

INVESTIGATION OF SULFATE-REDUCING BACTERIA IN PRODUCED WATER FROM THE “NEFT DASLARI” OGED WELLS

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Sulfate-reducing bacteria (SRB) are recognized as key contributors to microbiologically influenced corrosion (MIC), a major challenge in oil and gas industries. These anaerobic microorganisms are capable of reducing sulfate ions (SO_4^{2-}) to hydrogen sulfide (H_2S), thereby accelerating electrochemical corrosion processes on metallic surfaces. Moreover, SRB activity promotes biofilm formation, which further enhances the persistence and severity of corrosion. A wide range of substrates can serve as electron donors in their metabolism, including molecular hydrogen (H_2), lactate, acetate, pyruvate, malate, ethanol, propanol, and butanol. Some SRB species, such as those from the genera *Desulfovibrio* and *Desulfobacterium*, are also capable of utilizing nitrate as an alternative terminal electron acceptor. In this study, the presence of sulfate-reducing bacteria were investigated in produced water samples collected from the production wells of the “Neft Daslari” Oil and Gas Extraction Department. Microbiological analyses were carried out using selective culture media appropriate for each microbial group. The enumeration of sulfate-reducing bacteria was performed following the NACE TMO194-2014 standard protocol of Field Monitoring of Bacterial Growth in Oil and Gas Systems. For each sample, 1 mL of formation water was aseptically transferred into a sterile glass vial containing 9 mL of Postgate’s medium. The mixture was homogenized for 5 minutes to obtain the first serial dilution. Further serial dilutions were prepared at ratios of 1:100, 1:10,000, and higher, as required. All inoculated samples were incubated anaerobically at 32 °C for 15 days. Media was autoclaved at 121 °C for 20 min. The presence of SRB was determined based on visible changes in the medium, such as blackening due to the formation of iron sulfide precipitates. Microbiological analyses of the produced waters revealed detectable populations of sulfate-reducing bacteria, with cell densities ranging from 10^4 to 10^6 cells/mL, depending on the sampling location. Chemical characterization of the produced waters suggested favorable conditions for SRB proliferation, including high salinity, sulfate availability, and reducing redox potential. The sulfide production by SRB was monitored over a 7-day incubation period to evaluate the time course of SRB metabolic activity in the Postgate (B) medium. This medium is specifically designed to promote SRB growth and activity due to its balanced nutrient composition and the presence of sulfate as the terminal electron acceptor. The experiment was conducted in triplicate using sterile 250 mL serum bottles containing 100 mL of Postgate (B) medium inoculated with the SRB culture under anaerobic conditions. The cultures were incubated at 32 ± 2 °C for 7 days. At 24-hour intervals, samples were aseptically withdrawn to quantify the produced sulfide, reflecting the metabolic reduction of sulfate. The sulfide concentration was determined iodometrically following the standard procedure of the American Public Health Association (APHA, 1989). In this method, sulfide ions react stoichiometrically with iodine in an acidic medium, allowing quantitative determination of sulfide through titration. Briefly, a measured aliquot of the culture supernatant was acidified with 6 N HCl to release dissolved sulfide as H_2S gas, which was immediately trapped and titrated with 0.025 N iodine solution using starch as an indicator. The end point was observed as a color change from colorless to pale blue, indicating the complete oxidation of sulfide. The measured sulfide concentrations (mg L^{-1}) were plotted as a function

of incubation time (days), showing a gradual increase in sulfide accumulation corresponding to enhanced SRB activity. Maximum sulfide production was observed on the 7th day, reaching approximately 400 mg L⁻¹, indicating the active reduction of sulfate to sulfide under favorable anaerobic conditions. The obtained results contribute to understanding their ecology and metabolic potential and will facilitate the development of targeted biocorrosion mitigation strategies, thereby enhancing the safety, efficiency, and environmental sustainability of petroleum extraction activities.