Study on the Treatment Effect of the Oily Wastewater by the Biological Soil Aquifer Treatment System

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Abstract. Soil aquifer treatment with physical, chemical and biological functions for oily wastewater treatment was put forward and strain identification, best growth conditions was studied in this paper, including temperature, pH, diesel oil content and inorganic salt concentrations. Firstly, the oily wastewater treatment effect and the influence factors of the SAT was investigated. Then, the oil degradation bacteria were domesticated, screened and purified and its degradation characteristics were studied and build a biological SAT(bioSAT). Thirdly, the oily wastewater was treated by the bioSAT and the effect was investigated. At last, to confirm the optimal treatment conditions. The research results showed that: 5 bacteria strains were identified as following: Enterococcus faecalis-L1, Lysinibacillus-L2, Bacillus sp-L3, Rhodococcus equi-L4 and Ochrobactrum-L5. Based on their ability to degrade oil, L-2 and L-4 were selected and their growth conditions majorized. Optimum bacteria growth was recorded at 2% substrate (diesel) concentration, pH 7-8, temperature range between 25°C and 30°C and inorganic salt concentration range from 1 g/L to 3 g/L for the bacteria strains L2 and L4. The equilibrium concentration was reached on the 30th day, and the highest removal rates of 84.63% and 99.23% were reached on the 36th day.

Keywords: BioSAT; Oily wastewater; Biodegradation; Petroleum hydrocarbon.

1. Introduction
With the continued increase in oil production and consumption, the amount of petroleum hydrocarbon pollutants released into the environment is increasing day after day. Every year, about $1 \times 10^9$ tons of oil and its products enter the environment through various ways and continue to increase, resulting in more and more serious pollution of petroleum hydrocarbons in surface and ground water environments. On the other hand, with increased industrial development, there is increment in the amount of oil used but various technical and management developments lag behind reasons that are not in any way perfect and release a lot of oil into water causing pollution. Oil pollution is rampant due to the progressive emergence of industries and also lack of proper strategies in place to ensure that the oil produced is treated before it comes into contact with water and cause a more complex pollution. so, to treat the oily wastewater is very complex, because of the oily wastewater from production process of the oil industry, oil refining, oil storage, transportation and petrochemical industries.

Oil pollution is the resultant contamination of water/water bodies because of the introduction of excess oil amounts into these waters[1]. The key contaminants in oily wastewater are petroleum hydrocarbons,
all of which have "Triplet effect" (carcinogenic, teratogenicity and mutagenic) with the most toxic aromatic hydrocarbons, benzene, naphthalene, anthracene and their derivatives and these components have been identified as priority contaminants for monitoring[2].

At the same time, these substances through the long-term enrichment of the food chain once transferred into the human body, it will cause cytoplasmic membrane dissolution, and the human enzyme system caused interference and causing the pancreas and liver and other organ lesions, and thus endanger human health[3].

Therefore, to explore an efficiency oily wastewater treatment technology is an important study for several decades in the environment field. For the oily wastewater treatment, there are common single or mixed physical, chemical methods such as air flotation[4], adsorption[5], filtration[6], flocculation[7], and oxidation-reductionistic[8]. However, most of the study results showed that they can't completely remove oil pollutants by the above physical and chemical methods and we have to combine the biology technology and use the biodegradation process to remove pollutants thoroughly[9].

In this study, A biological Soil Aquifer Treatment (bioSAT) which is a method treating contaminants that adds the degrading bacteria into the popular SAT with the functions of physical adsorption, chemical reaction and biodegradation simultaneously[10]. Antibiotics[11], common organic compounds[12-13], heavy metals[14-15] and inorganic substances etc. can all be processed by the SAT system. K.H. Rekha et al.[16] studied the removal of different kinds of heavy metals (copper, nickel, zinc and hexavalent chromium) by SAT, the results showed that the effect is pretty good, all the heavy metals’ removal rates are above 97%. Xi Zhang et al. [17] isolated a PNP-degrading bacterium and enhanced by SAT, the results showed that bacterium have the ability to degrade PNP below 200 mg L/1and the degrade rate can reach to 99%. The treatment effect and treatment effect influence factors were studied in this study meanwhile. Omer Mienis et al.[18] studied the long-running SAT nitrogen removal project in the Dan area of Tel Aviv, Israel, the study showed total nitrogen removal rates in the upper and lower part of the aquifer was 47-63% and 49-83% respectively. E. V. Karpenko et al. [19] studied the oil degradation bacteria of the genus Rhodococcus enhanced with microbial surfactants, after 192 h, the removal rates of crude oil (2vol %) reached to 84%. From the study carried out by Boqun Liu et al. [20], a hydrocarbon-degrading bacterium Y-1 was isolated, the effect in biodegradation of crude oil was studied, the result showed that in the 5th days, degradation of crude oil reached to 60.2%.

All the above examples prove that the SAT system has a good ability to deal with pollutants, meanwhile, it is feasible to use microorganisms to degrade petroleum. Combining the SAT system with microorganisms will greatly improve the ability to treat petroleum wastewater.

2. Materials and Methods

2.1. SAT Device

In this study, a set of simulation device is simplified and designed as figure1. The SAT device was built using a polyvinyl chloride (PVC) column, with the internal diameter of 5 cm and overall length of 90 cm. Before packing the columns, the soil aquifer media was sieved through a 2 mm mesh device. By so doing, it ensured homogeneous soil column fill material which brought down flow distortions caused by larger soil material and minimized unrepresentative. Meanwhile, the device consists of two functional zones, the upper 40cm was physical adsorption zone and the lower 50cm was the biodegradation one which the oil degradation bacteria mobilization in it, and the two part are separated by double layer 200 mesh stainless steel wire mesh.

![bioSAT device](image)
2.2. Oil Degradation Bacteria Culture Purification Screening and Oil Degradation Characteristics

From the oil-long-term contaminated soil, 5 high efficiency petroleum Degrading Strains were cultured and screened, and the colony morphological characteristics of the them were shown in table 1.

| Strain NO. | Colonies form                     | Bacterial form | Colony Color     | Degradation rate (%) |
|------------|-----------------------------------|----------------|------------------|----------------------|
| 1          | Transparent and smooth            | Spherical      | White            | 30.1 (2)             |
| 2          | Transparent and smooth, pale yellow | Spherical     | Pale yellow      | 36.9 (3)             |
| 3          | Rough colonies, edge jagged state | Spherical      | White            | 29.7 (1)             |
| 4          | Smooth, irregular, purple         | Spherical      | Pale purple      | 38.2 (4)             |
| 5          | Smooth and have neat edges.       | Spherical      | Pale yellow      | 26.8 (0)             |

| Table 2. The Experimental Materials. |
|-------------------------------------|
| Materials & Reagents               | Chemical Formula |
| Magnesium sulfate                  | MgSO₄            |
| Calcium chloride                   | CaCl₂            |
| Potassium hydrogen phosphate       | K₂HPO₄           |
| Potassium dihydrogen phosphate    | KH₂PO₄           |
| Ammonium nitrate                   | NH₄NO₃          |
| Iron (III) chloride                | FeCl₃            |
| Sodium chloride                    | NaCl             |
| Hydrogen peroxide                  | H₂O₂             |
| Ethanol/Alcohol                    | CH₃CH₂OH         |
| Peptone                             |                  |
| Yeast powder                       |                  |
| Beef extract                       |                  |
| Agar                                |                  |
| Gram's Iodine solution             |                  |
| Phenol Red                         |                  |
| Safranin                           |                  |
| Crystal Violet solution            |                  |
| Soluble Starch                     |                  |
| Carbohydrate: lactose, glucose, fructose, maltose and sucrose |          |
| Distilled Water                    |                  |

The strains of 1, 2, 3, 4 and 5 are similar to Enterococcus faecali, Lysinibacillus, Bacillus sp., Rhodococcus equi and Ochrobactrum at 98%, 98%, 97%, 99%, and 98% respectively. So for the strains of 1, 2, 3, 4 and 5, respectively, they are named Enterococcus faecalis s-L1, Lysinibacillus-L2, Bacillus sp-L3, Rhodococcus equi-L4 and Ochrobactrum-L5. It was also established that the bacteria strain L4 (figure2) and L2(figure3) had much higher biodegradation rates for the diesel by 38.2% and 36.9% respectively. So, the L4 and L2 were selected as the experimental strains in this study.
2.3. Effect of the Oily Wastewater Treatment by the bioSAT

OD600 value was used to characterize the concentration of bacterial solution. The petroleum hydrocarbon content was measured by infrared spectrometry methods. The domesticated oil-degrading bacteria solution was slowly passed through the lower 50cm SAT at a flow rate of 0.1m/d until the OD600 value of the inflow and outflow fluids was very close even same which means the Microorganisms were immobilized in the SAT and the bioSAT was built successfully. Then, the oily wastewater flow through the bioSAT by the speed 0.1m/d and the oil concentration of the effluent was tested at any time.

2.4. The Experimental Materials and Instrument

Table 3. The experimental instrument

| Equipment                          | Model                     |
|-----------------------------------|---------------------------|
| Constant temperature incubator    | DNP-9082                  |
| Constant temperature culture oscillator | SKY-200B                |
| pH Meter                          | pH.S.3C                   |
| Ultraviolet-Visible (UV) spectrophotometer | UV-2102PC             |
| General refrigerator              | BCD-216SDX                |
| Ultrapure water machine           | QYSW-10D                  |
| Analytical balance                | AB204-L                   |
| Bio-clean workbench               | SWCJ-2F                   |
| Blast Drying Box                  | DHG9104                   |
| Microscope                        | MJ43                      |
| Low-speed centrifuge              | TGL-16G                   |
| Infrared oil meter                | JH-OIL-8                  |

3. Experiment

3.1. Determination of Growth Curve of Diesel Degrading Bacteria

Screen and isolate bacteria through the ability to metabolize petroleum. Minimal media for purification which the only carbon source is diesel was used. Before being suspended in the same solution, LB media were washed twice with 0.85% NaCl solution, then bacterial cells were cultured overnight in it previously. After autoclaving at 121°C for 15 minutes, aliquot the bacterial culture (100 μL) and transfer to a test tube with 5 ml of minimal medium. Diesel (2%) was then added and incubate at 37°C and shake it at 150 rpm. Minimal media composition (g/L): (NH4)2SO4 (1.0), MgSO4.7H2O (0.1), KH2PO4 (0.5), K2HPO4 (0.76). Composition of trace elements solution (mg/l): ZnSO4 (100), H3BO3 (300), CaCl2.2H2O (134.2), FeSO4.7H2O (2000), CuCl2.2H2O (10), NaMoO4.2H2O (30), NiCl2.6H2O (20), MnCl2.4H2O (30). 1 M NaOH solution was used to adjust the pH to 7. Pure bacterial cultures were streaked on Bushnell Haas agar plates with 2% diesel oil in a 25 ml beaker and incubated at 25°C for 100 days. The OD value was determined after every 10 days using a spectrophotometer reading at 600nm.

3.2. Study on the Best Conditions for Growth of Degrading Bacteria

The best conditions for growth of L-2 and L-4 was conducted in a medium with diesel oil. Spectrophotometer readings at 600 nm after culturing for 7 days were sued to determine bacterial growth.

3.2.1. The influence of temperature on the growth of degrading bacteria. The experiment was carried out using 2% diesel oil concentration at varying temperatures (15°C, 20°C, 25°C, 30°C, 35°C and 40°C) alongside control group at pH 7.

3.2.2. The influence of pH on the growth of degrading bacteria. The effect of pH on growth of L-2 and L-4 was determined with a 100 ml BHM which the only carbon source is 2% diesel. Bacterial inoculums
(100 µl) previously cultured overnight in LB media were inoculated in sterile BHM, which pH values were 5, 6, 7, 8, 9 and 10. 1 M NaOH and 1M HCl solutions were sued to adjust the pH values. The experiment was carried out at 25°C for 7 days in a shaker with a speed of 150 rpm.

3.2.3. The influence of different substrate concentration on the growth of degrading bacteria. The effect of substrate concentration on growth of L-2 and L-4 was determined using sterile BHM at pH 7 added with different concentration of diesel oil i.e., 1, 2, 3, 4, 5 and 6% at 25°C. After culturing for 7 days in a shaker at 150 rpm, bacterial growth was determined.

3.2.4. The influence of different NaCl concentration on the growth of degrading bacteria. To study the role of inorganic salt concentration on degradation, Minimal salt medium added with 2% of diesel oil and different concentrations of NaCl (1 g/L, 1.5g/L, 2.0g/L, 2.5g/L, 3.0g/L, 3.5g/L) for checking the salinity condition. Two hundred and fifty milliliter Erlenmeyer conical flasks were inoculated with 1.5 OD inoculum size of bacterial culture. Bacterial strains were incubated at 25°C along with a shaking condition of 120 rpm. As a control group, flasks without inoculation of strain were kept under similar conditions with inoculated flasks. Samples were collected at regular intervals of time range of 6-120 h.

4. Result and Discussion

4.1. Screening of Crude oil Degrading Bacteria
From the diesel contaminated soil, 5 strains of bacteria were screened. These were the bacteria with the highest capacity to degrade diesel.

It was also established that bacteria strain L2 and L4 had the highest biodegradation rates of diesel, 38.2% and 36.9% respectively. So L2 and L4 were selected as experimental strains in this experiment.

4.2. Identification of Diesel Oil Degradation Bacteria
The physiological and biochemical characteristics of L2 and L4, such as oxidase, starch hydrolysis, nitrogen hydrolysis, glucose oxidation fermentation, contact enzyme, and nitrite, were tested. The results are shown in Table 4.

| Test name         | L2   | L4   |
|-------------------|------|------|
| Aerobic or not    | +    | +    |
| Starch hydrolysis | -    | -    |
| Contact enzyme    | +    | +    |
| Nitrite reduction | +    | +    |

Note: "+" is positive, "-" is negative.

According to the growth morphological characteristics of oil degradation bacteria and physiological and biochemical experimental results, and "common bacterial system identification manual", the preliminary identification of L2 and L4 as Pseudomonas (Pseudomonas sp).

4.3. Growth Characteristics of Diesel Oil Degrading Bacteria
The growth of a population of bacteria under ideal conditions happens in several stages namely lag phase, log/exponential phase, stationary phase, and the death phase. Active metabolic activity occurs during the lag phase, involving DNA and enzyme synthesis, but with zero growth. Population growth occurs during the log or exponential period, when the most pronounced metabolic activity and the cell replication exceeds cell death. After the log phase, the rate of growth slows down, and the rate of cell death equals the growth of new cells. This stage, also known as the stationary phase, involves the creation of an equilibrium in population numbers and the slowing down of individual cells' metabolic activities. The stationary phase indicates a switch in growth conditions, for example, inadequacy of nutrients and/or the accumulation of waste products.
If the rate of cell deaths outnumbers the number of new cells produced, the population equilibrium shifts to a net decrease in numbers and the population enters either the process of death or decline. Population of bacteria can decline until only a few cells remain or die out altogether.

The OD value of the strain suspension was determined by the ultraviolet spectrophotometer, and the enrichment medium of the concentrated strain suspension was properly diluted, so that the OD value of the strain suspension was controlled in the 0-4 range and the OD value was then recorded. Using the strain enrichment culture time as the horizontal coordinate, the OD value measured as longitudinal coordinates, the growth curve of the oil degradation bacteria is shown in figure 4.

As can be seen in figure 4, all the bacteria strains are in the lag phase in the first 5 days. From 5th day, the cell metabolism of all the bacteria strains increased gradually. This resulted to the accelerated rate of synthesis of new cellular substances. For strain 1, 2 and 4, these exponential growths continued until the 40th day. During this time, the metabolic activity of microbial cells is in the best state, the growth and breeding rate of bacteria is accelerating, and the number of microbial cells increases significantly. After the 40th day, the cell metabolism of the 3 strains (1, 2 &4) gradually stabilized and the bacteria entered the stationary phase. It is at this point where the growth rate of bacteria is same as the rate of death and the number of bacteria remains constant. Between the 70th and 80th day, the bacteria entered the decay period where the death rate was higher than the growth of bacteria strains and the number of microbial cells showed a sharp decline trend.

On the other hand, for strain 3 and 5, their exponential growth is realized between 5th and 60th day. After which the two bacteria strains enter the stationary phase. This is where the growth rate of the bacteria is basically the same as the rate of death and therefore bacteria growth remains constant. After the 80th days, the bacteria strains 3 & 4 entered into the death phase. The death rate was significantly higher than the growth rate and therefore there was a sudden decline in the graph.

In summary, L-2 and L-4 had highest rates of biodegradation of crude oil and were used in the next experiment.

4.4. Study of the Influence Factors on the Oil Degradation by the Bacteria Strains

4.4.1. The effect of the bacteria combination on oil degradation. As can be seen in figure 5, the removal rate of both L2 and L4 at 1ml of each is slightly higher than the diesel removal rate for the mixed bacteria at a dosage of 1ml bacterial suspension. This indicates that, 1ml of the mixed bacteria isn’t a significant amount to use for diesel oil degradation. As the dosage of each of the isolates is increased, there is rapid increase the removal rate of diesel oil. Increase in dosage of each particular isolates suggests increase in bacteria allowed to degrade the oil which leads to higher removal rates. At a dosage of 4ml for each bacteria isolate, 100% removal rate of diesel oil is attained. Thus, from the experiment, the optimum dosage for each isolate to remove 2% diesel oil is 4ml bacterial suspension.

On the other hand, the mixed bacterial suspension of L2 and L4 continues to show a continued rapid increase with each dosage of the mixed bacteria added for the degradation of the diesel oil. The 100%
removal rate of diesel oil is achieved at 5ml dosage. Therefore, the optimum dosage for the removal of diesel oil by mixed bacteria is 5ml.

Despite the fact that these 3 bacterial proportions all have a 100% removal rate, it is evident that mono bacteria do not require higher dosages for them to complete degradation of diesel oil. In addition, at 1% bacteria isolate suspension, the mono bacteria had a higher removal rate of diesel oil than the mixed bacteria did.

4.4.2. The effect of the combination of bacteria on diesel degradation versus time. From the figure 6, it is evident that all the three bacteria isolates (L2, L4 and L2+L4) were all able to achieve 100% removal rate of the diesel oil. However, these bacteria had their respective number of days that saw them get there.

For isolate L2, it was able to degrade the diesel completely and thus attain 100% removal rate after 40 days. For isolate L4, it was able to attain 100% removal rate after 50 days. For the mixed bacteria L2+L4, the 100% removal rate was achieved after 60 days. Therefore, it is prudent to say that, isolate L2 has a higher degrading capacity as compared to L4 and the mixed bacteria. It is more effective bacteria for diesel degradation compared to the other two.

On the other hand, bacteria isolates L2 and L4 are more effective when working alone compared to them together. Their removal rates are achieved sooner compared to when they are mixed bacteria.

4.5. Study of the Influence Factors on the Oil Degradation by the Bacteria Strains

4.5.1. The influence of temperature on the growth of degrading bacteria. To determine the effect of temperature on the degradation of diesel oil by L-2 and L-4, experiment was carried out at temperatures between 15°C and 40°C, pH was adjected to 7. As the temperature increases, the biochemical reaction rate and growth rate within the cell increase. It showed a variation in microbial growth at 15°C, 20°C and 25°C for bacteria strain L2 after which the growth started declining as shown in figure 7.

The optimum growth temperature of L2 was 25°C. On the other hand, there was evident microbial growth of bacteria strain L4 at 15°C, 20°C, 25°C, and 30°C. After 30°C, the microbial growth of L4 started declining. The optimum growth temperature of L4 is 30°C according to the experiment.

Obviously, temperature range of between 25°C and 30°C is suitable for L2 and L4 reproduction.

4.5.2. The influence of pH on the growth of degrading bacteria. From the figure 8, there was significant microbial growth between pH 5 and pH 7 for bacteria strain L2 after which the growth started declining with every increase in pH. Strain L4 recorded a significant growth between pH 5 and pH 8 after which the growth of the bacteria strain started declining.

![Figure 8](image_url)

**Figure 8.** The influence of pH on the growth of degrading bacteria. Diesel oil 2%; Temperatures 25°C; pH range 5, 6, 7, 8, 9, 10; Natural salt concentration.

![Figure 9](image_url)

**Figure 9.** The influence of different substrate concentration on the growth of degrading bacteria. Diesel oil 1% and 2%; Temperatures 25°C; pH 7; Natural salt concentration.

![Figure 10](image_url)

**Figure 10.** The influence of different NaCl concentration on the growth of degrading bacteria. Diesel oil 2%; Temperatures 25°C; pH 7; Salt concentration (g/L): 1, 1.5, 2.0, 2.5, 3.0, 3.5.

![Figure 11](image_url)

**Figure 11.** Water inlet and outlet concentration of petroleum hydrocarbon in each cycle. Diesel oil 2%; Temperatures 25°C; pH 7; Natural salt concentration.
The optimum pH for microbial growth of strain L2 and L4 was 7 and 8 respectively. Above pH 7 the growth of L2 was reduced and above pH 8 the growth of L4 was reduced. Isolate L4 however, showed slight tolerance to alkaline pH compared to isolate L2 during the growth period. Therefore, the optimization of pH is very important for the enhanced growth of bacteria.

4.5.3. The influence of different substrate concentration on the growth of degrading bacteria. The results shown in Figure 9. There was a significant difference in bacteria growth at diesel oil concentration of 1% and 2%, after which the bacteria growth started declining. Both bacteria strains L2 & L4 recorded gradual growth observed via spectrophotometer after which the growth started declining. At 2% diesel content, the two strains recorded optimum growth of 3.50 and 3.60. The growth of the bacterial was increased with the increase of diesel concentration. Diesel oil concentration higher than 2% cannot promote the growth of bacteria. The reason of the decrease of bacteria growth may be attributed to stress of hydrocarbons on bacterial.

4.5.4. The influence of different NaCl concentration on the growth of degrading bacteria. Inorganic salt is a crucial part of cells and enzymes, but also for cells to provide desirable permeation pressure, high concentration of inorganic salt will increase the permeation pressure of cells, subdue the activity of various metabolic enzymes of microorganisms and bring down their ability to degrade pollutants. Inorganic salt is a nutrient that affects different organisms in different ways. Effect of NaCl concentration for growth of strains L2 and L4 with diesel oil was evaluated, the results are shown in Figure 10. With the increase of NaCl concentration gradually, the growth of the bacteria in minimal salt medium with 2% diesel oil increased to a certain point where the bacterial growth starts to decline. At 1g/L and 1.5g/L, there is elevated growth of bacteria strain L2. After this point, the growth of these bacteria begins to decline. On the other hand, at 1g/L, 1.5g/L and 2.0g/L, the bacteria strain L4 grows steadily. After this point, the bacteria growth starts to slow down and therefore there is evident decline in the trend. This indicate that variations in NaCl had a great influence on bacterial growth, bacterial increased by degrees when salinity increased until the best condition was achieved. After the optimum salinity was reached, the bacteria growth started to decrease.

5. Removal of Petroleum Hydrocarbon Polluted Wastewater by SAT System

The concentration of petroleum hydrocarbon pollution is measured from the groundwater around an oil field in Gansu Province, and its value is about 40 mg / L. The polluted water was simulated, the concentration was detected and the removal rate was calculated by sat system. The simulated treatment process has four cycles, each cycle of effluent as the next cycle of water. The difference between the water concentration before each cycle treatment and the effluent concentration after treatment is the removal amount, as shown in Figure 11. And calculated as the removal rate. The removal rate of petroleum hydrocarbon in each cycle is shown in Figure 12. In order to prolong the treatment time of SAT, the simulated groundwater temperature is 15 °C, and the simulated flow rate of petroleum hydrocarbon polluted wastewater is 0.12ml/min. Detect the concentration of petroleum hydrocarbon content in the effluent in different periods of each cycle, calculate the removal rate in that period, and make the effect diagram of time in each cycle on the removal rate as shown in Figure 12.

It can be seen from the figure that the petroleum hydrocarbon content will gradually decrease with time in each cycle. Because it is the circulating water to simulate the continuous long-term water inflow process, the concentration of petroleum hydrocarbon in the former stage is equal to that in the latter stage. It can be seen that, with the increase of cycle number, the removal rate of petroleum hydrocarbon decreases gradually, because in the later stage, most of petroleum hydrocarbon substances are removed by microorganisms, the concentration of reaction substrate decreases gradually, and the reaction rate decreases gradually, which leads to the decrease of removal rate. Another very important reason is that in the long-term reaction process, some microorganisms carry out internal respiration and consume microbial activity, resulting in the reduction of removal rate.
Figure 12. Oil hydrocarbon removal rate per cycle. (A. Cycle-1, B. Cycle-2, C. Cycle-3, D. Cycle-4). Diesel oil 2%; Temperatures range: 15°C, 20°C, 25°C, 30°C, 35°C and 40°C; pH 7; Natural salt concentration.

It can be seen from the figure 12 that each cycle of SAT system has different degrees of removal effect for petroleum hydrocarbon pollutants. If the removal rate of microorganisms for petroleum hydrocarbon pollutants in each cycle is superposed, the overall figure of simulated removal effect in the whole process can be obtained. As shown below.

Figure 13. Removal rate of petroleum hydrocarbon in the whole process. Diesel oil 2%; Temperatures25°C; pH 7; Natural salt concentration.

From the whole process chart, it can be seen that the SAT system under the action of L2 and L4 strains reduces the removal rate of petroleum hydrocarbon pollutants gradually, and the SAT system under the action of L4 is significantly stronger than the SAT system under the action of L2. At 30 days, it basically reached equilibrium concentration, and the removal rate of petroleum hydrocarbon pollutants by SAT system under the action of L2 and L4 reached 83.1% and 98.35%, respectively. At 36 days, 84.63% and 99.23% respectively. It is proved that the application of L2 and L4 strains in SAT system has excellent effect on the treatment of petroleum hydrocarbon polluted wastewater.

6. Conclusions

1) Using artificial enrichment cultures, it is possible to screen and isolate diesel degrading bacteria. This was possible as the 5 strains of bacteria were identified using culture media. The 5 strains of bacteria were identified and their morphological, physiological and biochemical characteristics established. Some bacteria strains are more efficient and more effective as compared to others; bacteria strain L2 and L4 stood out from the others. Their removal rates were significantly higher compared to the other strains.

2) The optimum growth condition of L2 and L4 are studied, under these conditions, all the bacteria growing well and indicators reached the best condition, equilibrium concentration on the 30th day, the degradation rate of the two strains reached the highest, reaching 84.63% and 99.23% on the 36th day.

3) Previous work only did research on degrading bacteria, and did not have a platform that can be combined and applied. This study combines biodegradable bacteria with the SAT system, which effectively improves the treatment effect and the breadth of application.

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