

# Magnesium alloys implants coated with 58S sol-gel bioactive glass to retard first stage corrosion

Sheila A. Omar,\* Yolanda Castro,\*\* Josefina Ballarre,\* Wido H. Schreiner,\*\* Alicia Durán,\*\* and Silvia M. Ceré.\*†

\*INTEMA, Universidad Nacional de Mar del Plata-CONICET, Juan B. Justo, 4302, B7608FDQ Mar del Plata, Argentina

\*\*LSI-LANSEN, Departamento de Física, UFPR, Curitiba, Brazil

\*\*\*Instituto de Cerámica y Vidrio, CSIC, Campus de Cantoblanco, Madrid, Spain

## ARTICLE INFO (THIS STYLE IS ARTICLE AND ABSTRACT HEADING)

*Article history: (This style is Article Info subhead)*

Received Day Month Year  
Accepted Day Month Year  
Available Day Month Year

*Keywords: (This style is Article Info subhead)*

A. magnesium alloys  
B. degradation  
C. sol gel glass coatings

†Corresponding author: E-mail: smcere@fi.mdp.edu.ar

## ABSTRACT

Two composition of 58S (60 mol% SiO<sub>2</sub>, 36 mol% CaO, 4 mol% P<sub>2</sub>O<sub>5</sub>) glass coatings were made by sol gel and applied by dip coating onto two commercial magnesium alloys (AZ31 and AZ91) as a strategy to retard the hydrogen evolution and accelerate the deposition of a hydroxyapatite layer to enhance bioactivity. Surface studies with XPS, Raman spectroscopy and SEM techniques are presented after coating deposition and after immersion in Hanks' Balance Salt Solution (HBSS) at 37 C. Electrochemical tests were also conducted to evaluate the evolution of the coating with immersion. It is observed that even the coating present defects, it is able to retard substrate degradation and this effect is more pronounced for AZ91 as substrate. Coating performance is mainly governed by the substrate due the defective nature of the film.

## INTRODUCTION

Surface modification of metallic materials constitutes a strong strategy applied in the biomedical field. It opens a wide world of possibilities oriented to develop the functional properties needed for an implant acceptance when the bare material is not enough to assure good biocompatibility and tissue regeneration. Moreover, chemical

surface modification developed from a potentially bioactive coating can determine the human body response. In this case, the interface between the implant and the physiological environment becomes the key factor which conditions the prosthesis success or failure.<sup>1, 2</sup>

Nowadays, magnesium alloys are in the spotlight as they have the attractive feature of being biodegradable. This fact eliminates the need of an implant removal surgery, meaning a reduction in sanitary costs and patient post-operative suffering. However, these alloys have the drawback of hydrogen evolution during its degradation which could generate tissue inflammation and pain.<sup>3,4,5</sup>

Bioceramics or bioglasses are produced in a variety of forms and phases to fulfill several number of medical strategies for tissue regeneration.<sup>6</sup> Bioactive glasses have the ability to react with physiological media faster than bioceramics, and to bond with living tissues forming an apatite layer. Typical bio-glasses such as Bioglass 45S5<sup>7</sup> are produced by melting. However, Sol-gel method is also a suitable route to produce glass, mainly as coatings in a variety of compositions with the further advantage of high degree of purity and good homogeneity.<sup>8</sup> The precursors of SiO<sub>2</sub> and P<sub>2</sub>O<sub>5</sub> in the sol-gel technique are usually alkoxides, whereas inorganic salts, such as nitrates, are generally used to generate the calcium component.<sup>8</sup> In fact, there are only occasional references about bioglass sol-gel materials prepared using calcium alkoxides or lactates.<sup>9-11</sup> However, the use of calcium nitrate (which is the standard reagent employed) presents some disadvantages such as the need of removal of nitrates from the glass structure at temperatures higher than 550 °C.

The aim of this work was to generate a chemically modified surface of two commercial magnesium alloys with a 58S sol-gel glass coating, using organic precursors such as silicon and phosphate alkoxides along with calcium lactate. The produced bioactive layer is sintered at low temperatures in order to maintain the substrate integrity. This 58S composition belongs to the SiO<sub>2</sub>-P<sub>2</sub>O<sub>5</sub>-CaO system and has proven bonding to soft and hard tissue,<sup>12</sup> becoming itself a good strategy to retard the hydrogen evolution and accelerate the deposition of a hydroxyapatite layer.

## MATERIALS AND METHODS

### Magnesium alloys

Two commercial magnesium alloys known as AZ31B (Al 3.0, Zn 1.0, Mn 0.2 in wt. %) and AZ91D (Al 9.0, Zn 1.0, Fe 0.005, Mn 0.33, Ni 0.002, in wt. %) were selected as substrates for this work. Plane sheets of 0.33 cm width were cut into rectangles of 3 cm x 1.5 cm exposing an area of 8 cm<sup>2</sup>. They were all polished with 1200 and 2500 grit SiC paper, rinsed with ethanol and dried with hot air prior to coating. This last procedure assures to have a standardized topography for all samples assayed.

### Sol-gel glass coating

The 58S glass (60 mol% SiO<sub>2</sub>, 36 mol% CaO, 4 mol% P<sub>2</sub>O<sub>5</sub>) was prepared by sol-gel method.

Tetramethyl orthosilicate (TMOS, 98% Sigma Aldrich), triethyl phosphite (TEPI, 98% Sigma Aldrich) and calcium L-lactate hydrate ([CH<sub>3</sub>CH(OH)COO]<sub>2</sub>Ca•xH<sub>2</sub>O) were used as precursors and sources of silicon, phosphorus and calcium, respectively. HNO<sub>3</sub> 1N was used as catalyzer. Ethylene glycol (EG: HOCH<sub>2</sub>CH<sub>2</sub>OH, 99.8% Sigma Aldrich) was added in a constant molar ratio EG/alkoxides=0.65 to improve the alkoxides condensation degree and increase the specific surface area<sup>13,14</sup> of the coating as porosity agent. Methanol (CH<sub>3</sub>OH, 99.8% Sigma Aldrich) was the solvent used to dissolve the calcium lactate. With a water/alkoxides ratio fixed at 3, the sol labeled as E1 was synthesized.

Keeping the same 58S glass composition but with a different precursors set, another sol was created and labeled as E2. The

difference with E1 was that methyltriethoxysilane (MTES, 98% Sigma Aldrich) was combined with TMOS as silicon source, as a way to improve the coating ductility and therefore its ability to copy the substrate roughness.<sup>15</sup> In this case a molar ratio TMOS/MTES=40/60 was selected based on previous studies<sup>16</sup> and the molar ratios water/alkoxides was increased to 4.9.

Coatings were performed by dip-coating with a withdrawal rate of 15 cm/min for E1 and 25 cm/min for E2 in a glove box with argon atmosphere and relative humidity lower than 8 ppm. A thermal treatment at 300 °C for 3 h was carried out to consolidate the coated systems, using a furnace inside the glove box.

For a better understanding and from now on, the samples are labeled first with the corresponding substrate and then the 58S sol-gel glass respective composition. Thus, the samples under study are: AZ31-E1, AZ31-E2, AZ91-E1 and AZ91-E2. Also, both bare alloys (AZ31 and AZ91) are used as controls in the experiments.

## Solution

The solution selected for experiments was the Hanks' Balanced Salts Solution (HBSS) to simulate the plasma composition in the implant surrounding environment<sup>17</sup>. HBSS was homemade prepared following the chemical composition summarized in Table 1. Hanks' solution was selected among the many possibilities of simulated body fluids, since it is the only one that seems not to influence the microstructure of the metallic surface, meanwhile others cause acceleration of micro-galvanic effects in the second phases.<sup>18</sup> Before every characterization where Hanks' solution was used as electrolyte, it was deaerated for 15 minutes using high purity N<sub>2</sub>.<sup>19</sup> The reason of this is that the oxygen concentration in plasma is lower than in HBSS without deaeration.<sup>20</sup> While the mimic O<sub>2</sub> concentration in plasma is 5.3x10<sup>-5</sup> mol/L, we measured 4.5x10<sup>-5</sup> mol/L after our protocol. All assays in this research were carried out at normal body temperature, 37 °C.

## Electrochemical tests

Electrochemical assays were performed in a GAMRY Ref 600 electrochemical unit (Gamry, USA) with a conventional three electrode cell, using a saturated calomel electrode (SCE, Radiometer Copenhagen) as reference electrode, a platinum wire of convenient area as a counter electrode and the coated and bare magnesium alloys as working electrodes.

Electrochemical impedance spectroscopy (EIS) tests were registered also at the E<sub>corr</sub> with amplitude of 0.01V rms sweeping frequencies from 20000 to 0.005 Hz. Polarization curves for each sample were recorded from -1.8 V to -1.0 V (vs. reference) and 1 mV/s of scan rate. In all these electrochemical assays, HBSS was the electrolyte used and each measurement was performed four times to assure the repetitiveness of the results.

## Superficial sample characterization

The thickness of 58S E1 and E2 coatings was measured with a profilometer KLA Tencor (Alpha-Step D-100, US). A minimum of three lineal measurements for each sample were performed to determine the step height between the coated and the uncoated part of each sample. Two specimens per condition were used to the statistical analysis. The tape test (instructed by ASTM D3359-B) was done to characterize coating adhesion to the substrates.

Final 58S sol-gel glass coating structures were analyzed through Fourier Transformed Infrared Spectroscopy (Nicolet 6700 FTIR Spectrometer, Thermo Electron Corporation) by Attenuated Total Reflectance (ATR). The FTIR spectra were collected for E1 and E2 coatings after thermal treatment.

With the aim of studying the evolution of the coated system with time and the possible apatite deposition the samples were analyzed *in*

*vitro* by immersion in HBSS at 37 °C, during 72 hours (72 h) and 17 days (17 d) of exposure to the electrolyte. In all cases, samples without immersion were used as control. Micro-Raman assays were performed before and after immersion together with their blanks using an Invia Reflex Confocal (Renishaw RM 2000, UK) with a 785 nm wavelength laser, at 5% intensity, with 2 accumulations and 10 second of exposition per spectrum. X-ray photoelectron spectroscopy (XPS) was selected to perform an elemental analysis of surfaces prior and after immersion of 72 h and 17 d in HBSS. Measurements were made using a commercial VG ESCA 3000 system. The base pressure in the experimental chamber was in the low 10<sup>-9</sup> mbar range. The spectra were collected using Mg K $\alpha$  (1253.6 eV) radiation and the overall energy resolution was about 0.8 eV. Survey spectra were recorded for the samples in the 0–1100 eV kinetic energy range by 1 eV steps. High resolution scans with 0.1 eV steps were conducted over Mg 2p, Ca 2p, P 2p, Si 2p and O 1s. In the entire cases surface charging effects were compensated by referencing the binding energy (BE) to the C 1s line of residual carbon set at 284.5 eV.

Coatings visual integrity and deposits produced during immersion tests were examined by scanning electronic microscopy (SEM, JEOL JSM-6460LV, Japan) after 72 h and 17 d of immersion in HBSS. In all cases, after the coating was consolidated and without any immersion the corresponding sample were observed by SEM (0 h).

## RESULTS AND DISCUSSION

Coating thickness was measured by profilometry, with errors lower to 3%. Results are summarized in Table 2. Coatings of composition E1 resulted thicker than E2 ones. Both E1 and E2 compositions deposited on AZ91 resulted in thinner than those deposited on AZ31, probably associated with the different viscosity of the sols and/or different wettability of E1 and E2 sols on AZ91 and AZ31. Regarding to the coatings adhesion measured by ASTM D3359-B, all the conditions showed excellent adhesion to the respective substrates.

Figure 1 shows FTIR spectra for both commercial alloys coated with the two compositions under study (58S E1 and E2), after heat treatment. The main difference between the two compositions is the presence of a sharp peak located at 1272 cm<sup>-1</sup> which is associated with the methyl (-CH<sub>3</sub>) group<sup>15</sup> in the E2 coated samples, evidencing the non-hydrolysable methyl group presence originally from the MTES used as precursor during the synthesis of E2 sol. The remaining spectra serve to give an explanation of the resulted structure of the coatings in further detail. It can be distinguished: a small peak at 450 cm<sup>-1</sup> associated with the rocking Si-O-Si vibration, a wider small band between 850~750 cm<sup>-1</sup> which can be attributed to the symmetric and asymmetric bending of Si-O-Si band. Additionally, the wide band between 900~1200 cm<sup>-1</sup> is related to the glass coating structure, being a consequence of the overlapping of several important peaks. The band around 940~960 cm<sup>-1</sup> can be associated with the P-OH stretching and Si-O-Ca vibration mode as well as Si-OH vibration modes. The band at ~1040 cm<sup>-1</sup> is associated to the asymmetric stretching vibration of Si-O-Si together with the P-O stretching band that appears at about 1056 cm<sup>-1</sup>.<sup>21</sup> Another band at 1190 cm<sup>-1</sup> assigned to Si-O-Si and/or P-O-P stretching is also included in the broad band. <sup>8,12,22-25</sup> The band located at 1300~1500 cm<sup>-1</sup> includes the -CH bending of lactate group and the vibration modes of non-bridging PO<sub>2</sub> in PO<sub>4</sub><sup>3-</sup> groups (at 1448 cm<sup>-1</sup>), and the band at around 1500~1700 cm<sup>-1</sup> corresponds to the C=O stretching due to the lactate group presence.<sup>26,27</sup> The thermal treatment used for these coatings (at 300°C) was not enough to degrade these groups (expected around 400°C), so they remain in the structure. This last feature is not considered a problem as it is not harmful to human body. Finally, the insert in Figure 1 shows a wide band located between 2500~3600 cm<sup>-1</sup>, generally associated with O-H stretching of absorbed water and Si-OH and P-OH groups.<sup>24,28,29</sup>

The composition and presence of the different compounds in deposits or degradation process products can be analyzed by Micro-Raman spectroscopy. The analysis was focalized on the different deposits and singular points present in both systems. The samples were studied without immersion and after 72 h and 17 d of immersion in HBSS. Figure 2a shows Raman spectra of AZ31-E1 and AZ31-E2 and Figure 2b shows the same for AZ91-E1 and AZ91-E2. Also spectra of bare AZ31 and AZ91 substrates after 72 h of immersion are shown for comparison in the respective figure. The presence of magnesium hydroxide and Mg-Al hydroxalicates (Mg and Al hydroxy-carbonates) can be noticed by the peak around  $440\text{ cm}^{-1}$ .<sup>30,31</sup> In AZ31 bare alloy after 72 h immersion in HBSS, a peak at  $956\text{ cm}^{-1}$  related to symmetric stretching of phosphate compounds<sup>24</sup> is present. Coated AZ31-E1 and AZ31-E2 samples, presented the same type of deposited compounds but at different immersion times. Moreover, both coatings applied onto AZ31 alloy presented a wide band centered at  $440\text{ cm}^{-1}$  related with magnesium hydroxides<sup>32</sup> and other around  $694\text{ cm}^{-1}$  related with  $\text{CH}_3\text{-O-Si-OH}$  vibrations.<sup>33</sup> These denote the sample degradation from early times which involves the dissolution of the coating in contact with the HBSS. Also at  $594\text{ cm}^{-1}$  a band associated with P-O symmetric stretching is present<sup>34</sup> related with the phosphorous oxide present in the bioactive glass coating. After 72 h immersion in HBSS, AZ31-E1 system still presents the characteristic peaks of the silica glass and substrate, but a band at  $961\text{ cm}^{-1}$  related with phosphates in hydroxyapatite compounds can be seen.<sup>35</sup> In AZ31-E2 composition after 72h immersion, there is a clear evidence of apatite related compounds deposited on the surface, noticed by the presence of  $963\text{ cm}^{-1}$  band.<sup>36</sup> At 17 d of immersion, phosphate related bands at 435, 592 and  $961\text{ cm}^{-1}$  are present in both coated samples (either with E1 or E2).<sup>34,37</sup> Also a band at  $1078\text{ cm}^{-1}$  is related with carbonates present in hydroxyl-carbonate apatite.<sup>35</sup>

Similar behavior is noticed in AZ91-E1 and AZ91-E2 in Figure 2b. For E1 composition after 72 h and 17 d of immersion, the apatite-like deposits are mixed with remains of the bioactive glass coating, while for E2 the deposits are just based on phosphates, denoting the *in vitro* bioactive behavior of the E2 coating. Table 3 summarizes the assignments of Raman vibration modes associated with the coated samples.

In order to complete the information about surface composition changes after immersion in HBSS, X-ray photoelectron spectroscopy (XPS) assays were performed. The analyzed area with XPS technique depends on instrument design, having a spot around  $0.3\text{ mm}^2$  in this case. Nevertheless, the obtained information is related with surface since the maximum depth reached is around 2 nm. Figures 3 and 4 show the detailed spectra for O 1s, Si 2p, Mg 2p, Ca 2p and P 2p for all systems under study, without immersion and after 72 h of immersion in HBSS. Table 4 shows the peak value assignments and the correspondence with the different binding energies and compounds.

The presence of magnesium compounds can be noticed with a peak around 49.5 eV corresponding to  $\text{Mg(OH)}_2$ .<sup>38</sup> Figure 3a evidences that Mg 2p signal is detected with and without immersion for E1 composition for both alloys (mainly for AZ31), while Figure 4a shows that for E2 is only measured after the 72 h in HBSS. This fact emphasizes the higher integrity for samples coated with E2, which correlates with the EIS results (see below). Moreover, the signal is stronger when AZ31 is the substrate denoting the less protective feature of E2 on AZ31 as corroborated with the electrochemical results that follows.

The glassy coating can be studied by the Si 2p signal peak in Figures 3b and 4b where the binding energy at 102.9 eV is related with typical Si-O-Si bonds in silica gels for all the conditions without immersion.<sup>39</sup> For all coated samples, it can be seen that after 72 h in contact with the electrolyte this signal almost vanishes and shifts to 101.5eV, evidencing the degradation of the coating and the Si-O<sup>-</sup> bonds present in first steps of apatite formation reactions.<sup>40</sup> In Figures 3c, 3d,

4c and 4d, the presence of a shift in P 2p and Ca 2p peaks and the increase of intensity denotes that after 72 h of immersion in HBSS all bioactive coatings form apatite related compounds<sup>31,32,41</sup>, as it was previously shown by the Raman spectroscopy results. Also the shift in O 1s peaks, shown in Figures 3e and 4e, to lower values of binding energies is related with oxygen in phosphates and reactive Si-O<sup>-</sup> species.<sup>30,41</sup>

Figure 5 shows the SEM images for coated samples before immersion and after 72 h and 17 d of immersion in HBSS. All the conditions without immersion appear to be homogeneous despite some flaws that are present on the surface. It is important to say that the presence of discontinuities on the surface at the initial stage is not considered as a failed coating. The purpose of the coating is to provide a bioactive surface to accelerate bone healing together with a retard (without inhibition) of the electrochemical reactions and thus a perfect coating is not a desired objective. The biodegradable feature of magnesium alloys is preferred to be kept and the generation of a perfect barrier coating can retard excessively the metal degradation. After 72 h in HBSS at 37 °C all coatings show delamination and also some deposits. After 17 d in contact with the electrolyte all coatings appear to be degraded and partially covered with deposits on the surface. It can also be observed that the deposits mainly nucleate over the sol-gel glass coating than on the  $\text{MgO/Mg(OH)}_2$  layer of the bare alloy, denoting the bioactive behavior of the thin sol-gel glass ceramic layer.

Figure 6 shows the EIS spectrum for the two coated alloys and the comparison with the bare material after 24 h and 17 d of immersion in HBSS. It can be observed two time constants at medium and high frequency and an inductive response at low frequency for both bared alloys and also for the coated AZ31 at all immersion times. The inductive behavior disappears for both coated compositions on AZ91 samples (Figure 6c and d). The nature of the inductive behavior is largely discussed in literature and it has been reported to correspond to charge effect, adsorbed species that may affect the electrochemical reactions and /or pitting of the alloy.<sup>42,43</sup> It can be observed that in all the systems a resistive component is depicted in the low frequency range and hence it can be assumed that the coating resistance is revealed, allowing the electrolyte to reach the substrate underneath the coating. Both AZ91-E1 and AZ91-E2 present higher total impedance than the bare alloy and in turn, the total impedance is even higher for E2 (Figure 6c). After 17 d of immersion there is a slight decrease of the impedance modulus and an increase in the angle for AZ91-E2 at high frequencies probably related to deposits onto the surface. The AZ91-E1 system shows a slight decrease in the total impedance modulus and a less capacitive behavior indicating the proliferation of defects in the coating. The EIS results for both coated AZ31 systems showed more defective coatings than the ones onto AZ91. AZ31-E2 showed the highest impedance modulus after 24 h of immersion and this parameter diminishes after 17 d of immersion (Figure 6a). A similar behavior is observed for AZ31-E1, both reaching values near the bare alloy as an indicative that the coating degrades in time. The decrease in phase angle at higher frequencies for coated AZ31 (Figure 6b) compared with coated AZ91 (Figure 6d) is directly related with the formation and extent of electrolyte pathways into the coating.

Kramer – Kronig relationship was verified for the EIS results. It was demonstrated that Kramer-Kronig transformation into real and imaginary components present low noise ratio (< 1% for the frequency domain under study), as observed for other authors for Mg in chloride containing electrolytes.<sup>44</sup>

To perform a deeper analysis of events taking place at the interface and also to correlate them with physical events, electrochemical impedance spectra were fitted using the electrical circuits shown in Figure 7a and b. In all cases where an inductive loop was present, it was necessary to use an inductance followed by a resistance in the equivalent circuits selected. In the circuits  $R_{\text{sol}}$

represents the electrolyte resistance;  $R_1$ ,  $R_2$  and  $R_3$  the resistance of the deposits, oxides/hydroxides onto the surface and through the film defects as charge resistance, respectively. An ideal capacitor ( $C_2$ ) was used in the capacitance associated to double layer at the bottom of the defects. Constant phase elements (CPE) were also used to represent non ideal capacitors associated with the presence of deposits and/or oxo-hydroxides on the surface and the deviation of the slope from -1 in the impedance modulus vs. frequency plot in the Bode plot.  $Z_{CPE}$  is given by:

$$Z_{CPE} = \frac{1}{Q(j\omega)^\alpha} \quad (1)$$

Where  $Q$  is a parameter independent of frequency and  $\alpha$  is a coefficient associated with system homogeneity. The inductor  $L$  is equivalent to a short circuit in steady state and is related to adsorbed species and reaction intermediates as stated above.<sup>45,46</sup>

Table 5 and 6 show the results of data fitting according the equivalent circuits presented in Figure 7. Polarization resistance ( $R_p$ ) values were calculated in an attempt to estimate the corrosion resistance of the systems under study by means of equations (2) and (3) and using the values obtained from the data fitting presented in Table 5 and 6 from the circuits a and b respectively.

$$\frac{1}{R_p} = \frac{1}{R_{sol} + R_1 + R_2} + \frac{1}{R_3} \quad (2)$$

$$R_p = R_{sol} + R_1 + R_2 \quad (3)$$

Assuming that, according to Stern Geary relationship given by equation 4, corrosion current density ( $i_{corr}$ ) is inversely proportional to  $R_p$  and that corrosion rate ( $V_{corr}$ ) is directly proportional to  $i_{corr}$  (equation 5), it can be said that from the analysis of  $R_p$ , a ranking of corrosion rates can be assented. In equation 5,  $V_{corr}$  is the instant corrosion rate that is an average over the surface sample,  $M_{eq}$  is the equivalent weight (12.15 eq/mol),  $n$  the number of electron interchanged,  $\delta$  is the magnesium density (1.74 g/ mL) and  $F$  the Faraday constant. There is a range of reported Tafel slopes in literature and a very complete analysis was made by Bland et al<sup>47</sup> in order to assess a reliable value of  $B$  in equation 4. In this work we prefer to base the analysis of facility to corrode of the different compositions used in the study on the  $R_p$  analysis and hence to establish a status among the samples and coating compositions. Since generalized corrosion is not the type of degradation observed in these samples and there is no decade of linearity or evidence of activation control, Tafel behavior cannot be assured. For this reason, the corrosion rate calculated by equation 5 would provide only an approximation which would be as useful as the  $R_p$  analysis by itself.

$$i_{corr} = \frac{\beta_a \beta_c}{2.3(\beta_a + \beta_c) R_p} = \frac{B}{R_p} \quad (4)$$

$$V_{corr} = \frac{M_{eq}}{n\delta F} * i_{corr} \quad (5)$$

It can be observed in the values shown in Table 5 that the films deposited onto AZ31 present lower values of  $R_1$  and  $R_2$  in time, giving account of wider pathways for the electrolyte to reach the base metal as the immersion proceeds. This is also observed for the AZ91 alloy with the exception of E2 coating where an increment in  $R_2$  is observed, probably related to deposits onto the surface, as seen in XPS and Raman results. It can be assumed that the corrosion rates expected should be higher for the AZ31 coated samples than for AZ91 coated ones. According to the  $R_p$  values calculated from equation 2 and 3 and

presented in Tables 5 and 6 it can be assumed that the corrosion rates expected should be higher for the AZ31 coated samples than for the coatings over AZ91. Equally, there is one magnitude order of difference between the  $R_p$  values of these two bare substrates. It is observed that, since the porous nature of the coating, the substrate commands the corrosion behavior of the system.

Figures 8a and b show the polarization curves of the samples when a raising potential is applied and density current is measured. It can be noticed that  $E_{corr}$  results in more active values for AZ91 systems, and that there is also a significant shift in the  $E_{corr}$  between conditions compared with the AZ31 system. This fact can be attributed to a more heterogeneous microstructure in AZ91 substrate, due to its higher aluminum (Al) content. The Al presence in magnesium alloys conditions significantly the formation of the  $Mg_{17}Al_{12}$  intermetallic ( $\beta$  phase) which acts as a cathode while the matrix ( $\alpha$  phase) remains as anode. When Al content is high, more intermetallic is present and local micro-galvanic cells takes place that might be the reason for more active  $E_{corr}$  values. Even though the  $E_{corr}$  values for AZ91 resulted more negative, it is not enough to consider that the overall alloy corrosion behavior is inferior. Figure 8a shows the polarization curves for the AZ91 alloy. It can be observed that this alloy presents a passive region that remains stable after immersion. In addition, E1 and E2 coatings show stable coatings with no sign of major degradation after immersion with lower current densities for the E2 composition. Polarization curves presented in Figure 8b show that bare AZ31 presents a zone of passivity that disappears after 17d of immersion. AZ31-E1 shows a defective coating denoting signs of degradation ever higher than the bare alloy after 17 d of immersion. AZ31-E2 shows a broad pseudo passive region after 24 h of immersion but the coating shows severe signs of deterioration after prolonged immersion reaching values comparable to the bare substrate as AZ31-E1. In general, corrosion rates for coated AZ31 after 17 d of immersion are very similar with the bare metal, showing no improvements to the corrosion behavior with the uncoated alloy. This correlates with the EIS results, which show minimal changes in the overall corrosive response when the AZ31 metal is coated.

Porosity (in this case consider as permeation degree since the coating is porous and also cracked) can be estimated according to Creus<sup>48</sup> by:

$$P = \frac{R_{ps}}{R_p} * 10^{-\left|\frac{\Delta E_{corr}}{\beta_a}\right|} * 100 \quad (6)$$

Where  $R_p$  is the polarization resistance of the coating,  $R_{ps}$  is the polarization resistance of the bare steel,  $\beta_a$  is the Tafel slope of the bare steel and  $\Delta E_{corr}$  is the difference in corrosion potential between the coated and bare substrate. Tafel slope was determined from the data of Figure 8 and a value of 0.162 V dec<sup>-1</sup> for AZ31 and 0.160 V dec<sup>-1</sup> for AZ91 was obtained.  $R_{ps}$  and  $R_p$  data can be obtained from Table 5 and 6. Table 7 shows the result obtained for the coatings after 24 h of immersion in HBSS. It can be observed that E1 coatings show higher initial porosity than the E2 on both substrates and in turn, coating applied onto AZ31 are more porous than the ones onto AZ91, in good agreement with the results observed in EIS and polarization curves.

Once again it can be noticed from the polarization curves that the substrate is controlling the electrochemical response. For both alloys, the E1 sol-gel glass coating has poor or null protective capacity, being also harmful when AZ31 is the substrate selected. This fact is not observed when the 58S E2 coating is applied, but it clearly lasts longer when is applied over AZ91 than over AZ31.

The different behavior between both coatings is likely due to the more organic composition of E2 respecting to E1, having a further

hydrophobic effect. The coating, although thinner, will be more ductile, showing lower tendency to cracking. It is worth noting that even the coatings applied onto AZ31 seems to be no effective in the long term, it accomplishes the goal of enhancing the corrosion resistance at the beginning of the immersion reducing the area where the reactions take place and then reducing the overall corrosion at first stages of implantation.

## CONCLUSIONS

- ❖ Nitrate-free sol-gel glass coatings of bioactive 58S composition were prepared and applied onto AZ31 and AZ91 magnesium alloys. The coating constitutes a versatile strategy to generate a chemically modified surface that can inhibit the degradation behavior of a magnesium alloy.
- ❖ The films shown bioactivity *in vitro* as demonstrated by Raman and XPS techniques. Both E1 and E2 compositions have evidence of formation of apatite-like compounds on the surface. The nucleation of the compounds is preferred over the coating than on the bare substrate.
- ❖ The substrates are the controlling part of the electrochemical response of the magnesium coated alloy. This fact was corroborated by EIS and polarization curves and related with the porous characteristic of the coatings developed.
- ❖ The E2 composition has a slow degradation rate at the beginning of immersion in a simulated body fluid, such as the Hanks' Buffered Salt Solution. After prolonged immersion AZ91-E2 present higher corrosion resistance than AZ31-E2, and the former remains with lower corrosion densities than the bare alloy.
- ❖ Due to the uncertainties in Tafel slopes determination,  $R_p$  values were considered as a preferred parameter to establish a ranking of corrosion resistance of the different conditions presented in this study.
- ❖ Coatings applied onto AZ31 and AZ91 effectively reduce the corrosion reactions, mainly at the beginning of the immersion, reducing the anodic and cathodic reactions and promoting the formation of phosphate related compounds on the surface.

## ACKNOWLEDGMENTS

The support from the Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET, PIP 752) and from the Universidad Nacional de Mar del Plata (Project 15G/449) is acknowledged. The authors also acknowledge Dr. Mariela Desimone for the Raman measurements and Mr. Sebastian Rodriguez, INTEMA, Argentina for his help in sample preparation.

Sheila Omar specially acknowledges Ursa Tiringier, Aritz Iglesias Ayestaran, Desirée Ruiz García, Eva Pereitado and Miguel Gómez for the valuable support and help during the stage at GlaSS group of CSIC in Madrid.

## REFERENCES

1. Nouri A, Wen C. Introduction to surface coating and modification for metallic biomaterials. In: *Surface Coating and Modification of Metallic Biomaterials*. ; 2015:3-60. doi:10.1016/B978-1-78242-303-4.00001-6.
2. Romonti DC, Iskra J, Bele M, Demetrescu I, Milosev I. Elaboration and characterization of fluorohydroxyapatite and fluoroapatite sol - gel coatings on CoCrMo alloy. *J.of Alloys and Compounds*. 2016;665:355-364. doi:10.1016/j.jallcom.2016.01.072.
3. Song G-L, Atrens A. Corrosion Mechanisms of Magnesium Alloys. *Adv Eng Mater*. 1999;1(1):11-33. doi:10.1002/(SICI)1527-2648(199909)1.
4. Atrens A, Cao F, Shi Z, Dargusch MS. Corrosion of Mg for biomedical applications. In: *Surface Coating and Modification of Metallic Biomaterials*. Elsevier Ltd; 2015:81-102. doi:10.1016/B978-1-78242-303-4.00003-X.
5. Hermawan H, Mantovani D. Degradable metallic biomaterials : the concept, current developments and future directions. 2009;21(4):207-216.
6. Hench LL, Wilson J. *An Introduction to Bioceramics (Advanced Series in Bioceramics)*. Vol. I. World Scientific Pub Co Inc.; 1993.
7. Li R, Clark AE, Hench LL. An investigation of bioactive glass powders by sol-gel processing. *J Appl Biomater*. 1991;2:231-239. doi:10.1002/jab.770020403.
8. Arcos D, Greenspan DC, Vallet-Regí M. A new quantitative method to evaluate the *in vitro* bioactivity of melt and sol-gel-derived silicate glasses. *J Biomed Mater Res A*. 2003;65:344-351. doi:10.1002/jbm.a.10503.
9. Rámila A, Balas F, Vallet-Regí M. Synthesis routes for bioactive sol-gel glasses: Alkoxides versus nitrates. *Chem Mater*. 2002;14(2):542-548. doi:10.1021/cm0110876.
10. Bosetti M, Cannas M. The effect of bioactive glasses on bone marrow stromal cells differentiation. *Biomaterials*. 2005;26:3873-3879. doi:10.1016/j.biomaterials.2004.09.059.
11. Rezabeigi E, Wood-Adams PM, Drew RAL. Synthesis of 45S5 Bioglass via a straightforward organic, nitrate-free sol-gel process. *Mater Sci Eng C*. 2014;40:248-252. doi:10.1016/j.msec.2014.03.042.
12. Sepulveda P, Jones JR, Hench LL. *In vitro* dissolution of melt derived 45S5 and sol-gel derived 58 S bioactive glasses. *Biomed Mater Res*. 2002;61:301-311.
13. Ștefănescu O, Stoia M, Ștefănescu M, Vlase T. Study on the influence of teos-diol molar ratio on their chemical interaction during the gelation process. *J Therm Anal Calorim*. 2009;97:251-256. doi:10.1007/s10973-009-0247-5.
14. Ștefănescu M, Stoia M, Ștefănescu O. Thermal and FT-IR study of the hybrid ethylene-glycol-silica matrix. *J Sol-Gel Sci Technol*. 2007;41:71-78. doi:10.1007/s10971-006-0118-5.
15. Innocenzi P, Abdirashid MO, Guglielmi M. Structure and Properties of Sol-Gel Coatings from Methyltriethoxysilane and Tetraethoxysilane. *J Sol-Gel Sci Technol*. 1994;3:47-55. doi:10.1007/BF00490148.
16. Durán A, Castro Y, Aparicio M, Conde A, de Damborenea JJ. Protection and surface modification of metals with sol-gel coatings. *Int Mater Rev*. 2007;52(2):175-192. doi:10.1179/174328007X160263.
17. Hanks JH, Wallace RE. Relation of oxygen and temperature in the preservation of tissues by refrigeration. *Proc Soc Exp Biol Med Soc Exp Biol Med*. 1949;71(2):196-200. doi:10.3181/00379727-71-17131.
18. Zainal Abidin NI, Atrens AD, Martin D, Atrens A. Corrosion of high purity Mg, Mg2Zn0.2Mn, ZE41 and AZ91 in Hank's solution at 37°C. *Corros Sci*. 2011;53(11):3542-3556. doi:10.1016/j.corsci.2011.06.030.
19. Sussmann HJ. An Assessment of ASTM F 2129 Electrochemical Testing of Small Medical Implants - Lessons Learned. *Nace Int*. 2007;(244):1-10.
20. Zheng Y. My notes on Oxygen concentration in plasma and tissue. i:2-4.

21. Luz GM, Mano JF. Preparation and characterization of bioactive glass nanoparticles prepared by sol-gel for biomedical applications. *Nanotechnology*. 2011;22:494014. doi:10.1088/0957-4484/22/49/494014.
22. Ma J, Chen CZ, Wang DG, Meng XG, Shi JZ. Influence of the sintering temperature on the structural feature and bioactivity of sol-gel derived SiO<sub>2</sub>-CaO-P<sub>2</sub>O<sub>5</sub> bioglass. *Ceram Int*. 2010;36:1911-1916. doi:10.1016/j.ceramint.2010.03.017.
23. Pereira MM, Clark AE, Hench LL. Calcium Phosphate Formation on Sol Gel Derived Bioactive Glasses in Vitro. *J Biomed Mater Res*. 1994;28:693-698.
24. Aguiar H, Serra J, González P, León B. Structural study of sol-gel silicate glasses by IR and Raman spectroscopies. *J Non Cryst Solids*. 2009;355:475-480. doi:10.1016/j.jnoncrysol.2009.01.010.
25. Huang K, Cai S, Xu G, et al. Sol-gel derived mesoporous 58S bioactive glass coatings on AZ31 magnesium alloy and in vitro degradation behavior. *Surf Coatings Technol*. 2014;240:137-144. doi:10.1016/j.surfcoat.2013.12.026.
26. Cheong SH. Physicochemical properties of calcium lactate prepared by single-phase aragonite precipitated calcium carbonate. *Res J Pharm Biol Chem Sci*. 2016;7:1786-1794.
27. Lee Y-K, Kim S-D. Preparation and Characteristics of Calcium Lactate from Black Snail. *Nutraceuticals Food*. 2003;8:166-172.
28. Innocenzi P. Infrared spectroscopy of sol-gel derived silica-based films: A spectra-microstructure overview. *J Non Cryst Solids*. 2003;316:309-319. doi:10.1016/S0022-3093(02)01637-X.
29. Petrovskaya TS, Borilo LP. Production of SiO<sub>2</sub>-P<sub>2</sub>O<sub>5</sub>-CaO-(Na<sub>2</sub>O) structured films. *Glas Ceram*. 2012;69:25-29. doi:10.1007/s10717-012-9408-2.
30. Kaabi Falahieh Asl S, Nemeth S, Tan MJ. Mechanism of calcium phosphate deposition in a hydrothermal coating process. *Surf Coatings Technol*. 2015;270:197-205. doi:10.1016/j.surfcoat.2015.03.003.
31. Klopogge JT, Hickey L, Frost RL. FT-Raman and FT-IR spectroscopic study of synthetic Mg/Zn/Al-hydroxalrites. *J Raman Spectrosc*. 2004;35:967-974. doi:10.1002/jrs.1244.
32. De Oliveira EF, Hase Y. Infrared study and isotopic effect of magnesium hydroxide. *Vib Spectrosc*. 2001;25:53-56. doi:10.1016/S0924-2031(00)00107-7.
33. Colomban P. Raman Studies of Inorganic Gels and their Sol-to-Gel, Gel-to-Glass and Glass-to-Ceramics Transformation. *J Raman Spectrosc*. 1996;27:747-758. doi:10.1002/(SICI)1097-4555(199610)27:10<747::AID-JRS38>3.0.CO;2-E.
34. Agathopoulos S, Tulyaganov DU, Ventura JMG, Kannan S, Karakassides MA, Ferreira JMF. Formation of hydroxyapatite onto glasses of the CaO-MgO-SiO<sub>2</sub> system with B<sub>2</sub>O<sub>3</sub>, Na<sub>2</sub>O, CaF<sub>2</sub> and P<sub>2</sub>O<sub>5</sub> additives. *Biomaterials*. 2006;27:1832-1840. doi:10.1016/j.biomaterials.2005.10.033.
35. Penel G, Leroy G, Rey C, Bres E. MicroRaman spectral study of the PO<sub>4</sub> and CO<sub>3</sub> vibrational modes in synthetic and biological apatites. *Calcif Tissue Int*. 1998;63:475-481. doi:10.1007/s002239900561.
36. Notingher I, Boccaccini AR, Jones J, Maquet V, Hench LL. Application of Raman microspectroscopy to the characterisation of bioactive materials. *Mater Charact*. 2002;49(3):255-260. doi:10.1016/S1044-5803(03)00029-9.
37. Rehman I, Hench LL, Bonfield W, Smith R. Analysis of surface layers on bioactive glasses. *Biomaterials*. 1994;15:865-870. doi:10.1016/0142-9612(94)90044-2.
38. Tie D, Feyerabend F, Hort N, Willumeit R, Hoeche D. XPS studies of magnesium surfaces after exposure to Dulbecco's modified eagle medium, Hank's buffered salt solution, and simulated body fluid. *Adv Eng Mater*. 2010;12(12):699-704. doi:10.1002/adem.201080070.
39. Vernè E, Ferraris S, Cassinelli C, Boccaccini AR. Surface functionalization of Bioglass® with alkaline phosphatase. *Surf Coatings Technol*. 2015;264:132-139. doi:10.1016/j.surfcoat.2015.01.001.
40. Cerruti M, Bianchi CL, Bonino F, Damin A, Perardi A, Morterra C. Surface modifications of bioglass immersed in TRIS-buffered solution. A multitechnical spectroscopic study. *J Phys Chem B*. 2005;109(30):14496-14505. doi:10.1021/jp050705t.
41. Santos JD, Jha LJ, Monteiro FJ. In vitro calcium phosphate formation on SiO<sub>2</sub>-Na<sub>2</sub>O-CaO-P<sub>2</sub>O<sub>5</sub> glass reinforced hydroxyapatite composite: a study by XPS analysis. *J Mater Sci Mater Med*. 1996;7:181-185. doi:10.1007/BF00121258.
42. Walter R, Kannan MB. Influence of surface roughness on the corrosion behaviour of magnesium alloy. *Mater Des*. 2011;32:2350-2354. doi:10.1016/j.matdes.2010.12.016.
43. King AD, Birbilis N, Scully JR. Accurate electrochemical measurement of magnesium corrosion rates; A combined impedance, mass-loss and hydrogen collection study. *Electrochim Acta*. 2014;121:394-406. doi:10.1016/j.electacta.2013.12.124.
44. Bland LG, Gusieva K, Scully JR. Effect of Crystallographic Orientation on the Corrosion of Magnesium: Comparison of Film Forming and Bare Crystal Facets using Electrochemical Impedance and Raman Spectroscopy. *Electrochim Acta*. 2017;227:136-151. doi:10.1016/j.electacta.2016.12.107.
45. Bland LG, Troconis BCR, Santucci RJ, Fitz-Gerald JM, Scully JR. Metallurgical and electrochemical characterization of the corrosion of a Mg-Al-Zn alloy AZ31B-H24 tungsten inert gas weld: Galvanic corrosion between weld zones. *Corrosion*. 2016;72:1226-1242. doi:10.5006/2078.
46. Orazem ME, Tribollet B. *Electrochemical Impedance Spectroscopy*. New Jersey: Wiley & Sons; 2008.
47. Bland LG, King AD, Birbilis N, Scully JR. Assessing the corrosion of commercially pure magnesium and commercial AZ31B by electrochemical impedance, mass-loss, hydrogen collection, and inductively coupled plasma optical emission spectrometry solution analysis. *Corrosion*. 2015;71:128-145. doi:10.5006/1419.
48. Creus J, Mazille H, Idrissi H. Porosity evaluation of protective coatings onto steel, through electrochemical techniques. *Surf Coatings Technol*. 2000;130(2-3):224-232. doi:10.1016/S0257-8972(99)00659-3.

## FIGURE CAPTIONS

FIGURE 1. FTIR spectra of 58S sol-gel glass coated treated at 300 °C for 3 h for both alloys.

FIGURE 2. Raman spectra of 58S coated samples over AZ31 (a) and AZ91 (b), without immersion and after 72 h and 17 d in HBSS.

FIGURE 3. XPS high resolution spectra for both materials (AZ31 and AZ91) bare and coated with E1 composition, after 0 and 72 h immersion in HBSS: (a) Mg 2p, (b) Si 2p, (c) P 2p, (d) Ca 2p, (e) O 1s.

FIGURE 4. XPS high resolution spectra for both materials (AZ31 and AZ91) bare and coated with E2 composition, after 0 and 72 h immersion in HBSS: (a) Mg 2p, (b) Si 2p, (c) P 2p, (d) Ca 2p, (e) O 1s.

FIGURE 5. SEM images for the 58S coated samples after 72 h and 17 d in HBSS. Samples without immersion are also shown for comparative purposes. (a) AZ31-E1, (b) AZ31-E2, (c) AZ91-E1, (d) AZ91-E2.

FIGURE 6. Bode (impedance modulus and phase angle vs frequency) plots for coated samples on both alloys and the comparison with the corresponding bare alloy after immersion in HBSS at 37 °C: AZ31 (a, b) and AZ91 (c, d). Solid lines show the fitting of the data by modeling with the equivalent circuits detailed in Fig. 6.

FIGURE 7. Equivalent circuits used for modeling for coated samples over: (a) AZ31 and (b) AZ91 alloy.

FIGURE 8. Polarization curves measured in HBSS at 37 °C for coated samples onto: (a) AZ91 and (b) AZ31 substrate.

## Tables

TABLE 1. Chemical composition of the Hanks' Balanced Salts Solution (HBSS) [mmol/L].

TABLE 2. Coatings thickness measured on AZ31 and AZ91 alloys [ $\mu\text{m}$ ].

TABLE 3. Assignments of Raman vibration modes associated with 58S sol-gel made glass coating.

TABLE 4. XPS binding energy assignment for both coated and bare alloys after 72 h immersion in HBSS.

TABLE 5. Fitting results calculated from statistical analysis of electrochemical impedance spectroscopy for AZ31 coated alloy after 72 h and 17 d in HBSS. The standard deviation is lower than 20% in all cases.

TABLE 6. Fitting results calculated from statistical analysis of electrochemical impedance spectroscopy for AZ91 coated alloy after 72 h and 17 d in HBSS. The standard deviation is lower than 22% in all cases.

TABLE 7. Calculated porosity percentage (by using eq. 6) of the coatings applied onto AZ31 and AZ91 after 24 h immersion in HBSS.