHs), and organic and inorganic compounds such as phenols, sulfur, and nitrogen-encompassing compounds. PRE formulas vary based on the type of crude oil and handling methods employed. PAHs, which are very hazardous and carcinogenic, have been shown to change the community framework and soil microbiology of watersheds. Oil leaks during shipping or discovery, tank spillage, or incorrect disposal of petroleum waste can contribute to soil pollution at petroleum refineries. Various refinery effluents contribute to soil pollution including dissolved minerals, radioactive substances, dissolved gases (CO<sub>2</sub>, H<sub>2</sub>S, and O<sub>2</sub>), and oil compounds like wax, asphaltene, and aliphatic hydrocarbons. Petroleum refineries are also substantial contributors to water contamination (Imam et al., 2021). Furthermore, the petroleum refinery effluents host microbial communities that are methanogenic, anaerobic, fermentative, and sulfate-reducing; these microbes are renowned for their efficiency in breaking down hydrocarbons (Roy et al., 2018).

## 3. Type of microbial communities exist in PRE

A diversity of microorganisms are found in PRE including hydrocarbon-degrading bacteria, fungi, yeast, and microalgae. The major role of these microbial communities is bioremediation and biodegradation of oil pollutants. The method of detoxification of environmental pollutants by the utilization of microbes is an innovative technique for the elimination of petroleum contaminants. Microbial degradation is the primary and definitive natural process to eliminate petroleum pollutants from ecosystems (Chunyan et al., 2023). The types of microorganisms found in PRE are described in detail below.

### 3.1. Bacteria

Bacteria are unicellular organisms that lack a nucleus, exhibit metabolic activity, and reproduce through asexual processes (binary fission) (*The Biology of Bacteria*, 2019). A different genus of bacteria was recognized in oil refinery sludge, effluent (wastewater), and oil refinery soil. Some were known for their biodegradation activity, and some were not. They exist as mixed microbial communities (consortium) to metabolize organic pollutants and no single bacteria can work and utilize all types of petroleum hydrocarbons alone (Table 1).

A study discovered that raw and treated samples isolated from oil refinery effluents had two dominant phyla which were *Proteobacteria* and *Firmicutes*. At the genus level, four genera were identified exclusively from crude effluents namely *Halomonas*, *Rhodabaculum*, *Bacillus*, and *Wenzhouxiangella*. In addition, *Pseudoxanthomonas*, *Rhizobium*, *Pseudomonas*, *Rhodabaculum*, and *Brevundimonas* were obtained from processed effluents. These bacteria can decompose a variety of organic and inorganic contaminants since they are halophilic or halotolerant (Lukhele et al., 2021).

Furthermore, numerous bacteria strains were recognized and investigated in several studies. Goveas (2020) isolated and identified thirty-two bacteria strains from PRE. Some bacteria including *Acinetobacter baumannii* (OCB1), *Pseudomonas aeruginosa* (WD23), *Pseudomonas aeruginosa* (WDE11), and *Bacillus cereus* (WD22) were shown to tolerate salinity and break down petroleum hydrocarbons in saltwater (Goveas et al., 2020). In addition, a more comprehensive study shows that 50 bacterial isolates from oil refinery effluent wastewater were identified and tested for resistance to heavy metals and PAHs. One of those isolates was *Enterobacter ludwigii* (KWB3) exhibited a maximum degradation efficiency of (66 %) (Khatoon & Malik, 2019). From the petroleum refinery wastewater in Nigeria, four main microbial strains were isolated namely, *Bacillus subtilis*, *Micrococcus luteus*, *Staphylococcus aureus*, and *Staphylococcus epidermidis* noted as (S1, S2, S3, S4), respectively. They were found to degrade hydrocarbons (Musa et al., 2015). Table 1 summarizes different microorganisms isolated from PRE.

The main role of bacteria in oil refineries is biodegradation of petroleum hydrocarbons. This occurs through aerobic and anaerobic pathways, with oxygen acting as an electron acceptor in the former. Bacteria initiate degradation catabolism of alkanes in aerobic mechanism via oxygenases by four pathways: terminal oxidation, oxidation,

**Table 1**

The microorganism isolated from different PRE.

No.	Microorganism	Type	Oil refinery	Source
1	<b>Bacteria</b>	<i>Pseudomonas aeruginosa</i>	Al-Najaf refinery wastewater, Iraq	(Sultan, et al., 2015)
2		<i>Alcaligenesfaecalis</i>		
3		32 bacterial strains including <i>Bacillus cereus</i> WD22, <i>Acinetobacter baumannii</i> OCB, <i>Pseudomonasaeruginosa</i> WD23 and <i>WDE11</i>	Petrochemical refinery industry in Mangalore, India	(Goveas et al., 2020)
4		50 isolates including <i>Enterobacter ludwigii</i> KWB3	Mathura oil refinery, U.P., India	(Khatoon & Malik, 2019)
5		<i>Pseudomonas</i> , <i>Bacillus</i> spp, <i>Burkholderia</i> , <i>Kocuria</i> , <i>Enterobacter</i> , and <i>Pandoraea</i>	Indian Oil Corporation Ltd. (IOCL) oil refinery, Digboi, India	(Sarkar et al., 2017)
6		<i>Bacillus</i> , <i>Wenzhouxiangella</i> , <i>Rhodabaculum</i> , <i>Halomonas</i> , <i>Rhizobium</i> , <i>Pseudomonas</i> , <i>Pseudoxanthomonas</i> , <i>Rhodabaculum</i> , and <i>Brevundimonas</i>	Oil refinery in Johannesburg, South Africa	(Lukhele et al., 2021)
7		<i>Micrococcus luteus</i> , <i>Staphylococcus epidermidis</i> , <i>Staphylococcus aureus</i> , and <i>Bacillus subtilis</i> .	Petroleum refinery wastewater, Nigeria	(Musa et al., 2015).
8	<b>Fungi</b>	Yeast, <i>Fusarium</i> , <i>Cephalosporium</i> , <i>Verticillium</i> , <i>Phytophthora</i> , <i>Aspergillus flavus</i>	Kaudana Petroleum Refinery, Area of Kaduna State, Nigeria	(Onyeiwu et al., 2022)
9		<i>Monilia</i> , and <i>Penicillium</i>		
10		<i>Chrysosporium tropicum</i> , <i>Aspergillus flavus</i> , <i>Aspergillus niger</i> , and <i>Rhizopus oryzae</i>	Oil refinery wastewater of Kaudana and Romi River, Area of Kaduna State, Nigeria	(Obukohwo et al., 2020)
11		<i>Penicillium</i> sp., <i>Aspergillus fumigatus</i> , <i>Aspergillus niger</i> , and <i>Aspergillus flavus</i>	Petroleum oil refinery wastewater, AINasiriya, Iraq	(Al-jawhari et al., 2015)
12		<i>Monocillium</i> sp.	Refinery effluent, Nigeria	(Odili et al., 2021)
13	<b>Microalgae</b>	<i>Scenedesmus obliquus</i>	Oil refinery effluent was collected from Ennore, a suburb of Chennai, India.	(Rajasulochana et al., 2009)
14		<i>Chlorella</i> sp. UMACC 322, <i>Chlorococcum</i> sp. UMACC 324, and <i>Oscillatoria</i> sp. UMACC 323	Oil refinery wastewater treatment pond (WWTP) on the West Shore of Peninsular Malaysia	(Yusof & Phang, 2012)
15		<i>Cyanophyta</i> , <i>Oscillatoria</i> , <i>Rhizochloris</i> , <i>Gleotrichia</i> , <i>Chlamydomonas</i> , <i>Cryptomonas</i> , and <i>Spirulina</i>	Oil refinery wastewater, Niger Delta Region, Nigeria	(Agwa et al., 2011)

subterminal oxidation, and a long-chain n-alkane oxidation pathway. These pathways produce primary alcohols, fatty acids, and  $\beta$ -oxidation, with the final product being n-alkyl hydroperoxide, peroxy acids, alkyl aldehydes, and fatty acids (Mekonnen et al., 2024).

The degradation of petroleum hydrocarbons via an anaerobic mechanism can occur in environments with restricted oxygen availability through the utilization of nitrate, ferrous iron, or sulfate as electron acceptors, or under methanogenic circumstances. Numerous anaerobic bacterial strains, such as sulfate-reducing and denitrifying strains, possess the capability to break down n-alkanes containing six or more carbon atoms into carbon dioxide (CO<sub>2</sub>) and chemicals that are comparatively less hazardous. The anaerobic breakdown of n-alkanes can be attributed to two established mechanisms: the fumarate addition pathway and the carboxylation pathway. Sulfate-reducing bacteria, denitrifying bacteria, and a consortium of nitrate-reducing bacteria all possess the fumarate addition mechanism. The sulfate-reducing bacteria strain (Hxd3) is responsible for the development of the carboxylation pathway. This route facilitates the conversion of a C-odd alkane into a C-even fatty acid using subterminal carboxylation at the C-3 position (Mekonnen et al., 2024).

### 3.2. Fungi and yeast

Fungi are unicellular or multicellular microorganisms that belong to the eukaryotic domain. Fungi can manifest as yeasts, molds, or as a hybrid of both kinds. Certain fungi can induce superficial, cutaneous, subcutaneous, systemic, or allergic illnesses. Yeasts are unicellular fungi that reproduce asexually through a process called budding. They use carbon as a main source of nutrients (Zhang & Wiederhold, 2016).

Many fungal species have also been isolated from petroleum refinery effluents. From the Kaudana petroleum refinery, several fungi have been isolated including *Verticillium*, *Phytophthora*, *Fusarium*, *Cephalosporium*, *Aspergillus*, *Penicillium*, *flavus* *Monilia*, and yeast. The measured oxidase enzyme activity of 87 % of the 60 isolates of fungi was shown to be positive. Among the most common fungal species were *Penicillium* and *Fusarium*. These fungi can change or break down pollutants due to their enzymatic capabilities, which highlights their prospective application in bioremediation procedures (Onyeiwu et al., 2022).

Similarly, another study revealed the presence of four fungal isolates including *Rhizopus oryzae*, *Aspergillus flavus*, *Chrysosporium tropicum*, and *Aspergillus niger* from the refinery effluents of Kaudana and Romi River (Obukohwo et al., 2020). All these fungal species were selected for bioremediation purposes. Al-Jawhari's group was able to isolate several fungi strains containing *Penicillium* sp., *Aspergillus fumigatus*, *Aspergillus flavus*, and *Aspergillus niger* from petroleum oil refinery wastewaters, AlNasiriya, Iraq (Al-jawhari et al., 2015). *Monocillium* sp. Also has been extracted from soil polluted with hydrocarbons from refinery effluent in Nigeria. According to the study's findings, *Monocillium* sp. Is a practical hydrocarbon degrader that can be applied to the bioremediation of areas damaged by petroleum (Odili et al., 2021). Several fungal species (21 isolates) have also been known from petroleum-polluted soil and water including *Aspergillus*, *Penicillium*, *Fusarium*, *Paecilomyces*, *Graphium*, *Neosartorya*, *Talaromyces*, and *Amorphoteca*. In addition, yeast genera have been identified as well such as *Pichia*, *Candida*, and *Yarrowia* originating from oil-contaminated soil and water (Chaillan et al., 2004).

There are two primary pathways that fungi use to break down hydrocarbons: enzymatic and non-enzymatic. Biochemical reactions such as biosorption, biomineralization, and stripping are involved in non-enzymatic processes. Bioremediation of petroleum hydrocarbons is aided by enzymatic procedures that employ certain fungal enzymes. Enzymes within cells, such as transferase and cytochrome P450 family peroxidases, transform hydrocarbon mixtures into intermediate metabolites and substances that are expelled as byproducts. Catabolizing hydrocarbons into their excreted derivatives is facilitated by extracellular enzymes such as laccases, peroxidases, and oxidases (Mahmud et al., 2022).

### 3.3. Microalgae

Microalgae are eukaryotic, single-celled, that develop and release biomass. This biomass has numerous potential uses, including as a source of hydrocarbons, biofuels, biofertilizers, and nutraceuticals. The microalgae depend on the phosphates and nitrates in wastewater that comes from farmland, cities, and businesses. Therefore, wastewater is an ideal culture medium for microalgae biomass production, because it supplies a variety of minerals, micronutrients, and organic and inorganic contaminants the algae need (Kukwa & Chetty, 2020).

Oil refinery effluents contain a wide range of microalgae, some of which have been found to have the capacity to break down contaminants. PAH-tolerant microalgae that could break down PAHs were isolated and discovered by Yusuf and Phang (2012). These microalgae included *Chlorella* sp. UMACC 322, *Chlorococcum* sp. UMACC 324 and *Oscillatoria* sp. UMACC 323. Within a reservoir for waste liquids in an oil processing facility, Joseph and Joseph (2002) discovered a resistant microalgal community with a clear seasonal variation in species composition. A previous study has shown that Nineteen genera and up to six phyla of microalgae were found in wastewater of a petroleum refinery in the Delta Region, Niger with *Cyanophyta* being the most prevalent type. Throughout the examination, *Oscillatoria* (12.73 %) also was the most common microalgae, whereas the least common microalgae at 1.82 % were *Rhizochloris*, *Gleotrichia*, *Chlamydomonas*, *Cryptomonas*, and *Spirulina* (Agwa et al., 2011). It has been discovered that the presence of microalgae, such as *Scenedesmus obliquus*, is beneficial for the bioremediation of oil refinery effluents (Rajasulochana et al., 2009). Together, previous studies demonstrate the variety and possible advantages of microalgae in oil refinery wastewater.

Microalgae have a role in bioremediation through two primary mechanisms: indirect participation by fostering the growth of degrading bacteria, and direct action on pollutants. Their primary contribution is derived from the activation of many processes, encompassing the synthesis of hydrocarbon-degrading enzymes, such as lipooxygenases, as well as the assault through the release of free radicals (Radice et al., 2023).

Although the exact mechanism by which microalgae bioremediate petroleum remains unclear, two main theories have been proposed: first, that they consume organic carbon derived from hydrocarbons; or second, that they store these toxins internally as a defense mechanism and treat them as an actual pollutant (Radice et al., 2023).

## 4. Roles of microbial communities in PRE

The diversity of microorganisms in PRE indicates distinctive roles of those microorganisms including but not limited to biodegradation and bioremediation. Here, we will demonstrate the potential different functions of microorganisms found in PRE.

### 4.1. Biodegradation and bioremediation of hydrocarbons

Numerous microbial communities, comprising bacteria, microalgae, fungi, and yeast, are well-known for their capability to break down and decompose oil. Biodegradation of hydrocarbons is a crucial technique for eliminating petroleum and hydrocarbon contaminants from the ecosystems. These microorganisms break down hydrocarbons and utilize them as the exclusive carbon source by using their metabolic capabilities (Chunyan et al., 2023). Bacteria have been recognized as a primary degrading agent and the most effective factor in the degradation of oil pollutants among microorganisms (Ganesan et al., 2022). Since bacteria can thrive in harsh conditions, they have been employed to process petroleum hydrocarbons. As a result of their exceptional efficiency, environmental friendliness, and cost-effective production, bacteria have demonstrated considerable potential as a biotechnique for the remediation of oil contamination (Xu et al., 2018). Table 1 shows examples of numerous bacterial species that have been isolated from PRE,

including *Klebsiella* sp., *Pseudomonas aeruginosa*, *Pseudomonas* sp., *Rhodococcus* sp., *Bacillus* sp., and *Bacillus pumilus* where most of them have been identified as hydrocarbon-degrading bacteria. The biodegradation process is influenced by various factors, including salinity, temperature, pressure, hydrocarbon type (length, aromaticity), pH, hydrocarbon concentrations, existence of other microorganisms, and presence of oxygen (Liu et al., 2024, Mekonnen et al., 2024).

A series of recent studies have indicated that the ability of bacteria to degrade petroleum contaminants varied among different strains. Prior research by Goveas's group isolated 14 bacteria strains from PRE that could use petroleum hydrocarbons as a sole source of carbon. Four of them could completely decolorize 2,6-dichlorophenol indophenol within 36 h namely *Acinetobacter baumannii* (OCB1), *Pseudomonas aeruginosa* (WDE11) *Pseudomonas aeruginosa* (WD23), and *Bacillus cereus* (WD22). Among these strains, *P. aeruginosa* (WD23) is shown to be the best degrader. It decomposed about 27.25 % of petroleum crude oil in seawater enriched in glucose and yeast extract in 15 days (Goveas et al., 2020).

Al-Marri's group identified a new bacteria strain *Marinobacter* sp. that has the highest biodegradation ability among other bacteria in the study. The study was conducted on samples contaminated with hydrocarbons from the oil and gas sectors' sludge, wastewater, and soil in Qatar. The results identified 26 bacterial isolates strains. Among those bacteria, *Marinobacter* sp. had the maximum growth rate and biodegradation ability. When this strain was examined for hydrocarbon biodegradation, it was revealed to be effective in decomposing 90–100 % of low and medium-MW hydrocarbons. On the other hand, HMW hydrocarbons ranging from C35 to C50 were degraded by 60–80 % by the same strain. *Marinobacter* sp shows the potential to be used as green biotechnology for the degradation of hydrocarbons in PRE (Al-Marri et al., 2023).

In addition, Ghorbannezhad's group demonstrated the ability of *B. subtilis* (HG01), obtained from contaminated soils, in the decomposition of high-molecular-weight petroleum hydrocarbons (tetracosane and pyrene). *B. subtilis* (HG01) was able to decompose 55.5 % of 500 mg/l pyrene and 77.2 % of 500 mg/l tetracosane at 5 % w/v NaCl. The study also investigates the impact of a combination of cultures (TMC mixed-culture) containing *B. subtilis* (HG01), *Pseudomonas putida*, and *Pseudomonas aeruginosa*. The results showed an enhancement of pyrene and tetracosane degradation by around 20 %. *B. subtilis* HG 01 has significant hydrocarbon elimination abilities under both saline and non-saline conditions, independently and in combined cultures (Ghorbannezhad et al., 2022).

Petroleum biodegradation involves the utilization of inherent biochemical mechanisms by bacteria to break down petroleum hydrocarbons. These mechanisms involve enzymatic stimulation and/or functionalization reactions. These mechanisms can be conducted under aerobic and/or anaerobic pathways. The resulting products include water, gasses such as methane and carbon dioxide (CO<sub>2</sub>) also metabolites, biosurfactants, organic, and inorganic compounds (Fig. 1). Metabolites are small molecules produced as intermediate or end products of metabolism that benefit the organism. Biosurfactants are compounds produced and excreted to the cell surface to increase solubility and biodegradability, in addition to decreasing the surface tension (Victor et al., 2020). The process of converting crude oil components frequently entails derivatization and preservation, as opposed to directly eradicating high molecular weight molecules (Oldenburg et al., 2017). This suggests that the biodegradation of petroleum components could be partial which would yield organic and inorganic compounds. To our knowledge, no prior studies have examined the organic or inorganic by-products resulting from oil biodegradation.

Few research has demonstrated methanogenic biodegradation of crude oil by bacteria. A methanogenic C22–C30 n-alkane-degrading enrichment culture was found to produce significant methane over a two-year incubation period (736 days). The maximum methane yield rate occurred from day 351 to 513. The biotransformation of paraffin to

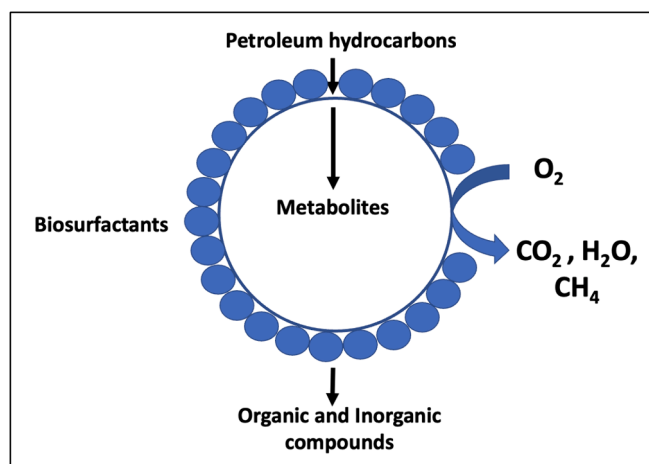


Fig. 1. Type of products resulting from utilizing petroleum hydrocarbons.

methane under anoxic conditions led to the dominant methanogen shifting from *Methanothermobacter* to *Thermoplasmatales*. Bacterial community analysis showed Proteobacteria and Firmicutes as predominant in control cultures, while Actinobacteria became the most dominant phylum in n-alkane-dependent cultures (Chen et al., 2020). Research conducted by Hogendoorn's group showed that *Methylacidiphilum fumariolicum* (SolV) was also able to convert methane to methanol under different growth conditions (Hogendoorn et al., 2020).

Fungi also are effective candidates for bioremediation because they can adapt to diverse environmental circumstances and produce a variety of hydrolyzing enzymes that help decompose waste materials (Singh et al., 2020). Furthermore, it is cost-effective, safe, and environmentally friendly, and produces large quantities of extracellular and intracellular polycyclic aromatic hydrocarbon degradative enzymes throughout the fungal bioremediation process. Fungi exhibit a significant production of enzymes that degrade polycyclic aromatic hydrocarbons, both inside and outside their cells such as manganese peroxidases, tyrosinases laccases, reductive dehalogenases, and cytochrome P450 monooxygenases. The cellular membranes of fungi are susceptible to less complex organic compounds including petroleum hydrocarbons, which are subject to extracellular enzyme oxidation. Moreover, it can proceed with metabolic processes such as  $\beta$ -oxidation, hydrolysis, tricarboxylic acid cycle entrance, and dehalogenation (Chaurasia et al., 2023).

One of the valuable fungi obtained from refinery effluent is *Fusarium proliferatum* WC416. It has been shown the ability to degrade naphthalene. The results indicate that *Fusarium proliferatum* WC416 degrades naphthalene into salicylamide and salicylic acid in an efficient manner. At concentrations of 100 ppm, the maximum degradation rate of 83 % was observed. The research also evaluates the capacity of sophorolipid-based fungi to produce biosurfactants, proposing *Fusarium proliferatum* WC416 as a viable degradant for environmental remediation (Gupta et al., 2023).

Besides bacteria and fungi, yeast is widely acknowledged as one of the most efficient microbial species with notable bioremediation properties. Their significant raises are from their ability to return the environment to its former state. In addition, they can remove heavy metals from polluted refinery waste. Moreover, they have also been capable of degrading hydrocarbons (Rana et al., 2023). Due to the hydrophobic nature and potent emulsification capability of biosurfactants derived from *Candida* genus yeasts, they are frequently utilized in the decomposition of petroleum and its component compounds, for example, poly aromatic hydrocarbons (PAHs) and alkanes (Priya et al., 2016).

An investigation by Benmessouad's group was conducted on oil-polluted soil samples wherein a new strain of *Candida parapsilosis* SK1 was detected. The strain exhibited a significant capability for decomposing crude oil. Upon 21 days of growing at 30 °C, the degradation

efficiency of the strain was evaluated to be 68 %. Furthermore, the research examined the potential effects of glucose supplementation on the strain's biodegradation efficiency. It was discovered that the removal rate increased to approximately 78.6 % within an equivalent period when 0.5 % glucose was added to the crude oil (Benmessaud et al., 2022).

A previous study by Gargouri and his colleagues discovered that a couple of yeast strains (HC1 and HC4) could biodegrade petroleum hydrocarbons including PAHs and alkanes after being isolated from wastewater from an industrial refinery. These strains are members of the *Trichosporon* and *Candida* genera. They demonstrated a high capacity for hydrocarbon biodegradation as well as a significant amount of biomass, with strain HC1 dominating strain HC4 in terms of hydrocarbon substrate degradation. These yeast strains may help reduce petroleum pollution in wastewater and be beneficial for bioremediation (Gargouri et al., 2015).

Further microorganisms that contribute to bioremediation indirectly are microalgae. It encourages the growth of microorganisms that break down substances and directly interact with them. Their direct involvement stems from the activation of many mechanisms, which include the creation of hydrocarbon-degrading enzymes such as lipoxygenases and the release of free radicals to initiate an attack (Radic et al., 2023).

Satpati discussed the ability of microalgae to both biotransform harmful PAHs into less toxic PAHs and destroy them through bioaccumulation. Nature harbors a diverse array of microalgal strains that possess the ability to convert or metabolize various polycyclic aromatic hydrocarbons (PAHs) such as naphthalene, anthracene, phenanthrene, and benzo [a] pyrene. *Ankistrodesmus braunii*, *Scenedesmus acutus*, and *Selenastrum capricornutum* are examples of microalgal species (Satpati et al., 2023a).

According to Ashwaniy's study, microalgae cultivated in petroleum refinery effluent (PRE) can lower the concentrations by 70 % of chemical oxygen demand (COD), 61 % total suspended solids (TSS), 61 % sulfide, and 81 % biochemical oxygen demand (BOD). It contains 67 % phosphorus and TDS and can serve as a growing medium for bacteria in a microbial desalination cell (MDC) to generate renewable electricity (Ashwaniy et al., 2020). According to Znad (2018), COD was reduced from 504 to 144 mg/L when oil effluent was treated with *Chlorella* spp. (Znad et al., 2018).

Microalgae can work synergistically with bacteria or fungi to reduce pollutants and participate in bioremediation. Ashwaniy's study shows in this manner a significant reduction of organic compounds in petroleum refinery effluents, with the microalgae *Scenedesmus abundans* and bacteria (Ashwaniy et al., 2020). Furthermore, Satpati reported that the combination of microalgae and fungi has been found to effectively eliminate various pollutants, such as heavy metals, from wastewater (Satpati et al., 2023b). Gods'gift and Fagade (2016), examined microalgal-bacterial consortia consisting of microalgae species *Chlorella minutissima* and *Aphanocapsa* sp. and bacteria species *Citrobacter* sp. SB9, *Pseudomonas aeruginosa* SA3, and *Bacillus subtilis* SA7. PAH degradation percentage of consortium BCC (bacteria inoculants and *Chlorella minutissima*) was the highest at 92.09 %. BCCA (all inoculants) and BCA (bacteria inoculants and *Aphanocapsa* sp.) had 67.76 % and 47.19 % respectively. These studies collectively emphasize the significant function of microalgae with bacteria and fungi in the bioremediation of pollutants of PRE, offering a sustainable and multifaceted solution.

#### 4.2. Elimination of heavy metals

Petroleum refinery effluents are among the primary sources of heavy metals pollution for example nickel, copper, and zinc (Daflon et al., 2017). The discharge of PRE into the environment will impact the ecosystems and the biological systems of living organisms. As demonstrated by Adebiyi, the soils surrounding a petroleum refinery products station were revealed to contain heavy metals such as (Fe, Cu, Cd, As, Pb, Ni, Zn, and Cr) at increased levels, these metals have the potential to

contaminate the surrounding ecosystem (Adebiyi et al., 2020). Also, as seen on surface soil near an oil refinery, high levels of Yb, Eu, Mo, Hf, Cd, and Tb presence can cause severe pollution (Alshahri & El-Taher, 2018). Excessive heavy metal exposure can lead to symptoms in humans such as weight loss, asthma, skin rashes, cancer, and others (Ungureanu & Mustatea, 2022).

One strategy that microorganisms can adapt to eliminate heavy metals is biosorption. It is defined as the biomaterials' capacity to capture heavy metals from effluent via ATP-dependent metabolic pathways or naturally physicochemical intake pathways, or as a characteristic of specific kinds of inert, non-living microbial biomass that attach and allocate heavy metals from extremely dilute water solutions (Derco & Vrana, 2018).

A study conducted by El-Bestawy shows that during the remediation of oil refinery effluent, *Pseudomonas* spp. demonstrated excellent effectiveness in eliminating oil (56.8 and 68.8 %) and Pb (90.97 %) from starting oil dosages of 500 and 25 mg/l (El-Bestawy et al., 2013). Another study by Oaikhena and his group investigated the impact of five isolates bacteria from PRE on heavy metal tolerant. Bacteria were identified as *Klebsiella pneumoniae*, *E. coli*, *Staphylococcus aureus*, *Proteus vulgaris*, and *Pseudomonas aeruginosa*. When compared to chromium, nickel, and zinc, these isolates exhibited a low level of resistance to cadmium. The mixed culture consortium (MCC) successfully remedied higher concentrations of zinc (90.1 %), chromium (33.4 %), cadmium (100 %), and nickel (73.9 %) in comparison to pure culture isolates in PRE (Oaikhena et al., 2016). *Bacillus* species and *Staphylococcus* species isolated from PRE were revealed to have significant chromium resistance. *Bacillus* species exhibited the highest resistance to heavy metals containing chromium (Dayan and Jeyachandran, 2019). Collectively, the previous research shows the proficiency of bacteria to tolerate heavy metals from PRE.

In addition to the bioremediation of petroleum hydrocarbons, scientists have examined the function of fungi in the elimination of heavy metals. Toxic metals can accumulate on the surfaces of fungal cells through the method of biosorption, which can cause the production of nuclei and successive accumulation of minerals. Certain metals may accumulate intracellularly via active transport or dispersion across the plasma membrane (Li et al., 2020). Prior research by Machido's group isolated six genera of filamentous fungus from PRE and tested against extraction of heavy metals for example nickel (Ni), cadmium (Cd), and lead (Pb). The results show that the six isolates were shown to be able to extract between 0–50 % of Cd, 30–58 % of Ni, and 36–64 % of Pb. By using *Nigrospora* and *Chaetophoma* sp., the removal efficiency of all mentioned metals was consistently at the lowest levels, whilst *Trichoderma* sp. exhibited the maximum level of efficiency. Additionally, compared to the other fungal species studied, *Trichoderma* sp. and *Microsporium* sp. showed the greatest capacity for bioaccumulating the metals. Therefore, it was determined that Cd, Ni, and Pb could be removed from the petrochemical industry's wastewater contaminated with heavy metals by using *Trichoderma* sp. and *Microsporium* sp (Machido et al., 2016).

Another study investigates the heavy metals (Pb, Zn, Cr, and Cd) removal efficiency of three tolerant fungus isolates (*Microsporium nanum*, *Aspergillus flavus*, and *Aspergillus niger*) separated from Kaduna Refinery and Petrochemical Company (KRPC) effluent. Of these, *A. niger* had removal efficiency of Cd, Zn, Pb, and Cr by 90.72, 72.40, 67.23, and 51.25 %, respectively. Similarly, *A. flavus* exhibited a significant removal efficiency in the same order as *A. niger* for Cd, Zn, Pb, and Cr by 87.63, 64.63, 64.195, and 49.66 %, respectively. Likewise, *M. nanum* was able to eliminate Cd with maximum efficiency of 87.83, followed by Pb, Zn, and Cr with percentages of 74.09, 64.51, and 46.99 %, respectively. The findings indicate that *A. niger*, *M. nanum*, and *A. flavus*, which are naturally present in refinery effluent, have the potential to be employed in bioremediation aimed at eliminating heavy metals (Abdulmajeed, Yusuf, & Abdullahi, 2016).

The application of indigenous fungi has been investigated in Egypt's

petroleum refining effluents for myco-remediating heavy metals. The heavy metals with the maximum concentrations were documented for Ferric ( $\text{Fe}^{3+}$ ) and Cobalt ( $\text{Co}^{2+}$ ). The study's findings indicate that heavy metals were removed using either biosorption (non-living) or bioaccumulation (living) in the biomass. The fungal isolates *Aspergillus* sp. (AHM69) and *Penicillium* sp. (AHM96) were demonstrated to be efficient in heavy metals removal in actual wastewater mixtures and PAHs in aqueous solutions. The results highlight the potential of fungal species to remediate heavy metals (El-Bondkly & El-Gendy, 2022).

Bacteria and fungi consortium has been utilized as a technique to remove heavy metals from industrial wastewater. Migahed's group employed the Statistical Design of Experiment (DOE) to assess the efficacy of microbial isolates in the removal of heavy metals. A combination of bacterial biomass and fungal spores was used to eliminate Chromium and Iron ions from industrial effluents via biosorption. The isolates of the biosorbent exhibited notable efficacy in the removal of heavy metals. The microbial consortia effectively eliminated Chromium ions and more than 50 % of Iron ions from both standard solutions and industrial effluents (Migahed et al., 2016).

*Candida* species have also been proven to be promising candidates for heavy metal bioremediation. Because of their rapid development rate, distinctive cell wall structure, and adaptability to growing in low-cost growing conditions, widespread research has identified yeasts as exceptionally efficient biosorbents for eliminating heavy metal ions from effluents. Under different environmental conditions, *Candida* species have shown that they may accumulate heavy metals of varying types. One study was conducted to examine the elimination of Cd from wastewater by using *C. tropicalis*. The result showed that *C. tropicalis* was capable of eliminating Cd by about 40 % after treatments for 6 days (Bhattacharyya et al., 2022).

Removal of heavy metals in *Candida* could be achieved by biosurfactants. This has been discussed by Da Rocha Junior et al as they show that biosurfactant produced by *Candida tropicalis* was able to remove metals. The biosurfactant was shown to be effective in eliminating Cu and Zn, with the elimination rate fluctuating from 35 – 80 %. The researchers hypothesized that the biosurfactant facilitates the elimination of heavy metals by forming complexes with the metals at the soil surface. Subsequently, the complexes separate from the soil due to a reduction in their tension, and ultimately, they attach to surfactant molecules (Da Rocha Junior et al., 2019).

Studies have shown that heavy metals can be effectively eliminated from petroleum refinery effluents via biosorption using microalgae biofilms. Using an *oscillatoria* culture, heavy metals including Cu, Co, Pb, and Zn metals were eliminated from sewage and oil industry wastewater by 37–50 %, 20.3–33.3 %, 34.6–100 %, and 32.1–100 %, respectively. *S. bijuga* was able to absorb Cu, Co, Pb, and Zn metals from sewage and petrochemical effluents with efficiency of 60–50, 29.6–66, 15.4–25, and 42.9–50 %, respectively. The two species are shown to be highly efficient in removing heavy metals. Several variables affected the microalgae's metal sorption effectiveness. These included the biosorbent kind, cell physiology, heavy metal concentration and availability, and effluent chemical composition. (Ajayan et al., 2011).

Microalgae and bacterial consortiums create a useful biological adsorption system for treating wastewater effluent with high volume and low content of heavy metals. A group of bacteria and microalgae exhibits symbiotic benefits. Using the photosynthesis process, microalgae absorb carbon dioxide, release oxygen, and transform inorganic materials into organic ones. After that, oxygen-utilizing bacteria metabolize organic substances and oxygen, resulting in the emission of carbon dioxide. To form a consortium, these two systems are required to occur (Zhao et al., 2023).

While numerous studies have investigated the effects of microalgae-bacteria consortiums on the removal of heavy metals from wastewater (Zhao et al., 2023; Bashir et al., 2023), there is a scarcity of study specifically focused on the utilization of microalgae-bacteria consortiums for the removal of heavy metals from petroleum refinery effluents (PRE).

#### 4.3. Production of biosurfactants and bioemulsifiers

The synthesis of biosurfactants and bioemulsifiers was mentioned as a possible explanation for why those microorganisms proved to be appropriate candidates for microbial development in oil recovery and hydrocarbon degradation. They are formed as secondary metabolites by various microorganisms for example bacteria, fungi, and yeast. Biosurfactants are considered amphiphilic biomolecules and are either secreted by microorganisms themselves or exist externally on them (Sharma et al., 2021). The exceptional surface activity of biosurfactants is well-documented, encompassing the ability to decrease surface and interfacial tension among various phases, such as liquid–air, liquid–liquid, and liquid–solid. Adsorption of biosurfactants at various phases enables the reduction of surface and interfacial tension, resulting in enhanced contact and mixing of dissimilar phases (Shaimerdenova et al., 2024).

Biosurfactants are promising techniques that may replace chemical surfactants as alternatives owing to their ecological sustainability, minimal toxicity, biodegradable characteristics, exceptional foaming capability, efficacy in promoting biodegradation and solubilizing compounds with low solubility, and lower critical micelle concentration (CMC) compared to their chemical counterparts. These potential benefits mark them valuable in several applications, including bioremediation, healthcare, food, and petroleum sectors (Jahan et al., 2020).

Forming biosurfactants and emulsifiers has been used as a biological approach that can increase the interaction between bacteria and hydrocarbons (hydrophobic). In addition, they assist in the oil dispersion process, augment the surface area accessible to bacteria for growth, and promote the separation of bacteria from oil droplets once the hydrocarbon resource is consumed (Karlupudi et al., 2018).

Bacteria produce biosurfactants characterized by low molecular weight (LMW), which effectively lower surface and interfacial tensions. Additionally, they produce large polymers with high molecular weight (HMW), which exhibit robust surface adhesion. Glycolipids, disaccharides linked to an aliphatic chain fatty acid, and lipopeptides, are commonly known as low molecular weight biosurfactants. Rhamnolipid, a glycolipid extensively studied, comprises of 2 molecules of fatty acids ( $\beta$ -hydroxydecanoic acid) and 2 molecules of rhamnose sugar. The genus *Pseudomonas*, which includes *Pseudomonas aeruginosa* and *Pseudomonas cepacia*, is the most prolific generator of Rhamnolipids, although other bacterial species including *Serratia rubidaea* and *Lysinibacillus sphaericus* can also create them (Miao et al., 2024).

Bacterial surfactants with high molecular weight (HMW) consist of polymeric surfactants, lipoproteins, lipopolysaccharides, proteins, or intricate combinations of various polymers. These surfactants have lower efficacy in lowering interfacial tension, but they demonstrate high efficiency in covering oil droplets and avoiding their adhesion. They act as an emulsion-stabilizing agent. Bioemulsions produced by numerous *Acinetobacter* species are the biosurfactants that have been studied the most (Fenibo et al., 2019).

As mentioned previously, biosurfactants can be used as a tool for bioremediation of crude oil and heavy metals. A study conducted on discharged wastewater containing oil from Kaduna Refinery and Petrochemical Company shows the formation of biosurfactants. Biosurfactant production has been confirmed by *Pseudomonas* spp., *Bacillus* species, and *Corynebacterium* spp. isolated from PRE. The emulsification activity of these isolates is pH-dependent and Temperature-dependent. Isolates BS1, BS2, BS4, BS6, BS7 and BS11 show to emulsify crude oil. These microorganisms can produce assayable biosurfactants and can be helpful as a bioremediation tool (Ndibe et al., 2019). It has been recognized that *Bacillus* and *Pseudomonas* are the most effective bioremediators. One of the reasons behind that is their ability to make biosurfactants. As a result, a combination of cultures of these bacteria may be capable of decomposing petroleum hydrocarbons, particularly HMW hydrocarbons (Mahmud et al., 2020).

Biosurfactants produced by microorganisms can eliminate heavy

metals from PRE. Chromium from PRE was removed by utilization of two biosurfactants. Two biosurfactants were successfully produced, sophorolipids biosurfactant (SB) which was derived from *Candida bombicola* on cassava, and the second one *E. coli* biosurfactant (EB) was derived from *Escherichia coli* on orange peels. The adsorption efficiency of the biosurfactants EB and SB in removing chromium from refinery effluent was evaluated to determine the maximum capacity. The results demonstrate that the (EB-Zn/AC) exhibited a much greater removal efficiency (99.60 %) compared to the (SB-Zn/AC) (53.98 %) due to its hydrophilicity (Menkiti et al., 2022).

Additionally, fungi can bioremediate contaminants such as total petroleum hydrocarbons (TPH) and naphthalene by using produced biosurfactants. A recent study by Othman et al. examined the efficacy of biosurfactants produced by fungi derived from petroleum sludge in breaking down total petroleum hydrocarbons (TPH). The fungus *Aspergillus fumigatus* (Shu2) exhibited superior potential contrary to *A. terreus* (Shu1). The fungi's breakdown of total petroleum hydrocarbons (TPH) reached approximately  $57 \pm 2$  % within the initial 16-day period and subsequently rose to  $63 \pm 2$  % when provided with more nutrients. The study also discovered that the total petroleum hydrocarbon (TPH) concentration in sludge polluted with hydrocarbons was notably diminished with the implementation of supplementary nutrition treatment (Othman et al., 2022). In addition, Gupta's group extracted *Fusarium proliferatum* WC416 from refinery wastewater and used it to study the decomposition of naphthalene at various doses. It was found that *Fusarium proliferatum* WC416 was able to synthesize a biosurfactant that possessed sophorolipid properties and degraded naphthalene at a maximum of 83 % at a 100 ppm concentration level. *Fusarium proliferatum* WC416 has the potential to be an effective bioremediation tool for naphthalene and ecological cleanup (Gupta et al., 2023).

A comparison of some microbial species involved in biosurfactant production in PRE is provided in Table 2. The comparison shows their unique characteristics, type of biosurfactant, strengths, and limitations. However, many factors could impact the production of biosurfactants such as microbial population ratio, pH, and temperature.

Several microalgae have been known for their biosurfactant

production such as *Arthrospira* sp., *Synechococcus nidulans*, *Chlorella minutissima*, and *Chlorella vulgaris* (Radmann et al., 2015). However, as far as we know, no previous research has investigated the characteristics of biosurfactant-producing microalgae in PRE.

#### 4.4. Nitrogen fixation

Different microorganisms have been known to have a vital function in eliminating nitrogen compounds from refinery wastewater. The existence of these bacteria in refinery effluents underscores their capacity for bioremediation and the management of nitrogen-rich industrial wastes.

Nitrogen-fixing bacteria from petroleum refinery effluents have been investigated by several studies.

*Pseudomonas mendocina* Y7 isolated from refinery wastewater has demonstrated high efficacy in the removal of nitrogen from refinery effluent characterized by a low (C/N) ratio. The bacteria demonstrated a significant capacity to remove nitrogen-containing compounds ( $\text{NO}_2^-$ -N,  $\text{NH}_4^+$ -N, and  $\text{NO}_3^-$ -N). The removal rate was up to 90 % in nitrogen source mediums, single (100 mg/L), and mixed (200 mg/L). The functional genes of the organism were responsible for its capacity to engage in both heterotrophic nitrification and aerobic denitrification processes. The bacterium can live cooperatively with other microbes in activated sludge and continuously remove nitrogen with a high degree of effectiveness (Hu et al., 2021).

Sarkar (2016) discovered a varied community of bacteria in refinery waste that can degrade hydrocarbons, reduce nitrate, and fix nitrogen. *Pseudoxanthomonas* and *Azovibrio* were the prevalence genera that are capable of lowering nitrates and fixing  $\text{N}_2$  within that bacteria community. It was found that the sludge community contained a gene encoding a portion of the nitrogenase enzyme responsible for  $\text{N}_2$  fixing (*nifH*) (Sarkar et al., 2016).

Zhang's study investigated the functional attributes of nitrogen removal bacteria, the community of microorganisms, and the primary nitrogen metabolism pathways in various sludges throughout a range of effluent water quality conditions (both stable and fluctuating). The

**Table 2**  
Comparison of different biosurfactant-producing microbial species including bacteria and fungi.

Microbial communities	Organism	Characteristics	Type of produced biosurfactant	Strength	Limitations	Source
Bacteria	<i>Pseudomonas</i> , <i>Bacillus</i> spp., <i>Burkholderia</i> , <i>Enterobacter</i> , <i>Kocuria</i> , <i>Pandora</i> , etc	–Utilization of aliphatic (C6-C22) and polyaromatic compounds. –Anaerobic growth with multiple terminal electron acceptors.	Not mentioned	–High total petroleum hydrocarbons (TPH) removal (98 %) –High biosurfactant production.	Focus on a specific environment (petroleum refinery waste).	Sarkar et al., 2017
	<i>Bacillus</i> , <i>Pseudomonas</i> , <i>Corynebacterium</i> , and <i>Streptococcus</i> species	Has emulsification activity	Not mentioned	Capable of producing measurable biosurfactants.	pH and temperature dependence	Ndibe et al., 2019
	<i>Candida bombicola</i> and <i>E. coli</i>	–Maximum adsorption efficiencies were indicated at pH 8.0 –Chromium removal –Process was feasible and spontaneous	Synthetic biosurfactant functionalized biocomposites (The sophorolipids biosurfactant (SB) and the <i>E. coli</i> biosurfactant (EB))	High affinity for chromium removal	The efficiency of hydrocarbons removal was not studied	Menkiti et al., 2022
Fungi	<i>Aspergillus terreus</i> isolate Shu1 and <i>Aspergillus fumigatus</i> isolate Shu2	–Produce biosurfactant –Removal of TPH	Not mentioned	<i>Aspergillus fumigatus</i> Shu2 has better TPH removal and biosurfactant productions than <i>Aspergillus terreus</i> Shu1	Nutrients-dependant	Othman et al., 2022
	<i>Fusarium proliferatum</i> WC416	Biodegradation of naphthalene	Sophorolipid	Highest degradation achieved was 83 %	Tested in laboratory scale	Gupta et al., 2023

research revealed that the removal of nitrite and nitrate by this process was comparatively stable, whereas the oxidation of ammonia fluctuated significantly. Nitrogen was predominantly eliminated from the A/O process via a nitrification–denitrification pathway, according to an examination of microbial communities. Most functional genes (~90 %) that were accountable for nitrogen removal were found to be involved in denitrification. Conversely, only 0.17 % of these genes were utilized in the nitrification process to convert ammonia nitrogen (Zhang et al., 2022).

Microalgae have been employed in numerous research investigations to mitigate or eliminate nitrogen from PRE. Znad et al. (2018) conducted a study wherein *Chlorella spp.* demonstrated a reduction of nitrogen by 78 % and a complete elimination of phosphorus within 13 days. Huo's group conducted a study on the filamentous microalgae *Tribonema sp.* They found that the NH<sub>3</sub>-N and P of the two treated PRE were nearly eliminated after 5–7 days (Huo et al., 2018).

The microalga-fungus consortium demonstrated efficacy in nitrogen removal for the treatment of petroleum effluent. In the study conducted by de Andrade et al. (2023), the selected species included *Tetrademus obliquus* LCE-01, a type of microalgae, as well as *Aspergillus niger*, *Penicillium oxalicum*, and *Cunninghamella echinulata*, which are filamentous fungus. The fungus was co-cultivated with microalga, resulting in the removal of larger percentages of nitrogen (in the form of nitrate). Specifically, the removal percentages were found to be 63.4 %, 44.4 %, and 31.7 % for starting concentrations of 25, 50, and 100 mg L<sup>-1</sup>, respectively. Upon testing the actual effluent, the microalga-fungus consortium demonstrated superior efficacy in removing nitrogen compared to the monocultures (de Andrade et al., 2023).

## 5. Challenges, limitations, and opportunities

A range of treatment technologies have been explored for petroleum refinery effluents, with electrochemical methods showing promise. The significance of physical, chemical, and biological treatment processes was underscored by Yuting (2021) and Elamari (2020). Elamari specifically highlighted the necessity of pre-treatment to mitigate the presence of grease, oil, and suspended materials (Yuting et al., 2021; Elamari et al., 2020). According to Ye (2016), the successful removal of phenol and COD was achieved through the utilization of direct electrochemical oxidation, indirect electrochemical oxidation, and electro-phenton methods (Ye & Li, 2016). In a study conducted by Varjani et al. (2019), the authors emphasized the potential of membrane technology, photocatalytic degradation, and improved oxidation processes. The utilization of adsorption, coagulation, chemical oxidation, and biological approaches was further examined by Singh (2018) and Jawad (2019). Notably, the latter study emphasized the application of electrochemical oxidation with graphite anodes (Singh & Shikha, 2019; Jawad & Abbar, 2019). Utilizing a combination of chemical, physical, and biological techniques to tackle the intricate characteristics of petroleum refinery effluents will ultimately aid in the process of remediation. However, it is more costly, challenging to oversee, and challenging to implement on an industrial scale.

Bioaugmentation is an approach that involves the addition of microbial strains or mixed cultures to the contaminated area to enhance the degradation of pollutants (Singh et al., 2020). It is a cost-effective and environmentally friendly process for the treatment of hydrocarbon-contaminated wastewater. This strategy has been used in PRE to enhance oily wastewater and petroleum refinery sludge bioremediation (Hussein et al., 2020; Sarkar et al., 2020). Bioaugmentation in under-regulated settings presents challenges stemming from factors such as biodiversity, competition, substrate competition, and climatic variables. Further research is needed to optimize tactics for bacteria resistance, enzyme activity, and system resilience. The combination of genetic engineering, nanotechnology, and systems biology has the potential to enhance bioremediation. (Muter, 2023).

Omic approaches including metagenomics, metatranscriptomics,

and metaproteomics have been implemented to understand the microbial diversity and function in PRE. By using these approaches, the identity of strains involved in biodegradation will be revealed, for example, key hydrocarbon-degrading microbial communities, such as *Proteobacteria*, *Firmicutes*, *Actinobacteria*, and *Bacteroidetes* (Pal et al., 2019). Those approaches also provided insights into the metabolic potential of these communities, including the degradation of xenobiotic aromatics (Bao et al., 2017; Mukherjee and Chattopadhyay, 2017). Integration of omics technologies helps in a better understanding of microbial physiology and eventually enhances the bioremediation process within PRE.

The bioremediation of petroleum refinery effluents using microbial communities presents a promising sustainable solution to environmental pollution (Imam 2021). However, this process faces numerous challenges. The complexity of petroleum refinery effluents, which contain a variety of organic and inorganic compounds, makes their treatment difficult (Pal et al., 2019). The hydrophobic nature and nutritional imbalance of petroleum refinery sludge further impact the growth of microbial communities and complicate the bioremediation process (Sarkar et al., 2020).

Climate change poses a serious challenge in the bioremediation of PRE. Bioremediation activities in general mostly depend on environmental conditions such as temperature, pH, salinity, pressure, and nutrition availability. Temperature is considered one of the main factors that influence the bioremediation process with higher temperatures generally leading to better degradation of petroleum hydrocarbons (Mekonnen et al., 2024). Although there are many studies on the effect of climate change on the bioremediation process, the research on the impact of climate change on the bioremediation of PRE remains limited. It is important to consider the potential impacts of climate change on the effectiveness of bioremediation in PRE and to develop strategies to mitigate these effects.

Microbial consortia can be designed and used as new biotechnology to serve several tasks in PRE such as enhancing the degradation rate of oil, conversion of chemicals into useful petrochemical products, removal of heavy metals, and nitrogen fixation. Numerous research, have been discussed in this review, investigated the construction and the function of different microbial consortia for example, bacterial consortium (Sarkar et al., 2020; Lukhele et al., 2021; Oaikhena et al., 2016), microalgal-bacterial (Gods'gift and Fagade, 2016; Zhao et al., 2023), bacterial-fungal (Migahed et al., 2016), algal-bacterial (Bashir et al., 2023), microalgal-fungal (de Andrade et al., 2023) consortia. That could be achieved also by genetically engineering microbes to elevate their ability in the bioremediation and conversion process. However, it is a complex task involving challenges and limitations such as monitoring microbial populations, emergence of mutants, and controlling gene expressions (Duncker et al., 2021).

In addition, different parameters in petroleum refinery sites may affect microbial consortia such as seasonal variations, the presence of undesirable bacteria, ecological interaction, and scaling up lab experiments to industrial scale. Long-Term Monitoring (LTM) of remediated petroleum refineries is a critical process that ensures sustainability, and assess the effectiveness of bioremediation. However, PRE requires intensive monitoring and it is time-consuming. LTM is a complex process that poses challenges and limitations when it comes to the cost, controlling the microbial populations, and changing native microbial communities. In addition, it is affected by surrounding environmental factors such as temperature and humidity. Also, frequent introduction of new batches of PRE to the existing field will result in changing the environment and disrupt the existing monitoring process.

## 6. Conclusion and future prospective

Petroleum refineries produce useful petroleum-derived products like gasoline, kerosene, and jet fuel. However, it is contributing to environmental pollution and human health issues by discharging effluents



containing toxic substances. Microbial communities are promising biotechnology that plays a crucial role in altering contaminants in petroleum refinery effluents which include PAHs, hydrocarbons, heavy metals, sulfur, phenols, inorganic complexes, and nitrogen-containing organic compounds. Numerous aspects impact the abundance of these microorganisms such as nutrients, temperature, pH, pressure, salinity, and electron donor/acceptor. This review explores the major roles of microbial communities in petroleum refinery effluents, focusing on bacteria, microalgae, fungi, and yeast, in improving wastewater treatment efficiency and mitigating environmental impacts. The microbial communities are known for their biodegradation ability, hydrophobicity, and strong emulsification capacity making them effective candidates for bioremediation. In addition, biosurfactants and bioemulsifiers are environmentally friendly, have low toxicity, are biodegradable, and are effective in enhancing biodegradation. In addition, the microbes' capacity to fix nitrogen and detoxify heavy metals shows their important role in the bioremediation of PRE.

Many studies have been done on bioremediation and the use of microorganism consortiums to degrade crude oil. However, some key questions and notions are still not discussed in the literature on the ability of microbial consortia to transform crude oil into useful petrochemicals. In addition, analyzing the by-products generated through oil biodegradation using spectroscopic methods. The current body of research also lacks sufficient investigation into the application of microalgae-bacteria consortiums to eliminate heavy metals from petroleum refinery effluents (PRE).

Ongoing investigation in this particular domain is of utmost importance to effectively tackle the urgent environmental issues presented by petroleum refinery effluents, as well as to propel the boundaries of biotechnology and sustainable resource management. This provides a good starting point in the future for petroleum industries to actively considering incorporate biotechnologies in the treatment of PRE. A question raised of future research to investigate the possibility of designing microbial consortia aligning with specific PRE requirements or eliminating one contamination only. In addition, develop bioassays and tools for the assessment of microbial populations directly in PRE. Gaining knowledge of microbial communities and their intricate functionality will provide opportunities for extensive research and the creation of advanced biotechnology tools. These tools may be used to convert crude oil into chemicals and effectively address the pressing environmental concerns arising from the release of waste materials from petroleum refineries.

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## CRediT authorship contribution statement

**Hayfa Habes Almutairi:** Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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