Research Article

Performance Evaluation of Bacterial Consortia from Low-Permeability Reservoir in Ordos Basin

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The combination of strains of different species and genera may enhance the effects of single bacteria, surpass the tolerance upper limit, and optimize the viscosity reduction and degradation. In this study, six strains were isolated in low permeability layers of the Ordos Basin and were combined to verify the effect of the consortium strains. The selected single strains have good emulsifying and viscosity-reducing effects, but their degradation components are different. SC4561 (Bacillus cereus), SC4551 (Bacillus sp.), and H-1 (Brevibacillus sp.) form consortium A, and SC4534 (2) (Bacillus sp.), SC4542 (Bacillus licheniformis), and A-3 (Bacillus licheniformis) form consortium B. The performance of the mixed strains was evaluated by the analysis of change in emulsification rate, crude oil composition, viscosity, and the tolerance (temperature, salinity, and pH) through GC-MS, rotational rheometer, and other methods. The results showed that the temperature tolerance of the consortium strains was improved by 5-7°C. Consortium B had higher emulsibility ($E_{24}$ was higher than 40% in average) and viscosity degradation (above 35%), and the crude oil in consortium B has almost no wall adhesion. The components of crude oil that consortia use were still diverse, including both long- and short-chain hydrocarbons. However, the proportion of long-chain n-alkanes is further reduced compared with that of single bacteria, and the highest ratio was reduced by 23.81% (B-ALL). At the same time, they also had effects on aromatic hydrocarbons with complex structures (phenanthrene and phenanthrene). This research confirms the enhanced effect of consortium bacteria on single bacteria, facilitating the implementation of microbial enhanced oil recovery technology in the future.

1. Introduction

After conventional primary and secondary oil recovery operations, more than two-thirds of the residual oil still remains in the reservoir [1]. Microbial enhanced oil recovery (MEOR) technology is one of the low-cost and high-efficiency methods in tertiary oil recovery. It utilizes microorganisms and their metabolites in the reservoir to alter the physical properties (viscosity, wettability, and interfacial tension) and composition of crude oil. This can facilitate the release of crude oil from the rock pores and enhance the recovery [2–4]. However, oil and gas reservoirs are unique deep underground ecosystems [5]. Additionally, crude oil is structurally complex and contains saturated hydrocarbons, aromatic hydrocarbons, nonhydrocarbons, and trace organic metallic substances, with compositions varying from one oilfield to another [6]. Numerous studies have shown that individual bacteria evaluated in MEOR exhibit varying degrees of preferential degradation towards different components in crude oil [7–10]. Therefore, some scholars pointed out the necessity of studying the bacterial consortium, which may strengthen their effects [3, 11].

In 2002, Rahman et al. [12] confirmed that a consortium of five mixed bacteria achieved a degradation rate of 78% at 1% crude oil concentration after 20 days of incubation, surpassing the degradation rates of individual bacteria. Hao
et al. [13] found that the consortium bacteria SF67, isolated from the Daqing oilfield, reduced viscosity by 35.6% and surface tension by 33.6% under conditions of 60°C. Aboelwafa et al. [14] utilized gas chromatography to analyze the effects of a composite bacterium composed of Pseudomonas aeruginosa, Bacillus subtilis, and Acinetobacter lwofii on Egyptian crude oil. After 28 days, the composite bacterium achieved a maximum degradation rate of 88.5%. Chen et al. [15] conducted displacement tests using natural core samples under conditions of 0.32 to 0.50 PV (pore volume) and achieved an incremental recovery rate of 5.3%, confirming the recovery-enhancing effects of composite bacteria. However, in recent years, some scholars have proposed that the effect of the bacterial consortium will be hindered by the influence of the environment and antagonistic interactions among bacteria [16]. There is still controversy on whether consortium bacteria can improve oil recovery.

In our previous research, we screened six single bacteria (mainly Bacillus sp.) in low-permeability oil wells. These bacteria effectively emulsify and reduce their viscosity, but the strains obtained are of various species, and the components used by the strains for crude oil are also different [17]. Therefore, it is important to study whether the mixture of strains will complement each other features, which has rarely been reported in low-permeability oil fields before.

Therefore, this study has two objectives: to evaluate the effects of composite bacterial strains composed of individual bacteria obtained from low-permeability oilfields on crude oil and to investigate whether there are antagonistic or synergistic interactions among these single bacteria. Based on the completed single-bacteria evaluation results, two consortia were designed, and each consortium consisted of three strains (SC4542, SC4534(2), SC4561) and one strain (SC4542, SC4534(2), SC4561). The performance of the mixed strains was evaluated by the analysis of changes in growth rate, emulsification rate, crude oil composition, viscosity, and tolerance (temperature, salinity, and pH) through GC-MS, rotational rheometer, and other methods. The obtained consortium bacteria can better enhance the single bacterial effect and apply it better in MEOR.

2. Methods and Materials

2.1. Bacteria and Oil Sources. The crude oil sample was obtained from Well Anjia 162-411, 154 Block in Hujianshan oilfield (36°49′-37°53′N, 107°35′-108°22′E) [18], Ordos Basin. The sampling layer is Chang 4+5, and the permeability is 3.6 mD. Low-permeability reservoirs are those with permeability less than 50 mD [19], making Chang 4+5 a typical low-permeability layer. The temperature of the sampled oil well is about 40-50°C, and the salinity is 37.48 g/L. The specific essential information on the geological and geochemical factors is shown in [17].

The strains used in this study were previously screened from produced water samples from Chang 4+5 and crude oil samples from Chang 6. We obtained strains through enrichment and streaking. Six strains in Table 1 were selected based on their ability to produce biosurfactants, temperature tolerance, and biodegradation abilities. The specific screening method is described in detail in Bian et al. [17]. The basic characteristics are shown in Table 1. All terms used in this study can be found in the Nomenclatures table (S1).

2.2. Culture Media. Two kinds of culture media were used in this study. One is the nutrient broth medium, which was used for the tolerance test, growth rate, and emulsification rate tests. The components were as follows: g/L, beef extract 3.6; peptone 10; CaCl₂ 5.6 (the amount of CaCl₂ in the salinity tolerance test was changed according to the experimental design). The other is MSM medium with crude oil as the sole carbon source, which was mainly used in tests such as viscosity, contact angle, and component change. The components were as follows: g/L, NaNO₃ 1.5; (NH₄)₂SO₄ 1.5;
K2HPO4 1; MgSO4 0.5; KCl 0.5; CaCl2 0.002; FeSO4 0.001. Crude oil as carbon source was added separately (10%, v/v) [20]. Errors during the preparation of the culture medium may lead to inaccuracies in the determination of growth curves and tolerance tests.

2.3. Strain Combinations. Two groups of strain combinations were designed by taking into account the biosurfactant-producing ability, degradation ability, and species of the single strains. Combination A mainly considers the effect of the combination of strains that degrade different components, while combination B verifies the superposition of the emulsification effect. Consortium A was composed of SC4561, SC4551, and H-1. Three strains of *Bacillus* belong to different species, *Bacillus cereus*, *Bacillus* sp., and *Brevibacillus* sp., respectively. H-1 degraded the short chain but had no effect on naphthalene, while SC4561 and SC4551 did the opposite [17]. Research has also shown that bacterial interactions can promote the production of biosurfactants [21]. Consortium B was composed of SC4534(2), SC4542, and A-3. These three strains have a strong ability to produce biosurfactants, and A-3 and SC4542 have the best degradation effects. Each consortium has four combinations. The strains in every combination were 1:1 and 1:1:1 (Table 2).

2.4. Growth Monitoring (Growth Rate). The monitoring method for growth was improved according to Zhang et al. [22]. Each combination was inoculated in 200 ml medium at 1% inoculum volume and then incubated for 48 hours.
Figure 3: Continued.
at 45°C and 160 rpm [22]. The optical density (OD$_{600}$) was measured via UV-vis spectrophotometry (Spectrumlab 752pro, USA) at two-hour intervals in the first 24 hours and at four-hour intervals in the second 24 hours.

2.5. Sensitivity Test. Temperature, salinity, and pH have been proven to have a great influence on microbial propagation in oil reservoirs [23, 24]. Testing the growth of strains at different temperatures, salinity, and pH can evaluate their tolerance to extreme environments. The testing methods were improved according to [25, 26].

The temperature test set eight values between 25°C and 60°C, including 25, 35, 40, 45, 50, 55, 57.5, and 60°C. The temperature interval between 40 and 60°C is smaller to obtain the optimal culture temperature. The culture temperature was fixed at 45°C and then change the value of salinity to test the salinity tolerance. The set range was wide, from 1 g/L to 50 g/L. The pH sensitivity test was also carried out at a fixed temperature of 45°C and CaCl$_2$ concentration of 5 g/L. The pH value is set from 4 to 10, with a step of one. All cultures were conducted for three days, and the concentration of the bacterial solution was measured using an OD$_{600}$ value. All OD$_{600}$ values were tested three times, using the mean ± standard deviation as the final value.

2.6. Evaluation of Emulsification

2.6.1. Emulsification Rate ($E_{25}$). Three milliliters of n-hexane and three milliliters cell-free supernatant (8000 rpm, 10 min) were vortexed and then ultrasonic shocked (Xinyi SB-4200D, China) for 10 min. The emulsified layer height was measured after 24 hours. The emulsification rate ($E_{25}$) was determined by dividing the emulsification layer height by the total height [27]. Each sample was tested three times, and the results were calculated as the mean ± standard deviation.

2.6.2. Crude Oil Emulsification. One hundred milliliters of MSM medium with ten milliliters of sterile oil and five milliliters of strain solution were inoculated at 160 rpm, 45°C for 7 days. The degree of emulsification and wall adhesion of crude oil were observed and recorded in detail.

2.6.3. Contact Angle. The mixed strains were inoculated into 100 ml of MSM medium at 5% inoculum at 160 rpm, 45°C for 7 days. Ten microliters of emulsified crude oil was taken and dropped into the center of the cover slide. The experimental device for measuring contact angle is self-designed and includes a fixed circular light source, a glass slide, and a micro lens (Kudianxing, 25X, China). The glass slide is made of glass material, and the light source was fixed behind the slide. A macro lens was used to photograph the oil droplets, and the edges of the droplets were clear and horizontal. The size of the droplets occupied at least 1/4 of the page. Each group took three pictures. Then, LB-ADSA and Drops-nake methods of drop analysis in ImageJ software (Figure 1) were used to measure the contact angle [28].

2.7. Viscosity. The mixed strains were inoculated into 100 ml of MSM medium at 5% inoculum at 160 rpm, 45°C for 7 days. The culture medium was set at 25°C for half an hour, and the upper layer of crude oil was taken by a sterilized weighing spoon and divided into a five-milliliter centrifuge tube with three parallel samples. Then, the crude oil was left overnight to separate oil and water under the action of gravity. The change in crude oil viscosity was measured by a rotational rheometer (Anton Paar MCR 302, Austria). About 1 ml of oil was placed in the sample tray. The test temperature of the rheometer was 42°C, the rotor was dpp25, the shear rate was 50/s, and the test time of each sample was 120 s. The viscosity reduction ratio was calculated through the formula in Bian et al. [17]. Due to the emulsification process, oil and water may mixed, resulting in impurities in the collected oil samples. This impurity may introduce errors in viscosity measurements.

2.8. Crude Oil Degradation. The crude oil after the action of strains was extracted by n-hexane. The family compositions of crude oil were separated according to the China oil and

![Figure 3: Tolerance of consortium A to temperature, salinity, and pH (a) 51+61; (b) 61+H-1; (c) 51+H-1; (d) A-ALL.](image-url)
Figure 4: Continued.
gas industry standard (SY/T 5110-2016). Then, the family components were determined by GC-MS (Thermo Fisher ISQ7000, USA). The methods used were derived from national standards (GB/T 18606-2017). The detailed process was shown in [17, 29]. The content of each component was analyzed after normalization, and the normalization formula (1) is as follows:

\[
\text{Relative abundance(\%)} = \frac{P_i}{P_A} \times 100\%.
\]

\(P_A\) is the sum of the peak areas of all components; \(P_i\) is the peak area of a certain component.

### 3. Results and Discussion

#### 3.1. Growth Rate

The growth of consortium strains for 48 hours was tested, and the results are shown in Figure 1. The strain grew rapidly in the first 2 hours, the OD\(_{600}\) value of consortium A reached 0.8 at 16 hours (Figure 2(a)), and the value of consortium B reached 0.8 at 14 hours (Figure 2(b)), indicating that they had entered the logarithmic growth phase in a short time. The OD\(_{600}\) value of consortium B began to decline after 48 hours of incubation, indicating that the incubation time should not be too long in the application. Compared with the growth curve of the petroleum-degrading single bacterium previously obtained by Chen et al. [25], the composite bacteria have almost no lag phase, indicating a shorter adaptation period to the environment. Additionally, they enter the exponential growth phase faster. Both of these characteristics indicated the rapid proliferation of the consortium bacteria.

The OD\(_{600}\) value of consortium B began to decline after 48 hours of incubation, indicating that the incubation time should not be too long in the application. Compared with the growth curve of the petroleum-degrading single bacterium previously obtained by Chen et al. [25], the composite bacteria have almost no lag phase, indicating a shorter adaptation period to the environment. Additionally, they enter the exponential growth phase faster. Both of these characteristics indicated the rapid proliferation of the consortium bacteria.

#### 3.2. Tolerance of Strains

The growth trend of strains growing in conical flasks was similar to that in tested tubes, but the growth rate of strains in conical flasks is significantly faster due to the increase of nutrients. Larger cultures in extreme environments were better for reproduction. The tolerance of combination A was significantly improved in three dimensions, while that of combination B was only improved in salinity tolerance. The four combinations in consortium A had higher OD\(_{600}\) values (≥1.0) when the temperature was higher than 55°C. Compared with single bacteria, the tolerance above 50°C was improved [17], and the range of the optimum growth temperature also increased (Figure 3). All combinations in consortium B had a good growth rate between 25 and 45°C, but the OD\(_{600}\) was low when the temperature was higher than 50°C (Figure 4). It had a poor temperature tolerance compared with consortium A. The average temperature of the oil wells in this region is 40-50°C. The experimental verification of temperature tolerance has demonstrated the favorable adaptability of the consortium bacteria to the oil well environment. Furthermore, most studies on petroleum microorganism screening have been conducted at relatively low temperatures (28-42°C) [30, 31]. However, in order to better simulate reservoir conditions, all subsequent tests and cultivations in this study were set at 45°C.

In the salinity experiment, all four combinations could survive when the salinity was less than 50 g/L. Among them, the optimum salinity of 51+61 and 61+H-1 was 30 g/L.
(Figures 3(a) and 3(b)), and the peak value of 51+61 and 61
+H-1 was later than that of single bacteria. For the other two
groups, 5 g/L was the best (Figures 3(c) and 3(d)). The opti-
mum salinity of 34+A-3 and B-ALL was 15 g/L (Figures 4(b)
and 4(d)), and the other two groups had the highest OD_{600}
value at 5 g/L (Figures 4(a) and 4(c)). Both groups of con-
sortium bacteria salt tolerance were superior to halophilic
microorganisms previously reported [32–34]. They can rap-
idly reproduce under high salinity conditions and can be
considered as salt-tolerant microorganisms. Under high
salinity conditions, these microorganisms consume more
energy to balance cell osmotic pressure, resulting in a
decrease of energy for growth and reproduction capacity
[35, 36], leading to lower OD values.

The consortium A preferred an alkaline environment,
and the optimum pH was 8. However, consortium B

<table>
<thead>
<tr>
<th>Combinations</th>
<th>Contact angle (°)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>93.27 ± 5.91</td>
</tr>
<tr>
<td>SC4561+SC4551</td>
<td>81.59 ± 0.68</td>
</tr>
<tr>
<td>SC4561+H-1</td>
<td>112.28 ± 1.12</td>
</tr>
<tr>
<td>SC4551+H-1</td>
<td>107.15 ± 0.74</td>
</tr>
<tr>
<td>A-ALL</td>
<td>87.82 ± 0.45</td>
</tr>
<tr>
<td>SC4534(2)+SC4542</td>
<td>90.68 ± 1.13</td>
</tr>
<tr>
<td>SC4534(2)+A-3</td>
<td>80.68 ± 0.27</td>
</tr>
<tr>
<td>SC4542+A-3</td>
<td>89.53 ± 1.21</td>
</tr>
<tr>
<td>B-ALL</td>
<td>72.79 ± 0.58</td>
</tr>
</tbody>
</table>

Figure 5: Wall adhesion of crude oil after action.

<table>
<thead>
<tr>
<th>Strains</th>
<th>Viscosity (mPa.s)</th>
<th>Viscosity reduction ratio (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>120</td>
<td>0</td>
</tr>
<tr>
<td>SC4561+SC4551</td>
<td>120</td>
<td>0</td>
</tr>
<tr>
<td>SC4561+H-1</td>
<td>120</td>
<td>0</td>
</tr>
<tr>
<td>SC4551+H-1</td>
<td>120</td>
<td>0</td>
</tr>
<tr>
<td>A-ALL</td>
<td>120</td>
<td>0</td>
</tr>
<tr>
<td>B-ALL</td>
<td>120</td>
<td>0</td>
</tr>
</tbody>
</table>

Figure 6: The change of viscosity after the action of combination strains.
Figure 7: Continued.
preferred a weakly acidic environment and grew fastest at pH 5 (Figure 4).

3.3. Emulsibility

3.3.1. $E_{24}$ The results of $E_{24}$ are shown in Table 3. The emulsification rate of bacteria combinations was higher than 30% except for SC4551+SC4561. A-ALL and B-ALL got the highest $E_{24}$ among the test combinations, 48.31% and 47.73%, respectively. The study conducted by Nayarisseri et al. [31] indicated that an emulsification rate exceeding 30% can be considered as a positive response to biosurfactant. On the whole, the emulsification rate of consortium B was higher than that of consortium A.

In addition, the culture media after the strain action were observed to obtain the emulsified state. The phenomenon videos were added to the supplementary material (S2). It was found that the color of the medium was brown, indicating that the crude oil and water phases had dissolved [37]. The emulsified crude oil droplets are smaller. Compared with single bacteria, the emulsification rate of complex bacteria did not increase significantly, but macroscopically, the oil droplets formed by emulsification were smaller, and the emulsification effect was better. This may be because, in the $E_{24}$ test, the supernatant was only mixed with n-hexane, while the crude oil is a mixture. The strains had effects on different components in the crude oil, which made the results different. Consortium B almost has no wall-hanging phenomenon after shaking, whereas consortium A, except for SC4561+H-1, still adhered to the bottle wall (Figure 5), indicating that the emulsification of consortium B was more obvious, which was consistent with the results of the $E_{24}$ experiment.

The emulsification of the consortium strains was due to the existence of biosurfactants. As amphiphilic molecules,
it was soluble in water and oil, resulting in emulsified crude oil on the macrolevel [38, 39]. *Bacillus* has been shown to produce lipopeptide biosurfactants [40, 41], and all single strains involved in the combinations have been shown to produce biosurfactants in previous experiments.

The combinations A-ALL and B-ALL had the best emulsifying effect, possibly because the three strains produced more biosurfactants than other combinations. Some researchers even used genetic engineering to increase the biosurfactant yield of single bacteria to achieve higher emulsifying efficiency [42, 43]. The combination of strains is another simple way to improve efficiency. Nnabuife et al. [11] pointed out that when all strains have the ability to produce biosurfactants, the consortium formulation was unaffected. However, in this study, combination B of single bacteria with a better ability to produce surfactant showed a better emulsification effect, and many studies also took this as a measure of the effect of consortium bacteria [44, 45]. Therefore, in the consortium formulation, strains with strong surfactant production ability should be combined to achieve the best emulsification effect.

### 3.3.2. Contact Angle

The contact angle can reflect the wettability of liquid to solid [46]. Wettability is closely related to surface tension, and the decrease in surface tension will reduce the contact angle [47]. Table 4 shows the results of the contact angle of emulsified oil. Compared with the control group, the contact angle of crude oil after 7 days of action was decreased except for combination 61+H-1 and 51+H-1, indicating a decrease in surface tension. After the treatment, the contact angle of B-ALL was measured to be 72.79° ± 0.58. Compared with the previously screened single bacterial contact angle by Sharma et al. [27], which can be changed to 76.51° ± 1.06, the reduction effect was more significant. The lower surface tension can reduce the adhesion work of crude oil on the core pore surface and make the oil droplets more easily deform and fall off [48]. The reduction of consortium B was higher than that of consortium A, which was consistent with the emulsification test results. However, the contact angle of the combination 61+H-1 and 51+H-1 increased, which was not consistent with the results of the emulsification experiment. The reason for this phenomenon was speculated to be related to the viscosity of crude oil. Although the viscosity decreased after the action of the strain, it still cannot reach the degree of free dripping. Therefore, there may be pulling in the process of adding the amount of crude oil to make the oil drops deformed.

### 3.4. Viscosity

The change in viscosity is shown in Figure 6. The green columns represented consortium A, and the yellow columns represented consortium B. The broken line was the viscosity reduction rate. Compared with the control group, the viscosity of the experimental group decreased. Except for combination 61 + 51, the viscosity reduction rate of other combinations was above 30%. Comparison between consortia showed that the viscosity reduction rate of consortium B (upon 35%) was higher than that of consortium A, and combination SC4542+A-3 promoted the highest reduction (41.54%).

The decrease of crude oil viscosity under the action of microorganisms has two mechanisms: emulsification and degradation [1]. Emulsion viscosity reduction mainly used the unique amphiphilic properties of biosurfactants to adsorb on the rock surface or oil-water interface to achieve the effect of wetting reversal, reducing the tension of the
Figure 9: Continued.

(a) Naphthalene
- Control
- 61+51
- 61+H–1
- 51+H–1
- A–ALL

(b) Phenanthrene
- Control
- 34(2)+42
- 61+51
- 61+H–1
- 51+H–1
- A–ALL

(c) Chrysene
- Control
- 34(2)+42
- 61+51
- 61+H–1
- 51+H–1
- A–ALL

(d) Phthalene
- Control
- 34(2)+42
- 61+51
- 61+H–1
- 51+H–1
- A–ALL

Relative abundance (%)
oil-water interface [41, 49, 50]. In the process of degradation, microorganisms converted long-chain and macromolecular components into short-chain and small-molecular components, reducing the composition of heavy components in crude oil and increasing fluidity [51, 52]. Therefore, consortium B with stronger emulsification ability (see Section 3.3.1) was superior to consortium A in viscosity reduction.

3.5. Change of Components

3.5.1. Saturated Hydrocarbon. After the action of combinations 61+51, A-ALL, 42+A-3, and B-ALL, the content of long-chain n-alkane \((C \geq 22)\) decreased, and the content of short-chain hydrocarbon increased (Figures 7(a), 7(d), 7(h), and 7(i)). A-ALL and B-ALL had the best degradation effect on long-chain hydrocarbons among all combinations, especially B-ALL, which could be reduced by 23.81\% based on single bacteria (Figure 7(j)). The other combinations mainly used short-chain hydrocarbons. In the single-bacteria experiment, SC4542 and H-1 mainly used short-chain alkanes, while in the mixed bacteria test, except 42+A-3, A-ALL, and B-ALL, the combinations including these two strains still mainly degraded short-chain alkanes (Figures 7(b), 7(c), 7(f), and 7(g)). The alkanes were not only consumed. Some scholars also pointed out that the increase in the content of short-chain alkanes may also be caused by the breakage of long-chain hydrocarbons [2, 53]. From the perspective of the content of long-chain hydrocarbons, the rate in the above combinations has decreased, indicating that the consortium bacteria still enhance the degradation of the strain (Figures 7(e) and 7(j)).

The combinations 42+A-3 and 34(2)+A-3 were special. Although 42+A-3 contains the SC4542 strain, the combination shows the degradation of long-chain hydrocarbons (Figure 7(h)). On the contrary, the combination 34(2)+A-3 mainly degraded short-chain hydrocarbons, while the single strains used long-chain hydrocarbons (Figure 7(g)). The results showed that the interaction between strains was complex, and there was mutual promotion and restriction. However, this paper only evaluated its performance and did not explore the mechanism. It needed to be studied from the perspective of its metabolism and metabolic pathway.

In addition to the degradation of n-alkanes, the consortium strain also has degradation effects on sterane (Figure 8(a)) and hopane in naphthenes (Figure 8(b)). Compared with single bacteria, compound bacteria had certain effects on hopane with a more complex structure, and B-ALL had the best effect.

3.5.2. Aromatic Hydrocarbon. The effect of consortium bacteria on aromatic hydrocarbons was mainly manifested in naphthalene, phenanthrene, and chrysene. After the action of single bacteria, the main degradation of aromatic hydrocarbons is dimethyl naphthalene, while the compound bacteria could degrade tetramethyl naphthalene \((C_{14}H_{16})\) with a more complex structure (Figure 9(d)) and make the content of dimethyl-naphthalene \((C_{12}H_{12})\) increase (Figure 9(a)), indicating that the components acted by consortium bacteria were more complex. In addition, the consortium bacteria also had effects on phenanthrene (Figure 9(e)) and chrysene (Figure 9(f)). Consortium bacteria could partially degrade methyl-phenanthrene \((C_{13}H_{12})\) (Figure 9(b)), and methyl-chrysene \((C_{19}H_{14})\) (Figure 9(c)). The increase in the relative content of dimethyl naphthalene may also be due to the degradation of more complex aromatic hydrocarbons. Results have confirmed that the synergism of strains could improve the degradation of aromatic hydrocarbons, and similar effects of consortium bacteria have been confirmed by Zhang et al. [54].

4. Conclusions

This research studied the performance of consortium bacteria from low-permeability reservoir. Based on the evaluation of single bacteria, two groups of consortium strains were designed. The specific conclusions and understanding are summarized as follows:

(1) Consortium B and consortium A have a higher tolerance to temperature \((\leq 55 ^\circ C)\) and salinity \((\leq 50 \text{g/L})\) compared with single bacteria

(2) Consortium B has better emulsification, and the viscosity reduction rate of consortium B was higher (upon 35%)

(3) The degraded components still are different, but the abundance of long-chain n-alkanes decreased, and the interactions between strains in the consortium strains were complex, not simply compensated

This study confirmed the advantages of the consortium strains. Consortium bacteria could enhance the role of
strains in a low-cost and easier operation way. This has great significance for improving the efficiency of applications in the field. Further research that focuses on cometabolism mechanism of consortium bacteria from the aspects of metabolic pathways is necessary to further understand the application of consortia.

Data Availability

The geological and geochemical data used to support the findings of this study are included within the article.

Additional Points

Highlights. (i) The consortium microbiota indeed enhances the effect of single bacteria, but the interactions between strains in the consortium strains were complex, not simply compensated. (ii) The temperature tolerance of the consortium strains increased by 5-7°C, and the emulsification was stronger. (iii) The degradation ability of long-chain hydrocarbon was increased by up to 23.8%. (iv) The more complex structure of aromatic hydrocarbons (chrysene and phenanthrene) was degraded.

Disclosure

Part of the material has been used in conference EGU (doi:10.5194/egusphere-egu23-1627).

Conflicts of Interest

The authors declared that they have no conflicts of interest in this work. We declare that we do not have any commercial or associative interest that represents a conflict of interest in connection with the work submitted.

Authors’ Contributions

Bian Ziwei was responsible for the conceptualization, formal analysis, methodology, visualization, writing—original draft, and writing—review and editing. Zhi Zena was responsible for the methodology, conceptualization, and writing—review and editing. Zhang Xiangchun and Qu Yiqian were responsible for the investigation and methodology. Wei Lusha was responsible for the investigation and funding acquisition. Wu Hanning and Wu Yifei were responsible for the conceptualization, project administration, funding acquisition, and writing—review and editing.

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Supplementary Materials

Supplementary 1. Nomenclatures table.

Supplementary 2. Videos of crude oil status after consortium strains action (uploaded on OneDrive) https://1drv.ms/f/s!Al7RYaN29O13gRoGywunYNMymsnsp9o?e=I3Ohc1.

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