

EFFECT OF INFLAMMATORY CONDITIONS AND ALBUMIN PRESENCE ON CORROSION RESISTANCE OF DENTAL AMALGAM IN SALIVA SOLUTION

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المخلص

تم استخدام ملغم الأسنان لأكثر من مائة عام كمادة ترميمية في طب الأسنان بسبب خواصه الميكانيكية المناسبة وتكلفته المنخفضة وسهولة استخدامه. لكن انبعاث المواد السامة مثل الزئبق ومركباته في تجويف الفم نتيجة للتآكل اثار قلقاً كبيراً اتجاه استخدام ملغم الأسنان في السنوات الأخيرة. في هذه الدراسة تم اختبار سلوك التآكل لملغم الاسنان عالي النحاس في محلول اللعاب الاصطناعي البيولوجي. تمت محاكاة تأثير إضافة كلاً من بيروكسيد الهيدروجين، وهو نوع من الأوكسجين التفاعلي وجوده يحاكي حالة التهاب للأسنان، وكذلك وجود الزلال (الألبومين)، وهو بروتين موجود بوفرة في البيئة البيولوجية. تمت دراسة سلوك التآكل عن طريق الاستقطاب الديناميكي الفعال والتحليل الطيفي للمقاومة الكهروكيميائية. تبين من الدراسة أن معدل تآكل الملغم السنني انخفض بمقدار النصف تقريباً في وجود بيروكسيد الهيدروجين والألبومين. أظهرت التجارب أن بيروكسيد الهيدروجين يعمل كمثبط تآكل أنودي، وهذا يتضح من تحول جهد التآكل نحو قيم موجبة وكثافة تيار التآكل نحو قيم أصغر. ويمكن اعتبار الألبومين مثبط تآكل عضوي وذلك لتحول كثافة تيار التآكل نحو قيم منخفضة، مع الحفاظ على جهد التآكل بقيم مماثلة.

ABSTRACT

Dental amalgam has been used for more than a hundred years as a restorative material in dentistry due to its adequate mechanical properties, low cost and ease of application. Unfortunately, the release of toxic materials such as mercury and its compounds in the oral cavity has raised a great concern towards the use of dental amalgam in recent years. In this study the corrosion behavior of high copper dental amalgam was investigated in biological artificial saliva solution. The effects of addition of hydrogen peroxide, a reactive oxygen species, simulating an inflammatory condition, and addition of albumin, a protein found in biological environment were simulated. The corrosion behavior was studied by potentiodynamic polarization and electrochemical impedance spectroscopy. It was found that the corrosion rate of dental amalgam decreased by half in the presence of hydrogen peroxide and albumin. Potentiodynamic polarization experiments showed that hydrogen peroxide served as an anodic inhibitor as indicated by the shift of corrosion potential towards more positive values and the corrosion current density towards lower values. Albumin was found to serve as an organic inhibitor as indicated by the shift of the corrosion current density towards lower values, while maintaining the corrosion potential at similar values.

KEYWORDS: Dental Amalgam; Corrosion; EIS; Polarization; Inhibitor.

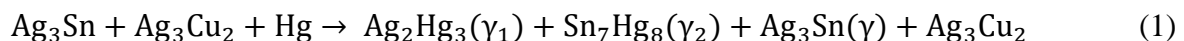
INTRODUCTION

Dental restoration also called filling is the process of repair of a damaged or decayed tooth to restore its shape, appearance and function. Dental restorative materials are metallic and nonmetallic biocompatible materials used in the restoration of damaged teeth. Dental amalgam has served as an excellent and versatile restorative material in dentistry. It has been

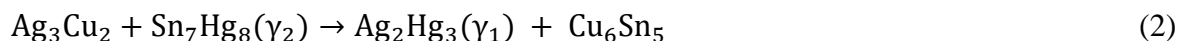
used for more than a century and it accounts for almost 75% of all restorative materials used by dentists [1]. Although the concern regarding the release of toxic mercury compounds, the demand for dental amalgam is still high due to the combination of reliable long term performance, low cost, low technique sensitivity, self-sealing property and longevity [2-4].

In general, dental amalgam alloys contain silver, tin and copper. However, zinc, indium, palladium and platinum are found in some alloys [5]. The various phases found in dental amalgam are the results of presence of the unreacted alloy and intermetallic compounds formed by the reactions of the alloy with mercury upon trituration. The silver-tin phase ($\text{Ag}_3\text{Sn} - \gamma$) of the alloy reacts with mercury to form a silver-mercury intermetallic compound ($\text{Ag}_2\text{Hg}_3 - \gamma_1$) and a tin-mercury intermetallic compound ($\text{Sn}_7\text{Hg} - \gamma_2$), with some alloy remaining unreacted. In high copper amalgams, the presence of silver-copper eutectic (AgCu) reacts with the tin-mercury intermetallic compound to form a copper-tin compound ($\text{Cu}_6\text{Sn}_5 - \eta'$) and more silver-mercury compound [6]. In high copper amalgam, eventually there is little or no tin-mercury (γ_2) in the amalgam [2]. The additional copper helps to increase the formation of γ_1 and prevents the formation of the weak phase γ_2 and thus enhancing the strength of the final product [7].

During trituration, the Ag and Sn in the outer portion of the particles dissolve into the Hg. Simultaneously; Hg diffuses into the alloy particle. In the setting reaction liquid mercury reacts with alloy amalgam in the form of powder particles producing a matrix of intermetallic compounds [8]. The following reactions occur in the high copper alloys which lead to the formation of a complex metallurgical structure consisting of up to 6 different phases:



Followed by:



One of the primary conditions of any metal used in vivo is that it must not produce corrosion products that will be harmful to the body [9]. Corrosion is a chemical or electrochemical process through which a metal is attacked by corrosive environment, resulting in partial or complete dissolution, deterioration, or weakening of any solid substance [10].

An important type of electrolytic corrosion occurs in the oral cavity when combinations of dissimilar metals are present, producing inhomogeneous areas (anodic and cathodic areas) and thus the net result would be an electric couple action known as galvanic corrosion of dissimilar metals. Galvanic corrosion occurs also due to the heterogeneous composition of the metal under study; for example, eutectic and peritectic alloys. If there is difference in the composition of the electrolyte itself another type of electrolyte corrosion occurs under the name of concentration cell corrosion or crevice corrosion. The accumulation of food debris inside the oral cavity produces one type of electrolyte while saliva produces another type. The coexisting of both electrolytes causes corrosion preferably underneath the layer of the food debris.

The corrosion process of dental amalgam is a complex process due to the complex metallurgical structure of the alloy consisting of up to 6 different phases. Each phase will contribute individually in the corrosion process in addition to the intergranular corrosion [11]. Due to their difference in chemical compositions, the different phases of an amalgam alloy have different corrosion potentials. Electrochemical measurements on pure phases

have shown that the Ag_2Hg_3 phase has the highest corrosion resistance, followed by Ag_3Sn , Ag_3Cu_2 , Cu_3Sn , Cu_6Sn_5 , and $\text{Sn}_7 - 8\text{Hg}$ [12].

It has been suggested that the use of amalgams should be discontinued but some researchers believe that amalgam restorations are quite safe. Amalgam restorations will continue to be, until esthetic can match amalgam's longevity, ease of placement, and versatility [13,14]. Many papers have been published on the corrosion of dental amalgam, the recording of polarization curves being one of the most widely used electrochemical methods for this kind of study. More information can be obtained by application of non-stationary techniques, e.g. electrochemical impedance to study the corrosion phenomena and references therein. Electrochemical impedance is a powerful tool in the study of the corrosion behavior of different kinds of metals and alloys [14-16].

The physiological environment into which a biomaterial is placed is a complex system containing different types of inorganic and organic components such as ions, amino acids, proteins and living cells. The composition of this environment varies from one site to another in the human body and can change according to health condition due to some physiological processes such as inflammation or due to the deterioration of the material [17]. The presence or absence of different physiological species in simulated physiological environments has been reported to influence corrosion behaviors of the different biocompatible materials such as Ti, CoCr, and High nitrogen steel. For example the effect of pH, presence of certain inorganic species (e.g. H_2O_2 or fluoride ions), presence of organic species (e.g. lipopolysaccharide (LPS)), and proteins such as albumin have been reported in literature [17,18].

In some medical conditions an associated activates of Reactive Oxygen Species (ROS) have been reported. The activities of these ROS are usually counteracted by the antioxidant system of the human body. Hydrogen peroxide (H_2O_2) is one of the ROS species present in vivo and has been used to simulate inflammatory conditions. H_2O_2 and superoxide are produced by activated phagocytes leukocytes with the subsequent formation of hydroxyle radicals, singlet oxygen and additional H_2O_2 . Albumin is the most abundant protein in the human body, which accounts of for about 4% of total protein content. The concentration of albumin in the blood has been used as a major index in health and disease over the years. Structural and functional impairments in albumin are attributed to various pathophysiological conditions like diabetes, osteoarthritis and advanced liver diseases [17].

The objective of this work is to investigate the effect of albumin and inflammatory conditions on the corrosion resistance of dental amalgam in saliva Fusayama Meyer biological solution. In order to simulate the inflammatory conditions, electrochemical in-vitro studies were conducted using artificial saliva with addition of hydrogen peroxide (a reactive oxygen species, found during inflammation) and albumin, the most typical protein present in biological fluids.

EXPERIMENTAL PROCEDURE

Materials Synthesis and Characterization

Dental amalgam alloy was provided in two-part plastic capsules separated by a membrane that must be broken by squeezing before they were immediately mixed with liquid mercury in an amalgamator for about 10 seconds in order to efficiently wet the surface of the alloy powder particles. The composition of the alloy powder particles is 29 wt. % Cu, 31 wt.% Sn, 40 wt.% Ag. After trituration, the dental amalgam was condensed into square shaped pieces of 1 cm^2 surface area. After setting, a copper wire was soldered to one face of

the square piece. This face including the contact together with the edges of the samples were covered with two-parts epoxy resin and dried in room temperature for at least 24 hours, leaving just one face (1 cm^2) exposed for electrochemical testing. The samples were then mechanically polished with set of SiC papers down to 1200 grit then degreased with acetone and alcohol and finally cleaned with di-water and air dried before conducting the electrochemical experiments. The microstructure of the dental amalgam was studied by etching a polished sample with 2% Nital solution (2 v.% nitric acids and 98 v.% ethanol) using Leica optical microscope.

Electrochemical Tests

All electrochemical measurements were performed using a Gamry Reference 600® potentiostat with the conventional three electrode configuration in naturally aerated and stagnant artificial saliva having a pH of 4.7. In order to simulate inflammatory conditions, the saliva solution was doped with 8 mL/L H_2O_2 (30% concentration). 8 mL/L human albumin (2% concentration) was used also to prepare the electrolyte. The chemical composition of artificial saliva in g/L is: 0.4 NaCl, 0.4 KCl, 0.8 CaCl_2 , 0.79 NaH_2PO_4 , 1 Urea. Electrochemical tests were carried out in a three electrode electrochemical cell configuration. The sample, graphite rod, and a commercial saturated calomel electrode (saturated KCl internal solution) were used as the working, counter, and reference electrode, respectively.

EIS tests were conducted after 60 min of immersion in the electrolyte at E_{oc} in the frequency range of 10 kHz to 10 MHz, 5 points per decade, and $10 \text{ mV}_{\text{rms}}$ sinusoidal potential excitation. Potentiodynamic polarization (PD) measurements were conducted at constant scan rate of 0.167 mV/s , starting from the open circuit potential (E_{oc}) to a cathodic potential (-300 mV to E_{oc}) up to anodic potential ($+300 \text{ mV}$ Vs. E_{oc}). The obtained data was then fitted using Gamry E-chem software. All electrochemical tests were conducted at room temperature and to ensure data reproducibility and accuracy, the results of minimum three samples are reported for each electrochemical test.

RESULTS AND DISCUSSION

Microstructure Characterization

Figure (1) shows optical microscope image of the surface morphology of dental amalgam alloy. The microstructure consists of different phases. The large, irregular particles are $(\text{Ag}_3\text{Sn}-\gamma)$, with dark Cu-Sn areas within them. The round particles are Ag-Cu, with surrounding areas containing Ag- Hg and Cu-Sn phases.

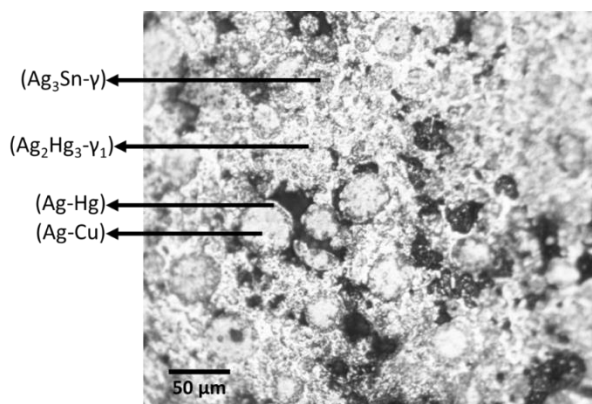


Figure 1: Optical microscope image of high copper dental amalgam etched with 2% nital solution.

This observed microstructure is similar to the findings of Marshal et al. [19] of an eight-year-old clinical amalgam restoration that showed up to 10 distinct phases. The phases that do not appear in the microstructure shown in Figure (1) are those phases that cannot be revealed when etched with the 2% nital and require the use of other etchant [20].

Electrochemical Impedance Spectroscopy (EIS) Study

To evaluate the corrosion mechanisms of dental amalgam, EIS measurements were conducted after E_{OC} stabilization for 1 hour in artificial saliva solution and artificial saliva solution doped with 8 ml/L H_2O_2 and albumin.

The Nyquist diagram for representative results is shown in Figure (2). The diagrams were generally similar in having a single capacitive loop shape. This shape is expected from a polarization resistance due to Faradaic processes coupled with an interfacial charge storage process. These findings are in good agreement to the results reported by Brett et al. [21] for the corrosion of Tytin dental amalgam in artificial saliva with and without lactic acid. They related the enhanced corrosion resistance to the rapid formation of oxide film due to the presence of dissolved oxygen. The presence of lactic acid increased the cell resistance by blocking parts of the amalgam surface.

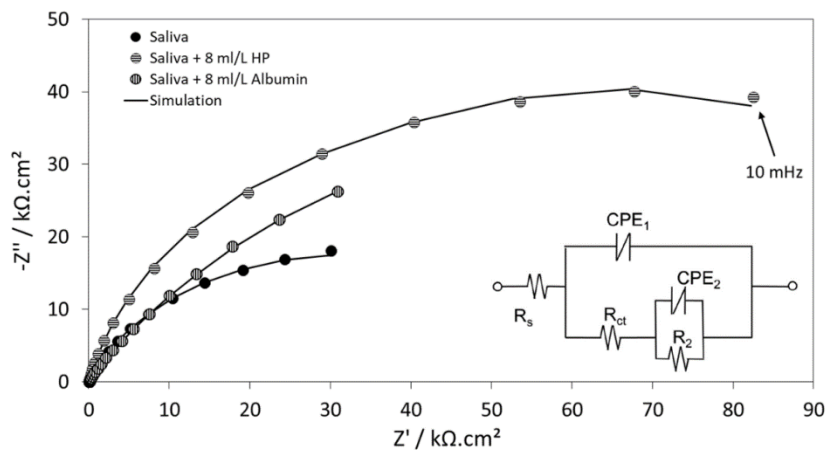


Figure 2: Representative Nyquist plots (scattered data) of dental amalgam conducted after E_{OC} stabilization for 1 hour in artificial saliva solution with and without the addition of HP and albumin. The bottom right inset shows the equivalent circuit model used to fit the experimental data, as represented by the solid lines in the Nyquist plots.

The experimental data were interpreted using the analog equivalent circuit model shown as an inset in Figure (2). A constant phase element (CPE) was used instead of a capacitor due to the expected depression of the Nyquist curve at low frequencies. R_s represents the ohmic solution resistance. In the simplest form, the solution/metal interface includes a polarization resistance (R_p) and a CPE. R_p here is the sum of R_2 and R_{CT} of the equivalent circuit model shown as an inset in Figure (2). The Nyquist results showed that the alloy exhibited similar corrosion mechanism in the three electrolytes, but the significantly larger capacitive loop when artificial saliva was doped with 8 ml/L albumin and H_2O_2 indicated an enhanced corrosion resistance.

The impedance of the non-ideal capacitance CPE is represented as:

$$Z_{CPE} = Y_o^{-1}(j\omega)^{-n} \quad (3)$$

where Y_o is a constant representing a base admittance, $j = (-1)^{1/2}$, ω is the angular frequency, and n is a frequency dependence exponent which measure deviation from ideal

capacitor behavior. The values of n are between 0 and 1. The CPE reduces to a simple capacitor with $C = Y_0$ when $n = 1$. The numeric value of Y_0 is roughly indicative of an effective interfacial capacitance, disregarding dimensional issues. As shown in Figure (2), the fitted results closely matched the experimental behavior for much of the frequency range for all results. The R_p of amalgam alloy is $130 \pm 25 \text{k}\Omega \cdot \text{cm}^2$ when artificial saliva was doped with albumin and $160 \pm 60 \text{k}\Omega \cdot \text{cm}^2$ when artificial saliva was doped with H_2O_2 . These values are significantly larger than that measured in pure artificial saliva ($65 \pm 11 \text{k}\Omega \cdot \text{cm}^2$), indicating that the addition of albumin and H_2O_2 reduced the electrochemical reactions occurring at the metal surface.

The corrosion current density (i_{corr}) from EIS tests was estimated from the R_p using Stern-Geary equation: [22, 23]:-

$$i_{\text{corr}} = B/R_p \quad (4)$$

where B is the apparent Stern-Geary coefficient which can be estimated from [22, 23]:

$$B = \frac{\beta_a \beta_c}{2.3(\beta_a + \beta_c)} \quad (5)$$

where β_a and β_c are the anodic and cathodic Tafel slopes, respectively measured from cyclic potentiodynamic polarization experiments. The validity of corrosion current densities estimated from Stern-Geary equation depends on the correct determination of the coefficient B and whether a metal (M) dissolves directly to M^{2+} or through an M^+ intermediate [23-25]. In this work, the value of B was chosen to be equal to 0.026 V, representing the condition of an active metal dissolution. i_{corr} was converted to corrosion rate (C.R) in micrometer per year as shown in Figure (3) using Faraday's conversion ($1 \mu\text{A} \cdot \text{cm}^{-2} \approx 10 \mu\text{m}/\text{y}$), a typical value measured for many engineering materials assuming uniform corrosion for the different components of the alloy [26].

As shown in Figure (3), the values of nominal corrosion rate estimated from EIS were lower by about half for the dental amalgam immersed in artificial saliva doped with 8 ml/L albumin and H_2O_2 as compared to the value when the dental amalgam was immersed in artificial saliva.

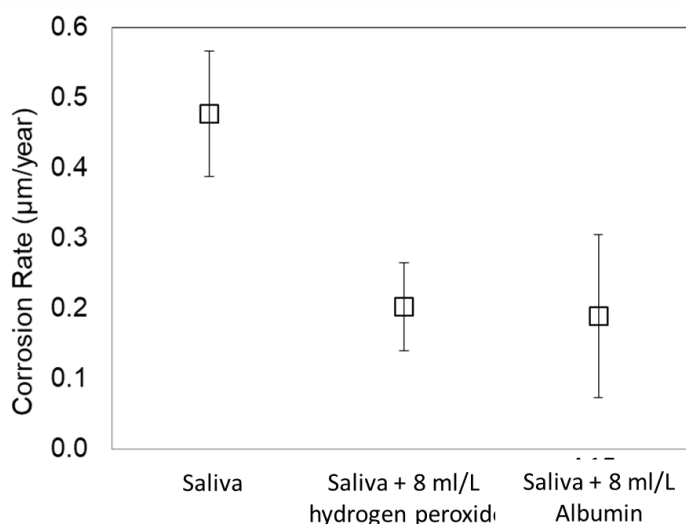


Figure 3: Corrosion rate of dental amalgam in artificial saliva solution and artificial saliva solution doped with albumin and H_2O_2 .

Figure (4) shows the values of parameters (n) and (Y_0) obtained from fitting the experimental data with the equivalent circuit shown as inset in Figure (2). These results show similar trends obtained with multiple replicate specimens.

The values of CPE exponents (n_1 and n_2) of dental amalgam immersed in the three electrolytes were in the range between 0.7 and 1. There is only moderate decrease in these values when saliva was doped with 8 ml/L albumin. The decrease in n_1 and n_2 is an indication of the depression of impedance semicircle at lower frequencies shown in Figure (2).

The values of Y_0 experienced significant increase when the saliva was doped with albumin and H_2O_2 .

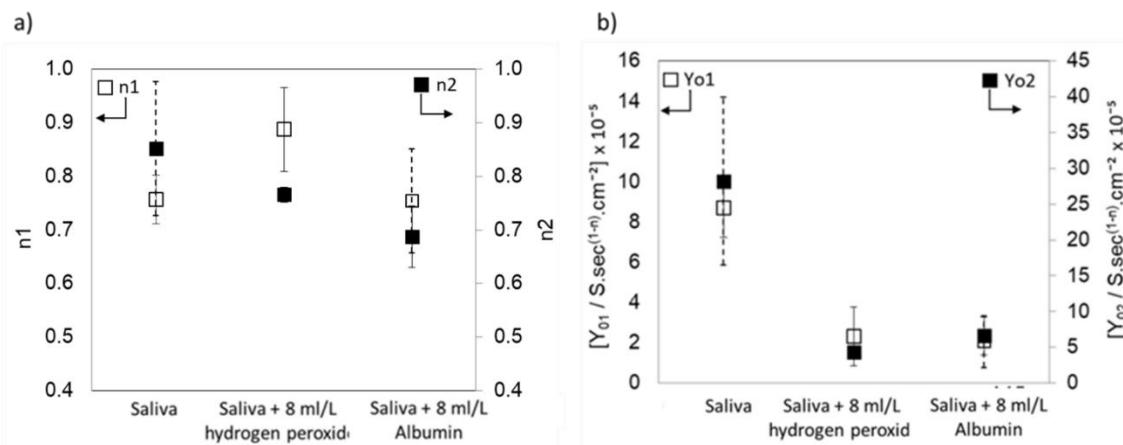


Figure 4: Equivalent circuit parameters (a) a, (b) Y_0 of dental amalgam after exposure for 1 hour.

Potentiodynamic Polarization Experiments

Figure (5) shows the representative potentiodynamic polarization curves of dental amalgam after 1-hour immersion in artificial saliva solution, artificial saliva solution doped with 8 ml/L albumin, and artificial saliva solution doped with H_2O_2 . The anodic branch of the PD curve represents the anodic dissolution of the alloy, while the cathodic branch represents the cathodic reaction of hydrogen evolution.

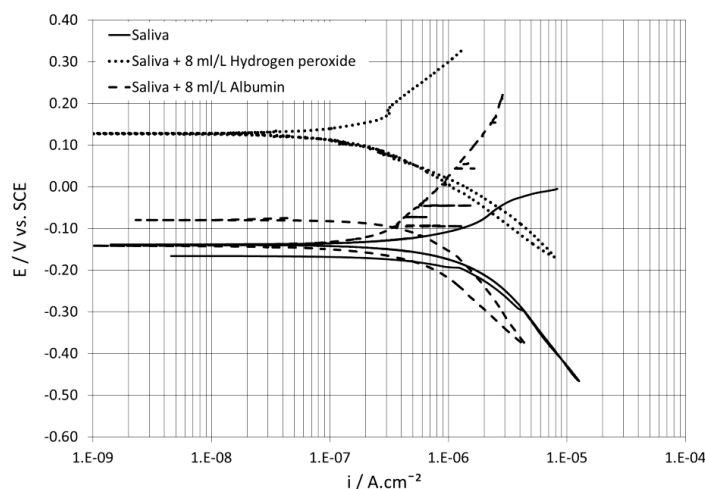


Figure 5: Typical potentiodynamic polarization curves of dental amalgam after 1 hour immersion in artificial saliva solution with and without the addition of HP and albumin

It can be seen from Figure (5) that dental amalgam exhibits a shift of both the anodic and cathodic branches towards lower corrosion current densities (i_{corr}) when artificial saliva solution was doped with albumin and with H_2O_2 , indicating a decrease in anodic and cathodic kinetics [27,28]. It should be noted here that (i_{corr}) values here are plotted in a logarithmic scale. In addition, the PD curves of the dental amalgam in all three solutions show relatively large Tafel slopes, indicating high polarizability of the alloy in these environments.

The values of the corrosion potential of dental amalgam immersed in artificial saliva solution doped with 8 ml/L H_2O_2 scatter around $+0.120 \text{ V}_{\text{SCE}}$. These values are significantly higher than the values when the alloy was immersed in artificial saliva solution ($-0.150 \text{ V}_{\text{SCE}}$), and when the alloy was immersed in artificial saliva solution doped with albumin ($+0.09$ to $-1.6 \text{ V}_{\text{SCE}}$). These findings are similar to those reported in literature for the effect of addition of albumin and H_2O_2 on the corrosion of biomaterials exposed to different physiological environments [17,18,29]. The shift of corrosion potential towards more positive values and the corresponding shift of corrosion current density towards lower values when artificial saliva solution was doped with H_2O_2 indicated that the latter has served as an anodic inhibitor. Anodic inhibitors act by reducing or blocking the anodic reaction and support the natural reaction of passivation of the metal surface as well as forming a film that is adsorbed on the metal. In general, the inhibitors react with the corrosion product resulting in a cohesive and insoluble film on the metal surface [30]. Albumin was found to serve as an organic inhibitor indicated by the shift of the corrosion current density towards lower values, while maintaining the corrosion potential at similar values. Organic inhibitors build up a protective hydrophobic film on the metal surface, providing a barrier to the dissolution of the metal in the electrolyte [30].

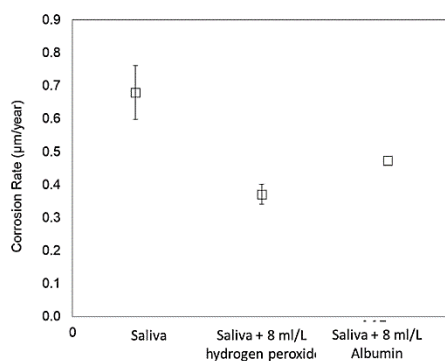


Figure 6: Corrosion rate of dental amalgam in artificial saliva solution and artificial saliva solution doped with albumin and H_2O_2 .

The corrosion rate in $\mu\text{m}/\text{year}$ shown in Figure (6) was estimated by Faradic conversion of the i_{corr} ($1 \mu\text{A}/\text{cm}^2$ corresponds to approximately $22.6 \mu\text{m}/\text{year}$), considering uniform corrosion of the metal (M) with the formation of M^{2+} . It was found that the corrosion rate of dental amalgam ($0.7 \pm 0.08 \mu\text{m}/\text{year}$) in artificial saliva was reduced to ($0.4 \pm 0.06 \mu\text{m}/\text{year}$) and ($0.3 \pm 0.03 \mu\text{m}/\text{year}$) due to the presence of albumin and H_2O_2 , respectively. These values are in good agreement with the results obtained from electrochemical impedance spectroscopy tests shown in Figure (3).

The inhibitor efficiency could be measured by the following equation [30]:

$$E = \frac{R_i - R_0}{R_0} \times 100 \quad (6)$$

where, E is inhibitor efficiency (percentage), R_i is corrosion rate of metal with inhibitor and R_o is corrosion rate of metal without inhibitor. The corrosion rate values obtained from both electrochemical impedance spectroscopy and potentiodynamic polarization experiments showed that the efficiency of albumin was ~ 30% whereas the efficiency of H_2O_2 was ~45%.

CONCLUSIONS

The aim of this work was to investigate the effect of albumin and inflammatory conditions on the corrosion resistance of dental amalgam in artificial saliva solution. In order to simulate the inflammatory conditions in vitro, studies were conducted by addition of hydrogen peroxide (which is a reactive oxygen species found during inflammation) or albumin (which is the most typical protein in biological fluids). The experimental results reveal that the addition of 8 mL / L albumin or H_2O_2 into artificial saliva solution decreased the corrosion rate. It was found that H_2O_2 serves as an anodic inhibitor while albumin serves as an organic inhibitor as indicated by potentiodynamic polarization curves. The corrosion rate values obtained from both electrochemical impedance spectroscopy and potentiodynamic polarization experiments showed that the efficiency of albumin was ~ 30% whereas the efficiency of H_2O_2 was ~45%.

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