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CORROSION INHIBITION OF CASEIN AS A NATURAL PROTEIN FOR 316L STAINLESS STEEL IN SIMULATED BODY FLUID

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ABSTRACT

The importance of biomaterials is increased steadily with the augmentation of population and aging. In this research, casein, a protein present in milk, used to modify the 316L stainless steel (316L SS) alloy surface, used in medical application, against corrosion in the corrosive body fluid. Due to the presence of hetero-atoms in its moiety, casein has the ability to form a protective thin layer. Self-assembled monolayers (SAMs) technique is a common tool providing a well ordered thin film on the metallic surface. Casein was assumed to form SAMs on the 316L SS surface and the corrosion inhibition efficiency was examined electrochemically. The results proved the formation of SAMs and displayed that casein reduce the corrosion rate and serves as a mixed type inhibitor. The inhibition efficiency is increased by increasing the casein̓s concentration and immersion time in the casein’s solution. A maximum inhibition efficiency obtained at 1000 ppm with immersion time equals 120 minutes. Further modification is undertaken to increase the inhibition efficiency.

Key words:

Biomaterial, corrosion inhibition, SAMs, natural protein, casein, 316L stainless steel, SBF

1. INTRODUCTION

The primary feature of biomaterials is that they are used in contact with the living body. Biomaterial can be defined as a biocompatible material, natural or synthetic, that used to replace or repair parts of an organ or tissue in the human body [1-3]. Biomaterials must display some requirements in their design such: biologically non-toxic, excellent biocompatibility, good mechanical properties, osseo-integration and high corrosion and wear resistance [1, 4-5].

Medical grade stainless steel 316L widely used as a metallic implant due to ease availability, lower cost, accepted biocompatibility and its corrosion resistivity is very high [1, 3, 6].

The formation of (SAMs) on the metal surface is one of the simplest method used to reduced the corrosion rate of metal by blocking the active sites present in the surface of metal [7-14].

Caseins are soluble milk proteins exist as micelles of four different types of protein; each one of them contains different types of amino acid besides minerals such as phosphorous and calcium [15-16]. These proteins have many usages such as manufacture of plastics and paints, and it was also used in food industries [17-19]. In surface science, few investigators have been inspected the effect of casein on the corrosion of metals [20-22]. Lately, the effect of casein on the rate of corrosion of mild steel in 0.1 M HCl was done by T. Rabizaden et al., 2019 [22].

In this work, casein is supposed to form SAMs on the surface of stainless steel 316L. These SAMs are investigated electrochemically by electrochemical impedance techniques and potentio-dynamic polarization.

2. EXPERIMENTAL METHODS

2.1. Preparation of the Working Electrode

A plate of Stainless steel 316L, with known chemical composition [23-24] was cutted into small samples. The surfaces of these samples were polished with emery papers have different degrees (320, 400, 500, 600 and 1200) to mirror finish. Only 2 × 1 cm^2 was exposed to electrolyte while the remaining area covered with Teflon tap and a coat material. The samples were then cleaned with acetone and washed several times with double distilled water then dried at ambient temperature before each measurement.

2.2. Self-assembled Monolayers Formation

The working electrodes were immersed for different time intervals (60, 90 and 120 min) in solutions of 0.1 M $NaHCO_3$ containing several concentrations (100, 300, 500, 700 and 1000 ppm) from casein at room temperature.

2.3. Electrochemical Measurements

The measurements performed by using three-electrode cell which are saturated Calomel electrode (reference electrode) , platinum electrode (counter electrode) and blank and treated samples of 316L stainless steel which used as working electrodes. The tests were done in 100 mL solution of SBF at 37 °C as electrolyte solution that was intended by to Kokubo [25].

2.3.1. Potentiodynamic Polarization Measurements

The electrodes immersed in simulated body fluid solution at open circuit potential to attain steady state before measurements. Potentiodynamic polarization studies performed in scan rate of 0.5 mVs^{-1} with a potential range from -250 to 250 mV. We used Potentiostat/Galvanostat (model PGZ 301, Voltalab, Radiometer Analytical – France) controlled with VoltaMaster software Version 4.0 for these measurements. The inhibition efficiency of casein (η%) can be calculated using the following equation [9, 13-14]:

$$(\% = (I\_1 - I\_2\ )/I\_1\ \times 100 \qquad (1)$$

where I_1 is the corrosion current density in the absence of inhibitor and I_2 is the corrosion current density in the presence of inhibitor.

The surface coverage θ can also calculate from equation [14, 26-28]:

$$\theta = ((\%)/100 \qquad (2)$$

2.3.2. Electrochemical Impedance Studies (EIS)

The measurements of electrochemical impedance were done in range of frequency from 0.1 to 10^5 Hz with amplitude 10 mV. A Potentiostat (model AutoLab 87070) is used for performed these measurements. EIS data were analyzed using a (FRA) impedance module which represented by Nyquist plot. The experimental data was analyzed and interpreted based on the equivalent circuits.

The following equation is used to calculated inhibition efficiency (η%) of the casein [9, 26, 29]:

$$(\% = (R\_(ct-\ )\ R\_ct\text{\^{}}()/R\_ct\ \ \times 100 \qquad (3)$$

where R°_{ct} is the values of charge transfer resistances of the untreated electrode and R_{ct} is the values of charge transfer resistances of the treated electrode .

3. RESULTS AND DISCUSSION

Corrosion behavior for bare and altered 316L SS in SBF were studied and analyzed by electrochemical techniques

3.1. Potentiodynamic Polarization (Tafel) Measurements

The Tafel polarization curves for blank and immersed 316L Stainless steel electrodes in various concentrations of casein's solutions at different immersion time in SBF at 37°C shown in figure 1.

In Figure 1, the curves exhibited that the values of E_{corr} obtained in the case of the treated electrodes are slightly moved to positive value than that of blank electrode proposed forming casein's SAMs and indicating its effect on the stainless steel performance. The electrochemical parameters derived from the polarization curves are putted in Table 1. The inhibition efficiency (η %) and surface coverage (θ) of electrodes are putted also in Table 1.

It was observed from table 1that the values of corrosion potential E_{corr} are shifted to positive values in case of covered electrodes suggesting the effect of casein on the corrosion reaction [11, 24, 26, 30]. In this study, the differences between E_{corr} of blank and treated electrodes are less than ± 85 mV. Thus, casein can be served as a mixed type inhibitor [9, 13,24,30]. It is also clear that the corrosion current density (I_{corr}) of untreated electrode higher than that of treated ones and polarization resistance (R_p) is lower. Additionally, with the increase of the concentration of the casein's solutions the values of (I_{corr}) are decreased consequently; the inhibition efficiency is increased. Moreover, as the immersion time of working electrodes in casein's solutions increased the corrosion rate is decreased resulting in maximum inhibition efficiency after 120 minutes immersion. This may be attributed to the formation of dense backed SAMs that require higher concentration and more immersion time to be achieved [8-9, 24, 30-31].

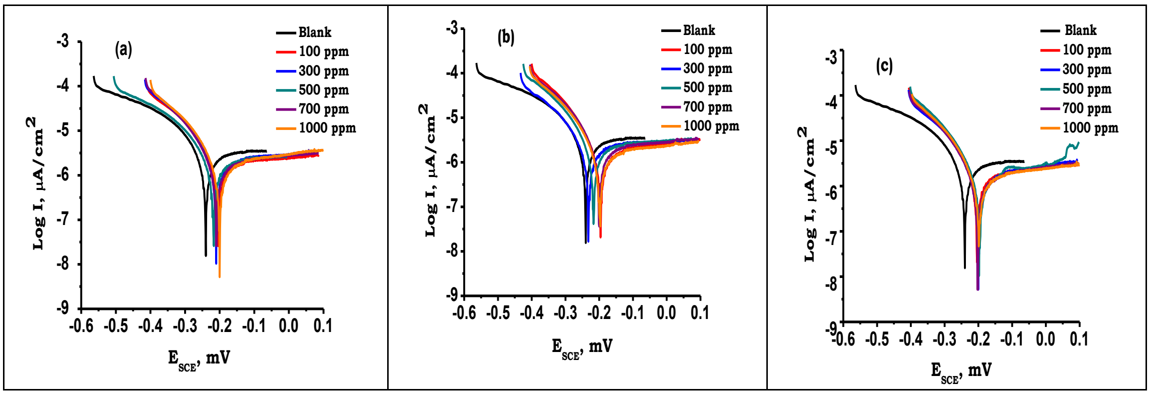

Figure 1: Tafel curves for blank and immersed 316L Stainless steel in various concentrations of casein's solutions for: (a) 60 (b) 90 and (c) 120 minutes, respectively.

Table 1: Electrochemical parameters for blank and immersed 316L Stainless steel specimens in various concentrations of casein's solution at different immersion time.

Time min	Conc. ppm	E_{corr} mV	R_p kΩ/cm^2	I_{corr} μA/cm^2	β_a mV	β_c mV	Corrosion rate (μm/Y)	θ	η %
0	Blank	-242	24.47	3.232	121	-231	37.92	---	---
	100	-206	29.82	2.053	147	-160	24.07	0.3648	36.48
	300	-212	32.92	2.043	156	-148	23.95	0.6379	36.79
60	500	-220	34.55	1.933	167	-176	22.67	0.4019	40.19
	700	-211	34.69	1.849	149	-141	21.68	0.4278	42.78
	1000	-202	36.64	1.805	152	-145	21.16	0.4416	44.16
90	100	-198	29.96	2.012	140	-127	23.59	0.3776	37.76
	300	-233	32.20	1.939	169	-176	22.73	0.4002	40.02
	500	-218	33.95	1.899	140	-180	22.26	0.4127	41.27
	700	-226	38.75	1.833	151	-161	21.49	0.4327	43.27
	1000	-200	41.06	1.799	144	-146	21.09	0.4434	44.34
120	100	-205	36.95	1.842	141	-150	21.60	0.4301	43.01
	300	-201	38.57	1.796	155	-140	21.06	0.4442	44.42
	500	-197	40.91	1.779	138	-128	20.86	0.4588	44.96
	700	-203	41.41	1.749	147	-136	20.50	0.4588	45.88
	1000	-199	44.37	1.640	139	-177	19.23	0.4924	49.24

3.2. Electrochemical impedance spectroscopy (EIS)

Figures 2 show the Nyquist plots for untreated and covered 316L Stainless steel electrodes with various concentrations of casein's solution at different immersion time in SBF at 37 ± 0.2 °C.

The obtained Nyquist plot of the blank electrode shows a depress semicircle indicating the charge transfer process [13, 23-24, 27, 31]. The imperfection of the semicircle assigned to harshness and non identity of the metal surface [31-32]. Additionally, arcs of the treated electrodes with casein have large diameters than that of the blank electrode. When concentrations of casein's solution increase increasing the probability of the interaction of its molecules with the active sites present in the surface of metal driving to more adsorption thus diameter of the semicircles of treated samples is increased [9, 13, 22, 26]. Moreover, by increasing the time of immersion of working electrode in casein's solutions the diameter of the obtained semicircle is increased this might be rationalize to the formation of more complete, regulate and denser SAMs on surface of the metal with increasing immersion time that increasing impedance of the metal surface consequently, reducing the corrosion rate [9, 26, 30, 31, 33, 34]. The equivalent electrical circuits used to analyze the EIS spectra for untreated and treated samples with casein's solution are shown in figure 3 [35-36].

where, Q is the constant phase element (*CPE*), R_s is the solution resistance, which used instead of the double-layer capacitor [9, 22, 37], W is the Warburg element that indicating diffusion process, R_{ct} is the charge transfer resistance, R_f is the resistance of the formed film (SAMs)and C_f is the capacitance of the formed film (SAMs).

The constant phase element (*CPE*) is mathematically expressed as [27, 31, 37]:

$$Z_{CPE} = Y_0^{-1}([Jw])^{-n} \qquad (4)$$

where w is the angular frequency, Y_o is the modulus of the *CPE* , n is the surface parameter and J is the imaginary root .

The fitted impedance parameters are presented in Table 2. The inhibition efficiency (η) was also putted in the same table.

As shown from Table 2, the charge transfer resistances (R_{ct}) in the case of treated samples with casein are higher than that of blank one. Additionally, the charge transfer resistance is increased by increasing the concentration of casein's solutions. Furthermore, with increasing the immersion time of working electrode in casein's solutions, the charge transfer resistances are increased. This could be attributed to the complete formation of regular and dense SAMs of casein on surface of the metal as mentioned before.

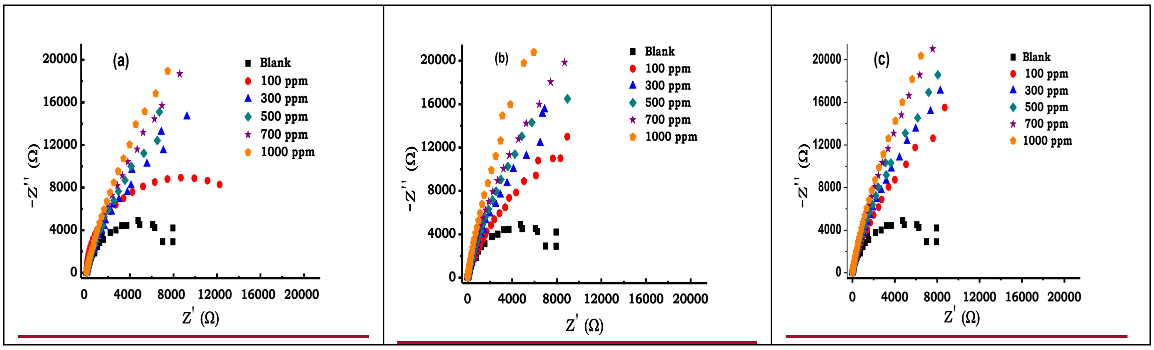

Figure 2: Nyquist plots for blank and immersed 316L SS electrodes in various concentrations of casein's solutions for: (a) 60 (b) 90 and (c) 120 minutes, respectively.

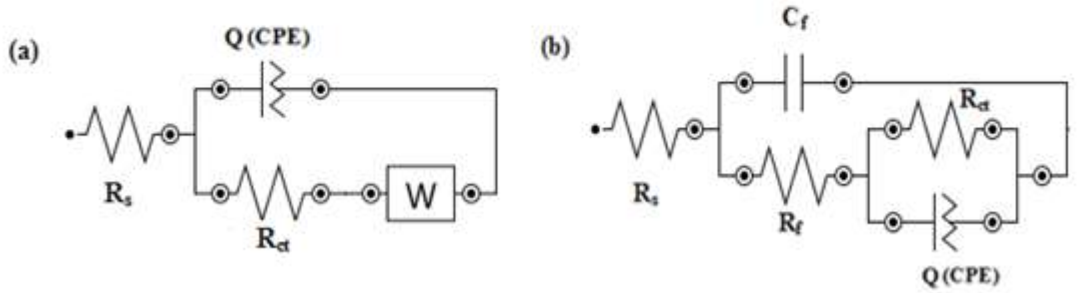

Figure 3: Equivalent circuits used to fit the EIS data of (a) blank and (b) treated stainless steel electrodes with casein's solutions.

Table 2: The fitted impedance parameters for blank and treated 316L SS electrodes

Compd.	Conc. (ppm)	R_s (Ω)	C_f (μF)	R_f (kΩ)	CPE		R_{ct} (kΩ)	W (μMho)	η
					Y_{dl} (μMho)	n			
	Blank	7.54	---	---	82.7	0.886	11.5	92.1	---
	100	11.4	16.6	5.54	82.5	0.793	19.2		40.10
	300	12.9	15.6	7.38	74.2	0.779	20.4		43.62
	500	10.6	13.3	8.93	57.1	0.869	23.2		50.43
	700	11.9	13.1	14.38	56.9	0.824	26.1		55.93
	1000	11.7	11.4	15.8	52.6	0.840	26.5		56.60
	100	9.54	15.3	7.23	85.4	0.832	20.2		43.06
	300	9.39	14.3	9.86	81.3	0.798	21.5		46.51
	500	10.0	13.8	13.7	80.2	0.823	25.3		54.54
	700	11.2	13.6	15.2	78.9	0.792	26.2		56.10
	1000	10.1	12.9	16.2	77.3	0.846	27.3		57.87
	100	8.83	16.0	10.6	79.6	0.875	21.8		47.24
	300	10.5	15.3	12.4	78.0	0.871	22.8		46.51
	500	8.87	12.3	16.6	76.8	0.829	27.0		54.54
	700	12.6	10.1	16.7	76.0	0.867	27.8		56.10
	1000	8.12	9.80	17.2	76.9	0.876	28.2		59.21

4. CONCLUSION

The self-assembled monolayers (SAMs) of casein which formed on stainless steel 316L surface was investigated electrochemically by electrochemical impedance spectroscopy and potentiodynamic polarization technique in simulated body fluid and the following conclusions could be taken from the study:

- The modification of surface of stainless steel with casein solutions improved its surface against the corrosive body fluid.
- Casein serves as a mixed type inhibitor
- As the concentration of the casein's solution is increased, the corrosion rate decreased.
- Additionally, by increasing the immersion time of the stainless steel in casein's solutions the inhibition efficiency of casein is increased .
- The inhibition efficiency of casein is found to be 50% subsequently, further modification is undertaken to increase the inhibition efficiency.

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الملخص العربى

تثبيط معدل تاكل الصلب المقاوم للصدا من نوع 316 L فى سوائل جسم الانسان باستخدام بروتين اللبن الكازين

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1- قسم الكيمياء كليه العلوم (بنات) جامعه الازهر
2- مدينه الابحاث العلمية والتكنولوجيا التطبيقية ببرج العرب

في هذا البحث تم استخدام الكازين وهو بروتين طبيعي موجود في الحليب لتعديل سطح سبائك الفولاذ المقاوم للصدأ من نوع 316L SS المستخدمة في التطبيقات الطبية ضد التآكل في سوائل الجسم المسببة للتآكل. نظرًا لوجود ذرات غير متجانسة في جزيئ الكازين فإن الكازين لديه القدرة على الامتصاص على السطح المعدني لتشكيل طبقة رقيقة واقية تسمى(SAMs). هذه الطبقه هى أداة بسيطة توفر تكوين طبقة رقيقة مرتبة جيدًا على السطح المعدني. تم افتراض أن الكازين يشكل SAMs على سطح سبيكة 316L SS وتم فحص كفاءة تثبيط التآكل كهربائيًا في سوائل الجسم عند 37 درجة مئوية. أكدت النتائج تكوين SAMs وأظهرت أن الكازين يقلل من معدل التآكل ويعمل كمثبط من النوع المختلط. تزداد كفاءة التثبيط بزيادة تركيز الكازين ووقت الغمر في محلول الكازين. أقصى كفاءة تثبيط تم الحصول عليها عند تركيز 1000 جزء في المليون مع وقت غمر يساوي 120 دقيقة. فى الدراسات المستقبليه سوف يتم إجراء مزيد من التعديل على جزيئ الكازين لزيادة كفاءة التثبيط.