

Effect of Wax-Degrading Bacteria on Wax Removal and Viscosity Reduction of Waxy Crude Oil: Postprint

Authors: Wang Weiqiang, Liu Yan, Wang Guofu, Li Jia, Zhang Xiaobo

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Abstract

Waxy crude oil exhibits increased viscosity and deteriorated flowability in low-temperature environments due to the presence of wax components. During pipeline transportation, the precipitation of wax crystals reduces the transport capacity of pipelines and even poses a threat of pipeline blockage. This study utilizes a previously discovered wax-degrading bacterium, employing its fermentation broth to degrade wax components in waxy crude oil. Experimental results demonstrate that after 5 days of treatment, the wax content of the crude oil was reduced and the apparent viscosity decreased. Polarized light microscopy observations revealed that the wax crystal structures in the oil became significantly smaller and more dispersed. Measurements obtained from processing the microscope images with ImageJ software showed that the maximum wax crystal size decreased from 609.1 μm to 72.1 μm , and the average Feret diameter decreased from 79.1 μm to 50.3 μm .

Full Text

The Effect of A Wax Strain on the Wax-removal and Viscosity-reduction in Waxy Crude Oil

WANG Wei-Qiang, LIU Yan, WANG Guo-Fu, LI Jia, ZHANG Xiao-Bo

(Liaoning Shihua University, Fushun 113001, China)

Abstract

The presence of wax components in waxy crude oil causes increased viscosity and reduced fluidity at low temperatures. During pipeline transportation, wax crystal precipitation decreases pipeline capacity and poses safety hazards such as

blockages. This study utilized a newly isolated wax-degrading bacterial strain to treat waxy crude oil with its fermentation broth. Experimental results demonstrated that after 5 days of treatment, the wax content and apparent viscosity of the crude oil decreased significantly. Polarized microscopy revealed that wax crystal structures became markedly smaller and more dispersed. ImageJ software analysis of microscopic images showed that the maximum wax crystal size decreased from 609.1 μm to 72.1 μm , while the average Feret's diameter dropped from 79.1 μm to 50.3 μm .

Keywords: waxy crude oil; wax strain; microstructure; viscosity reduction

0 Introduction

Waxy crude oil exhibits its most characteristic behavior when temperature drops below the wax appearance point, causing wax components to gradually precipitate. This precipitation imparts non-Newtonian fluid properties such as thixotropy and shear-thinning to the crude oil, resulting in increased viscosity. During pipeline transportation, as temperature decreases, precipitated wax deposits on pipe walls, reducing transportation capacity and creating safety hazards from potential blockages [1]. Reducing viscosity and preventing wax crystallization are key challenges in waxy crude oil pipeline transport. Current industrial methods primarily include heated transport and addition of drag-reducing or viscosity-reducing agents [2-3]. However, these measures do not fundamentally reduce wax content in crude oil, but merely inhibit wax crystallization. Using microorganisms to degrade wax components in waxy crude oil can fundamentally solve this problem by significantly reducing the impact of wax on crude oil properties. This paper demonstrates from multiple experimental perspectives that a wax-degrading bacterial strain can effectively remove wax and reduce viscosity in waxy crude oil.

1 Materials and Methods

1.1.1 Bacterial Strain Source

For heavy oil from Liaohe Oilfield, a wax-removing and viscosity-reducing bacterial strain was screened using paraffin as the sole carbon source. This facultative anaerobic strain was identified through genetic analysis as *Bacillus subtilis* and designated L-5. Its optimal culture temperature is 40 °C with an optimal culture period of 5 days.

1.1.2 Culture Medium Composition

Enrichment medium: yeast extract 1 g, NaCl 5 g, NH₄Cl 1 g, MgSO₄ · 7H₂O 0.25 g, KH₂PO₄ 5 g, NaNO₃ 2 g, pH 7.0-7.2.

Fermentation medium: peptone 10 g, yeast extract 5 g, NaCl 5 g, pH 7.2.

Screening medium: sucrose 1 g, Na₂EDTA 0.6 g, KH₂PO₄ 0.2 g, NaNO₃ 2 g, CaCl₂ 0.01 g, MgSO₄ · 7H₂O 0.3 g, yeast extract 0.5 g, liquid paraffin 4 ml, pure water 1000 ml, pH 7.5.

Sterilization conditions: 0.103 MPa, 121 °C, 20 min.

1.1.3 Oil Sample

Waxy crude oil from Liaohe Oilfield with density of 966 kg/m³ at 35 °C and wax content of 29.45%.

1.2 Experimental Methods

Viscosity was measured using an RV2 rotational viscometer across shear rates of 0.4-2.5 s⁻¹ and temperatures of 40-70 °C. Oil composition was analyzed using Fourier transform infrared spectroscopy. Wax content was determined using a Q2000 DSC differential scanning calorimeter. Wax crystal microstructure was observed using polarized microscopy, with quantitative analysis performed using ImageJ software.

2 Results and Discussion

2.1 Crude Oil Viscosity Measurement

The bacterial strain was activated in enrichment medium and inoculated into fermentation medium at 10% volume fraction. After culturing to logarithmic growth phase, the fermentation broth was added to 100 ml of waxy crude oil at a 1:1 ratio, then incubated at 40 °C, 150 rpm for 5 days.

Viscosities of treated and untreated crude oil were measured using an RV-2 rotational viscometer (Table 1), with viscosity-temperature curves shown in Figures 1 [Figure 1: see original paper] and 2 [Figure 2: see original paper].

Comparison of Figures 1 and 2 reveals that after 5 days of treatment with bacterial fermentation broth, the crude oil's abnormal point decreased by 2 °C. Viscosity in the non-Newtonian fluid temperature range was significantly lower than that of untreated oil. The viscosity range in the non-Newtonian region decreased from (800-3600) mpa.s to (750-3200) mpa.s, representing a reduction rate of 6.25%-11.1%. Viscosity in the Newtonian fluid temperature range remained essentially unchanged.

2.2 Fourier Transform Infrared Spectroscopy Analysis

FTIR analysis of crude oil composition before and after treatment is shown in Figure 3 [Figure 3: see original paper].

The broad, strong peak in the 3700-3100 cm⁻¹ range corresponds to O-H bond stretching vibrations, which is important for identifying alcohols, phenols, and organic acids. The broad peak shape results from high concentration causing

multimolecular association: alcohol/phenol O-H symmetric stretching at 3400-3200 cm^{-1} , free amine $-\text{NH}_2$ at 3550-3300 cm^{-1} , and associated amine $-\text{NH}$ at 3460-3420 cm^{-1} . In the 1500-600 cm^{-1} range, secondary alcohol C-O vibrations appear around 1100 cm^{-1} with broad peaks and strong intensity, primary alcohol C-O vibrations near 1050 cm^{-1} with broad peaks, and NH out-of-plane rocking at 750-700 cm^{-1} [4-5].

These results indicate that bacterial fermentation broth treatment produces organic substances similar to alcohols, phenols, and amines, which can act as organic solvents in crude oil to dissolve heavy components and reduce viscosity [6-7]. This finding aligns with the viscosity-temperature curve results.

2.3 DSC Measurement of Wax Content

Wax content, wax appearance point, and peak wax crystallization temperature were measured using a Q2000 DSC differential scanning calorimeter. After obtaining the thermogram, the following processing was performed: the point corresponding to the wax appearance point was connected to the point at $-20\text{ }^\circ\text{C}$ to create an interpolated baseline for the exothermic peak. The area enclosed by this baseline and the DSC curve represents the heat released during crystallization [8]. The average crystallization heat of wax was taken as 210 J/g [9]. Wax content was calculated using Equation (1).

Results are shown in Figures 4 [Figure 4: see original paper] and 5 [Figure 5: see original paper].

Comparison reveals distinct differences in wax appearance point, peak wax crystallization temperature, and crystallization heat before and after treatment. Using 210 J/g as the average crystallization heat [9], the wax content decreased from 29.45% to 10.75% after 5 days of treatment. The wax appearance point decreased from $42.02\text{ }^\circ\text{C}$ to $41.57\text{ }^\circ\text{C}$, while the peak wax crystallization temperature increased from $23.33\text{ }^\circ\text{C}$ to $25.62\text{ }^\circ\text{C}$.

Since paraffin alkanes primarily consist of C16-C28 saturated hydrocarbons [10], these results indicate that strain L-5 significantly degrades C16-C28 saturated hydrocarbons, converting high-carbon straight-chain alkanes into low-carbon compounds [11], confirming its suitability for wax removal in waxy crude oil.

2.4 Polarized Microscopy Observation of Wax Crystal Microstructure

Wax crystal microstructure in crude oil was observed at room temperature under $50\times$ magnification using polarized microscopy. ImageJ software was used for measurement and statistical analysis. Microscopic images are shown in Figures 6 [Figure 6: see original paper] and 7 [Figure 7: see original paper]. Measurement statistics after threshold processing are presented in Table 2, with processed images shown in Figures 8 [Figure 8: see original paper] and 9 [Figure 9: see original paper].

Feret's diameter is used to describe irregular particles by measuring the distance between parallel tangents to the particle projection profile in a given direction (perpendicular to the viewing direction in microscopic images).

The results show significant differences in wax crystal size before and after treatment. In untreated oil, wax crystals were large and aggregated into stable structures, with maximum crystal size reaching 609.1 μm . After dispersion analysis using ImageJ, the average Feret's diameter was 79.1 μm , with a minimum of 6.1 μm . After 5 days of treatment with ZL-7 bacterial fermentation broth, wax crystals became noticeably smaller and more dispersed, with maximum size decreasing to 72.1 μm . The calculated average Feret's diameter was 50.3 μm , with a minimum of 5.6 μm .

3 Conclusions

Using the discovered bacterial strain at 10% volume fraction, fermentation broth was added to waxy crude oil and incubated at 40 $^{\circ}\text{C}$, 150 rpm for 5 days. The results show: (1) Rotational viscometer measurements revealed that viscosity in the non-Newtonian fluid temperature range decreased from (800-3600) mpa.s to (750-3200) mpa.s, a reduction of 6.25%-11.1%, while Newtonian fluid range viscosity remained essentially unchanged, and the abnormal point decreased by 2 $^{\circ}\text{C}$. (2) FTIR analysis detected the production of alcohol, phenol, and amine-like organic substances after bacterial treatment, which can act as organic solvents to dissolve heavy components and reduce viscosity. (3) DSC analysis showed wax content decreased from 29.45% to 10.75%, the wax appearance point dropped from 42.02 $^{\circ}\text{C}$ to 41.57 $^{\circ}\text{C}$, and the peak wax crystallization temperature rose from 23.33 $^{\circ}\text{C}$ to 25.62 $^{\circ}\text{C}$. (4) Polarized microscopy revealed that large, aggregated wax crystals became smaller and more dispersed after treatment. ImageJ analysis showed maximum wax crystal size decreased from 609.1 μm to 72.1 μm , average Feret's diameter from 79.1 μm to 50.3 μm , and minimum Feret's diameter from 6.1 μm to 5.6 μm .

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