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Preliminary studies on microbial induced corrosion of ferrous materials (EN-8 and 411143 steels) in the presence of *Acidithiobacillus ferrooxidans*

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An electrochemical investigation was conducted to evaluate the corrosion behaviour of Iron alloys such as EN-8 and 411143 in absence and in presence of microbes *Acidithiobacillus ferrooxidans.* Linear polarization technique was employed to measure the Polarization Resistance and corrosion rates of two different ferrous metal samples. Polarization resistance (Rp) values of EN-8 were initially higher in the presence of *Acidithiobacillus ferrooxidans* than the values of 411143. Growth rate of the *Acidithiobacillus ferrooxidans* was more and sustained longer period with EN-8 in the presence of media than in the case of alloy '411143'. It was observed that there was a 1.5 fold increase in corrosion rate in both the cases of EN-8 and 411143 in the presence of bacteria.High corrosion rates were recorded with the steel 41143 rather than the En-8 with respect to the optical density of the microbes. It was concluded that EN-8 was exhibited high resistance to the microbial attack.

Keywords: *Acidithiobacillus ferrooxidans*, Corrosion, Linear polarization, Microbes

Corrosion is one of the most complicated and costly problems facing in municipal drinking water utilities as well as industries. Corrosion has become part of the human life includes human body due to the anthropogenic activities (pollution) in the form of swift industrialization which is causing for generation and release of more effluents into the environment/ surroundings. In addition to that the life of animals such as microorganisms, fungi, algae is also playing vital role to deteriorate materials of the industrial equipment. These are the main reasons to create imbalance in the systems which lead to decrease the longevity of the materials used in a particular systems (water pipelines, materials used in particular industrial equipment). The way causing deterioration on the surfaces of metallic and non-metallic materials to explain the kinetics of corrosion processes through the adhering (biofilms) of microorganisms such as *Acidithiobacillus ferrooxidans etc*., is defined as Microbial Induced Corrosion (MIC). It is a slow process to access the damage caused by the microorganisms in the environment, where they are sustaining. It has been explained by many workers suggesting different methodologies depend on the characteristic features of the microorganisms (Little *et al*., 1990[1]; Videla, 1996[2]; Heitz *et al*., 1996[3]; Borenstein, 1998[4]; Geesey *et al*., 2000[5]).

Authors have chosen sulphur reduced bacteria, *Acidithiobacillus ferrooxidans* which is a chemolothoaurtrophic using energy from the oxidation of iron- and sulphur-containing minerals for its growth [Colme and Hinckle[6]]. It thrives and more active at extremely low pH (1-2 units) and fixes both carbon and nitrogen from the atmosphere. Exceptional property of this microbe is its ability to aerobically oxidize solid substrates such as pyrite (FeS_2). Although, immeasurable work on studies on bioleaching of metal content from the low grade ores with *Acidithiobacillus ferrooxidans*[7-13]. Corrosion of metal alloys in particular iron affected by these microbes has not been studied well[14-16]. Metals will find a way to rid themselves of the excess electrons. This means that anything that can help metals in this respect will actually accelerate corrosion. Some of these factors are excess internal stresses, coupling dissimilar metals, bacteria and so on. The parameters initiate corrosion in any system is excess electrons in the materials, chloride ions (alkalinity), sulphate ions, humidity (60-70%), pitting on the surface (smoothness), gases such as carbon dioxide, chloride, temperature, salinity of soil and its composition

MIC can enhance the corrosion of metal by reducing the metal's susceptibility to environmental

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fracture and increasing the risk of mechanical failure. The corrosivity of the polluted marine environment increases with the concentration of the dissolved oxygen, while that of the control one does with its growing biota determining the biomass and intensity of the metal fouling. The study on corrosion of mild steel was first induced by *Klebsiella rhinoscleromatis*. The effect of the presence of bacteria in corrosion of more commonly used ferrous material equipment in such environment need a close study the thriving mechanism of these bacteria which increases the corrosion rate. In view of this, experiments were conducted with two different materials EN-8 and 411143 in a corrosion cell to obtain relevant corrosion data in the presence of *Acidithiobacillus ferrooxidans*.

The appearance of the cleaned metal surface can also provide a clued to the nature of the cause of corrosion. Pitting is indicative of bacterial attack, although some aerobic bacteria produce flask-shaped cavities below a pinhole penetration (pope ad Morris, 1995[17]). Some of the myths surrounding bio corrosion have been reviewed by Little and Wagner (1997[18]). The corrosion relevant microbes that initiate biofilm formation and, at the same time, excrete aggressive metabolites (*e.g.* sulphide ions and acids) as well as exo-polymeric substances (EPS) that are the most important component of a gelatinous biofilm, are dangerous v*ia* the biofilm the protective (passivation) film can be removed from the metal surface. Literature survey revealed that no work has been reported on the microbial induced corrosion (MIC) of EN-8 and 411143 steels (ferrous metals) using the *Acidithiobacillus ferrooxidans* using linear polarization technique. Hence, preliminary studies in this direction have initiated to estimate the effect of microbial action on these steels.

Experimental Procedure

In the present study, authors have adopted one of the electrochemical method to calculate the effect of corrosion on the metal surface by the microorganism was "linear polarization method or "linear resistance method" which has been already implemented by some authors Pesic *et al*., 2001[16]; Nivens *et al*., 1986[19]; Mansfeld *et al*., 1990[20]; King *et al*., 1986[21]. The combination of Tafel and linear polarizations was used in the present study to estimate the effect of microbial corrosion on the surface of two alloy steels EN-8 and 411143.

The PAUTOSTAT used to record polarization data is Sycopel make, fully portable, software controlled potentiostat system. Tafel polarization measurements were carried out at a sweep rate of 0.2 mV/s in the potential range of –250 mV to +250 mV with regard to open circuit potential. The experiments were conducted at the seep rate of 0.166 mV/s in the potential range of –50 mV to + 50 mV. Exposed surface area of the test specimens are given in (Table 1). Platinum electrode and saturated calomel electrode (SCE) were used as counter electrode and reference electrodes respectively. Every time the test specimens and electrodes used in the experiments have been cleaned using acetone through ultrasound after that 98% ethyl alcohol to eliminate the bacterial remnants and dried in the presence of hot air prior to the immersion them into the test fluid. Stearn-Geary equation was used to calculate I_{corr}.

$$I_{corr} = \frac{\beta_a \beta_c}{2.303(\beta_a + \beta_c)R_p}$$

Preparation of bacterial growth medium

Acidithiobacillus ferrooxidans (MTCC-2361) procured from the Institute of Microbial Technology of Council of Scientific and Industrial Research, Chandigarh was cultured in the prescribed medium containing 0.4 g.l^{-1} of KH_2PO_4; 0.4 g.l^{-1} of $MgSO_4.7H_2O$; 0.4 g.l^{-1} of $(NH_4)_2SO_4$ and 33.3 g.l^{-1} of $FeSO_4.7H_2O$ at a pH adjusted to 1.4 units with 0.1 N H_2SO_4. Stock of the isolate was cultured in 100 mL medium inoculated with 10% *v/v* of the bacterium at a constant temperature of 30°C under continuous shaking at 120 rpm for 2 h, taking care not to lose water through evaporation. The stock and pre-inoculums were stored in the said medium at 4°C. The inoculums was sub-cultured from the stock as and when required, usually once in every four weeks; for experimentation. 10% (*v/v*) inoculum expressed as cell protein concentration estimated by Lowry method[22] and 1 g of cadmium sulphide.

Specimen preparation

Test specimen was made from metal rods machined to the desired size and was polished for obtaining smooth surface and washed to remove any traces of impurities. The specimen was then fixed rigidly to the connecting rod, which served as the

Table 1 — Specifications of the test specimens

Specimen	EN-8	411143
Length (l), cm	1.0000	1.0000
Diameter (d), cm^2	0.7196	0.6207
Exposed Area (A), cm^2	0.4070	0.3025
Density(ρ), gm/cm^3	7.8900	7.8900
Eq. wt, gm	27.9235	27.9235

working electrode. The electrode was coated with a thin was film except on the bottom surface of the working electrode as the bottom area was exposed to the medium. The experiments were carried out in the bacterial growth medium both in the presence and absence of bacteria for EN-8 and 411143. The specifications of test specimen (Table 1) and their chemical compositions obtained by analysis of X-ray fluorescence are shown in the tables (Table 2).

Results and Discussion

In the present study, experiments were conducted on the strain *Acidithiobacillus ferrooxidans* interacting with ferrous metals such as EN-8, 411143 and quantified the corrosion affects using Linear Polarisation Technique with respect to the Bacterial growth. The samples were placed in a laboratory model cell, charged with the specified media and the current-potential data for definite time periods of every one hour were obtained. The total period of exposure was 120 days (5 days). In industrial operations, when sulphuric acid is in immense use as an ingredient or as reagent, the process streams flow out with a pH ranging from 1.2 to 1.4. The acid environment was likely to give an opportunity to the bacterial growth such as *Acidithiobacillus ferrooxidans* as they can thrive well in such environments. Generally, the effect of microbial induced corrosion is very much depending on the retention time of the microorganism in the electrochemical corrosion cell. This is significantly controlled by the growth of the population over time in the media. It is seen from the figure (Fig. 1), abundance of microbes gradually increased up to nearly 63 h and 56 h in the presence of alloys EN-8, 411143, respectively, which support the availability of maximum count of microbial cells and surface area of the organisms for interaction or binding with the metal surface to get the maximum corrosion rates.On the other hand, microbial counts remained more or less constant after 55-60 h, and is reached stationary phase. This phase is called as Stationary phase and after that it will reach die-off phase.

Effect of microbe on Polarization Resistance

Polarization resistance was measured for the alloys EN-8 and 411143 after 1 h 40 min of its exposure to the bacteria and without bacteria by scanning through a potential range which is very close to the corrosion potential (E_{corr}). The potential data against current were recorded, plotted and shown in the (Figs 2 & 3) for comparison. It is clearly observed from the Potential values are significantly influenced by the presence of the *A. ferrooxidans*. Range of the potential values is condensed in the presence of microbial. In the case of alloy 41143, an increase 1.25 times in the potential values recorded to the pure solution is noted. It is almost parallel to the initial values.

Polarization behaviour in the presence of microbes

The Tafel polarization curves were obtained through the plots of (Figs 2 & 3) for both the alloys, in absence and presence of microbial for the determination of βa and βc to calculate the I_{corr} values and shown in the (Figs 4 & 5). The same kind of observations was also noticed in the shifting of

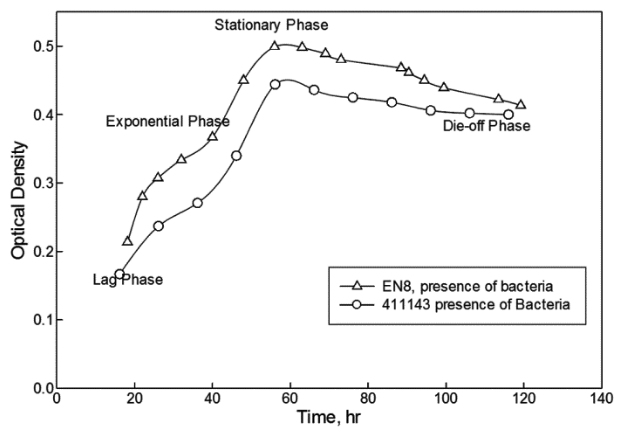

Fig. 1 — Optical growth curve of metals in the presence of bacteria

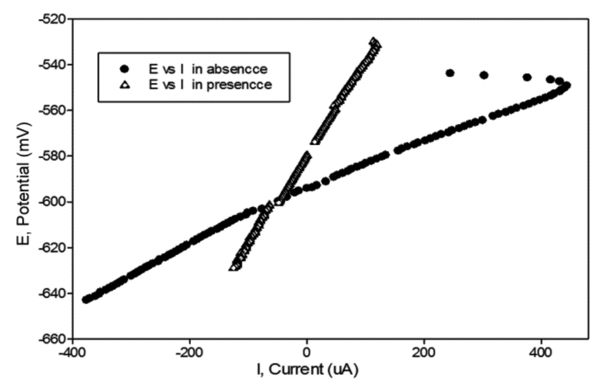

Fig. 2 — E-I with and without bacteria EN-8 steel

Table 2 — Elemental composition of these specimens

Specimen	Elements						
	Fe	C	Mn	P	S	Si	Al
EN-8	99.05	0.35-0.45	0.6-1.0	-	-	-	-
411143	98.95	0.16	0.61	0.029	0.019	0.210	0.022

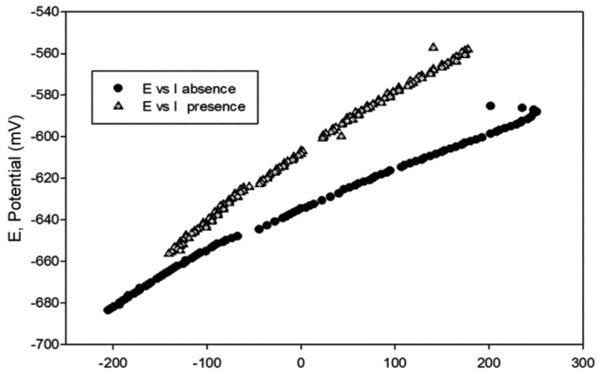

Fig. 3 — E *vs* I, with and without bacteria for 411143 steel

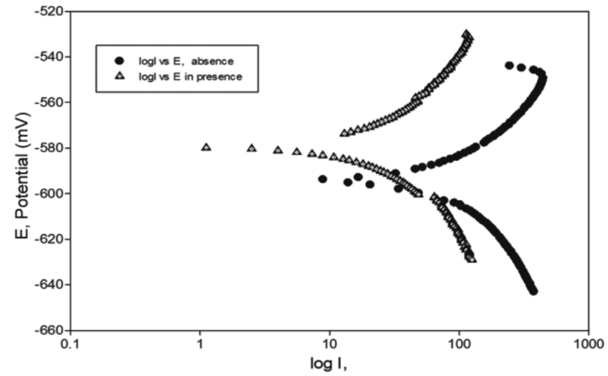

Fig. 4 — Log I *vs* E, with and without bacteria EN-8 steel

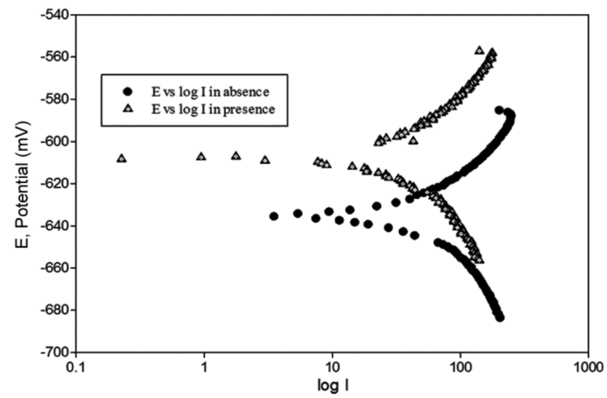

Fig. 5 — Log I *vs* E, with and without bacteria for 411143 steel

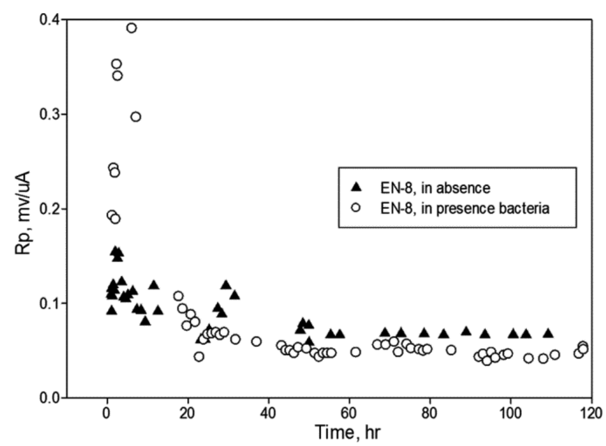

Fig. 6 — Variation of Rp (EN-8) against time - in absence and presence of bacteria

potential values (E).In the present case, βc values obtained were found to be in the range of 70-110 mV and βa values in the range of 45-92 mV.

Effect of polarization Resistance(R_p)in the presence of microbes

The data for polarization Resistance (R_p) for EN-8 both in the presence and absence of *Acidithiobacillus ferrooxidans* were plotted against time and shown in (Fig. 6). The plot revealed that polarization resistance in the presence of bacteria was found to be high in early stages and gradually decreased till it attains nearly a constant value at 53rd h onwards while the data in the absence of bacteria shows high R_p in the early stages and gradually decreased and increased as the time passes. In case of absence of bacteria, no specific trend was recorded due to formation and breaking of the protecting layer by the interaction between steel and media owing to the progression of the corrosion.

Data on Rp against time for 411143 steel coupons were graphically represented in the (Fig. 7) for two cases that are in presence and absence of *Acidithiobacillus ferrooxidans*. Rp values were veryhigh in the early stages and then decreased rapidly and attained almost nearly a constant value for both cases with and without bacteria in the corrosion media. Rp values could be reached nearly constant in between the time period of 45 h to 57 h. A comparison between the two plots of (Figs 6 & 7) revealed that the Rp values were found relatively high in the case of EN-8.

Effect of Corrosion current Density in the presence of microbes

Figures 8 & 9 depicted that the data on Corrosion current density plotted against time for both the metals with and without bacteria in the media. These plots revealed that the current density is high in the presence of bacteria by about 50% in case of EN-8 whereas, 25% increase was noted with 411143.

Effect on Corrosion Rates

The corrosion rates were evaluated for all the experimental runs for both the cases and these rates were expressed in terms mpy. A close inspection of the data (Figs 10 & 11) revealed that the presence of bacteria has increased the corrosion rate by 1.5 times than that observed in the absence of bacteria. It is concluded from the data, the presence of microbes has shown a positive increase in the corrosion rate for the cases of steels.

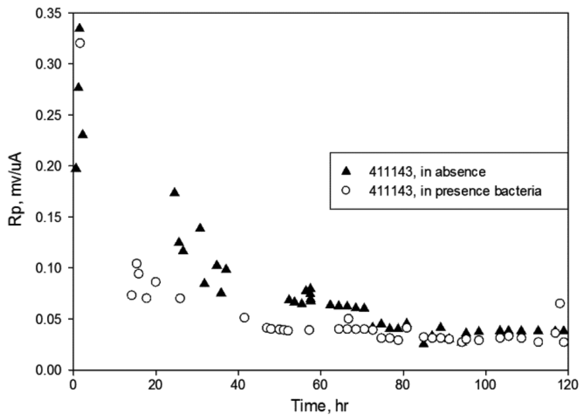

Fig. 7 — Variation of Rp (411143) – in absence and presence of bacteria

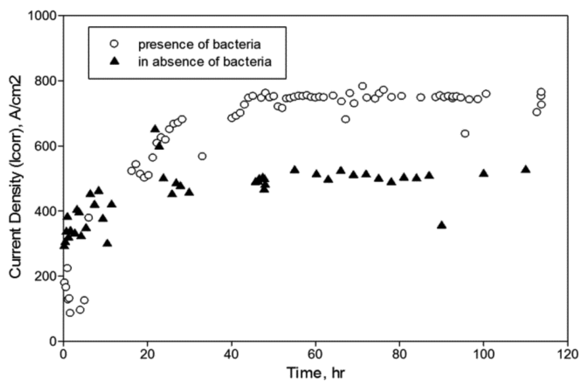

Fig. 8 — Current density (EN-8) comparison

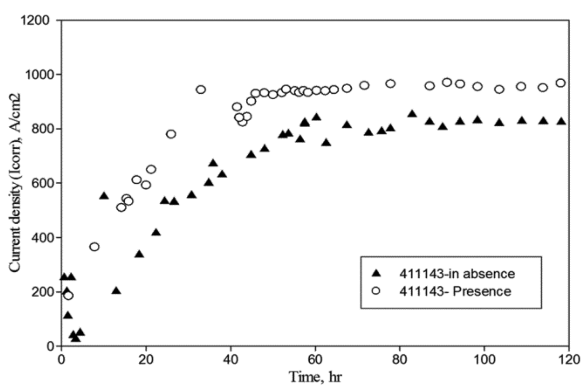

Fig. 9 — Current density comparison

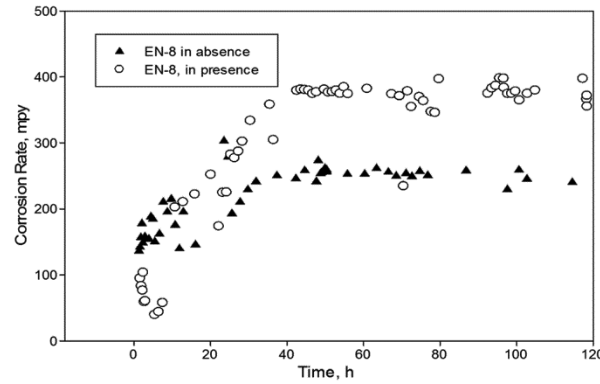

Fig. 10 — Comparison of corrosion rates EN-8 with and without bacteria

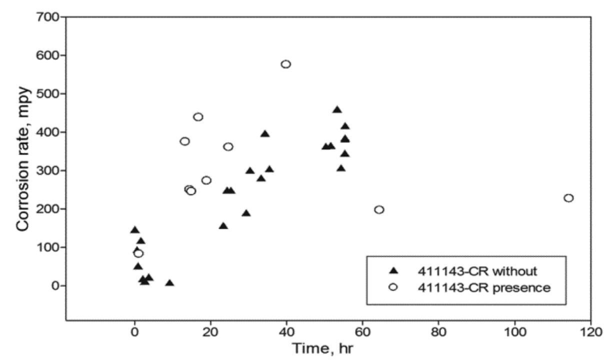

Fig. 11 — Corrosion rates of 411143 steel - Comparison

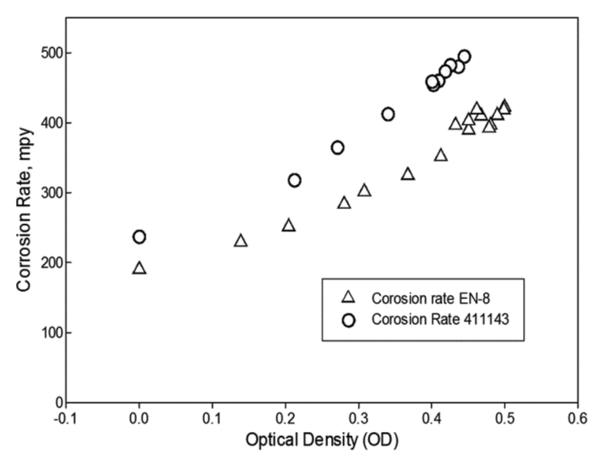

Fig. 12 — Variation of corrosion rate with bacterial growth – Comparison between EN-8 and 411143

Optical Density

As noticed from the optical density curves of the bacteria for both steels, there are four phases have been identified for sustainability of the microbial in the given environment (media) in the presence of steels. Among the four phases, stationary phase is more important to explain the mechanism of corrosion at a particular period where maximum growth of bacteria could observed. Corrosion rates of two steels were compared with respect to the optical density in the (Fig. 12). Both have attained maximum values at the stationary phases where maximum number of growth is available. It is evidenced by the recording of maximum values of optical density. For all the cases, higher values of corrosion are recorded with 411143 steel rather than the EN-8 steel. This is because of formation of excess of acid that would have been formed due presence of P, S (FePO4, H2SO4) in the case of 411143.

Conclusion

Experiments were conducted with two different materials EN-8, and 411143 (both ferrous based) in a corrosion cell to obtain relevant corrosion data in the presence of microbial. Based on about 130 experimental runs the following conclusions are drawn: (1) In case of EN-8, in the presence of *Acidithiobacillus ferrooxidans* the polarization resistance showed a very high value of Rp in the early stages of exposure gradually fell a minimum at 53rd h of exposure, and then onwards the values remain nearly constant. In case of 411143, fall in Rp values was rapid up to 46th h of exposure; beyond this they remain nearly constant. Rp values are found to be lower for the material 411143. (2) In the case of EN-8, the corrosion current density were found to increase to a maximum upto 53rd h of exposure time are then stabilize while in case of 411143, the current density values show a maximum at 46th h of exposure. (3) (i) The growth of bacteria *Acidithiobacillus ferrooxidans* in the medium with the exposure period was identified by the optical density values of the medium taken at regular periods of exposure. (ii) In the case of 411143, the log phase has reached its maximum at 46th exposure time beyond which the growth rate was stabilized. (4) The stabilized corrosion current data beyond these periods of exposure both for EN-8 and 411143 may be attributed to the attainment of stabilization phase and hence approach to constant growth rate. (5) The increase in corrosion current density is due to the bacteria, was found 1.5 times in both the cases of EN-8 and 411143 over that in absence of bacteria in the medium. (6) En-8 has shown relatively low corrosion rate compared to 411143. Between the two materials, EN-8 exhibited relatively high resistance to attack of *Acidithiobacillus ferrooxidans.*

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Conflict of interest

All authors declare no conflict of interest.

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