Improved cytocompatibility of Mg-1Ca alloy modified by Zn ion implantation and deposition

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ABSTRACT

The Mg-1Ca alloy has been modified by Zn ion implantation and deposition (II&D) using metal vapor vacuum arc plasma source (MEVVA). The surface characteristics, corrosion behavior and cytocompatibility were investigated. The auger electron spectroscopy (AES) results showed that a uniform Zn layer was formed on the surface of Mg-1Ca alloy. The electrochemical measurements revealed that the corrosion potential has been increased, and a comparable corrosion current density in the simulated body fluid (SBF) was obtained for the Zn-modified Mg-1Ca alloy. The indirect cell viability evaluation and direct cell culture results indicated that the cytocompatibility of MC3T3-E1 cells on the Mg-1Ca alloy was improved by the Zn layer on the surface.

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1. Introduction

As the potential biodegradable materials for implants, Mg-Ca alloys have gained a lot of attention in recent years due to their excellent mechanical properties, good biocompatibility and biodegradable behavior [1,2]. It is reported that the binary Mg-Ca alloy is qualification as orthopedic biodegradable materials and Ca is able to accelerate the healing of broken or damaged bones by producing hydroxyapatite during the degradation of the alloy in the body [3,4].

Recently, many methods such as magnetron sputtering [5], micro-arc oxidation [6] and ion implantation [7,8] have been used to improve the corrosion resistance and biocompatibility of Mg and Mg alloy. It is known that both Fe and Zn are members of biodegradable metals with lower degradation rate than Mg and Mg alloy [9]. Fe and Zn has been used as source elements to perform surface modification on Mg alloy and thus to influence the corrosion or mechanical behavior. It was reported that a biodegradable pure Fe thin film with the thickness from 2.73 μm to 6.36 μm was incorporated on the surface of ZK60 alloy using ion implantation and deposition (II&D) and the modified alloy exhibited better corrosion resistance [10]. Some studies indicated that the degradation rate of Mg and Mg-Ca alloy in SBF increased greatly after Zn ion implantation, which may be ascribed to galvanic effect between Zn rich area and matrix, although the surface mechanical properties kept stable [7,8]. However, there is seldom research on the cytocompatibility of Mg alloy surface modified with Fe or Zn film. It is known that Zn is a necessary trace element for cell activities and is specifically important in the formation and maintenance of bone structures [11,12]. Therefore, a uniform modified Zn film with proper thickness may be beneficial to the biocompatibility of Mg and Mg alloy. In the present work, Zn II&D has been performed on Mg-1Ca alloy to form new surface film and the improved corrosion behavior and cytocompatibility have been investigated.

2. Materials and methods

The as-extruded Mg-1Ca (wt%) rod same as reported in Ref. [13] was cut into wafer with 10 mm in diameter and 1.5 mm in thickness. The samples were mechanically polished and then ultrasonically cleaned in ethanol.

Zn II&D was performed using the compound coating machine with MEVVA ion source (Beijing Normal University, Beijing, China). The voltage during implantation was 8 keV, and the goal of ion implantation before deposition was to enhance the combination ability of deposition film with substrate. The deposition process continued for 60 min at the current of 25 mA.

The elemental depth analysis was performed with auger electron spectroscopy (AES, PHI 700, ULVAC-PHI, Japan). The sputtering rate was 38 nm/min for SiO₂.

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The electrochemical corrosion behavior was evaluated on an electrochemical workstation (CHI 660e, China) based on the traditional three-electrode system in SBF at 37 ºC. The scanning rate was 1 mV/s.

The MC3T3-E1 cells were cultured in α-MEM with 10% FBS at 37 ºC in a humidified atmosphere of 5% CO2. Before the experiment, the samples were sterilized and then immersed in α-MEM with the ratio at 1 cm2/ml for 24 h. The original extracts supplemented with 10% FBS were used at experiment.

The cells were seeded in 96-well plates at a density of 3000 cells per 100 µl and incubated for 24 h to allow attachment. Then the media were substituted by extracts with concentration of 100% and 50%. MTT assay was used to evaluate cell proliferation after culturing. The absorbance was measured at a wavelength of 570 nm on the enzyme-link meter (MultiSkan MK3, Thermo).

The cells were seeded on each sample in 12-well plates at a density of 30,000 cells ml⁻¹. The seeded samples were washed with PBS and then fixed with 2.5% glutaraldehyde. Then the cells were dehydrated in sequential concentrations of ethanol. The Hexamethyl disilylamine was added and then dried in air. SEM was used to analysis cell images.

3. Results and discussion

The AES depth profile of Zn-modified Mg-1Ca alloy (treated) is depicted in Fig. 1. The surface structure of the treated sample is divided into deposition zone and implantation zone based on the elemental distribution. In the deposition zone, the distributions of Zn and O form a plateau from 10 nm to 150 nm with a concentration of Zn and O form a plateau from 10 nm to 150 nm with a concentration of ZnO, indicating the occurrence of oxidized reaction of the samples in air. In the implantation zone, the concentration of Zn decreases almost in a linear manner. For the Mg-1Ca alloy, the outmost surface composition of Mg is almost the same as that of O and the concentration of O decreases with increasing depth [13].

Fig. 2 displays the potentiodynamic polarization curves of Mg-1Ca alloy and treated sample in SBF at 37 ºC. The Ecorr of Mg-1Ca alloy is about −1.92 V/SCE and increases to −1.58 V/SCE after Zn II&D. It indicates that the treated sample has a higher Ecorr due to the presence of ZnO, indicating the occurrence of oxidized reaction of the samples in air. In the implantation zone, the concentration of Zn decreases almost in a linear manner. For the Mg-1Ca alloy, the outmost surface composition of Mg is almost the same as that of O and the concentration of O decreases with increasing depth [13].

Fig. 2. Potentiodynamic polarization curves of Mg-1Ca alloy and treated sample.

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4. Conclusions

The Mg-1Ca alloy has been modified by Zn ion implantation and deposition. The corrosion potential increases and the corrosion current density is comparable for the modified Mg-1Ca alloy in the simulated body fluid. The treated sample shows similar cell viability with Mg-1Ca alloy at 100% concentration extracts and better cell growth when the extract is diluted to 50%. Better cell adhesion and growth are exhibited on the treated sample with short term cell culture, showing the improved cytocompatibility of Mg-1Ca by Zn ions.

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