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Anti-corrosion and Anti-bacterial properties investigation of GrapheneOxide coating on AISI 304 stainless steel

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Abstract

In this work, the corrosion resistance and antibacterial properties of Graphene Oxide were investigated. Hummer's method was used for production of Graphene Oxide on AISI 304 stainless steel, and its formation was tested by Raman spectroscopy. Electrochemical Impedance spectroscopy (EIS) and Polarization tests in 3.5% NaCl solution were carried out for corrosion inhibition studies of the produced samples. Results of corrosion measurements revealed that coating of GrapheneOxide on stainless steel greatly improves its corrosion resistance. Surface morphology of the samples after corrosion tests was examined by scanning electron microscope analysis (SEM). The equivalent circuit of all samples was obtained, using the EIS data. Japanese Industry Standard test Methods was used for antibacterial properties investigation against gram negative E. coli bacteria. The results of antibacterial analysis showed that Graphene Oxide with anti-corrosion property, is promising material for antibacterial applications.

Keywords:GrapheneOxide; Corrosion; Electrochemical impedance spectroscopy; Equivalent circuit; E. coli.

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Introduction

Stainless steel (e.g., types AISI 304 and AISI 316) has many applications in industry and technology (such as oil and gas industry, offshore platforms, nuclear reactors, aerospace) due to its good mechanical properties and good corrosion resistance[1,2]. However, when exposed to environments consisting of aggressive ions such as $Cl^{-}$and S^{-2} (particularly at higher temperatures and environments with high or very low pHs) it does not show a high degree of corrosion resistance [3-5]. Therefore, improving the corrosion resistance of this highly used material in different environments is very necessary. The corrosion behavior of AISI 304 and AISI 316 types of stainless steels for various applications is evaluated in different studies, using different methods namely ion implantation [6], sol gel deposition [7, 8], chemical deposition [9] and physical deposition [10]. Variation of the ratio of different elements in the structure of stainless steels results in different corrosion behavior in each corroding medium [11, 12].

In recent years, researchers' efforts in improving the corrosion inhibition of materials has led to the use of Graphene as a good candidate which as a two-dimensional carbon structures and because of high electrical conductivity and transmittance [13-16] Graphenecan act as a good protection against corrosion without any change in its physical properties. In 2008 Bunch et al. [17] showed that Graphene is a non-penetrable material for gas molecules. Later, Chen et al. [18] reported that Graphene coating prevents oxidation of copper. Recently, it is shown that production of Graphene by chemical vapordeposition can provide a resistant coating against corrosion [19-22].

GrapheneOxide is a structure consisting of Graphene layers that oxygenated groups such as hydroxiles, carboniles, carboxiles and epoxies are attached to it [23]. Graphene Oxidealso acts as a suitable material for corrosion protection [24, 25]. Although, Graphene Oxideis a soluble material in water and the performance of Graphene itself is much better than GrapheneOxide, but due to easier and more economical methods of mass production of GrapheneOxide in large dimensions,Graphene Oxidecan be a good substitute for Graphene. In recently years, Graphene Oxide is used for corrosion inhibition of copper [24] and aluminum [25].

Antimicrobial coatings inhibit the ability of microorganisms to grow on the surface of a material. Such materials widely investigated for possible use in various applications in clinics, industry, and home. The most important use of antimicrobial coatings has been in the healthcare setting for sterilization of medical devices to prevent hospital associated infections [26]. In addition to medical devices, linens and clothing can provide a suitable environment for many bacteria, fungi, and viruses to grow when in contact with the human body which allows for the transmission of infectious disease [27].different materials such as Cu, Ag, Au [28], Zn [29], ZnO [30], Mn, Fe, CdS [31], Al, Si, Ti, V, Cr, Mn, Co, Ni, , Zr, Nb, Mo, Pd, Sn, Ta, W, Pb, Pt [32], different polymers [33-

35], Graphene [36] and graphene Oxide [37] have been used in the antibacterial field, But a coating which is economical and low-cost in addition to its antibacterial properties, is one of the most interesting coating in the medicine investigation. The Hummers method is a cheap method of the production of graphene oxide coating. This method can easily be used to mass-production of Graphene Oxide at low cost for antibacterial applications.Many researchers have studied the antibacterial properties of Graphene and Graphene oxides [38-41], but there is no research that has examined the antibacterial properties with anti-corrosion properties using the Hummers method.

In this work, Graphene Oxide was used as a cheap and easy mass production material (Hummers method) FOR corrosion protection with antibacterial properties. The reason for examining the antibacterial properties along with the anti-corrosion properties is that an antibacterial anti-corrosion agent can be very useful in medical applications.

Experimental

After cutting the substrates to the desired size of (20 × 20 × 1 mm), the polyethylene sheet that was covered the surface of the stainless steel for scratching protection, was removed by soaking in ethanol. No mechanical polishing or any other disturbance was applied on the produced steels by the production factory and their surface roughness was measured as received materials. Prior to beginning work all substrates were ultrasonically cleaned in heated acetone then ethanol.

Hummer's method was used for production of Graphene Oxide. In this method, 27 ml of sulfuric acid ($H2SO_4$) and 3 ml of phosphoric acid ($H3PO_4$) (volume ratio 9:1) were mixed and stirred for several minutes. Then 0.225 g of graphite powder was added into mixing solution under stirring condition. 1.32 g of potassium permanganate ($KMnO_4$) was then added slowly into the solution. This mixture was stirred for 6 hours until the solution became dark green. To eliminate excess of $KMnO_4$, 0.675 ml of hydrogen peroxide (H_2O_2) was dropped slowly and stirred for 10 minutes. The exothermic reaction occurred and let it to cool down. 10 ml of hydrochloric acid (HCl) and 30ml of deionized water (DIW) was added and centrifuged using Eppendorf Centrifuge 5430R at 5000 rpm for 7 minutes. Then, the supernatant was decanted away and the residuals were then rewashed again with HCl and DIW for 3 times. The washed Graphene Oxide solution was dried using oven at90 °C for 24 hours to produce the powder of Graphene Oxide.To produce graphene oxide solution,1 mg of Graphene Oxide wasdissolved in 1 ml of ethanol solution (volume ratio 1:1) under ultrasonic for 1 hour.

In order to coating on the surface of AISI304 SS, a drop of solution was positioned on the AISI304 SS and heated to 40 ˚C for half an hour until Graphene Oxide was fixed on the substrate. Raman spectroscopy with a wavelength of 785 nm (Senterra, Bruker, Germany 2009) was used to examine the formation of Graphene Oxide on the substrate.

The electrochemical tests were performed using potentiodynamic method with a potentiostat coupled to PC (273A, EG & G, USA). In order to carry out this analysis only an area of 1.0 ± 0.05 cm^2 was exposed to the 3.5 wt% (0.6 M) NaClcorroding medium. Electrochemical impedance spectroscopy (EIS) was performed using a potentiostat coupled to PC (Ivium, De Zaale 11, 5612 AJ Eindhoven, Netherlands) with reference to the open circuit potential (OCP) and in the frequency range of 100 KHz to 0.01 Hz with a voltage amplitude of 0.01 V.In order to carry out this analysis only an area of $1cm^2$ was exposed to the 3.5 wt% (0.6 M) NaCl corroding medium. The rest of the surface area and back and edges of the sample were coated with a thick resin of lacquer paint. The samples were polarized in a 0.6 M NaClsolution made of analytical grade reagent and double distilled water. The samples were put into an inert mounting fixture (polyamide). This allowed an electrical contact to be supplied to the sample, without being influenced by undesirable effects on the working electrode.

A saturated calomel electrode was used as a reference electrode and a platinum electrode used as auxiliary one while the test sample was mounted in an inert fixture (polyamide) acted as the working electrode. Before starting the EIS test, the samples were immersed in the solution and the OCP measurement was carried out until it was stabilized [42].The polarization potential was applied via a copper wire contacted to the back side of the sample in the fixture, while a saturated calomel reference electrode (SCE) and a platinum counter electrode were used in a three-electrode setup. The potential sweep rate was 1 mV s^{-1}, and was scanned for each sample starting from -1.0 V vs. open circuit potential. The ends of the scans were selected 1.0 V. All of the potentials presented in this work are as a function of SCE. All measurements were performed at 298 K. The corrosion current density j_{corr} and the corrosion potential E_{corr} were calculated from the Tafel extrapolation of polarization curves of semi-logarithmic plot using the method prescribed in [43]. In order to make an accurate Tafel extrapolation we started our extrapolation at least 50-100 mV away from E_{corr}, while as prescribed in the literature [44, 45], our polarization curves also contain at least one linear (semi-logarithmic) scale which facilitates accurate extrapolation. After corrosion test the surface morphology of the samples was analyzed using scanning electron microscope (SEM).

Anti-bacterial properties of the Graphene Oxide film against gram negative Escherichia coli bacteria were investigated using the JIS Z 2801 method [46]. The AISI 304 substrate without Graphene Oxide coating, was used as the control sample bacteria cells cultured in test inoculums were placed on the surface of the AISI304 SS and Graphene Oxide on AISI 304 SS(GO/AISI304 SS) samples. The samples were incubated for 48 hours in a 40°C incubator. Immediately after inoculation and after incubation for 48 hours, the number of viable bacteria cells was determined by using the agar plate culture method. The number of viable cells of bacterial on each sample was calculated to be 500000 CFU/mL.For this, the sample was thoroughly washed by ethanol wiper to clean both surfaces before being placed in different labelled sterile Petri dishes and 0.5 mL of the test inoculum was dropped, using a pipette, on the surface of each sample. The Petri dishes were capped and incubated for 48 hours at 40 C and 90 % relative humidity.

For enumeration of the number of viable bacteria cells 10 mL washing broth of each inoculated sample was used to completely wash out the bacterial cells from the sample surface. And then 1 mL of each washing broth was used to make 10-fold serial dilutions in phosphate buffered physiological saline. From each of the washings and their respective dilutions, 1 mL was added to different Petri dishes and then followed by adding 15 mL of the plate count Agar. Then these dishes were incubated at 40°C and 90% relative humidity for 48 hours. After incubation, the colonies on each of the dishes were counted for calculating the number of viable cells.

Results and discussion

Raman spectroscopy

The Raman spectroscopy of the AISI304 SS coated with Graphene Oxide was carried out in 200 cm^{-1} to 3500 cm^{-1} range and the obtained result is given in Figure 1. Three peaks at 1289 cm^{-1}, 1565 cm^{-1} and 2601 cm^{-1} in this figure belong to D, G and 2D structures, respectively. The ratio of intensities G/2D>>1 confirms the formation of multilayer Graphene and the presence of D peak is indicative of Graphene Oxide formation.

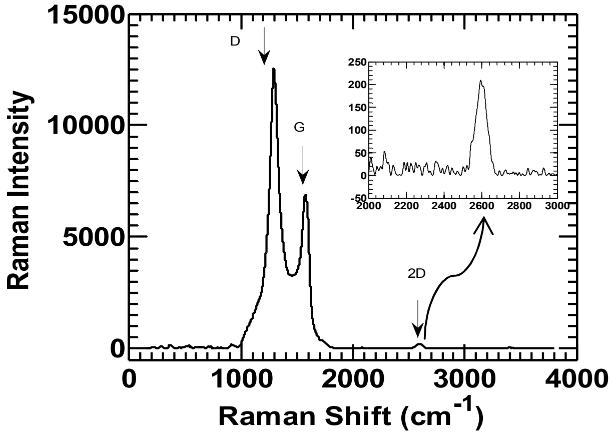

Figure1. Raman spectrum of AISI304 stainless steel coated with Graphene Oxide.

Corrosion tests

The potentiodynamic and Nyquist curves for the bare AISI304 SS substrates and the coated AISI304 SS are given in Figures 2 and 3, respectively.

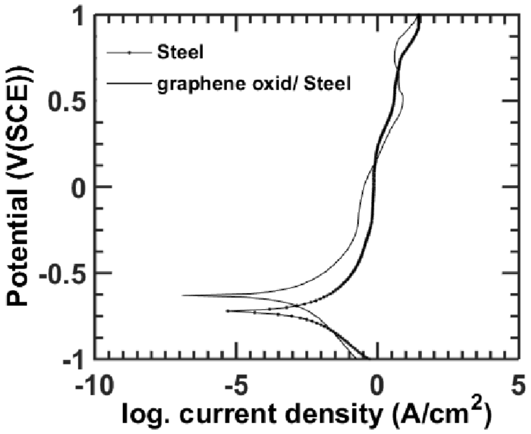

Figure 2. Potentiodynamic polarization curves for bare AISI304 stainless steel and Graphene Oxidecoated AISI304 stainless steel.

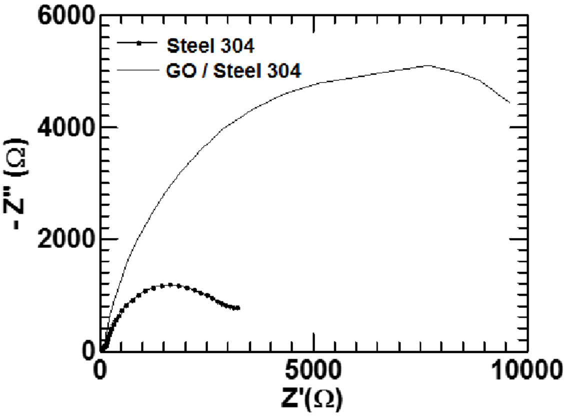

Figure 3. Experimental Nyquist diagrams for the bare AISI304 stainless steel and the Graphene Oxide coated AISI 304 SS.

The results of polarization test show that the sample with Graphene Oxidecoating has less corrosion current and more corrosion potential than the sample without coating, which indicates less corrosion tendencyof this sample. The corrosion current and corrosion potential of the coated sample and the sample without coating are given in Table 1.It is also clear from Figure 3 that samples with Graphene oxide coating have high electrical impedance and therefore high corrosion resistance

Table 1. Corrosion current density and Corrosionpotential for bare AISI304 and **Graphene Oxide**on AISI 304 stainless steel in 3.5 wt% NaCl medium.

Sample	**I_{corr}(A)**	**E_{corr}(V)**
AISI 304	**32×10^{-4}**	**-0.7**
GO/AISI 304	**15×10^{-5}**	**-0.6**

To better evaluate the corrosion resistance of the samples, the equivalent circuit of the samples was obtained and analyzed. Figure 4 shows this equivalent circuit.

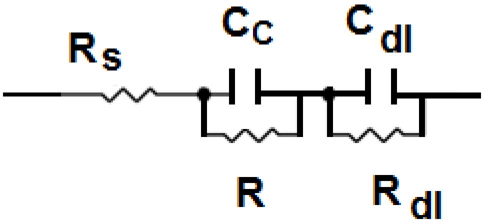

Figure 4. Electrical equivalent circuit of the experimental EIS data for both bare AISI304 stainless steel and the Graphene Oxide coated AISI 304 SS.

In these circuits R_S is the solution resistance, C_c is the capacitance of the coating that is obtained for incomplete coatings that may be formed when the solution is penetrated into the coating. C_{dl} is the capacitance of double layer which forms at the interface of metal-coating. Due to in homogeneity of the formed layer structure and the roughness of the coating surface, C_c and C_{dl} capacitors are not in an ideal state. Therefore, we may introduce parameters that identify the deviation of their capacitance from the ideal state as α_1 and α_2. Bod and Phase diagrams obtained from the best fit procedure between experimental and simulation results discussed in above for both bare and coated AISI304 SS samples are compared in Figure 5.

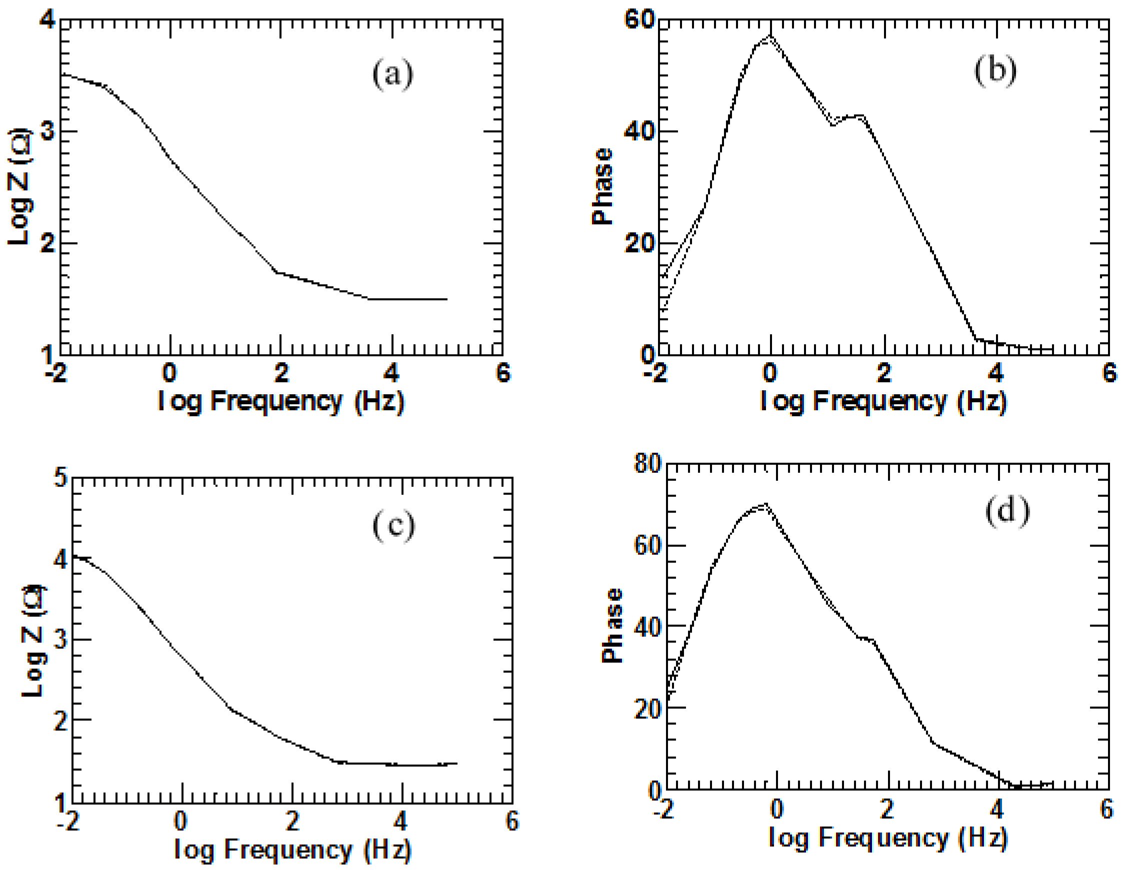

Figure 5. a and b) Bode and phase diagrams for the bare AISI304 SS, respectively. C and d) Bode and phase diagrams for the Graphene Oxide coated AISI304 SS, respectively.

Table 2 shows the quantities obtained from the fit between the experimental results and the equivalent circuit simulation (with Zview software).Reduction of capacitance and increase of resistance in the equivalent circuit by coating of substrate with Graphene Oxide is indicative of improvement of corrosion inhibition of the AISI304 sample.

Table 2. Polarization parameters of AISI 304 and Graphen Oxide/AISI 304.

Parameters	**AISI 304**	**GO/AISI 304**
R_s (Ω)	30	29
R (Ω)	211	87
R_{dl} (Ω)	3073	12750
C_{dl}(F)	4×10^{-4}	3×10^{-4}
C_C (F)	1.3×10^{-4}	7.3×10^{-5}
α 1	0.85	0.93
α2	0.85	0.83

SEM results

In order to observe how the samples are affected in the corroding medium, after completion of the potentiodynamic test, SEM analysis was performed. Figures 6(a) and 6(b) show the SEM images of bare AISI304 SS and the AISI304 SS coated with graphen oxide samples after corrosion tests. From the comparison of the SEM images of these samples it can be clearly concluded that the coated samples are almost remained intact with no obvious damage on its surface.

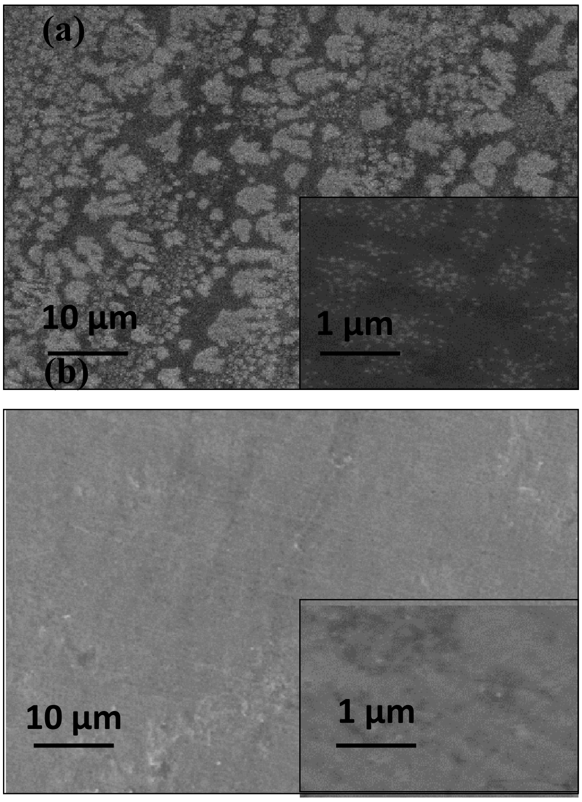

Figure 6. SEM images from the surface of; a) bare AISI304 SS and b) Graphene Oxidecoated AISI304 SS, after corrosion tests.

Antibacterial results

The antimicrobial behavior of Graphene Oxide coating against E. coli cells was tested and the results are shown in Figure 7. The number of E. coli cells on the surface of samples , immediately after inoculation, was 5.45×10^4 CFU mL^{-1}.the number of viable cells on the surface of AISI 304 and Graphene Oxideon steel after 48 hours incubation was 5.05×10^4 CFU mL^{-1} and 7.2×10^2 CFU mL^{-1} respectively. These results indicate that the Graphene Oxidecoating has strong antibacterial activity to the gram negative E. coli bacteria.

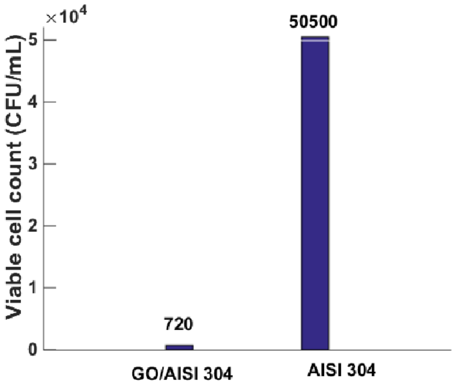

Figure 7. The number of E. coli cells on AISI 304 and GO/AISI 304, after incubation at 40°C for 48 hours.

Antimicrobial activity of Graphene Oxide film against E. coli bacteria was calculated from equation 1:

$$R = \log \left(\frac{B}{C}\right) \qquad (1)$$

Where B is the average number of viable cells on the control sample (AISI 304) after 24 hours and C is the average number of viable cells on the antibacterial sample (GO/AISI 304) after 24 hours. The results are shown in Figure 8. It can be seen from the results that Graphene Oxide exhibited very strong antibacterial activity against the gram-negative E. bacteria, and the blank sample (AISI304) exhibited no significant antimicrobial activity.

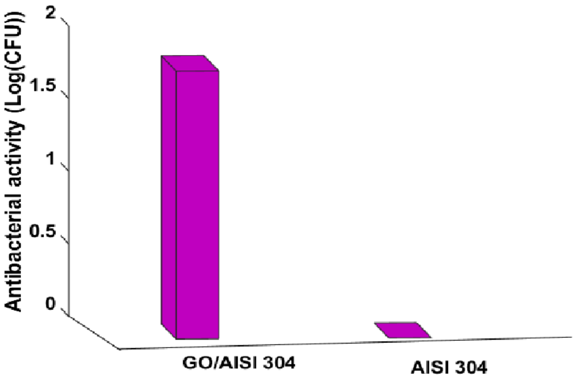

Figure 8. Antibacterial activity of graphen Oxide against E. coli bacteria.

Conclusions

A simple and economical method was used for production of Graphene Oxide which was also easily coated on the AISI304 stainless steel. Influence of the Graphene Oxide coating on the corrosion inhibition of steel in 3.5 wt% NaCl solution was investigated using both EIS and potentiodynamic methods. Results of these tests together with equivalent circuit simulation results showed that corrosion resistance of steel greatly increases once it is coated with Graphene Oxide. Equivalent circuit parameters showed that capacitance of capacitors decreases and electrical resistances increases by coating of Graphene Oxide which leads to improvement of corrosion resistance of the sample. The antibacterial properties investigation of Graphene Oxide against gram negative E. coli bacteria showed that graphene Oxide is a good and low cost material for anticorrosion applications. Therefore, it can be concluded that graphene oxide is suitable material for anti-corrosion applications with antibacterial properties.

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