Exploring the biodegradation of pure Zn under simulated inflammatory condition

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ABSTRACT

The study concerning the effects of inflammation on the corrosion behaviour of biodegradable Zn was carried out for the first time. Simulated inflammatory solutions containing H2O2 or Fenton’s reagent were prepared for electrochemical measurements and immersion tests. It turned out that H2O2 can significantly influence the mechanism underpinning Zn degradation, the degree of which varied with H2O2 concentration, and acted differently in PBS and Hank’s solution. Meanwhile, Fenton reactions also exerted a noticeable impact on Zn corrosion and the corrosion product formation. A proposal is thus put forward that degradation as biological consequences should be considered in evaluating biodegradable Zn.

1. Introduction

Metallic biomaterials such as cobalt-based alloys, titanium-based alloys, and the stainless steels, with a superior combination of high mechanical strength and fracture toughness, are commonly used as orthopaedic implants, cardiovascular interventional devices, and tissue engineering scaffolds. Despite of excellent wear and corrosion resistance, permanent implants fabricated from traditional inert metals may corrode and release wear debris in the human body during use [1]. In the physiological environment, immune reactions might take place in response to foreign materials and are very likely to cause inflammation. Inflammatory cells, particularly leukocytes and macrophages, can generate and release highly oxidative chemicals known as reactive oxygen species (ROS), such as hydrogen peroxide (H2O2), hypochlorous acid (HOCl), superoxide (O2−) and nitric oxide during the inflammatory response [2–4]. It’s reported that inflammatory cells could use ROS secretion and acid to attack foreign bodies [5], which not only caused the tissue destruction but also created a locally corrosive environment around implants [2]. The oxidative effect of ROS was found to cause the polymer chain scission and contribute to their degradation ultimately [3].

For traditional metallic biomaterials, inflammatory response as a result of adverse reactions to metal debris raised serious concerns and numerous studies were carried out to evaluate the corrosive effect of inflammation. It turned out that inflammation related reactions indeed affected the metal corrosion [6–8]. On the surface of human implanted and retrieved CoCrMo implants, the evidence of apparent inflammatory cell-induced corrosion (ICIC) was reported [5]. The cells migrated, attached, and then sealed down on the surface, and released chemical species under cell membranes between cell and metal, or perhaps into the solution, to provide the driving force for the metal corrosion. Regarding the stainless steel and titanium-based alloys, the direct corrosion of activated inflammatory cells such as osteoclasts on the surface can be identified, as well [9–13].

The major driver of the attack on metals caused by inflammation was claimed to be ROS and their byproducts [5]. H2O2, a major ROS produced by activated inflammatory cells and also known as a strong oxidant [14], can introduce a highly oxidizing condition into the solution, and alter the corrosion susceptibility of CoCrMo alloy [15], Ti6Al4V alloy [16] and the stainless steel [11–13]. Moreover, for ROS like O2− and H2O2, in the presence of transition-metal ions such as iron, the highly toxic radicals like hydroxyl radicals (OH•) can be produced. The involved reactions are so-called Fenton reactions and the Fe ion/H2O2 mixtures are known as Fenton’s reagent [17]. The study of retrieved CoCrMo implants confirmed the occurrence of Fenton-like reactions in the cell ruffled-border regions and a streak of corrosion...
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Inflammation-induced corrosion processes, accelerating the corrosion of biodegradable metals owing to its biodegradable characterise, offer a frame for the new tissue formation before completely replaced by natural tissues emerge as the viable alternative [19-21]. A great number of studies have been performed to thoroughly investigate the corrosion behaviour of biodegradable metals owing to its biodegradable characteristics. However, till now, the interactions between implants and inflammatory cells put much emphasis on the one-way viewpoint that the degradation of implant might lead to adverse biological reactions, and less attention was paid to the effect of inflammatory condition might have on the metal corrosion. The inflammation induce corrosion on biodegradable metals is poorly understood and how inflammatory cell-released chemical species affect the degradation behaviour of metals has not been studied yet, which is a whole new field of research.

Recently, biodegradable Zn alloys, with promising biocompatibility, mechanical strength, and more matched degradation pace to that of the tissue healing have received tremendous interest. Actually, Zn $^{2+}$ can induce the cellular H$_2$O$_2$ production and act as an oxidant stressor by triggering the mitochondrial production of H$_2$O$_2$, and cause multiple oxidative effects through H$_2$O$_2$-dependent and independent mechanism [22]. Moreover, during 12-month implantation for pure Zn stent in abdominal aorta of rabbit, Yang et al. [23] proposed that the decreased thickness of corrosion layer after 3 months might be associated with inflammation as the stent were surrounded by macrophages. The inflammatory reactions might lower the local pH and thus cause the dissolution of corrosion product [24]. After 6 months, the inflammation significantly alleviated, which lead to the formation of ZnO and triggered the precipitation of calcium phosphate phase accordingly. The evolution of degradation mechanism of pure Zn in vivo seemed to be strongly connected with inflammation and the inflammation-induced corrosion played a crucial role in Zn degradation. The research deficiency in this field needs to be filled up as it can help precisely understand the biodegradation process of Zn in vivo, better simulate the natural biochemistry in vitro, and might provide profound insights on the design and optimization of materials in the future.

In general, the immune cells can generate extracellular H$_2$O$_2$ at concentration in μM range [25-28], and the concentration of H$_2$O$_2$ in the local micro-environments can reach mM [29]. The exact concentration of ROS in vivo remains unclear and is likely to vary with the severity of the inflammatory response [6]. Therefore, there is a great variation in replicating the ROS exposures in vitro corrosion tests. The range of concentrations adopted herein is 0.1 mM ~ 1.0 mM, most of which can be potentially achievable at least in local cellular regions [6]. Phosphate buffer saline (PBS) and Hank’s solution, added with a varying concentration of H$_2$O$_2$, as well as Fenton’s reagent (H$_2$O$_2$ and iron ions), are separately prepared. The 14-day immersion test, corrosion product characterization and electrochemical approaches are performed on pure Zn. The degradation behaviour of Zn in simulated inflammatory conditions is thoroughly investigated and how H$_2$O$_2$ and Fenton reactions affect the Zn degradation is illustrated.

2. Experimental

2.1. Sample preparation

The 99.99 wt.% pure zinc rod was extruded from ingots at 250 °C, with a reduction ratio of 16, and cut into 1.0 mm thick discs with 10.0 mm diameter. All specimens were ground with the SiC abrasive paper up to 2000 grits, followed by ultrasonically cleaning in the acetone for 5 min and another 5 min in the ethanol. After that, samples were dried in air under the sterile condition.

2.2.Immersion tests

The simulated inflammatory solutions comprising of phosphate buffer saline (PBS) and hydrogen peroxide (H$_2$O$_2$) with the concentration about 0.1 mM, 1.0 mM, and 30.0 mM were prepared. The immersion tests were conducted at 37 °C for 1, 3, 7, and 14 days, with a ratio of solution volume to surface area at 20 mL/cm$^2$. The initial medium pH was adjusted to 7.40 with NaOH and HCl, and the pH value was recorded at specific time points during the 14-day immersion.

Hank’s solution was selected as another base medium and prepared by dissolving electrolyte powder in distilled water with stirring at 37 °C. The simulated inflammatory solutions were created by adding 0.1 mM, 1.0 mM, 10.0 mM, 0.1 % and 1.0 % H$_2$O$_2$ into Hank’s solution, respectively, to cover physiologically possible levels reported in the previous studies [15]. With respect to Fenton reactions, 10.0 mM H$_2$O$_2$ was added to Hank’s solution first and then 0.1 mM FeCl$_3$ were added. The initial pH of electrolyte was adjusted to 7.40 with NaOH and HCl. The immersion tests were carried out at 37 °C for 1, 3, 7, and 14 days, with the ratio of solution volume to surface area at 20 mL/cm$^2$. More importantly, the designated electrolyte was completely renewed every 3 days. The medium pH was monitored and recorded at different time intervals.

The immersion tests were performed in triplicates for each time point to verify the reproducibility, and after immersion, all specimens were cleaned with distilled water and dried at the room temperature. The corrosion product on the surface was removed by using solution containing 200 g/L CrO$_3$. The samples were then taken out, washed by distilled water and ethanol, and dried in air. The weight prior to the immersion tests and after the removal of corrosion product were determined, respectively. The mass loss (W) was obtained and the corrosion rate (C) was derived according to ASTM-G31-12a:

$$C = \frac{(K \times W)}{(A \times T \times D)}$$  \hspace{1cm} (1)

C is in units of mm/yr, the coefficient $K = 8.76 \times 10^4$, A is the exposed surface area (mm$^2$), T is the immersion time (h), and D is the density of material (g).

2.3. Corrosion product characterization

The environmental electron microscopy (ESEM, Thermal Fisher Quattro S, USA) coupled with energy-disperse spectrometer (EDS) was employed to characterize the surface morphology of pure Zn after immersing and identify the chemical composition of corrosion product on the surface. The X-ray diffractometer (XRD, X-Pert3 Powder, Netherlands) was conducted to analyse the constituents of corrosion product, with 2θ in the range of 10° to 90° at a speed of 4°/min. To investigate the chemical groups in the corrosion product, Fourier transform infrared spectroscopy (FTIR, Nicolet iS50, USA) was conducted and a series of scans were performed from 400 to 4000 cm$^{-1}$.

2.4. Electrochemical experiments

Electrochemical experiments were performed with an electrochemical workstation (Autolab, Metrohm, Switzerland) at the room temperature. The three-electrode cell set-up was utilized, with pure Zn as the working electrode, the saturated calomel electrode (SCE) as the reference electrode and a platinum electrode as the counter electrode. The open circuit potential (OCP) measurements were carried out over a time period of 3600 s. Electrochemical Impedance Spectroscopy
measurements (EIS) were performed at the open circuit potential with AC amplitude of 10 mV in a frequency range from 10^5 Hz to 10^-2 Hz. The potentiodynamic polarization (PDP) curves were measured with a scan rate of 1 mV/s. All the measurements were repeated at least three times.

3. Results

3.1. Immersion test

3.1.1. Solution pH

As shown in Fig. 1, the presence of 0.1 mM and 1.0 mM H2O2 slightly increased the pH of PBS in 7 days and the pH value in three solutions spawned a continuous increase. Afterthat, the increase in solution pH accelerated and H2O2 moderately raised the value. Lower the concentration, less the effect of H2O2 on pH variations. At the same time, the addition of 30.0 mM H2O2 remarkably altered the pattern pH values changed with time in PBS and significantly raised the pH. The pH value in PBS went up sharply in 3 days and reached 10.84. Afterthat, the solution pH marginally diminished and sustained at the value about 10.5 after 14 days.

The change in solution pH in Hank’s solutions as a function of time is shown in Fig. 2. Each electrolyte was completely renewed every 3 days, so the solution pH was adjusted to 7.40 repeatedly. Generally, the pH of Hank’s solution manifested a growth trend slowly with time, and moved towards higher value in presence of 0.1 mM and 1.0 mM H2O2. With H2O2 > 1.0 mM added, the pH of Hank’s solution increased initially and then dropped significantly as the immersion time extended. The decrease in solution pH occurred repeatedly after electrolyte renewal. With the introduction of 0.1 % H2O2, the pH of Hank’s solution exhibited the lowest value. At the same time, as depicted in Fig. 2b, Fenton’s reagent slightly decreased the pH value of Hank’s solution. The solution pH displayed higher value in general than that with 10.0 mM H2O2 added.

3.1.2. Weight loss and corrosion rate

As depicted in Fig. 3, the weight loss of pure Zn in PBS with or without H2O2 added continually increased, and the Zn corrosion slowed down with time. The evolution of Zn degradation in PBS and PBS with 0.1 mM or 1.0 mM H2O2 added was comparable and the effect of H2O2 on the corrosion rate seemed inconspicuous. However, after adding 30.0 mM H2O2 into PBS, the degradation of pure Zn got noticeably accelerated throughout the test, especially on Day 1, and the corrosion rate decreased with extended time, as shown in Fig. 3b.

In Hank’s solution, the addition of H2O2 with the concentration identical to or lower than 0.1 % retarded the degradation of pure Zn during 14 days of immersion, and the corrosion rate decreased as immersion prolonged. The inhibition effect of H2O2 on Zn degradation became more significant as its concentration increased. On the contrary, the degradation of Zn was substantially encouraged in Hank’s solution by 1.0 % H2O2. The corrosion rate decreased in 3 days and then went up after 7 days.

In the meantime, as shown in Fig. 4b and d, in Hank’s solution added with 10.0 mM H2O2 and FeCl3, the weight loss and corrosion rate of pure Zn exhibited the lowest values throughout the whole immersion. It turned out that 10.0 mM H2O2 inhibited the Zn degradation, and the mixture of H2O2 and FeCl3 further reduced the corrosion rate.

3.2. Surface morphology

The SEM images and optical images in Fig. 5 showed that after 14-day immersion in PBS with or without H2O2, clusters of white corrosion products were accumulated on the surface of pure Zn. In PBS, the sample surface was almost completely covered by the thick layer of corrosion product, and the surface without coverage was dispersed with white precipitations, as shown in the picture inside the red box. The EDS spectrum performed on selected corroded areas to characterize corrosion products indicated that the white precipitations and substrate after 14-day immersion were mainly composed of Zn, C, O, P and Ca. For pure Zn immersed in PBS with 0.1 mM or 1.0 mM H2O2 added, the corrosion morphology was slightly affected by H2O2 that the area of product layer decreased and less corrosion product were formed on the substrate. Zn, C, O and P were identified on the surface, as listed in Table 1. More importantly, after adding 30.0 mM H2O2 into PBS, the area of surface covered by white corrosion product layer decreased. The holes suggesting the occurrence of the severe corrosion were identified under white precipitation clusters. EDS results indicated that the corrosion product and surface both comprised of Zn, C, O, P and Cl. The corrosion product formed with 30.0 mM present contained high amount of Cl element.

Fig. 6 depicts the SEM surface morphology images of pure Zn during 14 days of immersion in simulated inflammatory solutions based on Hank’s solutions. For pure zinc in Hank’s solution only, white corrosion product was formed after 1 day and continually accumulated on the sample surface. As shown in Table 2, the layer and precipitations primarily consisted of Zn, O, C, Ca, P and Mg. After adding 0.1 mM or 1.0 mM H2O2 into Hank’s solution, the surface morphology of pure Zn barely changed for the whole immersion, and the EDS revealed the similar element composition of corrosion product to that in Hank’s solution. In the presence of 10.0 mM H2O2, the formation of corrosion product on pure Zn was inhibited in Hank’s solution, and EDS analysis identified only Zn, O, C and P on the surface. Regarding pure Zn exposed to Hank’s solution containing 0.1 % H2O2, the precipitation of corrosion product was significantly depressed and the corroded surface was recognized after 14 days, with small holes present. The corrosion product and the surface were both mainly composed of Zn, O, C and P. Conversely, with the addition of 1.0 % H2O2, white precipitations were surprisingly formed on the surface again after 14 days, and the corrosion product consisted of Zn, O, C, P and Ca.

The change of surface morphology of pure Zn during 14-day immersion in Hank’s solution with 10.0 mM H2O2 and FeCl3 is shown in Fig. 7. It turned out that almost no corrosion product was dispersed on the surface throughout the test, and small pits can be distinguished after 14 days. The formation of precipitations in Hank’s solution can be slightly depressed by 10.0 mM H2O2, and greatly restrained by Fenton’s reagent.
3.3. Corrosion product characterization

As depicted in Fig. 8, XRD were utilized to investigate the chemical composition of corrosion products on pure Zn after 14 days of immersion in PBS and Hank’s solution. It turned out that ZnO, Zn\textsubscript{8}(OH)\textsubscript{8}Cl\textsubscript{2}⋅H\textsubscript{2}O and Zn\textsubscript{3}(PO\textsubscript{4})\textsubscript{2}⋅2H\textsubscript{2}O were formed during the immersion in PBS with or without H\textsubscript{2}O\textsubscript{2}. And the peak of Zn\textsubscript{8}(OH)\textsubscript{8}Cl\textsubscript{2}⋅H\textsubscript{2}O exhibited the highest intensity on pure Zn immersed in PBS with 30.0 mM H\textsubscript{2}O\textsubscript{2} added.

With respect to pure Zn exposed to Hank’s solution with or without H\textsubscript{2}O\textsubscript{2}, as shown in Fig. 8 b and c, XRD spectra identified ZnO as the main crystallized products. To further confirm the surface chemical composition of precipitations, the FTIR spectra of pure Zn immersed in Hank’s solution with or without H\textsubscript{2}O\textsubscript{2} are analysed, as illustrated in Fig. 9. The FTIR spectra of Zn in all Hank’s solutions exhibited similar patterns and the corresponding bands suggested the existence of ZnO [30], OH bond [31], water molecule [32], carbonates [32–34] and phosphates [23, 35]. With increased concentration of H\textsubscript{2}O\textsubscript{2} added in Hank’s solution, the intensity of peak PO\textsubscript{4}\textsuperscript{3–} decreased first and then increased. Moreover, the content of phosphates in the corrosion product with Fenton’s reagent present displayed the lowest value in three solutions, as demonstrated in Fig. 9b. The existence of 10.0 mM H\textsubscript{2}O\textsubscript{2} lowered the intensity of peak PO\textsubscript{4}\textsuperscript{3–}, and the mixture of H\textsubscript{2}O\textsubscript{2} and FeCl\textsubscript{3} further reduced its value.

3.4. Electrochemical measurements

3.4.1. Open circuit potential

In Fig. 10, the evolution of open circuit potential (OCP) as a function of time during 3600 s of exposure in PBS is recorded. During the initial stage of immersion in all PBS, the OCP of pure Zn increased significantly and then dropped abruptly with time, which might be attributed to the breakdown of the surface film [36]. After that, the potential manifested a nearly constant value with prolonged test. The addition of 0.1 mM or 1.0 mM H\textsubscript{2}O\textsubscript{2} exerted marginal influence on the OCP of pure Zn in PBS. Nonetheless, 30.0 mM H\textsubscript{2}O\textsubscript{2} considerably changed the potential in PBS during the open-circuit exposure. The overall OCP notably moved towards the noble direction and fluctuated during the later stage, indicating the formation and breakdown of metastable surface on Zn [37]. For pure Zn immersed in Hank’s solution, likewise, the addition of 0.1 mM or 1.0 mM H\textsubscript{2}O\textsubscript{2} slightly differentiated the OCP values. With H\textsubscript{2}O\textsubscript{2} concentration identical to or higher than 10.0 mM, the overall OCP of Zn in Hank’s solution increased immensely, and larger increase appeared with higher H\textsubscript{2}O\textsubscript{2} concentration. As depicted in Fig. 10b, OCP of pure Zn sustained almost constant during the later stage of exposure, except the one in Hank’s solution with 1.0 % H\textsubscript{2}O\textsubscript{2} present. The potential drifted continuously to more positive direction owing to high concentration of H\textsubscript{2}O\textsubscript{2} and it might be associated with the deposition of corrosion product on the surface [38]. With the introduction of H\textsubscript{2}O\textsubscript{2} and FeCl\textsubscript{3} mixture, the overall OCP of Zn remained almost constant and exhibited higher value than that in
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3.4.2. Potentiodynamic polarization

To gain more thermodynamic and kinetic information about the degradation of Zn, potentiodynamic polarization test was performed in simulated inflammatory solutions. The corrosion potential ($E_{corr}$), current density ($i_{corr}$) and corrosion rate ($P_i$) estimated by Tafel extrapolation are listed in Tables 3 and 4.

The potentiodynamic polarization curves for pure Zn immersed in PBS media are demonstrated in Fig. 11a. The addition of 0.1 mM $H_2O_2$ or 1.0 mM $H_2O_2$ moderately altered the corrosion potential of Zn in PBS. On the contrary, the corrosion potential of pure Zn significantly moved towards the positive direction in PBS with the introduction of 30.0 mM $H_2O_2$. For pure Zn exposed to PBS only and PBS with 0.1 mM or 1.0 mM $H_2O_2$, the cathodic curves comprised of several cathodic reactions. With the addition of 30.0 mM $H_2O_2$, the PDP curve of pure Zn completely changed that the cathodic branch moved towards positive direction progressively. The cathodic current decreased with growing potential smoothly, and the anodic current displayed a rapid increase directly from the corrosion potential. The corrosion rates of pure Zn in four PBS were calculated and it turned out that pure Zn corroded fastest in PBS with 30.0 mM $H_2O_2$ added, and the addition of $H_2O_2$ with lower concentration mildly affected the corrosion rate.

The potentiodynamic polarization curves of Zn in Hank’s solution based simulated inflammatory solutions are present in Fig. 11b and c. The introduction of 0.1 mM $H_2O_2$ barely influenced the corrosion potential of pure Zn in Hank’s solution. However, with $H_2O_2$ equal to or more than 1.0 mM added, the corrosion potential considerably shifted to higher values. Besides, the corrosion potential exhibited higher value as the concentration of $H_2O_2$ increased. In the meantime, pure Zn displayed faster cathodic reactions and anodic reactions in Hank’s solution with the addition of $H_2O_2$. Similarly, the effect of $H_2O_2$ on cathodic reactions became more pronounced as its concentration increased. Furthermore, the cathodic curves of pure Zn in Hank’s solutions except the one in media with 1.0 % $H_2O_2$ contained exhibited similar shapes and constituted several cathodic reactions. The cathodic process of Zn in Hank’s solution was notably altered by 1.0 % $H_2O_2$. The cathodic current decreased smoothly and nearly linearly as potential increased, and the change of anodic current displayed the similar trend. With the mixture of $H_2O_2$ and $FeCl_3$ added, the corrosion potential of pure Zn in Hank’s solution moved towards noble direction, but exhibited more negative value compared to that in the media with 10 mM $H_2O_2$ only.

The corrosion rates of pure Zn in all Hank’s solution were calculated by the Tafel extrapolation. For samples immersed in Hank’s solution with $H_2O_2$ equal to or less than 10.0 mM, the corrosion rate was insignificantly affected by $H_2O_2$. With respect to media added with 0.1 % or 1.0 % $H_2O_2$, the dissolution of pure Zn got remarkably accelerated by $H_2O_2$ and the corrosion rate displayed the highest value in presence of 1.0 % $H_2O_2$. After adding 10.0 mM $H_2O_2$, the corrosion of Zn in Hank’s solution was encouraged. With the addition of Fenton’s reagent, the degradation of Zn slowed down but was still faster than that in Hank’s solution.

3.4.3. Electrochemical impedance spectroscopy

To better understand the electrochemical corrosion processes involved in corrosion media, electrochemical impedance spectroscopy (EIS) was carried out on pure Zn in simulated inflammatory solutions.

Hank’s solution, and lower value compared to that with only 10 mM $H_2O_2$ added.

Fig. 4. (a, c) weight loss and (b, d) corrosion rate of pure Zn during 14-day immersion in Hank’s solution.
Fig. 5. The surface morphology of pure Zn after 14-day immersion in PBS.
prepared in PBS or Hank’s solution to provide data for identifying the occurrence of multistep charge transfer, coupling of the interfacial reactions, various intermediate adsorption steps and so on [39]. The impedance response of pure Zn after 1 h of open-circuit exposure are illustrated in Fig. 12.

For samples immersed in PBS and PBS with 0.1 mM or 1.0 mM H$_2$O$_2$, the nyquist plot acquired was composed of two capacitive loops. The semicircle at high frequency corresponded to the charge transfer process and electrochemical double layer, and the capacitive loop at low frequency might be associated with the passive film on the surface. Meanwhile, for pure Zn in Hank’s solution with 1.0 % H$_2$O$_2$ present, aside from two capacitive loops, the inductive loop implying the adsorption of intermediate species also appeared in the impedance response [44]. The corrosion process of Zn was altered significantly and the severe corrosion might take place owing to the highly oxidizing environment. Furthermore, three nyquist plots in Fig. 12c demonstrated similar patterns. The plot in the presence of Fenton’s reagent displayed

<p>| Table 1 |
| Results of EDS analysis of sample morphology in PBS solutions (at. %). |</p>
<table>
<thead>
<tr>
<th>Day 14</th>
<th>Zn</th>
<th>O</th>
<th>C</th>
<th>P</th>
<th>Cl</th>
</tr>
</thead>
<tbody>
<tr>
<td>PBS - surface</td>
<td>44.6</td>
<td>33.4</td>
<td>12.4</td>
<td>5.7</td>
<td>3.8</td>
</tr>
<tr>
<td>PBS - corrosion product</td>
<td>17.9</td>
<td>56.4</td>
<td>7.0</td>
<td>18.6</td>
<td>0.1</td>
</tr>
<tr>
<td>PBS +0.1 mM H$_2$O$_2$ - surface</td>
<td>58.0</td>
<td>20.2</td>
<td>20.4</td>
<td>1.4</td>
<td></td>
</tr>
<tr>
<td>PBS +0.1 mM H$_2$O$_2$ - corrosion product</td>
<td>21.1</td>
<td>49.5</td>
<td>9.3</td>
<td>16.7</td>
<td>3.5</td>
</tr>
<tr>
<td>PBS +1.0 mM H$_2$O$_2$ - surface</td>
<td>45.9</td>
<td>31.1</td>
<td>16.1</td>
<td>6.9</td>
<td></td>
</tr>
<tr>
<td>PBS +1.0 mM H$_2$O$_2$ - corrosion product</td>
<td>21.1</td>
<td>49.0</td>
<td>9.3</td>
<td>17.0</td>
<td>3.6</td>
</tr>
<tr>
<td>PBS +30.0 mM H$_2$O$_2$ - surface</td>
<td>65.2</td>
<td>13.1</td>
<td>15.5</td>
<td>3.8</td>
<td>2.3</td>
</tr>
<tr>
<td>PBS +30.0 mM H$_2$O$_2$ - corrosion product</td>
<td>24.1</td>
<td>45.6</td>
<td>11.1</td>
<td>10.5</td>
<td>8.7</td>
</tr>
</tbody>
</table>

To gain a deeper insight about the corrosion from EIS measurements, the equivalent circuit in Fig. 12d and e were utilized to interpret the nyquist plots of pure Zn. Herein, the constant phase element (CPE) is used to replace the capacitance to better fit the experimental data and compensate the non-homogeneity induced by factors such as surface roughness and heterogeneities. The EIS spectra in the PBS with 30.0 mM H$_2$O$_2$ added can be fitted with the equivalent circuit drawn in Fig. 12e, which might be associated with the localized corrosion such as pitting process [42] or formation of intermediate products [43].

Table 2
<p>| Results of EDS analysis of sample morphology in Hank’s solutions (at. %). |</p>
<table>
<thead>
<tr>
<th>Day 14</th>
<th>Zn</th>
<th>O</th>
<th>C</th>
<th>Ca</th>
<th>P</th>
<th>Mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hank’s solution - surface</td>
<td>27.8</td>
<td>43.6</td>
<td>14.2</td>
<td>5.9</td>
<td>6.9</td>
<td>1.7</td>
</tr>
<tr>
<td>Hank’s solution - corrosion product</td>
<td>15.5</td>
<td>51.7</td>
<td>16.3</td>
<td>7.1</td>
<td>7.5</td>
<td>1.8</td>
</tr>
<tr>
<td>Hank’s solution +0.1 mM H$_2$O$_2$ - surface</td>
<td>41.9</td>
<td>32.1</td>
<td>17.0</td>
<td>3.6</td>
<td>4.2</td>
<td>1.2</td>
</tr>
<tr>
<td>Hank’s solution +0.1 mM H$_2$O$_2$ - corrosion product</td>
<td>13.4</td>
<td>55.4</td>
<td>13.3</td>
<td>7.5</td>
<td>8.9</td>
<td>1.5</td>
</tr>
<tr>
<td>Hank’s solution +1.0 mM H$_2$O$_2$ - surface</td>
<td>52.7</td>
<td>26.2</td>
<td>16.4</td>
<td>1.7</td>
<td>3.0</td>
<td></td>
</tr>
<tr>
<td>Hank’s solution +1.0 mM H$_2$O$_2$ - corrosion product</td>
<td>20.9</td>
<td>49.4</td>
<td>11.2</td>
<td>7.5</td>
<td>9.9</td>
<td>1.1</td>
</tr>
<tr>
<td>Hank’s solution +10.0 mM H$_2$O$_2$ - surface</td>
<td>68.8</td>
<td>7.8</td>
<td>22.5</td>
<td></td>
<td>0.9</td>
<td></td>
</tr>
<tr>
<td>Hank’s solution +10.0 mM H$_2$O$_2$ - corrosion product</td>
<td>38.0</td>
<td>37.0</td>
<td>16.2</td>
<td>2.9</td>
<td>5.3</td>
<td>0.7</td>
</tr>
<tr>
<td>Hank’s solution +0.1% H$_2$O$_2$ - surface</td>
<td>66.7</td>
<td>9.0</td>
<td>23.5</td>
<td></td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>Hank’s solution +0.1% H$_2$O$_2$ - corrosion product</td>
<td>69.3</td>
<td>5.8</td>
<td>24.4</td>
<td></td>
<td>0.5</td>
<td></td>
</tr>
<tr>
<td>Hank’s solution +10.0% H$_2$O$_2$ - corrosion product</td>
<td>50.5</td>
<td>26.8</td>
<td>18.2</td>
<td>0.9</td>
<td>3.5</td>
<td></td>
</tr>
<tr>
<td>Hank’s solution +1.0 mM H$_2$O$_2$ +FeCl$_3$ - corrosion product</td>
<td>72.5</td>
<td>5.3</td>
<td>21.8</td>
<td></td>
<td>0.4</td>
<td></td>
</tr>
</tbody>
</table>

Fig. 6. The surface morphology of pure Zn during 14-day immersion in Hank’s solution.
the largest diameter, implying the enhanced corrosion resistance.

4. Discussion

4.1. Corrosion mechanism of Zn in PBS and Hank’s solution

4.1.1. Zn degradation in PBS

The investigation on Zn corrosion in PBS has been carried out in recent years [45, 46], and the corrosion process can be generally described as follows (Fig. 13): in the initial stage, Zn was oxidized to $\text{Zn}^{2+}$, and the dominated cathodic reaction proceeding by means of reduction of oxygen dissolved in water gave rise to $\text{OH}^-$.

The unceasingly released $\text{OH}^-$ raised the pH value of PBS and reacted with $\text{Zn}^{2+}$, leading to the formation of insoluble $\text{Zn(OH)}_2$. Subsequently, $\text{ZnO}$ was formed due to its higher thermodynamical stability than $\text{Zn(OH)}_2$. The hydrogen phosphate ions abundant in PBS might also react with released $\text{Zn}^{2+}$ and $\text{Zn}_2(\text{PO}_4)_3\cdot4\text{H}_2\text{O}$ was generated. Moreover, the Cl$^-$ from PBS might attack $\text{Zn(OH)}_2$ and the simonkolleite was therefore precipitated. The accumulation of precipitations contributed to the formation of the corrosion product layer, inhibiting the further dissolution of Zn and lowering the corrosion rate consequently.

$$\text{Zn} \rightarrow \text{Zn}^{2+} + 2\text{e}^- \quad (1)$$

$$\text{O}_2 + \text{H}_2\text{O} + 4\text{e}^- \rightarrow 4\text{OH}^- \quad (2)$$

$$\text{Zn}^{2+} + 2\text{OH}^- \rightarrow \text{Zn(OH)}_2 \quad (3)$$
4.1.2. Zn degradation in Hank’s solution

The chemical composition of PBS deviated a lot from sophisticated physiological environment, and utilization of singular corrosive medium without renewal throughout the whole test might result in the depletion of H$_2$O$_2$ and free ions, the accumulation of released ions, and diverged solution pH. To better evaluate the degradation behaviour of Zn, Hank’s solution containing all essential inorganic components in the human body fluid was adopted and the electrolyte renewal was conducted every 3 days to simulate the physiological homeostasis and restore the setting pH$_2$O$_2$ concentration.

For pure Zn in Hank’s solution, likewise, the anodic reaction was the dissolution of zinc and the oxygen reduction reaction was the primary cathodic reaction. OH$^-$ and Zn$_{2+}$ in the aqueous solution contributed to the precipitation of Zn(OH)$_2$ and then ZnO. The solution pH increased gradually due to the continuing corrosion of Zn. Released Zn$_{2+}$ might react with ions such as HPO$_4^{2-}$ or HCO$_3^-$, forming less soluble corrosion products. As immersion time extended, the phosphate and carbonate agglomerates dispersed on the surface and continued to grow, and thus inhibited the corrosion of Zn. It’s reported that ZnO, Zn$_5$(CO$_3$)$_2$(OH)$_6$ and Zn$_3$(PO$_4$)$_2$.4H$_2$O were formed as the primary corrosion product on pure Zn during the immersion in Hank’s solution [47], which might also be precipitated in this study. Moreover, Ca$^{2+}$ and HPO$_4^{2-}$ in Hank’s solution might give rise to the formation of Ca-P salts, and further reduced the corrosion rate of pure Zn. Each time after the electrolyte renewal, pH level restored to the original value at 7.40. As the degradation of Zn continued proceeding, the accompanied formation of OH$^-$ raised the solution pH again. The concentration of essential components such as Ca$^{2+}$, phosphate, and carbonate species changed due to the renewal and

\[ \text{Zn}^{2+} + 2\text{HPO}_4^{2-} + 2\text{OH}^- + 2\text{H}_2\text{O} \rightarrow \text{Zn}_5(\text{PO}_4)_2.4\text{H}_2\text{O} \]  

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\[ \text{Zn}^{2+} + 2\text{HPO}_4^{2-} + 2\text{OH}^- + 2\text{H}_2\text{O} \rightarrow \text{Zn}_5(\text{PO}_4)_2.4\text{H}_2\text{O} \]  

\[ 3\text{Zn}^{2+} + 2\text{HPO}_4^{2-} + 2\text{OH}^- + 2\text{H}_2\text{O} \rightarrow \text{Zn}_5(\text{PO}_4)_2.4\text{H}_2\text{O} \]  

\[ 5\text{Zn}^{2+} + 2\text{Cl}^- + 2\text{H}_2\text{O} \rightarrow \text{Zn}_5\text{(OH)}_3\text{Cl}_2.2\text{H}_2\text{O} + 2\text{OH}^- \]
it can better emulate that in the physiological environment [48].

\[ 5Zn^{2+} + 2HCO_3^- + 8OH^- = Zn_5(CO_3)_{2}(OH)_6 + 2H_2O \]  

(7)

4.2. The influence of \( H_2O_2 \) on the corrosion mechanism of \( Zn \)

4.2.1. \( Zn \) degradation in PBS-based simulated inflammatory solutions

\( H_2O_2 \) is the major ROS released by activated inflammatory cells in living organisms to attack some biologic targets [14]. It’s known as a strong oxidant and can act to modify the corrosion resistance of metals [29]. It’s reported that inflammatory cell-based media containing \( H_2O_2 \) can alter the corrosion susceptibility and the surface oxide impedance of biomedical metals [6], and drive the corrosion in a biologically relevant manner [2].

During 14-day immersion in PBS with 0.1 mM or 0.1 mM \( H_2O_2 \) present, pure \( Zn \) corroded almost the same as that in PBS, and the degradation behaviour characterized by corrosion rate, corrosion product and corrosion morphology, as well as the variations in solution pH, was insignificantly influenced by \( H_2O_2 \). Nonetheless, after adding 30.0 mM \( H_2O_2 \) into PBS, the oxidative environment substantially
increased the corrosion susceptibility of Zn. The potentiodynamic polarisation test tends to give information on the instantaneous corrosion rate, and thus the derived corrosion rates differed from that in the immersion test. However, analysis of PDP curves also allows the quantification of relative rates of the anodic reaction and cathodic reaction over a range of potentials, unraveling the mechanistic aspects of Zn corrosion [49]. Herein, the cathodic current considerably drifted towards higher values in the presence of 30.0 mM H₂O₂. It can be speculated that cathodic reactions involved as such as the oxygen reduction were immensely facilitated by H₂O₂. And the reduction of H₂O₂ might also play a vital role in the positive shift of cathodic current during the polarization test.

\[
\text{H}_2\text{O}_2 + 2e^- \rightarrow 2\text{OH}^-
\] (8)

More importantly, encouraged oxygen reduction and the reduction of H₂O₂ might be associated with the dramatic increase in pH values in the initial stage of immersion, and spontaneously promote the dissolution of pure Zn in PBS, resulting in the striking upsurge of corrosion rate in the immersion test and electrochemical tests [50]. Similarly, Gilbert et al. [15] demonstrated that the addition of H₂O₂ into PBS greatly increased the corrosion current and reduced the corrosion resistance of CoCrMo alloy, and H₂O₂ also reduced the corrosion resistance of Ti6Al4V in PBS about three orders of magnitude and the i corr increased [16]. Moreover, H₂O₂ can accelerate the corrosion of the stainless steel in varying degree at different concentrations [11-13]. The influence of H₂O₂ on the corrosion behaviour of tradition biomedical metals and biodegradable Zn seemed to be alike in PBS and were inclined to promote the corrosion of metal in general.

Furthermore, during 14-day immersion, the continuing dissolution of Zn released a large quantity of Zn²⁺ into PBS, however, the deposition and accumulation of corrosion product on the sample surface was notably inhibited by 30.0 mM H₂O₂. Interestingly, the amount of Zn₉(OH)₆Cl₂·H₂O in the corrosion product increased after 14 days. In addition, the electrochemical approaches revealed the conspicuous increase in overall OCP of pure Zn due to 30.0 mM H₂O₂, consistent with its effect on CoCrMo alloy [51] and Ti6Al4V alloy [16] immersed in PBS, and even 316 L stainless steels in the natural seawater [52]. By using in situ electrochemical atomic force microscopy (ECAFM), Gilbert et al. [51] confirmed that the addition of H₂O₂ raised the OCP value, altered the oxide film behaviour on CoCrMo alloy and significantly changed the oxide film topography. It turned out that in H₂O₂ contained PBS, less oxide dome coarsening took place on the surface of pure Ti and Ti6Al4V alloy, as well [50]. Hence, the sharp increase in the OCP value might indicate the dramatic changes in the structure and properties of the oxide film on the surface of Zn [14]. Furthermore, in this study, the impedance response of pure Zn in PBS was notably altered by 30.0 mM H₂O₂, which might send similar messages about the prominently affected chemical reactions occurring on the metal/electrolyte interface. The differences of nyquist plots appeared at lower frequency, which might also indicate that the sample surface was mainly influenced by oxide deterioration, likely caused by corrosive H₂O₂ attack [53]. Thus, it can come to a conclusion that properties of the passive film on pure Zn was greatly influenced by H₂O₂, leading to the restrained formation and deposition of oxides, and preferred precipitation of Zn₉(OH)₆Cl₂·H₂O as H₂O₂ can affect the type of oxide formed [53]. The conjecture on the oxide film might not be solid or convincing enough, and the hypothesis need to be validated carefully in the future. Efforts will be made to comprehensively elucidate the corrosion mechanism.

Moreover, the pitting corrosion was considerably promoted by 30 mM H₂O₂, which can be ascribed to the insufficient protection on the surface. With extended immersion, the accumulation of corrosion product enhanced the corrosion resistance of pure Zn, and thus the Zn degradation gradually slowed down. Notably, the peroxide consumption in PBS might also be the reason for the decreasing corrosion rate [53].

4.2.2. Zn degradation in Hank’s solution-based simulated inflammatory solutions

In Hank’s solution, thanks to the electrolyte renewal performed
every three days, the concentration of H$_2$O$_2$ and essential components in simulated body solution can be roughly sustained at desired values, and might be able to describe the effect of simulated inflammatory condition on the degradation behaviour of pure Zn more precisely.

The introduction of H$_2$O$_2$ varied the corrosion susceptibility of pure Zn in Hank’s solution. In the initial stage of exposure, the degradation of Zn was inevitably affected by H$_2$O$_2$. With concentration higher than 1.0 mM, H$_2$O$_2$ raised the OCP values of pure Zn in Hank’s solution. Besides, in the presence of H$_2$O$_2$ less than 1.0 %, the EIS spectra of Zn in Hank’s solution barely changed, but exhibited diverse dimensions with varying H$_2$O$_2$ concentration. It can be deduced that the chemical reactions involved in the very beginning of exposure were slightly influenced by H$_2$O$_2$. However, in Hank’s solution with 1.0 % H$_2$O$_2$, the inductance appeared in the EIS spectrum, which signified the massive change in chemical reactions involved at the electrode/electrolyte interface and implied potentially unsteady surface/interface reactions induced by 1.0 % H$_2$O$_2$ [54].

The potentiodynamic polarization results showed that the cathodic current of pure Zn in Hank’s solution was elevated by H$_2$O$_2$ of any concentrations. The $i_{corr}$ derived from the cathodic branch of the polarization curves was raised by H$_2$O$_2$, and exhibited higher value with increasing concentration of H$_2$O$_2$. The addition of 0.1 % or 1.0 % H$_2$O$_2$ immensely elevated the corrosion rate, just like that in PBS with 30.0 mM H$_2$O$_2$. More importantly, it can be reasonably conjectured that the reduction of oxygen, and possibly H$_2$O$_2$ as well might be promoted during the corrosion of Zn. The increase in overall pH value of Hank’s solution induced by H$_2$O$_2$ can thus be ascribed to the by-product OH$^-$ of facilitated cathodic reactions. However, for long-term immersion, the effect of H$_2$O$_2$ on the media pH varied at different concentrations. In Hank’s solution containing H$_2$O$_2$ > 1.0 mM, the evolution of pH values completely changed and manifested a downward trend with time. It should be noted that the glucose, widely accepted as the primary nutrient to maintain and promote cell function with a concentration about 1.1 g/L in human blood plasma, existed in Hank’s solution with a similar concentration to that in the human body [55]. The glucose can transform into the gluconic acid, inducing the acidification of solution during the immersion and greatly affecting the degradation of materials [56–58]. High concentration of H$_2$O$_2$ in Hank’s solution contributed to the oxidative environment, with high possibility to motivate the transformation of glucose into gluconic acid and therefore lowered the solution pH in consequence.

During the immersion test, H$_2$O$_2$ equal to or less than 0.1 % added to Hank’s solution inhibited the corrosion of Zn, and the influence of H$_2$O$_2$ was different from that in the potentiodynamic polarization test. The Zn corrosion in the immersion test manifested as the mass loss determined the amount of cumulative corrosion of Zn without being accelerated, while the PDP test gave kinetic and thermodynamic information on the Zn corrosion [49]. Despite different corrosion rates from immersion and potentiodynamic polarization, two tests together provided necessary information to better understand the corrosion behaviour of Zn. More importantly, the effect of H$_2$O$_2$ on the corrosion of Zn significantly differed between Hank’s solution and PBS, and was more complicated in Hank’s solution as in most studies, H$_2$O$_2$ tended to promote the corrosion of metals. It’s speculated that the solution chemistry might play an essential role in the depressed corrosion of Zn. The presence of inorganic ions such as Ca$^{2+}$, Mg$^{2+}$ and HCO$_3^-$, and glucose in Hank’s solution might not only affect the formation of corrosion product, the evolution of solution pH, the degradation rate of pure Zn, but also change the way H$_2$O$_2$ participating in the Zn degradation through chemical interactions, and contribute to the totally different degradation of Zn from that in PBS, with or without H$_2$O$_2$. The detailed explanation on the mechanism was out of reach in this study and great efforts will be made in the future to illustrate it correctly and precisely.

Moreover, the formation of corrosion product, including ZnO, Zn$_2$(CO$_3$)$_2$(OH)$_4$ and Zn$_5$(PO$_4$)$_3$-4H$_2$O, on the surface was suppressed by H$_2$O$_2$ equal to or less than 0.1 % during 14 days of immersion, and the precipitation of Ca-P salts was retarded. With increasing concentration, the inhibition effect of H$_2$O$_2$ on the corrosion product became more visible. It’s reported that H$_2$O$_2$ could prevent the formation of the protective CaH(PO$_4$)$_2$-2H$_2$O layer on 316 stainless steel immersed in Hank’s solution. The study demonstrated that H$_2$O$_2$ could clean the surface from the easily oxidative species and oxidation of the surface components, and resulted in the lower amount of precipitates on the surface [11]. The addition of H$_2$O$_2$ into Hank’s solution also increased the oxide film instability of pure Ti and generated defects in the oxide layer [53]. Likewise, the dynamic oxide film interacted with the complex aqueous solution during the immersion, and the surface property of pure Zn in Hank’s solution might be altered by H$_2$O$_2$, leading to retarded formation of corrosion product in consequence. Accordingly, the pitting corrosion took place on the surface in the absence of protective layer, which was also recognized on the surface of pure Ti [59].

However, 1.0 % H$_2$O$_2$ remarkably favoured the degradation of pure Zn in Hank’s solution and raised the corrosion rate immensely. The formation of corrosion product was still restrained by H$_2$O$_2$, but the inhibition effect diminished, and precipitations were formed and deposited on the sample surface. The oxidizing properties of H$_2$O$_2$ of high concentration prevailed, increased the oxidation reaction rate and produced the oxide consequently [53]. The enhanced Zn degradation induced by 1.0 % H$_2$O$_2$ matched with the effect of 100 mM H$_2$O$_2$ on 316 stainless steel exposed to Hank’s solution [11].

4.3. The influence of Fenton reaction-based solution chemistry on Zn degradation

It’s known that a variety of ROS released from cells during inflammatory activation can engage in the Fenton reactions and generated significant amounts of radical species. The reactive oxygen intermediate species capable of oxidizing organic compounds can further accelerate corrosion, stimulate the cells and thus be hypothesized to exert impact on the corrosion of titanium and CoCrMo alloy [60], maybe biodegradable metal as well. The simulated inflammatory solution containing Hank’s solution and Fenton’s reagent was prepared in this study to replicate the Fenton reaction condition.

\[
\begin{align*}
\text{O}_2^- + \text{H}_2\text{O}_2 & \rightarrow \text{HO}^+ + \text{OH}^- + \text{O}_2 \\
\text{O}_2^- + \text{Fe}^{3+} & \rightarrow \text{O}_2 + \text{Fe}^{2+} \\
\text{H}_2\text{O}_2 + \text{Fe}^{2+} & \rightarrow \text{HO}^+ + \text{OH}^- + \text{Fe}^{3+}
\end{align*}
\]

It turned out that the OCP of pure Zn exhibited higher values under the Fenton condition. It’s reported that Fenton reactions could increase the oxidizing power of PBS medium [14] and lead to a significant rise in OCP of CoCrMo alloy, as well [15]. More importantly, it’s claimed that Fenton reactions could alter the oxide film characteristics and the film became less protective, resulting in the faster metal corrosion in PBS-based simulated inflammatory solutions [14]. Nonetheless, the degradation of Zn in Hank’s solution was differently affected by Fenton reactions. The corrosion rate of pure Zn decreased in presence of Fenton reactions and the EIS spectra suggested the enhanced corrosion resistance. The formation of corrosion product was notably depressed and the content of phosphates on the sample surface diminished. In comparison to Hank’s solution with 10.0 mM H$_2$O$_2$, pure Zn exposed to Fenton reaction condition degraded faster, with less corrosion product formed and less phosphates contained. Likely, it can be deduced that the relatively complicated chemical composition of Hank’s solution compared with PBS might be ascribed for the differently functioned Fenton reactions.

4.4. Outlook

The corrosion behaviour of biodegradable pure Zn under simulated inflammatory condition was determined in this study, by introducing
H₂O₂ and Fenton reactions into PBS and Hank’s solution. However, the physiological environment is more complex than designed simulated inflammatory conditions. The immune system possesses the ability to rapidly alter the chemistry of the local environment, and can give rise to the corrosion damage on the implant surface. Aside from H₂O₂ and Fenton reactions, there are other powerful reactive chemical species such as superoxide (O₂⁻), hypochlorous acid (HOCl) and nitric oxide (NO) also existing and possibly getting involved in the inflammatory cell-induced corrosion of biodegradable metals, and which species is the prime factor for the induced corrosion remains unknown [5,61]. For instance, HOCl is a potent oxidizing agent and can increase the capacity of reduction reaction in the solution, which may further serve to drive the corrosion by raising the rate of oxidation. In the human body, the neutrophils might unceasingly generate HOCl near implants as long as active inflammation continues [2]. In the studies of tradition medical metals, Gilbert et al. [2] found that HOCl raised the open circuit potential and the corrosion current of CoCrMo alloy in PBS, affected the oxide film, and caused extensive surface damage on it. By contrast, the effect of HOCl on Ti6Al4V alloy was less severe and resulted in slight corrosion damage. The influence of HOCl on the corrosion behaviour of biodegradable metals arouses much curiosity but the research is almost blank. What’s more, it should be noted that the acidity accompanying the inflammation can also be a big issue for metal corrosion. In H₂O₂ containing PBS, the increase of acidity turned out to raise the oxide resistance but lower the corrosion susceptibility of CoCrMo alloy, counterintuitive in that normally lower pH resulted in accelerated corrosion [15]. The complexity of the inflammatory environment made the corrosion of metals, especially for biodegradable metals dissolvable in the human body, quite difficult to be predicted. Multiple ROS, chemical reactions, pH and so on might not only cause a simple additive effect, but also combine in a complicated manner to amplify or suppress the corrosion. 

Due to the complexity of peri-implant environment with a large range of factors, the in vitro corrosion characterization using the simply simulated body fluids to mimic the physiological environment without considering the inflammatory condition are very likely to induce a significant misestimation of corrosion behaviour of materials. The interplay among the implants, ROS and cells could greatly affect the chemical reactions and potentially physical interactions at the implant surface. And there might exist the positive-feedback loop in which inflammation and corrosion processes of materials feed off each other. The inflammation may provide driving force or hinder for corrosion, and in turn, the corrosion might exacerbate or promote the immune response [2]. It’s thus of great importance to consider designing the in vitro corrosion tests in a way that can more closely simulate the natural biochemistry. This study, firstly targeting on this topic in the research field of biodegradable Zn, investigate and evaluate the capacity of H₂O₂ and Fenton reactions to cause corrosion attack on pure Zn. It will help better understand the corrosion process of biodegradable metals in vivo, provide necessary information on designing and developing materials and raise concerns regarding the biodegradation induced by biological consequences.

5. Conclusion

In this study, for the first time, the degradation mechanism of pure Zn in H₂O₂ or Fenton reactions contained simulated inflammatory solutions were evaluated. The vital roles H₂O₂ or Fenton reactions played in the Zn degradation were revealed and the great importance of taking inflammatory cell induced corrosion into consideration during the evaluation of biodegradation processes of Zn was thus highlighted.

(1) High concentration of H₂O₂ created an aggressive environment in PBS and Hank’s solution, altering the surface properties of Zn, retarding the formation of corrosion product, and remarkably promoting the Zn corrosion. (2) In PBS with H₂O₂ less than 30.0 mM, the degradation behaviour of pure Zn and the solution pH were insignificantly affected by H₂O₂. In Hank’s solution with H₂O₂ identical to or less than 0.1 %, the corrosion behaviour of Zn was notably affected and dependent on the concentration of H₂O₂. The inhibition effect on the corrosion rate and corrosion product formation increased with more H₂O₂ present. (3) The Fenton reactions retarded the corrosion of Zn in Hank’s solution and inhibited the formation of corrosion product on the surface.

CRediT authorship contribution statement

Xiao Liu: Conceptualization, Methodology, Investigation, Writing - original draft, Data curation. Wei Yuan: Conceptualization, Methodology, Investigation, Writing - review & editing. Danni Shen: Conceptualization, Methodology, Investigation. Yan Chen: Conceptualization, Methodology, Investigation. Dafu Chen: Conceptualization, Methodology, Investigation, Writing - review & editing, Project administration, Funding acquisition. Yufeng Zheng: Conceptualization, Methodology, Resources, Supervision, Writing - review & editing, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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