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Review Article

Recent Advance of Microbial Enhanced Oil Recovery (MEOR) in China

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Compared with other enhanced oil recovery (EOR) techniques like gas flooding, chemical flooding, and thermal production, the prominent advantages of microbial enhanced oil recovery (MEOR) include environment-friendliness and lowest cost. Recent progress of MEOR in laboratory studies and microbial flooding recovery (MFR) field tests in China are reviewed. High biotechnology is being used to investigate MFR mechanisms on the molecular level. Emulsification and wettability alternation due to microbial effects are the main interests at present. Application of a high-resolution mass spectrum (HRMS) on MEOR mechanism has revealed the change of polar compound structures before and after oil degradation by the microbial on the molecular level. MEOR could be divided into indigenous microorganism and exogenous microorganism flooding. The key of exogenous microorganism flooding was to develop effective production strains, and difficulty lies in the compatibility of the microorganism, performance degradation, and high cost. Indigenous microorganism flooding has good adaptation but no follow-up process on production strain development; thus, it represents the main development direction of MEOR in China. More than 4600 wells have been conducted for MEOR field tests in China, and about 500 wells are involved in MFR. 47 MFR field tests have been carried out in China, and 12 field tests are conducted in Daqing Oilfield. MFR field test's incremental oil recovery is as high as 4.95% OOIP, with a typical slug size less than 0.1 PV. The input-output ratio can be 1 : 6. All field tests have shown positive results in oil production increase and water cut reduction. MEOR screening criteria for reservoirs in China need to be improved. Reservoir fluid, temperature, and salinity were the most important three parameters. Microbial flooding technology is mature in reservoirs with temperature lower than 80°C, salinity less than 100,000 ppm, and permeability above 5 mD. MFR in China is very close to commercial application, while MFR as quaternary recovery like those in post-polymer flooding reservoirs needs further study.

1. Introduction

Oil and gas remain the main primary energies in the world. Enhanced oil recovery involves how to recover as most original oil in place (OOIP) as possible economically. According to the development stage, it can be divided into primary recovery (natural energy development), secondary recovery (water injection or gas injection to main reservoir pressure), and tertiary recovery [1]. Tertiary recovery is also known as enhanced oil recovery (EOR), which includes polymer flooding, surfactant flooding, gas flooding, thermal production, and microbial enhanced oil recovery (MEOR). The EOR process has two basic features: (a) effectiveness of recovery of more oil and (b) relatively low cost. MEOR is believed to be the cheapest EOR process. To get the goal of highest economical recovery, sound understanding of the basic mechanisms of enhancing oil recovery is necessary, which is sometimes not available to nonpetroleum engineers. Thus,

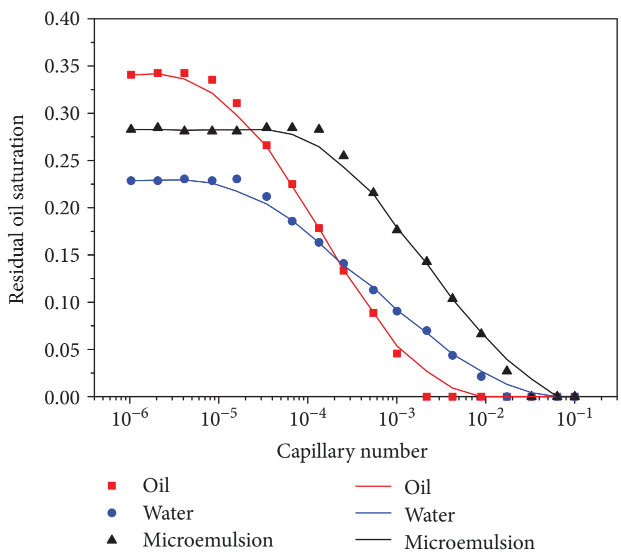


FIGURE 1: Example of a capillary desaturation curve (CDC) [2].

a brief introduction about the basic mechanisms of oil recovery is necessary.

The overall displacement efficiency of any oil recovery displacement process can be considered conveniently as the product of microscopic and macroscopic displacement efficiencies [1]. In equation form,

$$E = E_D \cdot E_V, \tag{1}$$

where E = the overall displacement efficiency (oil recovered by process/oil in place at start of process), E_D = the microscopic displacement efficiency expressed as a fraction, and E_V = the macroscopic (volumetric) displacement efficiency expressed as a fraction. Many factors affect microscopic displacement efficiency, like pore structure and distribution, microscopic heterogeneity, wettability, and interfacial tension. E_D is reflected in the magnitude of residual oil saturation (Sor) in the region contacted by the displacing fluid [1]. Factors affecting Sor significantly determine E_D. The most significant parameter to affect or determine Sor is the capillary number, which is defined as the ratio of viscous force to capillary force. The capillary number has many expressions, and the following is the most frequently used one.

$$N_c = \frac{v\mu}{\sigma}, \tag{2}$$

where N_c = capillary number, σ = interfacial tension (IFT) between displaced and displacing fluid, in mN/m, v=velocity of displacing fluid, in m/s, and μ = viscosity of displacing fluid, in mPa·s.

Many laboratory tests investigated the relationship between N_c and Sor and gave well-correlated curves between N_c and Sor, which are called capillary desaturation curves (CDC). An example of CDC is shown in Figure 1 [2]. Obviously, the larger the capillary number, the lower the residual oil saturation. From the CDC perspective, the highest displacing viscosity and the lowest IFT are beneficial to the reduction of Sor and thus to the increase in displacement efficiency. A higher velocity resulting from a great pressure gradient can also contribute to Sor reduction. This is the most basic mechanism of chemical flooding EOR techniques and other techniques.

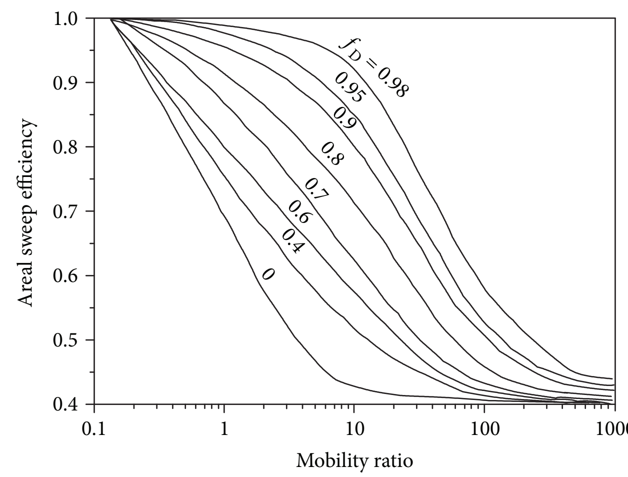


FIGURE 2: Areal sweep efficiency affected by the mobility ratio [1].

Macroscopic (volumetric) displacement efficiency (E_V) is also known as sweep efficiency. It can be further expressed as the product of areal sweep efficiency (E_A) and vertical sweep efficiency (E_I).

$$E_V = E_A \cdot E_I, \tag{3}$$

where E_A = area sweep efficiency and E_I = vertical sweep efficiency. Sweep efficiency is determined by reservoir heterogeneity, formation property, and fluid property. For a given reservoir, sweep efficiency is significantly, if not completely, determined by mobility ratio M, which is defined as follows:

$$M = \frac{\lambda_w}{\lambda_o} = \frac{k_w/\mu_w}{k_o/\mu_o} = \frac{k_{rw}}{k_{ro}} \cdot \frac{\mu_o}{\mu_w}, \tag{4}$$

where λ_w=water phase mobility; λ_o=oil phase mobility; k_w and k_o refer to water and oil phase effective permeability, respectively, in D; μ_w and μ_o refer to water viscosity and oil viscosity, in mPa·s; k_{rw} and k_{ro} refer to water and oil phase relative permeability, respectively. Areal sweep efficiency and vertical sweep efficiency affected by the mobility ratio is shown in Figures 2 and 3 [1], respectively. Obviously, as reflected in these two figures, the smaller the mobility ratio, the larger the sweep efficiency, especially when $M < 1$. When $M > 1$, flow becomes unstable and sweep efficiency decreased as M increased. Typically, the mobility ratio M is larger than unity due to the large contrast of oil and water viscosity, especially for heavy oil. To increase the sweep efficiency, the most important way is to reduce the mobility ratio, which can be attained by increasing the water phase viscosity, or reducing the oil viscosity, or improving the relative permeability. This is the key idea of mobility control, which is of vital importance in all EOR techniques.

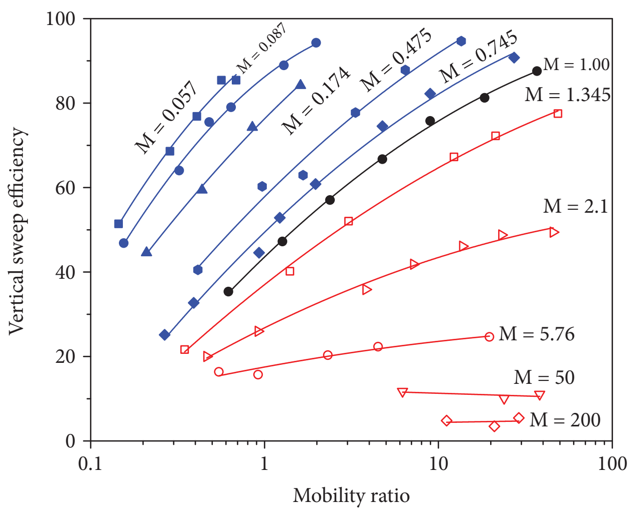


FIGURE 3: Vertical sweep efficiency affected by the mobility ratio [1].

Microbial enhanced oil recovery (MEOR) is a main topic of interest in energy researches as an environment-friendly and low operating-cost treatment technology [3, 4]. MEOR is a general designation of a series of technologies to increase oil production by propagation and metabolites of microbes [5]. The MEOR diagram can be seen in Figure 4 [6]. MEOR also follows the basic EOR principle of enlarging the sweep efficiency and increasing the capillary number. In the current situation of low oil price, MEOR is very promising, especially for the marginal reservoir and/or uneconomical reservoir, and microbial flooding was a potential alternative to other EOR/IOR methods, since it has high success ratio (as high 90% positive effects) according to a worldwide field test survey [4]. MEOR is very environmentally friendly and has no negative environmental impact [7, 8]. Since the implementation of the revised China Environment Protection Law in 2015, environment protection in petroleum exploration and development has never been given more emphasis than ever, which obviously adds to the total cost. Compared with thermal flooding and gas flooding, the preeminent advantages of microbial flooding were environmentally friendly characteristics and the lowest cost for increasing oil production [5, 9]. Compared with other technologies of EOR, the distinct features of microbial flooding include the low energy consumed by microorganisms, the combination of multiple mechanisms, and the reduced loss caused by degradation by some of the endogenous microorganisms [9]. Figure 5 shows different EOR cost estimations [10]. Table 1 [5] also summarizes the cost of different EOR techniques reported in 2002. These data show the relative cost advantages of MEOR compared with other EOR techniques. Although MEOR progress is well reviewed [4, 8], due to the language barrier, much MEOR progress in China is not included. MEOR progress in China is reviewed from both theoretical and practical aspects.

Microbial flooding was used in a wide range, including high water cut, heavy oil [11], marginal reservoir, and post-polymer flooding reservoir [12–16]. It could be applied to sandstone [17], carbonate [18, 19], light oil, heavy oil [11, 20, 21], and medium/high-permeability and low-permeability reservoirs [20, 22, 23].

2. Fundamentals of MEOR

2.1. MEOR Types. According to application, MEOR processes can be classified into four types [4]: microbial flooding recovery (MFR), cycle microbial recovery (CMR), microbial selective plugging recovery (MSPR), and others. According to a worldwide implemented field trial survey [4], MFR ranks first in the world among all MEOR trials judged from trial types, as can be seen in Figure 6 [4]. However, the MEOR application in China is quite different. MEOR in China can be divided into microbial flooding recovery (MFR), cycle microbial recovery (CMR), microbial selective plugging recovery (MSPR), and microbial wax removal (MWR). According to our survey of previous MEOR field tests and application in China, if judged from the application of well numbers, the total MEOR well number in China is more than 4600, while there are more than 3000 wells (producers and injectors) for MWR, accounting for about 65%, as can be seen in Figure 7. This figure is a summary of various field tests in China, and it is the first figure to describe MEOR types according to field test well numbers. Up to present, about 500 wells have been involved in MFR in China. These processes often involve more than one mechanism; thus, this classification is general. Since some MEOR data is not public or fully public, our survey involves most but not all MEOR field tests in China.

According to the source of the strains, microbial flooding could also be divided into indigenous microbial flooding and exogenous microbial flooding [9]. Exogenous microorganism indicated that the suitable microbes screened on a similar condition but not in the reservoirs were injected underground and increased oil production by using its propagation and metabolites. Indigenous microorganism means microbes were developed by remaining/residual oil as the carbon source on the basis of the active matter existing in formations and introducing the air and the inorganic salt with phosphorus source and nitrogen source when injecting water [5]. Indigenous microorganism flooding was the development trend with the advantages of good adaptability and avoiding of microbes' culture development and production process.

As introduced before, oil recovery was mainly determined by the displacement efficiency and sweep efficiency. MEOR had a double mechanism of improving the displacement efficiency and sweep oil efficiency [5, 9]. In the laboratory experiments, oil recovery could be increased by 10% by microorganism in a tertiary model [24] and increased by 5% by using microbial flooding and by 16% by combined microbial-chemical flooding in post-polymer flood reservoirs [25]. In another high-permeability (1400 mD) test, oil recovery could be increased by 18.4% [26]. A lot of experiments about microbial flooding and field had been studied in China [27]. The experience of field was not only beneficial to deeply understand microbial flooding mechanisms but also would provide evidence and guideline for industrial application to microbial flooding [5].

2.2. Microbial Community. The study on microbial community in the oil reservoir, especially the accurate analysis of the complex structure of the microbial community and

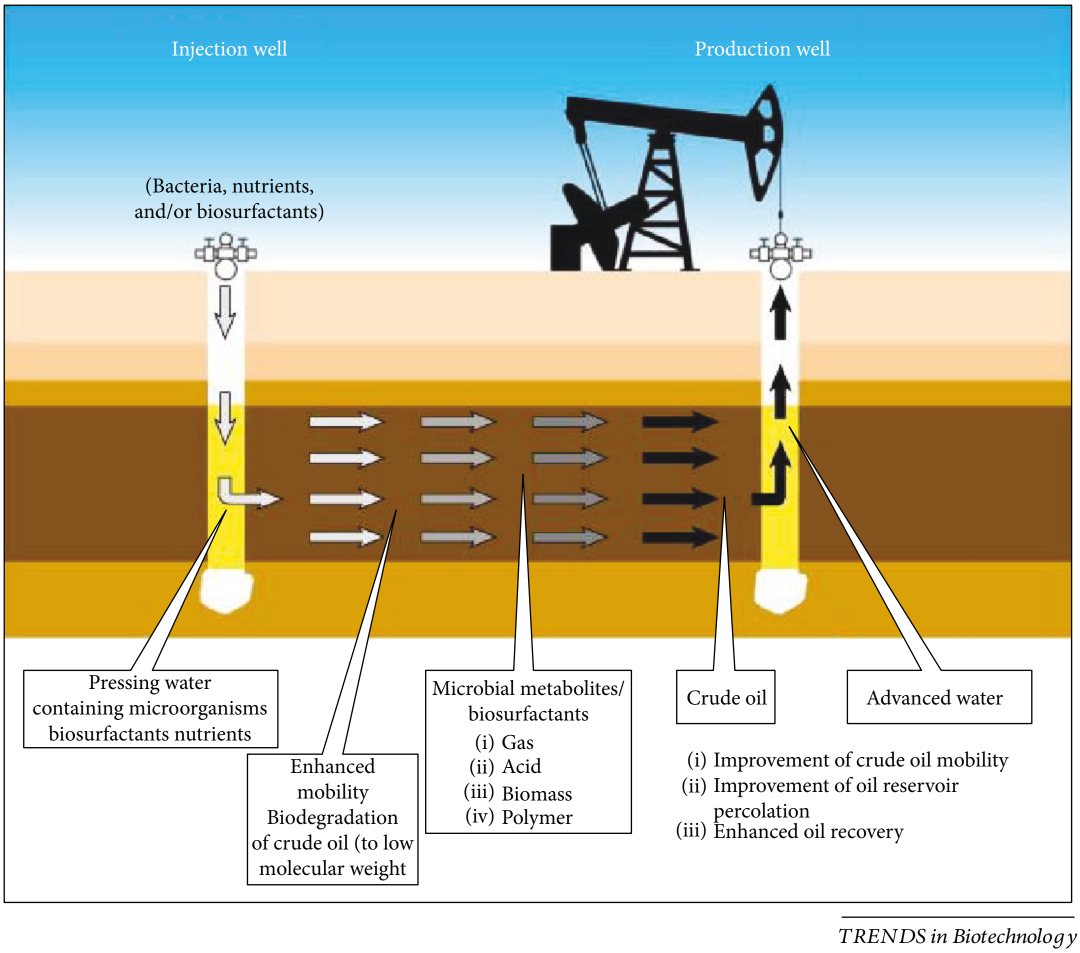


Figure 4: MEOR diagram [6].

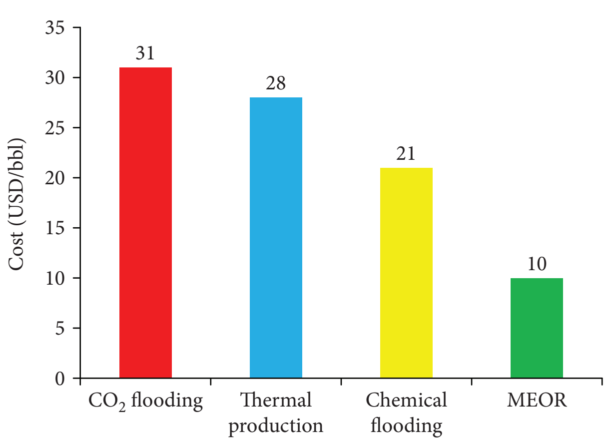


Figure 5: Different EOR technique cost estimation [10].

changes in monitoring, had significant importance for microbial flooding [28]. Different methods may be used to classify microbes according to research area or purpose. According to the growth dependence on oxygen, there are aerobic bacteria, facultative bacteria, and anaerobic bacteria in formations [29]. According to influence on oil production, some microbes in the oil reservoir were favorable for oil production, while others were not. Detecting microbes involves complex high-tech biotechnology, such as terminal restriction fragment length polymorphism (T-RFLP) [11, 16, 23, 30], gene bank [31], denaturing gradient gel electrophoresis (DGGE) [30], and most probable number (MPN) [32], which are beyond the scope of this paper. Microbial community structures and diversity characterization in Shengli Oilfield by such technologies are well documented [30, 33]. Figure 8 [33] shows microbe diversity in the second largest oil production reservoir in China. DGGE application in analyzing microbial diversity and community structure is explained elsewhere [30]. A new way of relating microbes that could not be cultured in extreme environments was provided by the molecular fingerprint technology with 16S rDNA as the main aspect [15, 34], which is important but beyond the understanding of petroleum engineers. Thus, MEOR requires close collaboration of different disciplines like petroleum engineering, chemistry, biology, and physics. The concrete microbes' names and characterization ways in MEOR are available elsewhere [4].

In studies of indigenous microorganism for improved oil recovery, the DGGE method was valuable for analyzing microbial community structures and monitoring community dynamics at the molecular level [35]. The analysis of microbial colony with T-RFLP technology in the pilot tests [36]

Table 1: Cost per incremental oil for different EOR techniques [5].

EOR techniques	Steam flooding	Combustion in situ	Gas flooding	Chemical flooding	MEOR
Cost (USD/bbl)	3-6	5-10	2-8	8-12	1-4

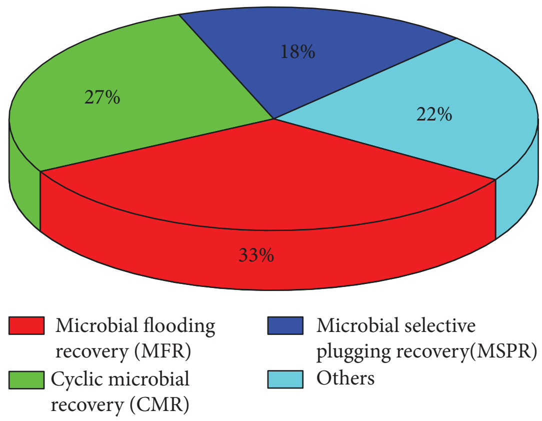


Figure 6: Worldwide MEOR trials [4].

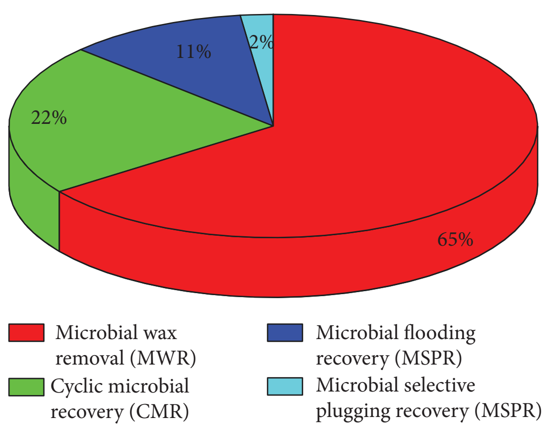


Figure 7: MEOR applications in China.

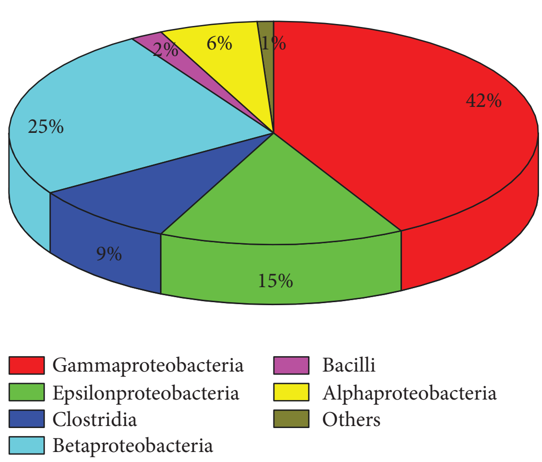


Figure 8: The diversity of bacteria in oil reservoirs [33].

in Shengli Oilfield showed the real situation changes of microorganisms in reservoirs, and the changes of diversity of indigenous microbial colony were promoted by the microbial flooding technology, which are verified by the fact that the diversity of the microorganism in the reservoir was negatively correlated with oil production in general. The core flooding tests [37] showed that the different distribution of crude oil and other metabolites in the core was the key factor affecting the microbial diversity in the reservoir. Studies on Daqing Oilfield [38] showed the number of microorganisms in water after polymer flooding was two orders of magnitude lower than that after water flooding. The field tests also proved that the substrates could be offered from metabolites of aerobic organisms in the formations after being activated by oxygen [29, 34].

2.3. Exogenous Microorganism and Indigenous Microorganism. According to the source of the strains, microorganisms can be divided into exogenous microorganisms (EM) and indigenous microorganisms (IM). Thus, microbial flooding recovery could be divided into indigenous microbial flooding recovery (IMFR) and exogenous microbial flooding recovery (EMFR) [9]. Exogenous microorganism indicated that the suitable microbes screened on a similar condition as the reservoir condition but not in the reservoirs were injected underground, increasing oil production by using its propagation and metabolites. Indigenous microorganism meant microbes were developed by using the remaining oil as the carbon source on the basis of the active matter existing in formations and introducing the air and the inorganic salt with phosphorus source and nitrogen source when injecting water [5]. Indigenous microorganism flooding was the development trend with the advantages of good adaptability and avoiding of microbes' culture development and production process. In recent years, the studies in China were generally about indigenous microorganisms [12, 16, 29, 37–40]. As for the carbon source of indigenous microorganisms, many scholars worked on microorganisms that took hydrocarbon in crude oils as the only carbon source [13, 24, 41], while some studied the microorganisms that took both polyacrylamide and hydrocarbon as the carbon source in post-polymer flooding reservoirs [12, 16, 25, 42]. It is worth mentioning that polymer flooding is very mature in China and has the largest commercial scale and production in the world [43].

Some microbes had significant influence on polymer which reduced the viscosity and molecular weight of polymer [12, 44]. The viscosity of polymer decreased by 52.1% in 7 days when microbes were cultured for 7 days without additional nutrition. After sucrose was added, the viscosity reduction of polymer reached 92% [12, 25]. Microbes also had influence on the polymer molecules, hydrolyzing the amide group into carboxylic acid [12, 44]. NMR tests showed that the polymer amide group content decreased from 74.6% to 60.8%, while the carboxylic group had an evident increase [12].

3. EOR Mechanisms

3.1. Oil Degradation by Microorganisms. Reducing oil viscosity is one of the main mechanisms in MEOR [8]. In macroscopic view, oil viscosity reduction is related with oil degradation, while in microscopic view, it is caused by the oil composition change, which are often detected by gas chromatography [5]. Laboratory experiments in Daqing Oilfield showed that the content of long-chain hydrocarbon content in crude oil was relatively decreased after microbial effect, and short- and medium-chain hydrocarbon content was relatively increased, which led to the light component of crude oil increasing by more than 30% [24]. Organic acid was produced to reduce the pH value from 7 to 6 to 5.5, and active material was produced to decrease the viscosity by more than 36% and interfacial tension was reduced from 35.67 mN/m to 8.1 mN/m. Further study [25] showed that after different microbes took effect with Daqing crude oil, the composition of crude oil was changed obviously and the wax and gum chicle content was decreased by 48% and 9.68%, respectively. The average acid value of Daqing crude was 10 times higher than that before and after the effect of suitable microbes [42], which indicated that bioproducts like acids are produced with significant amount.

The application of the high-resolution mass spectrum (HRMS) on the MEOR mechanism has revealed the change of polar compound structures before and after oil degradation by the microbe on the molecular level [45]. The study shows that this degradation mainly involves polar heteroatomic compounds changing from a high-molecular-weight compound into a small compound, and the alkyl chains of nitrogen compounds are easy to be degraded. This degradation produces small-molecular-weight organic acid dissolved in water, and some of this organic acid contains nitrogen, sulfur impurity atoms, and many aromatic rings. The number of organic acids even increases by one or two orders of magnitude [45].

Almost all the microbial laboratory and filed experiments had proved that microorganisms could be used to reduce the viscosity of crude oil, but the viscosity reduction content varied [41]. The viscosity and interfacial tension respectively decreased by 40% and 50% when Bacillus sp. was adopted [42]. The offshore heavy oil viscosity could be reduced by 66% by using one microbe. With the compound use of two microbes, the viscosity was decreased from 1146 cp to 5.11 cp, decreasing by 99% [12]. A study also showed that after a 14-hour reaction between streptococcus and crude oil, the oil viscosity decreased from 4000 mPa.s to 500 mPa.s, which are caused by both microbes and metabolites, with metabolites playing a major role [46]. As for viscosity-reducing causes, in addition to pectin degradation and biological emulsion, CO_2 produced by microbes in a supercritical state (high temperature and high pressure in reservoir) resolved in crude oil and thus reduced oil viscosity [41]. A field test in Dagang indicated that wax and gel contents respectively decrease by 2.2% and 3.7% [42]. Samples from five well groups in Daqing Oilfield also showed that the content of saturated hydrocarbon increased, while that of nonhydrocarbon decreased, which was consistent with the results of laboratory experiments [16]. Oil degradation by microorganisms accounts for MWR and MFR.

3.2. Biosurfactants in Microbial Metabolism. The microbial surfactant could be metabolized [5]. Surfactants included biosurfactants [41, 47], organic solvent [42], acids [41], and gas [41]. Gas was mainly CO_2, CH_4, and a small quantity of ethane [47]. The composition of biosurfactants mainly consisted of rhamnolipid [46, 48], as well as a mixture of paraffin ester and glyceride [46] which did not belong to sugar ester but phospholipid and polyketones. The main composition of acids was fatty acid [41, 49], acetic acid, propionic [41], and butyrate [42]. The mass fraction of fatty acid increased from 1% to more than 60% after the microbial effect [42]. Dagang Oilfield MEOR field tests showed that the content of low fatty acid increased notably with formic acid and acetic acid increasing 10 times and isobutyric acid increasing 7 times after nutrient solution was injected one year [35]. Organic solvent included alcohol, such as ethane [42]. It is notable that although biosurfactants are produced in MOER to reduce IFT, only few were reported to reduce IFT to an ultra-low level (10^{-3} mN/m).

3.3. Emulsification. Emulsification was one of the main mechanisms of MEOR [4, 7, 50, 51]. However, there is no satisfactory criterion to characterize the capacity of microbes on crude oil emulsification in China. At present, the simplest way to study and apply microbial flooding is the five-level classification method involving the direct observation of oil-water emulsion [5]. This method is simple and practical, but the disadvantage includes lack of quantitative characterization, which is well included in other much more complex ways of emulsion coefficient EI24 and the Tub-scan emulsion stability parameter measurement method [5]. The microscopic model flooding experiments used by the oil-water emulsion visual observation method found that crude oil was emulsified by microorganisms [24]. Microtransparent simulation models showed that the degraded crude oil was emulsified in different degrees in the form of various oil droplet sizes, and the oil droplets were tensile and deformed and had seepage flows [52]. The biogas produced was beneficial for emulsifying crude oil [5]. The degree of crude oil emulsion was seen with high correspondence to the growth rate of microorganisms [41]. The emulsification effectiveness for heavy oil was obviously improved by the effect of complex formulation of two types of microorganisms, as supported by the fact that as the emulsion stability increased, the average particle size decreased by 67.3% with notably reduced heavy oil viscosity [11]. The composition and structure of organic acid change lead to wettability alternation, and it also makes water-in-oil emulsion into oil-in-water, thus reducing the viscosity of oil, which in turn improves the flow rheological property [45]. This accounts for the common phenomenon of emulsification in MEOR on the molecular level and is considered to have notable progress on MEOR mechanism [45].

3.4. Altering Reservoir Physical Properties. Physical properties of the reservoir could be changed by the metabolism product of microbes, and the porosity was likely to be increased.

According to one laboratory test, the permeability was reduced from 284 mD to 24 mD, and the viscosity might be reduced by as large as 10 times due to the effects of acid [9]. The ability of reducing reservoir permeability is the main mechanism in MSPR as well as MFR. Two microbes were selected for cultivation from more than one hundred bacteria in post-polymer flooding reservoirs in Daqing Oilfield, and laboratory experiment indicated that the plugging rate of the profile of control bacteria was over 70% [13]. The field test in Daqing also indicated that reservoir permeability was decreased after profile control and the water injection profile was significantly improved. Microbes can produce biosurfactants through metabolism and change its wettability, and much attention has been given to this mechanism. The surfactants produced by microbes were adsorbed on the surface of porous media; the wetting state of the surface of porous media was thus changed due to the effects of the amphiprotic group in surfactants [40]. Microbes could make the reservoir wettability change from oil-wet to water-wet [9]. Most researches in China focused on the surfactant produced by microbes, while quantitative characterization of the wettability index after microbial mediation, contact angle, and other methods was seldom used. The most frequently used method is to test IFT by using the spinning drop method.

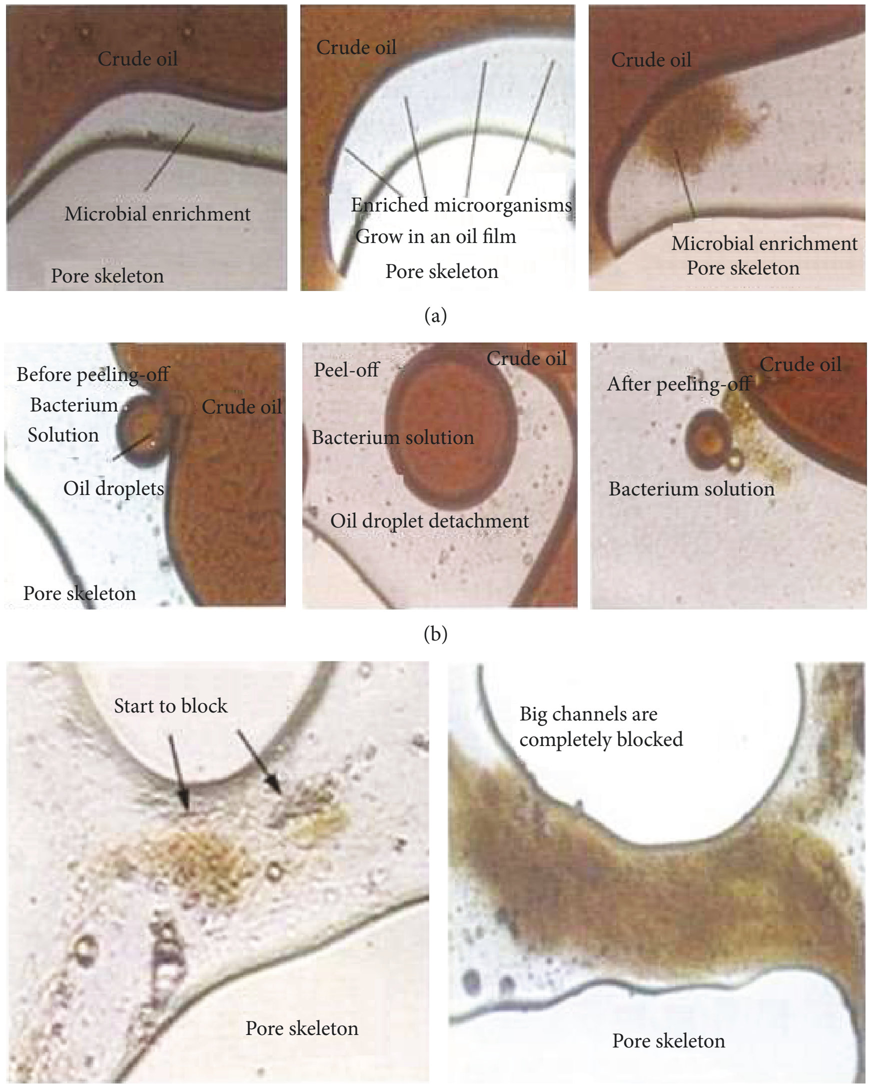
3.5. Microscopic Mechanism. Microscopic mechanisms of microbes contacting with crude oil and changing the pore surface as well as crude oil properties to enhance oil recovery help to clarify MEOR mechanisms. An experiment with a microscopic visible physical model [3, 13] indicated that microorganisms consuming crude oil could migrate directionally to crude oil and contact directly with crude oil and make the microbe colony highly concentrated towards carbon source and crude oil. The basis for microorganisms consuming crude oil to enhance oil recovery was its ability to automatically search for carbon source and directionally migrate. Microbes automatically migrate to crude oil, concentrate, and multiply constantly. The distribution regularity of the concentration of the surfactant as well as the acid in its metabolites was exactly the same with that of bacteria. The automatic directional migration of microbes was attributed to its chemotaxis. The microscopic mechanism of peeling off oil film and oil droplets by microbes could be seen in Figures 9(a)–9(c) [13]. Although hydrophilic, the bacteria could be hydrophobic at one end. Due to the synergistic effect with its metabolites, the bacteria entered into the space between pore surface and oil film or oil droplets, grew and reproduced massively, and entered deeply inward, and finally the oil film was peeled off. Their experiments also indicated that a proper time was required for the migration and concentration of microbes and metabolites, and it would be better to adopt huff and puff or intermittent methods during field MEOR. A microscopic photoetching physical model [3, 40] also indicated that due to the growth and metabolism of microbes attached to the oil-water interface, the oil-water IFT was reduced, the interface was softened, and the flow ability of the remaining oil was enhanced. This is because the surfactant produced by the microbial metabolism concentration of the surfactant at the interface was increased, generating a surface tension gradient. Once the gradient exceeded the viscous force, spontaneous interface deformation and movement, namely, Marangoni convection, would appear, and together with migration behavior of microbes and disturbance action of high pressure, positions of droplets in pores would be changed.

4. Microbial Flooding Recovery Designing

The system of microbial flooding involves activation of material composition in the system, injection of slugs, nutrition concentration, and the size of nutrition-injecting slug. Lots of studies had been conducted about the activation system. The feasibility of using corn starch as the activation system was studied [32]. The ultimate recovery increase could be significantly influenced by the cultivation time of injecting slugs, nutrition concentration, and the size of nutrition-injecting slug. It had been proved that injecting 0.4 PV slugs with a corn starch concentration at 10-20 mL/L and cultivating for 15-20 d was optimum for the condition. However, in many actual field tests of microbial flooding in China, slug size was no larger than 0.1 PV [44]. In view of the extra-low permeability reservoir in Dingbian, Changqing Oilfield, bacteria concentration of 10% and slug size of 0.5 PV were selected according to the laboratory core flooding test experiment with a tertiary recovery of 8% OOIP [53]. Actual MEOR design parameters in Daqing Oilfield in China were well summarized [54].

4.1. Reservoir Screening Criteria. Screening criteria for a reservoir varied greatly [9]. Different criteria existed due to different reservoir conditions and research progress. Safdel et al. [4] made a critical review on different MEOR screen criteria in different countries, although the data used for China are not latest. When microbial flooding was conducted, factors that must be taken into account [9] involved remaining oil saturation, hydrocarbon compositional analysis, fluid chemistry and composition, depth of reservoir, salinity of formation water, formation water sample analysis, estimated net oil increment, and economic aspects. According to laboratory research and field tests in China and with consideration of researches abroad, 8 major parameters were selected for reservoir screening and evaluation in microbial flooding could be seen in Table 2 [5]. Screening criteria by Shengli Oilfield in China could be seen in Table 3 [14]. Considering the MEOR research history and field test scale and number, as well as being the largest branch company of Sinopect, Shengli Oilfield screen criteria represent the standard of Sinopec. Major oil companies and their production share in China are available in a publication [43].

Temperature has direct influence on the growth of microorganisms [41]. There is optimum temperature for the growth of microorganisms, which could be largely affected when the optimum temperature was exceeded. Previous studies [41] also showed that 8 facultative anaerobes could grow well at 45-60°C, while they cannot grow when the temperature is higher than 75°C. Laboratory tests showed that for the same microorganism, when the temperature increased from 37°C to 73°C, the bacterial concentration



(c)

Figure 9: (a) The microbial growth and peeling-off oil film [13]. (b) The peeling-off course of oil droplet [13]. (c) A large amount of modular/floss stopping big orifices [13].

Table 2: CNPC MEOR reservoir screening parameter [5].

Parameter	Value range	Optimum
Formation temperature (°C)	20-80	30-60
Crude viscosity (mPa·s)	10-500	30-150
Permeability (mD)	≥50	≥ 150
Porosity (%)	12-25	17-25
Brine salinity (g/L)	≥300	≥100
Wax content (%)	≥4	≥7
Water cut (%)	40-95	60-85
Total bacterial concentration in produced fluid (number/mL)	≥100	≥1000

Table 3: MEOR reservoir screen parameter in Shengli oilfield [14].

Formation temperature (°C)	Permeability (mD)	Formation brine (pH)	Formation brine salinity (mg/L)	Dead oil viscosity at 50°C
≤80	≥50	6~8	≤150000	≤3000

dropped remarkably [20]. 80°C was the critical reservoir temperature for MEOR, and if the temperature exceeded 80°C, the microorganism's growth rate was very slow [14]. One pilot in the HKL-801 block (reservoir temperature 80°C) in Shengli Oilfield was effective while the one in the BNL-32 block showed no obvious effects because the temperature was 91°C [14]. In addition to reservoir temperature, reservoir heterogeneity also affected microbial flooding recovery significantly [14]. However, there were no reservoir heterogeneity criteria for MEOR yet. Although Jianghan Oilfield in central China reported cultivated thermophilic bacteria (Geobacillus kaustophilus) that could grow at 100°C and

Table 4: Chao 50 block microbial flooding field test in Daqing Oilfield [61–63].

Area of block (km^2)	2.43
OOIP (tons)	1667000
Reservoir depth (m)	989
Reservoir thickness (m)	7.9-9.5
Reservoir temperature (°C)	55
Injectors/producers	2/10
Formation brine salinity (ppm)	4450
Formation brine divalent (ppm)	14
Average permeability (mD)	25
Average porosity (%)	17
Dead oil viscosity (cP)	20.2
Formation oil viscosity (cP)	9.7
Original oil saturation (%)	57
Water cut	95%
Implementation time	June 2004-Sep 2005
Injection slug (PV)	0.005
Microbe concentration	5% (first slug), 2% (second slug)
Effective well ratio	74.2%
EOR (% OOIP)	3%
Water cut reduction (%)	30.3
Input-output ratio	1 : 6
Effective duration time	3 years
Expansion test	Yes

salinity of 350000 ppm and the paraffin removal pilot test in a well with 117°C temperature and 250000 ppm salinity verified satisfied paraffin and plug removal effects [55–57], the MEOR reservoir temperature criteria in China remains at 80°C, the highest reservoir temperature with use of MEOR 155°C for Norwegian fields [4].

Salinity was another key screen parameter affecting microbial flooding [5, 14]. High-salinity and high calcium concentration formation water was not suitable to the application of the microbial flooding technology [55]. It was reported that two microorganisms separated from produced fluid grew well in the salinity range of 100000-200000 ppm, while when the salinity was higher than 200000 ppm, the growth rate of these two microorganisms got slower [58]. By using 16S rDNA technology, these two microorganisms were proved to be Pseudomonas aeruginosa and Bacillus subtilis [58]. A pilot test [55–57] indicated that microorganisms cultivated at a salinity of 350000 ppm could remove paraffin in a well of 250000 ppm salinity with good performance.

4.2. Typical Field Tests. Field tests are a necessary stage for any EOR technology going from laboratory to real application; this is especially crucial for MEOR. The physical simulation methods learned from chemical flooding practice have some limitations, such as the impossibility of simulating oxygen-free and endogenous environments like real reservoir condition, and the short length of cores resulted in the exclusion of microorganisms without full growth and propagation [32, 59]. Shengli Oilfield MEOR tests showed that results between laboratory experiments and field tests varied greatly, which may be attributed to the complex reservoir conditions and/or physical simulation method limitations. Therefore, it was necessary to improve the evaluation method like choosing the low injection rate and the suitable core length to keep microorganisms staying in the core for at least 14 days [32]. More importantly, it is necessary to conduct field tests to check the technique effect and avoid risk and to learn from previous field tests to reduce costs in the low oil price era. Field trial data on global microbial flooding is available online [4, 54, 60]. Only a few typical MEOR field tests in China are selected to provide more operational information, such as cycle microbial recovery (CMR) and microbial flooding recovery (MFR), which are not available in the previous publication [60]. According to our own survey, up to present, there have been more than 47 MFR field tests in China, involving more than 500 wells (injectors and producers) and 15 oilfields in China. Different from previous studies focused on microbial huff and puff, or CMR, which are not real microbial flooding tests, this paper focuses on real MFR to show what progress and experience have been made in China. Below are some typical MFR projects based on latest references available. To better help possible reservoir screening and field application in similar reservoirs, the key parameters of incremental oil recovery and economic parameters are given. It is, to the best of our knowledge, the most detailed operational learning in view of the EOR scope from previous field tests in China.

4.2.1. Daqing Oilfield. Up to present, more than 12 MFR field tests have been conducted in Daqing Oilfield. Some MFR tests are available in reference [54]. Among these field tests, Chao 50 in Chaoyanggou Oilfield is very prominent. Two microorganisms (Brevibacillus brevis and *Bacillus cereus*) were selected from indigenous microorganisms to conduct field tests of single well simulation and microbial flooding in ultra-low-permeability reservoirs in Daqing Oilfield [22, 61, 62]. From 2002 to 2003, 60 wells were put into CMR tests. The average formation permeability was 10 mD, and the formation temperature was 55°C. Among the 60 wells, formation permeability of 28 wells was 15-25 mD, and that of 22 wells was 5-15 mD, and formation permeability of 10 wells was below 5 mD. 71.7% wells were seen as having positive results, and the input-output ratio was 1 : 8. Based on previous single-well MEOR success, microbial flooding recovery (MFR) tests were carried out in 50 blocks with 2 injection wells and 10 production wells [61]. The reservoir data and field test performance are given in Table 4 [61–63]. Well patterns and field test performance are given in Figure 10 [63] and Figure 11 [63], respectively. The liquid-producing capacity increased from 43.6 to 79.6 tons, daily oil production increased from 24.7 t to 40.8 t, and water cut decreased by 30% and the incremental oil recovery was 3% OOIP with an effective duration of three years. Considering the low injection slug (0.005 PV) compared to chemical flooding slug, the incremental oil recovery is very prominent. Another very successful microbial flooding field test was reported to have an incremental oil recovery of 4.45% OOIP by 0.05

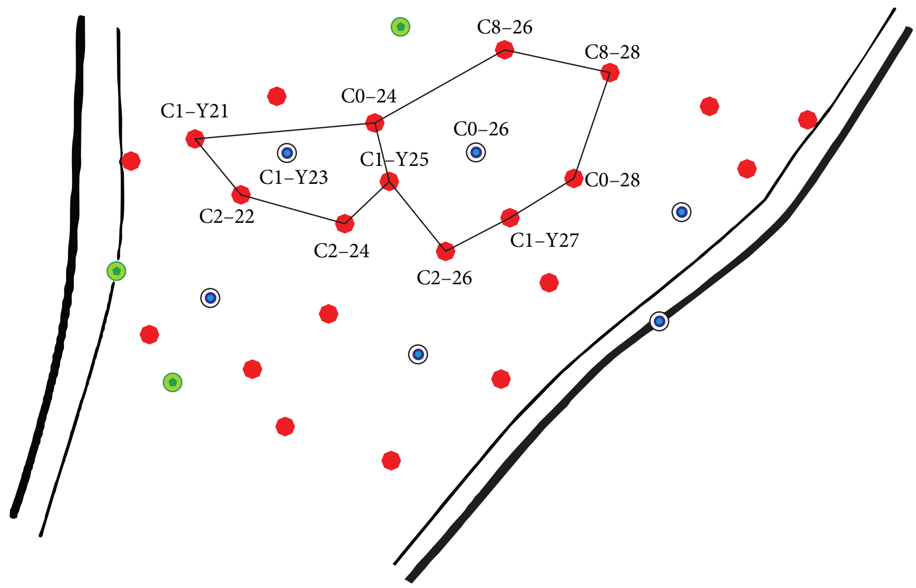


Figure 10: Chao 50 MFR field test well pattern in Daqing Oilfield [54].

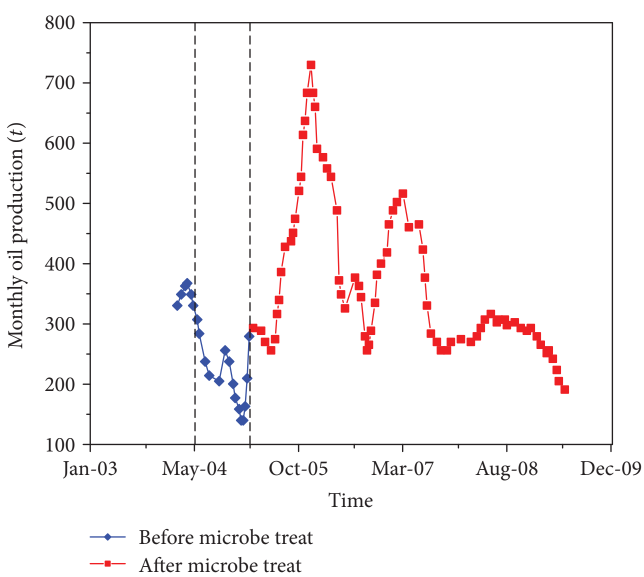


Figure 11: Chao 50 MFR field test production performance [54].

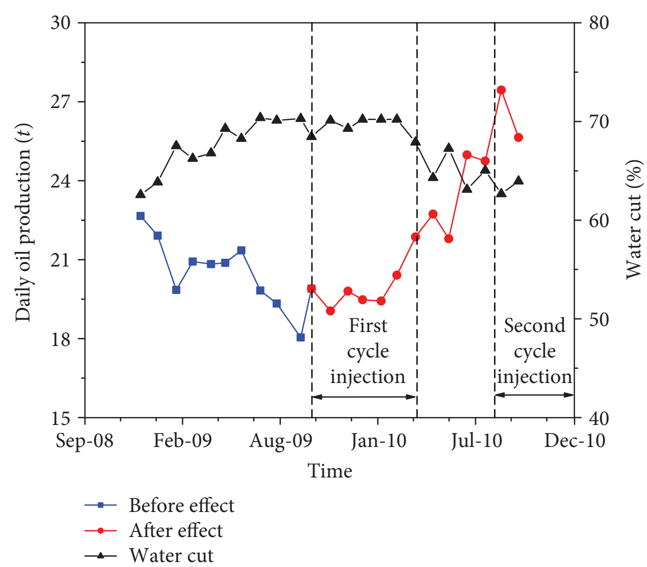


Figure 12: Chao 50 expansion MRF field test production performance [54].

PV bacteria slug [54]. The Chao 50 input/output ratio was 1 : 6. This successful pilot test indicated that MFR can succeed in the reservoir with permeability lower than present criteria at 50 mD, seen in Table 2 and Table 3. This test also showed that microbial flooding could set an effective displacement system which made the dead oil well remobilized. This field test verified that the injection-production relationship significantly affected microbial flooding effects. Based on the success of MRF in Chao 50, expansion microbial flooding tests with 9 injectors and 24 producers were conducted in 2009 [54]. The production performance of the expansion test can be seen in Figure 12 [54]. Detailed information of the expansion field test is not made public yet, but it is reported that microbial flooding makes the block production turn from decreasing to increasing.

4.2.2. Shengli Oilfield. Since Shengli Oilfield has been the second largest oil producer for a long time, MEOR in Shengli provides for the industry a valuable experience. MEOR research in Shengli Oilfield started since 1995, and MEOR field tests have been conducted since 1997 [14]. Although more than one thousand wells have been used in MWR and CMR in Shengli Oilfield, only 9 blocks have been conducted for MFR. Table 5 [14, 36, 64, 65] is a summary of 7 microbial flooding field tests in Shengli. The MRF test in Shan 12 is well introduced in a previous publication [30]. Among these field tests, only Guan 3 Block is not of fault block type, while the other 6 are all fault block reservoirs. And these 6 blocks are water flood reservoirs, while Guan 3 block is a post-polymer flood reservoir. In other words, the first 6 tests in Table 5 are all in tertiary recovery stage, while the last is in

Table 5: Recent microbial flooding recovery field tests in Shengli Oilfield.

Case	Block	Implement time	T (°C)	*Perm* (mD)	Salinity (ppm)	Dead oil viscosity at 50°C (mPa·s)	Inj./Pro	Area (km^2)	Type	Incremental oil (ton)	Water cut↓[1]	EOR↑[2] (%)
1	ZNXQ	1998.3-1999.09	54	477	1100	48	3/8	0.9	EMFR	5090	5	
2	Li 32	1998.06-2002.2	91	525	4600	88	4/7	1.8	EMFR	2001	slight	
3	Pan2-33	2000.08-2002.11	67	436	43900	1100	4/11	0.9	EMFR	7800	1.3	
4	Luo 801	1999.07-now	80	231	7790	353	5/13	1.25	Air EMFR	122800	7.3	4.95
5	Shan 12	2005.08-2008.06	66	263	20000	38	1/7	0.31	Air IMFR	8520	2	
6	Zhan 32	2011.11-now	63	682	9000	1885	3/12	0.69	Air IMFR	22855	5.5	*5.5*
7	Guan 3	2008.11-2012.12	69	2500	5920	1000	6/17	0.84	Air IMFR	21000	0.7	1.27

Note: T = reservoir temperature; Perm = permeability; Inj. = injectors; Pro = producers. [1]↓ means water cut reduction; [2]↑ means incremental oil recovery.

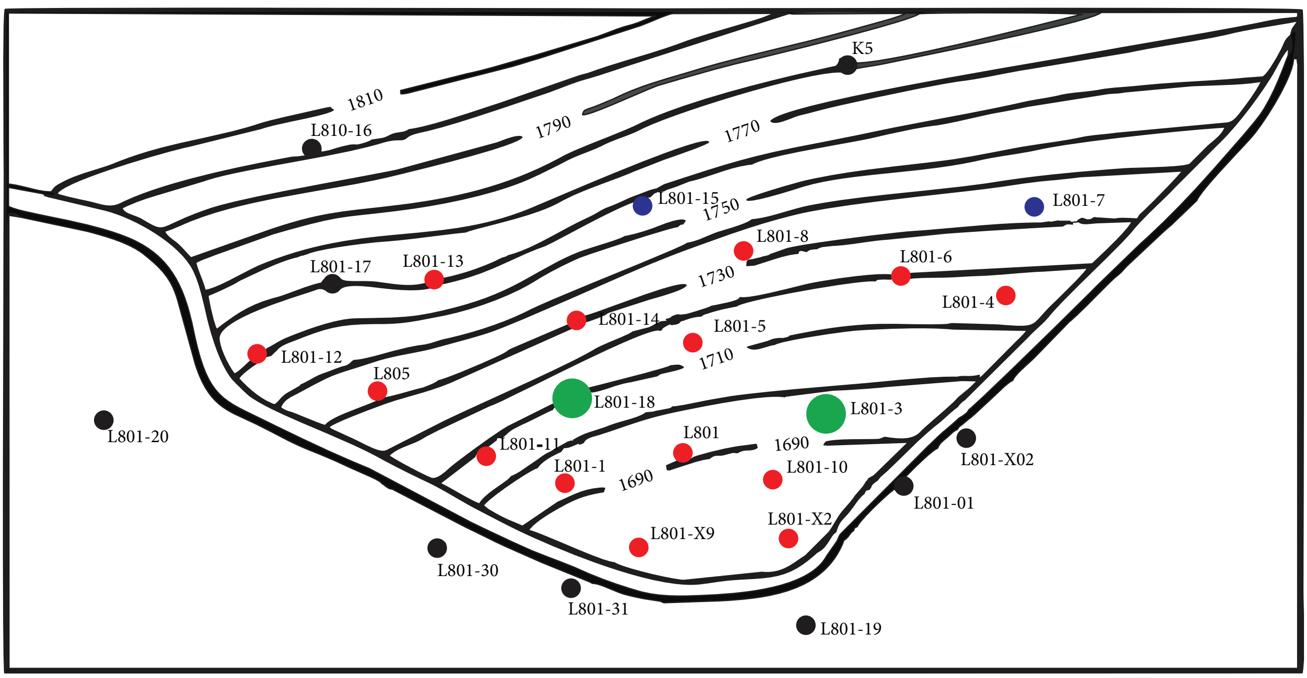


Figure 13: Luo 801 Block in Shengli Oilfield MEOR field test [65].

quaternary recovery stage. Since Shengli Oilfield has the second largest polymer flooding commercial use in China, the MFR test in Guan 3 is worthy of special attention. Polymer flooding in this block started in December 1994 and entered into the post-water flooding stage in April 1997 [14]. MFR started in November 2008. Although profile control measures have been taken before bacteria injection, injected bacteria broke through 4 days after injection in the latter stage. This test indicated the difficulty of MEOR in the post-polymer flooding reservoir with high heterogeneity. Among the 7 MFR field tests in Table 5, only three were reported with obvious enhanced oil recovery. In this block, MFR field tests have been enlarged from five wells (1 injector, 4 producers) in 2011 to 15 wells (3 injectors, 12 producers) in 2014 [14, 64]. In 2015, the field test has been enlarged, but the data has not been made public. Incremental oil recovery in Zhan 32 is a predicted recovery. Among all the blocks that are conducted for MEOR, Luo 801 deserves the most attention for several reasons. First, it has the longest MEOR application lasting time in China, probably in the world. Second, it has currently the highest field proven enhanced oil recovery in MEOR. The staged actual enhanced oil recovery is 4.95% OOIP, higher than the best one in Daqing Oilfield [54]. Finally, two kinds of microbial flooding (IMFR, EMFR) are both tested in the same block. The production history of Luo 801 is well introduced in reference [64, 65]. Figure 13 [65] shows the well pattern of MFR field tests. In Figure 13, green represents the two injectors from 2002 to 2011, while blue represents 3 injectors operated from July 1999 to August 2002, and red represents producers operated from 1999 to present [65]. The production performance of Luo 801 is shown in Figure 14 [65]. This data shows that microbial

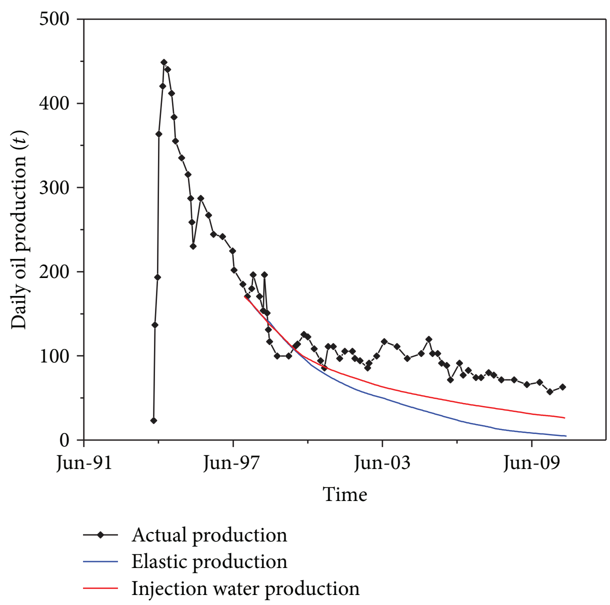


Figure 14: Luo 801 production curve and decline forecast [65].

flooding indeed improved oil recovery. For potential reservoir screening consideration, reservoir parameters and MFR field test performance are summarized in Table 6 from various references [30, 33, 60, 64, 65]. After air-assisted MFR, the annual water cut increase rate changed from 9% to 0.53%, and it has been maintained lower than 1.5% for 8 years [14]. The input/output ratio was not reported and is estimated to be 1 : 4 according to a comparison with some similar MRF projects in Shan 12. MFR success in Luo 801 paved a way towards enlarging MEOT tests in other blocks like Zhan 32 in Table 5. The cost for incremental oil from Luo 801 microbial flooding blocks is as low as 7 USD/bbl (339.56 yuan/ton) [65]. Some latest MEOR projects in Shengli Oilfield have not been made public.

Table 6: Luo 801 block microbial flooding field test in Shengli Oilfield.

Area of block (km^2)	1.25
OOIP (tons)	2910000
Reservoir depth (m)	1680-1800
Reservoir thickness (m)	15.5
Reservoir temperature (°C)	75-80
Injectors/producers	5/13
Formation brine salinity (ppm)	9794
Formation brine divalent (ppm)	NA
Average permeability (mD)	218
Average porosity (%)	23.4
Dead oil viscosity (cP)	221.7
Formation oil viscosity (cP)	12.8
Original oil saturation (%)	60
Water cut	86.5
Implementation time	July 1999-
Injection slug (PV)	0.25
Microbe concentration	NA
Effective wells ratio	NA
EOR (% OOIP)	4.95
Water cut reduction (%)	7.3
Input-output ratio	≈1 : 4
Effective duration time	>15 years
Expansion test	Yes

4.2.3. Changqing Oilfield. Changqing Oilfield is the largest oilfield if judged by production oil equivalent. Almost all reserves of Changqing Oilfield are from low-permeability reservoirs, and more than half are of ultra-low-permeability formation. Since the reservoir permeability bound given by CNPC and Sinopec is 50 mD, whether ultra-low-permeabilityreservoirs are suitable to use MOER draws attention. A pilot in an ultra-low-permeability reservoir was conducted in 2009 in Ansai in Changqing Oilfield [31, 66–68]. The average permeability of Ansai Oilfield is 1.29 mD, and the average porosity is 12.4%. The pilot was conducted to check the microbial flooding effect, which contains one well group with 1 injector and 6 producers. Oil production in this block started from March 1990, and the daily oil production per well before MFR is 1.48 tons [31]. The well pattern is shown in Figure 15 [31, 68]. Oil production before and after MRF is given in Table 7 [31, 68] while reservoir parameters are given in Table 8 [31, 66–68]. Oil production indicates that microbial flooding can reduce water cut and increase oil production. The water cut increase rate was reduced from 10.86% to 4.42%, and the comprehensive production decline rate was changed from 2.34% to -2.58%,

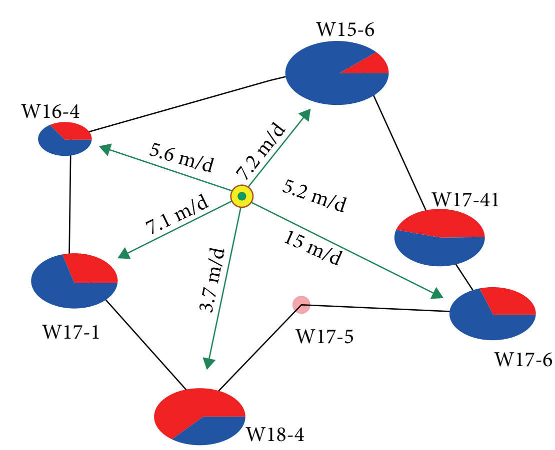


Figure 15: MRF pilot well pattern and bacteria flow velocity [31, 68].

which means oil production was significantly increased [68]. This pilot also shows that production performance has a positive relation with microbe movement. This is in agreement with other field tests in Daqing and Shengli. MEOR is a water-enhanced improved oil technique. Only when an effective injection-production relationship is formed can effective oil production be attained. In other words, if water injection is difficult, MEOR is likely ineffective. The economic performance is very good, with an input-output ratio of 1 : 5.9. This indicated that the permeability ground should be lower.

Table 7: Well performance of Ansai MFR pilot [31, 68].

No	Before MFR Daily fluid (m^3)	Before MFR Daily oil (ton)	Before MFR Watercut (%)	After MFR (December 18, 2009) Daily fluid (m^3)	Daily oil (tons)	Watercut (%)	Daily oil increase (tons)	Cumulative incremental oil (tons)
1	2.13	0.67	62.7	4.02	2.2	34.9	1	169.7
2	5.37	1.43	68.2	4.28	1.86	48.4	0.45	57.93
3	5.73	1.59	67	5.19	12.17	50.2	0.46	34.37
4	5.01	1.04	75.6	4.65	0.52	86.6	0.5	33.56
5	7.54	1.1	82.6	5.45	0.92	80	0.07	2.09
6	5.36	3.04	32.6	3.53	2	32.5	0	0
Total	31.14	8.87	66.1	27.12	19.67	57.6	0.66	297.65

Table 8: Ansai MFR pilot in Changqing Oilfield [31, 67, 68].

Area of block (km^2)	NA
OOIP (tons)	NA
Reservoir depth (m)	1220-1241
Reservoir thickness (m)	15.8
Reservoir temperature (°C)	45
Injectors/producers	1/6
Formation brine salinity (ppm)	92600
Formation brine divalent (ppm)	NA
Average permeability (mD)	5.22
Average porosity (%)	14
Dead oil viscosity (cP)	10.5 at 20°C
Oil saturation (%)	60
Water cut before test (%)	67.1
Implementation time	June 28-August 21, 2009
Injection slug (PV)	0.003
Microbe concentration	3%
Effective well ratio	67
EOR (% OOIP)	NA
Water cut reduction (%)	8.5
Input-output ratio	1 : 5.9
Effective duration time	1 year
Expansion test	Yes

5. Conclusions

Compared with thermal production, gas flooding, and other enhanced oil recovery methods, the prominent advantages of MEOR are much lower costs and more environment friendliness compared to other EOR techniques. Field tests show that the input-output ratio of microbial flooding recovery is as high as 1 : 6, with a much lower total cost than all the other EOR techniques like polymer flooding, gas flooding, and thermal production.

Indigenous microorganism flooding is the development trend with the advantages of good adaptability and avoiding of microbes' culture development and production process compared with exogenous microbial flooding.

Both laboratory and field tests have verified that the crude oil composition changed remarkably as the saturated hydrocarbon proportion increased; aromatics, nonhydrocarbon, and asphaltene proportion decreased; and the acid value increased while wax and pectin proportion decreased.

The microbial metabolism produced surface active compounds including biosurfactants, alcohol, acid, and biogases. The most common and desired biosurfactant was rhamnolipid which could reduce interfacial tension. Biogases were mostly carbon dioxide and methane, and little ethane. The acid was mainly fatty acid like methanolic acid, acetic acid, and propanoic acid.

The crude became emulsified with different extents due to effects of microbes.

Microbial products could change the wettability toward more water being wet and also reduce formation permeability remarkably. The microbial profile control mechanism could be accounted into one or all the mechanisms including microbes forming a reticular biofilm in porous media, precipitation of the colony, and formation of a bridge plug due to absorption of other microbes, the biogas block effects.

The basis for microorganisms consuming crude oil to enhance oil recovery was its ability to automatically search for carbon source and directionally migrate. Microbial effects on remaining oil could be ordered ranked in a descending order as island remaining oil, membranaceous remaining oil, columnar remaining oil, blind end remaining oil, and cluster remaining oil.

Application of a high-resolution mass spectrum (HRMS) on MEOR mechanism has revealed the change of polar compound structures before and after oil degradation by the microbe on the molecular level.

The reservoir screening parameters include temperature, salinity, oil viscosity, permeability, porosity, wax content, water cut, and microorganism concentration in which production fluid, temperature, and salinity were the three most important parameters. It is possible to use MFR in a reservoir with permeability as low as 5 mD.

Microbial flooding recovery field tests in China show that MRF is close to commercial application, since a high incremental oil recovery of 4.95% OOIP was attained with a typical 0.1 PV slug. Three typical reservoirs with detailed MFR field tests data were reviewed for possible guide for similar reservoirs.

Conflicts of Interest

The authors declare no conflict of interest.

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