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INFLUENCE OF MOLYBDATE ON MICROBIOLOGICALLY INFLUENCED CORROSION OF MILD STEEL

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ABSTRACT

The effect of molybdate ion on the corrosion of mild steel in an aerobic medium containing a Pseudomonas biofilm and sulfate reducing bacteria has been investigated. Changes in specimen open circuit potentials correlated with changes in microbial activities. Formation of a microbial film by Pseudomonas on the metal surface could be characterized by changes of phase angle at the lowest frequency; and molybdate functioned as a corrosion inhibitor in the presence of sulfate reducing bacteria.

INTRODUCTION

Microbiologically influenced corrosion (MIC) is recognized as a problem in number of environmental and industrial settings (1-3). In aquatic environments, microorganisms can colonize to metals and form biofilms causing localized changes in pH, oxygen gradients and inhibitor levels at the metal surface. These changes may alter the electrochemical behavior of the metal and induce localized corrosion (4).

The role of sulfate-reducing bacteria (SRB) in the corrosion of metals has long been

noticed. A comprehensive review of SRB (5) in MIC indicated that SRB generally cause a pitting corrosion of mild steel characterized by the presence of black iron sulfide corrosion products. In the absence of oxygen, SRB use sulfate as the terminal electron acceptor for their respiratory metabolism, thereby producing significant amounts of sulfide. This sulfide will depolarize the anode and precipitate iron sulfide. The consumption of hydrogen will depolarize the cathode and drive the corrosion reaction (6). Hamilton (7) suggested that the deposition of iron sulfide might offer an increased surface area for the formation and subsequent oxidation of cathodic hydrogen. Therefore, SRB interacting with the iron sulfide and enhancing pitting corrosion is apparent.

Molybdate based corrosion inhibitors have been the materials of choice in a wide range of applications including cooling water systems. Previous studies (8-10) suggested that the formation of ferrous and/or ferric molybdates on the steel surface at passive potentials accounted for the inhibitive effect of molybdate. Ogura et al (11) also proposed that molybdate enhanced the pitting resistance with increasing the concentration, which was attributed to the polymolybdate's ability to adsorb or precipitate as a salt. However, there is no investigation of molybdate as a corrosion inhibitor in solutions containing SRB. In the present study, electrochemical techniques associated with viable bacteria cell counts were employed to characterized the efficacy of molybdate as a corrosion inhibitor in the presence of SRB.

EXPERIMENTAL PROCEDURE

Specimens. The test specimens (AISI C1020 carbon steel), 16 mm diameter disks, were supplied by Metal Samples (Munford, Alabama). The specified composition of this steel was 0.17 C, 0.42 Mn, 0.09 P, and 0.006 S, wt%. A multi-electrode probe (12) was fabricated to simplify experimental design by combining four steel disks into one probe as illustrated in Figure 1. The surfaces were wet polished in sequence with 240, 400 and 600 grit SiC paper, ultrasonically cleaned with distilled water, degreased with acetone and sterilized with 70% alcohol for 30 minutes.

Electrochemical Cell. A sterilizable, flow-through electrochemical cell, as shown in Figure 2, consisted of a 600 ml glass beaker which included 1) a four sided working electrode probe, 2) a Pt coated Nb mesh counter electrode, 3) a saturated calomel reference electrode, 4) a 0.2 μm sterile filter ventilation port, 5) a magnetically driven, Teflon-coated stir bar and 6) test solution inlet and outlet. The electrolyte was simulated to cooling water with conductivity of 3.5E-04 Siemens and contained (in g/l) NH_4NO_3 0.05, Na_2SO_4 0.12, KH_2PO_4 0.038, K_2HPO_4 0.124, $FeCl_3$ 0.33 ml of a 10 mM solution, Hutner's salt solution 1.0 ml, $MgCl_2 \cdot 6H_2O$ 0.154 and NaCl 0.073. Total organic carbon content of the electrolyte was controlled at about 0.35 g/l by adding sodium lactate 0.8 and sodium succinate 0.5 g/l. Electrolyte pH was adjusted to 7.2 with NaOH. During the test period the solution temperature ranged from 23 to 25 °C. A dual- channel peristaltic pump was used to control flow rate through the cell at 60 ± 5 ml/hr.

Bacteria. Two organisms were used: Pseudomonas fluorescens (lux), hereafter referred to as 5RL, and Desulfovibrio gigas. 5RL was selected on the basis of its ability to bioluminesce

when induced, and its adhesive characteristics in liquid culture (13). D. gigas is an anaerobic, dissimilatory sulfate-reducer. Sterile enrichment medium for D. gigas was prepared using sulfate-API broth (American Petroleum Institute), containing (in g/l): bacto yeast extract 1.0, ascorbic acid 0.1, sodium lactate 5.2, $MgSO_4$ 0.2, K_2HPO_4 0.01, $Fe(NH_4)SO_4$ 0.1 and NaCl 10.

Test Procedure. Prior to the start of each experiment the electrochemical cell was sterilized with ethylene oxide using the precautions defined previously (14). All lines to and from the cell were autoclaved to achieve sterility. The electrolyte was also autoclaved at 121 °C for 4 hours, and allowed to cool with air equilibration by a 0.2 μm filter vent. Experiments involving the addition of 0.025 g/l molybdate ions were achieved by adding filter sterilized sodium molybdate solution after the electrolyte was cooled. Inocula of 5 ml of 5RL and D. gigas from culture cells into the electrochemical cells were added at specimen exposure times of 0 and 96 hours, respectively. Upon termination of an experiment, viable bacteria cell counts from bulk solution and specimen surface were determined by acridine orange direct counts (AODC) after fixation in 2.5% glutaraldehyde (15). In addition, viable plate counts (16) were used to enumerate Pseudomonas on YEPG plates with tetracycline, and the most probable number's technique (MPN) (17) was employed to estimated the total number of D. gigas.

Electrochemical Analyses. Open circuit potential (OCP) of test specimens was monitored at an interval of 1 hour by a HP 3458A multimeter via a Keithley 706 scanner controlled by a computer. Electrochemical impedance spectroscopy (EIS) analysis was performed by a microcomputer using the Zplot software (Scribner Associates Inc.), a Solartron 1255 HF frequency response analyzer, and a potentiostat/ galvanostat 273 (option 92) from EG & G Princeton Applied Research. The applied voltage amplitude was 5 mV at frequencies between 5 mHz and 10 KHz. Five frequencies were examined per decade. An Etec Autoscan Scanning Electron Microscope (SEM) was used to identify the pitting formation.

RESULTS AND DISCUSSION

Molybdate. Preliminary experiments were performed to study the inhibiting effects of molybdate in sterile solutions. Four-sided electrode probes were placed in a sterile electrolyte and a 0.025 g/l sterilized molybdate solution, respectively. Figure 3 presents the results of open circuit potential (OCP) (mV vs. SCE) versus time plot for these specimens up to 240 hours. In the sterile control solution a steady decrease in OCP was observed after 50 hours of exposure, while in the molybdate solution, a slight increase in OCP followed by a relatively constant value was obtained. Subsequent examination of the specimens indicated that the pitting formation had occurred on the specimens which immersed in the sterile control solution. Apparently molybdate inhibits the aqueous corrosion of steel in sterile conditions.

The average corrosion rate in terms of the corrosion current density, i_{corr}, can be determined from the polarization resistance R_p as defined by the Stern-Geary equation (18). Values of R_p measured by electrochemical impedance spectroscopy (EIS) for specimens in the sterile solutions are shown in Figure 4, which reveals that this parameter behaves in a similar manner for each of the OCPs investigated. It is generally recognized that molybdate enhances

the pitting resistance, which is attributed to polymolybdate's ability to adsorb or precipitate as a salt (11). Restricted geometries of pits provides the limited mass transport conditions to prevent the diffusion of molybdate ions into the bulk solution (9). Thus, inhibition from the formation of protective layer by molybdate may act as a physical barrier to slow the metal dissolution process.

5RL and D. gigas. Pseudomonas has been frequently cited as a typical genus of slime forming bacteria (19). Therefore, in the present study an anaerobic environment was created by the addition of 5RL in the solution to investigate the influence of sulfate reducing bacteria (SRB) upon metal corrosion. A four-sided electrode probe was immersed in the solution which contained 5RL initially; but an inoculum of 5 ml SRB enrichment medium into the solution was also added after 96 hours of exposure. A comparison of OCP versus time plots for specimens in the presence and absence of bacteria is given in Figure 5. In the presence of 5RL, a rapid potential drop occurred after about 30 hours of exposure and maintained at -750 mV (SCE) for 10 hours, followed by an abrupt increase in potential toward -300 mV (SCE) and kept increasing steadily until SRB medium was inoculated. At 96 hours, the addition of SRB enrichment medium resulted in changes of solution chemistry so that a transient perturbation in potential was observed. However, a gradual decline in potential from approximately -300 mV to -500 mV (SCE) was apparent throughout the rest of the experiment.

It is generally recognized that localized corrosion occurs when environmental effects induce heterogeneities on the metal surface. The physical presence of microbial cells on the surface, in addition to their metabolic activities, modifies electrochemical processes. Adsorbed cells grow, reproduce and form colonies that are physical anomolies on a metal surface, resulting in local anodes and cathodes. Under aerobic conditions, areas under respiring colonies become anodic and surrounding areas become cathodic (20). Therefore, it is clear that the colonization of 5RL to the metal surface, followed by nucleation of localized attack caused a sudden drop in OCP (Figure 5), and subsequent potentials at about -750 mV (SCE) may indicate the propagation of local attacks. However, a mature biofilm could prevent the diffusion of corrosive species such as oxygen to the metal surface thereby reducing the metal corrosion (20). Pederson (21) has shown that the effect of Pseudomonas on metals can be dominated by protective factors under certain conditions. Hence, a relatively sharp increase in OCP (Figure 5) could imply a mature biofilm formation. Changes in the OCP in the presence of SRB could be related to changes in the localized corrosion activity. It has been proposed (6) that as the SRB metabolize organic compounds and sulfate, sulfide is produced. This sulfide will depolarize the anode and precipitate iron sulfide. The consumption of hydrogen will depolarize the cathode and drive the corrosion reaction.

Figure 6 presents the R_p versus time curves for specimens exposed to 5RL+SRB and sterile control solutions. These data suggest that although 5RL induced corrosion, formation of a 5RL biofilm might reduce the corrosion rate. In other words the 5RL biofilm may serve as a physical barrier to prevent the diffusion of oxygen to cathodic sites and the diffusion of aggressive anions, such as chloride, to anodic sites. However, under anaerobic conditions, the fact that SRB remove atomic hydrogen accumulated at the cathode and force iron to dissolve at the anode results in a increase in corrosion rate. Figure 7 shows the phase angle versus time plot at the lowest frequency (5 mHz) from EIS analysis. In the case of 5RL+SRB a steady

increase in phase angle at 5 mHz was observed, which could indicate the formation of biofilm by 5RL. This was confirmed by viable counts, which showed 8.7E+07 5RL and 1.0E+05 SRB present on the specimen surface. It has been reported (22) that phase angle indicates whether one or more times constants occur and can be used to determine the capacitances of the intact coating layer. Dowling et al (23) also proposed that indications of local inhomogeneities in the biofilm formation will be pronounced as the sweep frequency decreases. Therefore, it is apparent that EIS techniques are helpful to interpret metal-electrolyte systems involving microbial films.

SRB and Molybdate. To investigate the effects of molybdate upon SRB an electrode probe was exposed to a 5RL+SRB solution for 192 hours, followed by continuous addition of 0.025 g/l molybdate medium for time up to 280 hours. Figure 8 presents the open circuit potential versus time plot for specimens in the solutions with and without molybdate. In both cases OCP was consistent during 5RL exposure. However, the addition of SRB resulted in different responses of OCP (Figure 8). A possible explanation could be that SRB modified the biofilm produced by 5RL so as to affect SRB colonization rates, since established biofilms provide prerequisite anaerobic habitat for SRB metabolic activity and growth. In other words, SRB could change the nature of the overall corrosion mechanism and thus change the OCP. Nevertheless, a return of the OCP to levels near those of an uncorroded metal was observed (Figure 8) after addition of molybdate. It could be reasoned that molybdate rapidly depleted adenosine triphosphate (ATP) pools in sulfate-respiring bacteria, thereby blocking the formation of adenosine-5'-phosphosulfate (APS) and thus causing death (24). Viable counts support this hypothesis, since no SRB were observed on the specimen surface after addition of molybdate.

Corrosion rate, in terms of R_p, versus time plot is given in Figure 9, suggesting that molybdate serves as a corrosion inhibitor in the presence of SRB. Figure 10 reports the phase angle versus time plot for specimens in solution with and without addition of molybdate. It seems to confirm that addition of molybdate results in a reduction of SRB, and 5RL biofilm can form. Moreover, it has been held that SRB are often characterized by localized activity and results in pitting corrosion. The effect of molybdate on the localized corrosion is to enhance pitting resistance by formation of an insoluble molybdate compound (10). Kodama and Ambrose (8) have shown that molybdate ion would inhibit propagation of pits but not their initiation. Nevertheless, at pH $<$ 6 polymerization of molybdate ions results in consumption of hydrogen ions (25); hence, if pits are induced by SRB, these reactions may retard hydrogen atom formation in order to reduce a metabolic energy source for SRB. Therefore, it is clear that pits induced by SRB could be inhibited by addition of molybdate.

Visual and SEM Observations. Figures 11-13 present photographs of specimens exposed in sterile control, 5RL+SRB, and 5RL+SRB+Mo solutions. In all cases, pitting corrosion was observed after 280 hours of exposure. Figures 14-15 show micrographs for bacteria formed in 5RL+SRB and 5RL+SRB+Mo solutions, respectively. This is consistent with the viable count data which demonstrated higher bacteria densities in the 5RL+SRB solution than in the 5RL+SRB+Mo solution.

CONCLUSIONS

1. Molybdate inhibits the aqueous corrosion of mild steel in sterile conditions.

2. Changes in specimen open circuit potentials correlated with changes in microbial activities.

3. Formation of a microbial film by 5RL on the metal surface could be characterized by changes of phase angle at the lowest frequency measured by electrochemical impedance spectroscopy.

4. Molybdate functioned as a corrosion inhibitor in the presence of sulfate reducing bacteria.

ACKNOWLEDGEMENT

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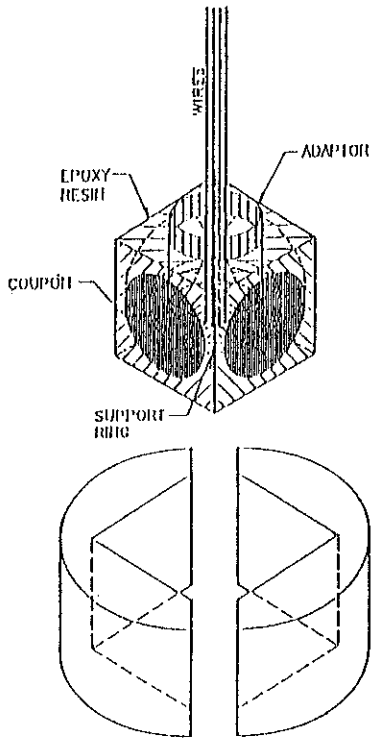

Figure 1. Schematic illustration of a four sided electrode probe.

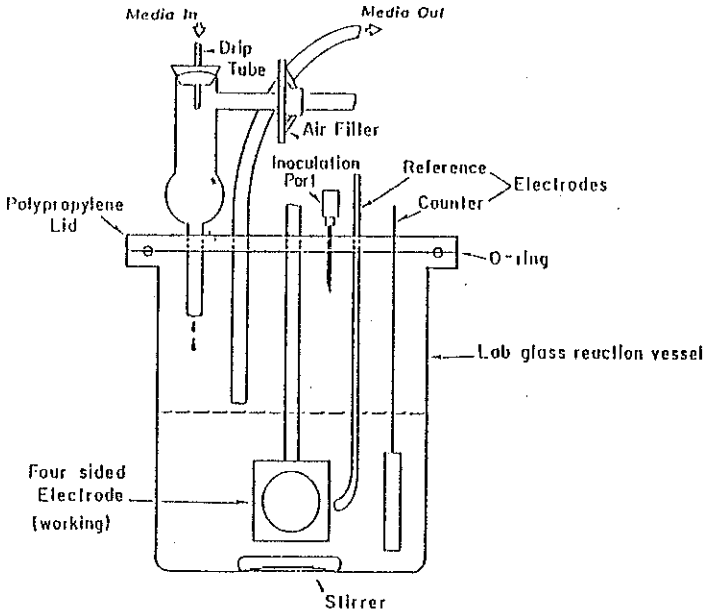

Figure 2. Electrochemical cell arrangement.

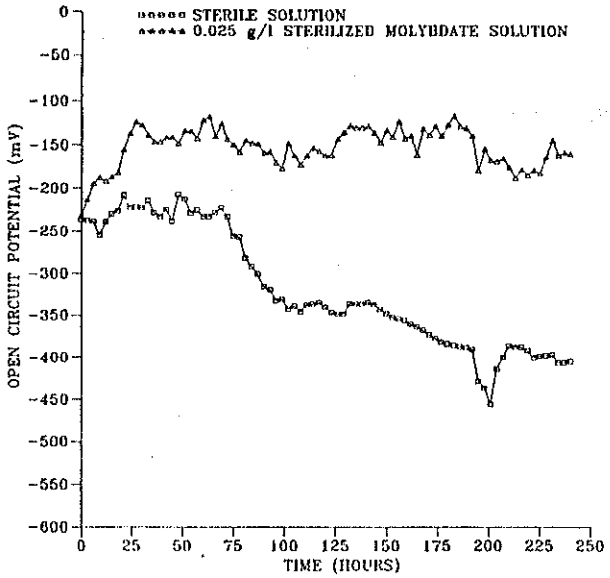

Figure 3. OCP versus time plot for specimens in the sterile solutions.

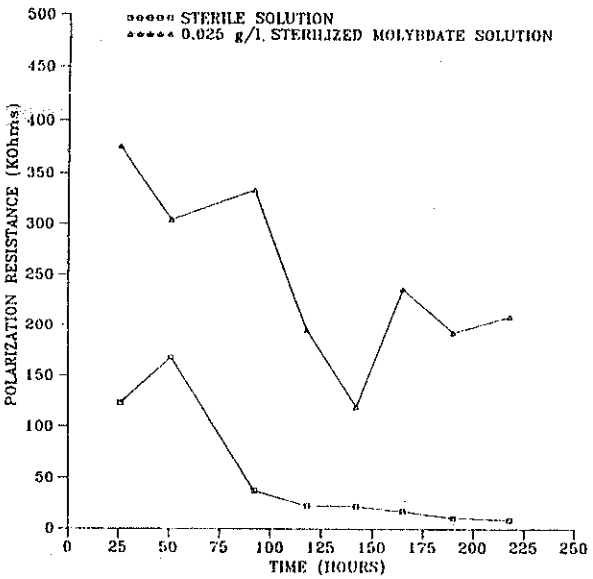

Figure 4. Polarization resistance versus time plot for specimens in the sterile solutions.

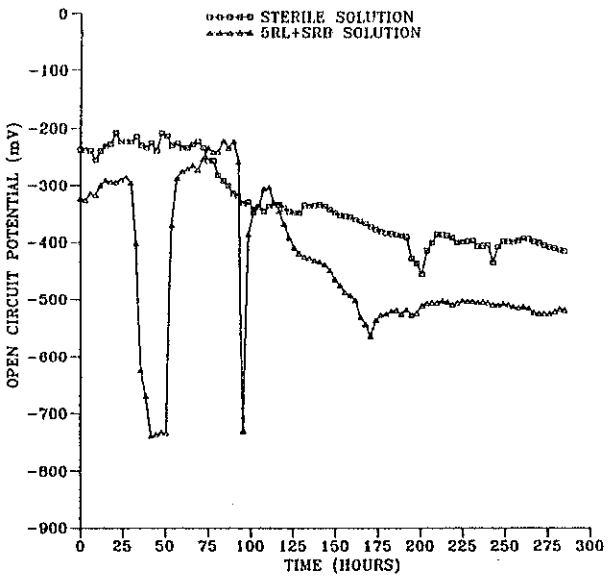

Figure 5. OCP versus time plot for specimens in the sterile solution and the 5RL+SRB solution.

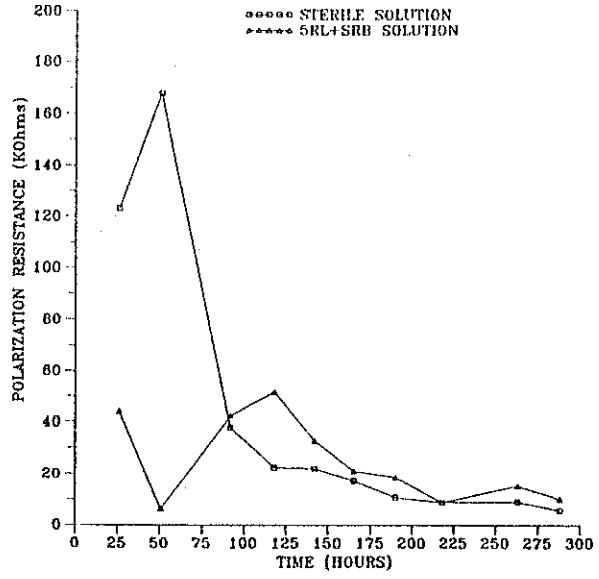

Figure 6. Polarization resistance versus time plot for specimens in the sterile solution and the 5RL+SRB solution.

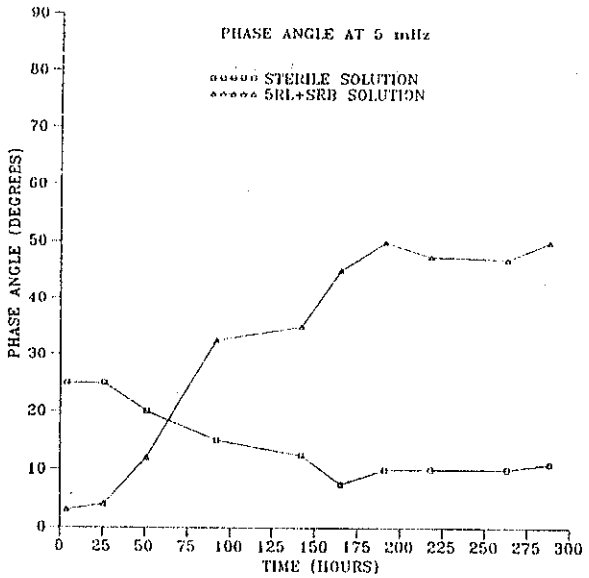

Figure 7. Phase angle (at 5 mHz) versus time plot for specimens in the sterile solution and the 5RL+SRB solution.

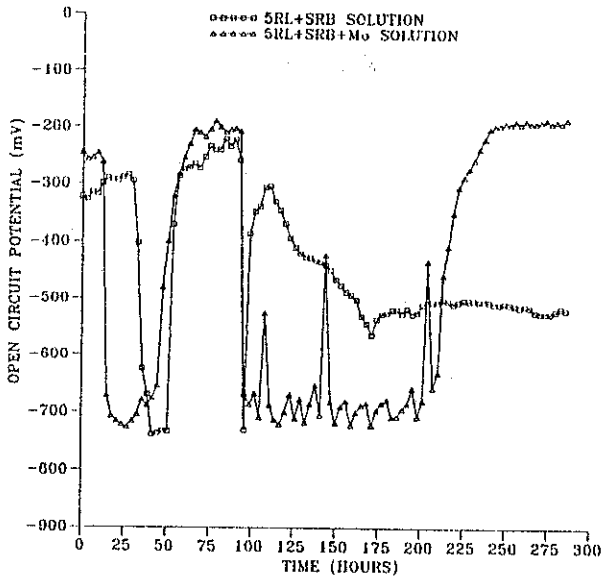

Figure 8. OCP versus time plot for specimens in the 5RL+SRB solutions with and without addition of molybdate.

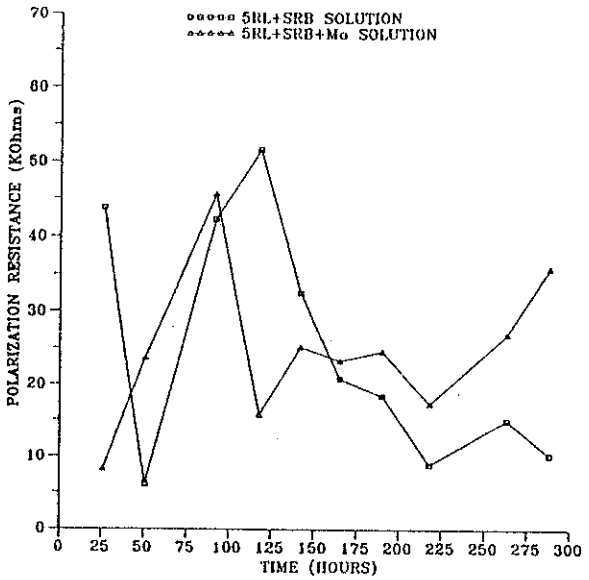

Figure 9. Polarization resistance versus time plot for specimens in the 5RL+SRB solutions with and without addition of molybdate.

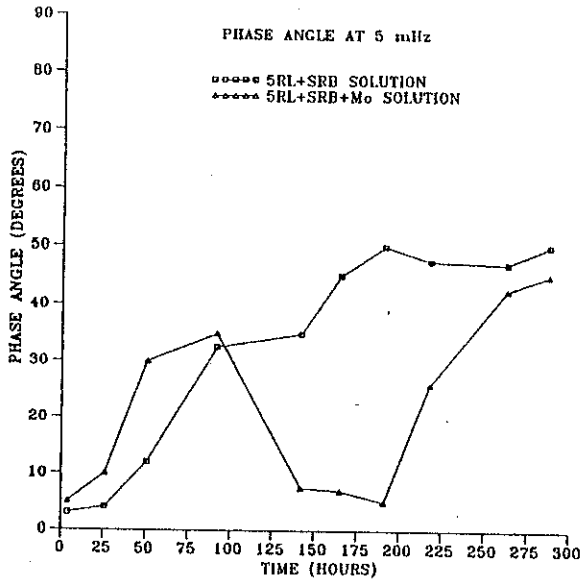

Figure 10. Phase angle (at 5 mHz) versus time plot for specimens in the 5RL+SRB solutions with and without addition of molybdate.

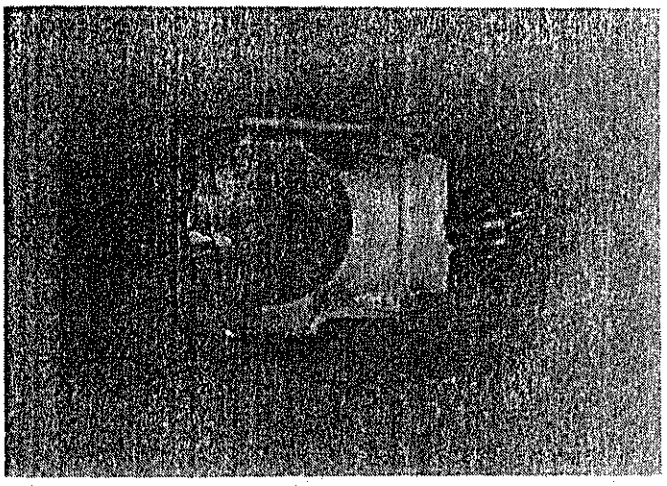

Figure 11. Specimen was exposed to the sterile solution for time up to 280 hours.

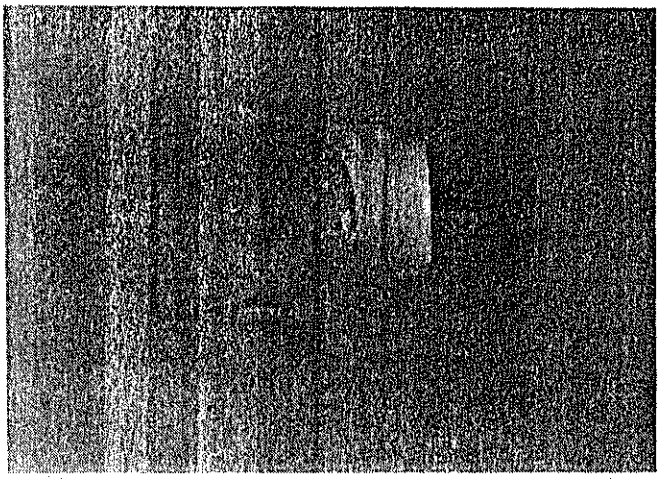

Figure 12. Specimen was exposed to the 5RL+SRB solution for time up to 280 hours.

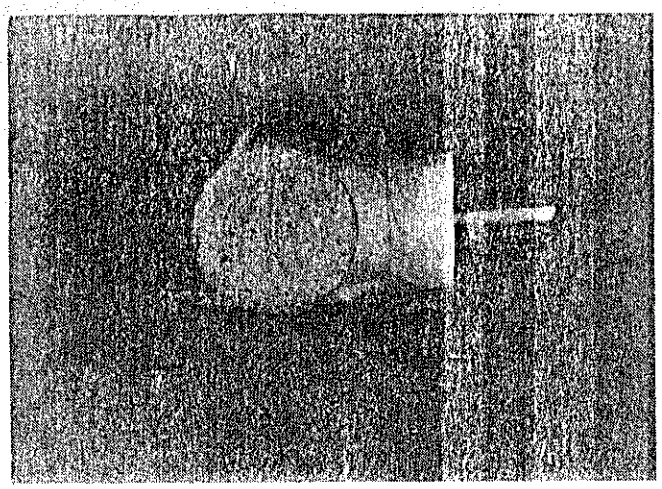

Figure 13. Specimen was exposed to the 5RL+SRB+Mo solution for time up to 280 hours.

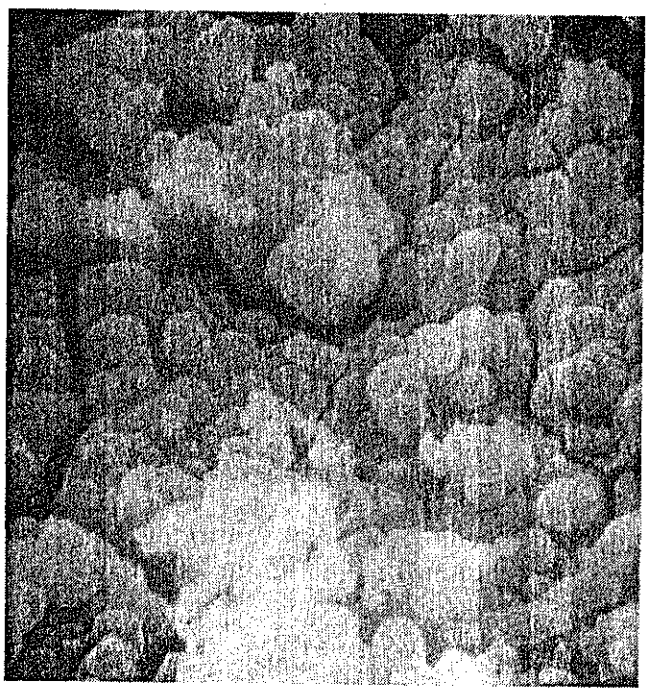

Figure 14. SEM micrograph of a specimen in the 5RL+SRB solution without addition of molybdate. Sample exposure time is 280 hours. Magnification X4000.

Figure 15. SEM micrograph of a specimen in the 5RL+SRB solution with addition of molybdate. Sample exposure time is 280 hours. Magnification X5000.