In situ decomposition of Ti$_2$AlN promoted interfacial bonding in Zn7Al-Ti$_2$AlN biocomposites for bone repair

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Abstract

In this study, in situ decomposition of Ti$_2$AlN was used to obtain strong interfacial bonding in Zn7Al-Ti$_2$AlN composites prepared via laser melting. During the preparation process, the Al atoms in Ti$_2$AlN could diffuse out of the lattice due to the weak bonding between Al and Ti, followed by easily diffusing into the liquid Zn7Al matrix. Consequently, the diffused Al could bond with the Al in Zn7Al matrix owing to their inherent chemical affinity, leading to a strong interfacial bonding in Zn7Al-Ti$_2$AlN composites. This significantly improved the load transfer ability and prohibited the motion of dislocations in the composites. As a result, the hardness and compressive strength of Zn7Al-Ti$_2$AlN composites were enhanced from 74 HV and 155 MPa to 80 HV and 205 MPa, respectively, which were more suitable for bone repair application. What’s more, the composites also showed improved accelerated degradation and cytocompatibility.

1. Introduction

Zn alloys are introduced as a new class of bio absorbable metals for biomedical application owing to their good biocompatibility as well as nearly ideal biodegradation rate [1–5]. In terms of biocompatibility, as an abundant element in the human body, Zn plays a vital role in cell proliferation and the metabolism of human body [6–9]. As for biodegradability, it was reported that Zn alloys exhibited moderate degradation rate that could meet the requirements for bone repair [10–14]. What’s more, the degradation products show good biocompatibility [15]. Nevertheless, the main limitation of Zn alloys laid in their low strength that is insufficient for bone implant applications.

Reinforcing phase is a feasible way to enhance the mechanical performance of metals [16, 17]. Therefore, some researchers attempted to add reinforcing phase to Zn in the hope of enhancing its mechanical properties. For instance, Sharma et al [18] found that the zircon reinforced Zn matrix composites exhibited improved strength, modulus and hardness. Girish et al [19, 20] have studied the graphite particles reinforced Zn matrix composites and found that the hardness and strength were enhanced. However, they found it was difficult to form good interfacial bonding in metal matrix composite due to the large physical and chemical differences between the matrix and the reinforcing phase [21–24]. This will reduce the load transfer capacity of the enhancement phase [25–28]. Hence, it is imperative to get better interfacial bonding between the Zn matrix and the reinforcements to improve the mechanical properties.

Ti$_2$AlN is a promising reinforcement which has high strength (670 MPa) and hardness (408 HV) [29]. More importantly, the Al atom in Ti$_2$AlN is weakly bonded to Ti compared with the much stronger covalent Ti-N bonds [30, 31]. Consequently, the weakly bonded Al atoms might be out-diffused from the lattice of Ti$_2$AlN under certain conditions. This property motivated our inspiration that the diffused Al atoms from the Ti$_2$AlN in situ decomposition, Zn7Al-Ti$_2$AlN composite, interfacial bonding, mechanical properties, cell viability

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might form a good bond with the Al atoms inside the ZnAl alloy owing to the inherent chemical affinity between the Al atoms, and the strong interfacial bonding would be obtained. In addition, it was reported that Ti2AlN exhibited better cell proliferation and differentiation ability than Ti6Al4V alloy and pure Ti [32], which endowed it potential in biomedical applications.

In the present study, in situ decomposition of Ti2AlN was used to obtain strong interfacial bonding in Zn7Al-Ti2AlN composites fabricated by selective laser melting (SLM). The microstructures, mechanical and degradation properties as well as cytocompatibility have been studied to estimate the possibility of Zn7Al-Ti2AlN composites as biomedical materials. What’s more, the strengthening mechanism of interfacial bonding has also been discussed.

2. Materials and methods

2.1. Materials

The Zn7Al alloy powder (7 wt% Al, purity >99.9%, average particle size 70 μm, obtained from Naioiu Nano technology Co., Ltd, China) and the Ti2AlN powder (purity >99.9%, average particle size 1 μm, Laizhou Kai Kai Ceramic Materials Co., Ltd, China) were used as original materials. In order to mix the powder evenly, these two powders were ball milled in a ball grinder at a speed of 250 rpm min⁻¹ for 1 h under an argon atmosphere. Afterwards, the Zn7Al alloy and the mixed Zn7Al-Ti2AlN composites powders (denoted as Zn7Al-x%Ti2AlN, where x = 0, 5, 10, 15 wt%, respectively) were used to produce samples via SLM. Besides, the particle size distributions of the Zn7Al and representative composite powders were obtained via a laser particle size analyzer (Malvern E-3000, Malvern Panalytical Co., Ltd, Britain).

2.2. Sample preparation

A home-made SLM machine was used to fabricate Zn7Al alloy and Zn7Al-Ti2AlN composite. The concrete parameters have been mentioned in our previous study [33]. During the process, the argon atmosphere was used to prevent oxidation. The SLM process parameters used in this work were as following: the laser power 80 W, the spot diameter 150 μm, the scanning speed 15 mm s⁻¹, and the scanning line interval 120 μm.

2.3. Microstructural characterization

The phase compositions of the Zn7Al alloy and Ti2AlN ceramic powders and the SLMed Zn7Al-Ti2AlN composites were determined by an x-ray diffractometer (XRD, Germany) in a step of 8 degrees per minute and a scan angle range of 20–80 degrees. The microstructure of the Zn7Al-Ti2AlN composites were observed via a scanning electron microscopy (SEM, Phenom pure, Netherlands) equipped with an energy dispersive spectrometer (EDS). And the SEM worked in backscattered electrons (BSE) mode. In addition, a field-emission electron probe micro-analysis (FE-EPMA; JXA-8530F, JEOL Ltd, Japan) equipped with wavelength-dispersive x-ray spectrometers (WDS) was used to measure compositions in the composite. All samples were polished by 1 μm SiC abrasive paste and washed in alcohol before the microstructure was observed.

2.4. Mechanical tests

The mechanical properties of Zn7Al alloy and Zn7Al-Ti2AlN composites were evaluated by compressive and hardness tests. A universal test machine (ZLC-50M, Jinan Zhongluchang Testing Machine Manufacturing Co., Ltd, China) was used to carry out the compressive test and the compression loading rate was 0.5 mm s⁻¹. A Vickers hardness tester (Shanghai Taiming Optical Instrument Co. Ltd, China) was used to measure Vickers hardness at a loading force of 9.8 N and dwell time of 15 s. Each group was measured at least 5 times.

2.5. Electrochemical measurements

Biodegradable materials should have good degradable behavior. And the degradation of metal in human environment was in the form of electrochemical reaction. Therefore, electrochemical measurements were commonly used to study the degradation behavior of Biodegradable metal implants. The simulated body fluid (SBF) was usually used as electrolytes to simulate the degradation in physiological environment. In this study, the electrochemical measurements were executed at room temperature in the SBF. The SBF were consisted of 0.225 g l⁻¹ KCl, 0.231 g l⁻¹ K₂HPO₄·3H₂O, 0.292 g l⁻¹ CaCl₂, 0.311 g l⁻¹ MgCl₂–6H₂O, 0.354 g l⁻¹ NaHCO₃ and 8.036 g l⁻¹ NaCl [34, 35]. An electrochemical workstation (MUL TI AUTOLAB M204, Switzerland) was utilized for electrochemical tests, which equipped with a platinum sheet as counter electrode and a saturated calomel electrode (SCE) as the reference electrode. During the test, each sample with a surface area of 1 cm² served as the working electrode. Before the actual experiment, each sample was monitored for 1 h to reach a steady open-circuit potential. Afterwards, the potential-dynamic polarization experiments were carried out at a scanning rate of 3 mV s⁻¹. The corrosion potential (Ecor) and corrosion current density (Icor) were analyzed by
linear fit and Tafel extrapolation to the cathodic and anodic parts of polarization curves. The corrosion rates could be calculated according to ASTM-G102-89 [36].

2.6. Immersion tests
According to ASTM G31-72 [37], all samples were immersed in SBF at 37 °C for 21 days to measure the degradation properties. The samples were gradually polished to 3000 grits before being placed in SBF. After immersing for 21 days, the samples were taken out from SBF solution and lightly washed with distilled water followed by dried in air. The degradation morphologies of the samples were observed by SEM in BSE mode. Thereafter, the samples were washed in a 200 g l⁻¹ CrO₃ solution to remove the degradation product. After removing degradation product, the mass loss of samples was obtained. The degradation rate would be calculated according to the following equation:

\[
C = 8.76 \times 10^7 \times m / (\rho \times A \times T)
\]

Where \( C \) was the degradation rate in \( \mu m \)/year, \( m \) was the mass loss (g), \( \rho \) was the density of the composites, \( A \) (cm²) was the initial surface area of the samples and \( T \) was the immersion time (h).

2.7. Cell experiments
Biodegradable materials should have good cytocompatibility to ensure they were harmless to humans and promoted cell/tissue growth. Cell experiments were common way to assess the biosecurity of biodegradable materials. The human osteosarcoma MG-63 cells (American Type Culture Collection, USA) were adopted to evaluate the cytocompatibility of Zn7Al-Ti₂AlN composites. Before the test, all the samples were polished to 3000 grits and subjected to high temperature and high pressure to sterilize. MG-63 cells were cultured in Dulbecco’s modified eagle medium (DMEM) supplemented with 10% fetal bovine serum (FBS), 100 U ml⁻¹ penicillin and 100 mg/ml streptomycin in an incubator at 37 °C in a humidified atmosphere (5% CO₂). The cytocompatibility evaluation was conducted by indirect contact method. The extracts were prepared by separately immersing samples in DMEM cell free medium. According to ISO 10993-5:1999 [38], for each 1.25 cm² of sample, 1 ml of culture medium was used and then incubated for 72 h.

For Cell Counting Kit-8 proliferation test: firstly, the MG-63 cells were seeded in a 96-well plate with a density of 1000 cells/well, and then cultured for 24 h to allow attachment. After that, the medium was added with the above extracts. After cultured for 1 and 5 days, 10 \( \mu l \) of CCK-8 (5 mg ml⁻¹, Sigma-Aldrich, St. Louis, MO, USA) solutions were added to each well and then incubated for 2 h at 37 °C. Then the absorbance was measured at 450 nm by paradigm detection platform (BECK MAN, S. Kraemer Boulevard Brea, CA). The cells number was represented by the measured absorbance.

For LIVE and DEAD viability test: the cells were first cultured in DMEM for 4 h. After that, the cell culture media were substituted by the above extracts of Zn7Al-Ti₂AlN composites. MG-63 cells were cultured for 1 and 5 days, respectively. Afterwards, the cells were gently washed with PBS and then stained with Calcein-am and Ethidium homodimer-1 reagents for 15 min at 37 °C. After washing twice with PBS, cells were placed onto glass slides for fluorescence microscopy observation (BX60, Olympus, Japan).

2.8. Statistical analysis
The SPSS 18.0 software (SPSS Inc. Chicago, USA) was used to perform statistical analysis. One-way analysis of variance (ANOVA) was used to evaluate the statistical significance of differences among groups. It was considered statistically significant when \( P < 0.05 \).

3. Results and discussions

3.1. Characterization of original powder
The characteristic morphologies of the Zn7Al, Ti₂AlN and the mixed powders are shown in figures 1(a)–(c). It could be seen that the Zn7Al particles were spherical and Ti₂AlN particles were flake shape. The representative image of the mixed powders in figure 1(c) showed that the Zn7Al and Ti₂AlN powders were mixed evenly. As shown in figure 1(d), the size distribution revealed the normal distribution. The average particle size was measured as 70 \( \mu m \) for Zn7Al powder, 1 \( \mu m \) for Ti₂AlN and 8–70 \( \mu m \) for the mixed powder. The XRD patterns of the Zn7Al and Ti₂AlN powders were shown in figure 1(e). It could be seen that the Zn7Al pattern presented peaks corresponding to Zn and Al phases. The peaks in Ti₂AlN pattern confirmed that the Ti₂AlN was pure and didn’t decompose.
3.2. Microstructure

There were several kinds of interfaces in the metal matrix composites, such as physical bonding, wetting combination, and reaction bonding. Among them, reaction bonding is the strongest interfacial bonding because of the formation of chemical bonding at the interface [39]. In order to confirm the chemical bonding appeared in the Zn7Al-Ti2AlN composites, the line scanning analysis was performed through SEM on Zn7Al-10Ti2AlN as a representative. The line scanning analysis result was shown in figure 2(a). The red dotted line marked the position of the signal and the green solid line represented the atomic concentration of Al element. The result manifested that the Al element enriched in the junction of the reinforced phase and the matrix displayed an obvious increase. The result of phase composition obtained by XRD analysis in figure 2(b) indicated that besides the main phases of Zn enrich phase, Al enrich phase and Ti2AlN phase, two new phases of TiN and Al0.64Ti0.36 were detected in the Zn7Al-10Ti2AlN. In addition, the EDS result of the feature region in figure 2(a) was presented in figures 2(c), (d). In order to determine the element composition more accurately, the FE-EPMA equipped with WDS was applied to accurately identify the Ti2AlN phase. FE-EPMA was a kind of instrument which used high energy electron beam to bomber the solid surface and generate characteristic x-ray, and then qualitatively and quantitatively analyze the micro-region of the solid surface according to its wavelength and intensity. It could analyze the elemental composition of particles with a diameter of 1 μm. And the WDS was used to accurately quantify the elemental composition of material [40–42]. The FE-EPMA-WDS results were shown in figure 3. The atomic ratio of Ti2AlN in the reinforced phase area was approximately equal to 2:1:1. Based on the XRD result, it could be confirmed that the reinforced phase was Ti2AlN.

The appearance of the Al0.64Ti0.36 and TiN phase could be deduced that a handful of Ti2AlN in situ decomposed into TiN, Al and Al0.64Ti0.36 during the SLM progress under an effect of liquid Zn7Al matrix. Ti2AlN was a ‘211’ phase ceramic in the MAX phase ceramic. It was composed of strong covalent Ti–N weakly connected Al atom. Under certain condition, the Al atoms could diffuse easily of Ti2AlN owing to the weak combination. Thereafter, the liquid Zn7Al matrix provided a diffusion pathway and the Al atoms in Zn7Al matrix provided a strong chemical affinity for the Al atoms from Ti2AlN. As a result, the in situ decomposition of Ti2AlN was promoted and the interfacial bonding was improved. What’s more, due to atom diffusion, a part of Ti atom separated from Ti2AlN and reacted with Al to form Al0.64Ti0.36. The in situ decomposition reaction at the interface was beneficial to improve the interfacial bonding.

The surface morphologies of the Zn7Al-Ti2AlN composites were presented in figure 4. It was obvious that the Ti2AlN particles uniformly distributed in Zn7Al matrix. But when the Ti2AlN content was excessive to 15 wt%, the Ti2AlN was aggregated, which might be detrimental to mechanical properties.
3.3. Mechanical properties

The stress-stain curves of the Zn7Al-Ti2AlN composites obtained from compressive tests were shown in figure 5. The results suggested that Ti2AlN content had a significant effect on the compressive properties of the Zn7Al-Ti2AlN composites. With the increase of Ti2AlN content, the compressive stress-stain curve moved up obviously, which means that the compressive strength was increased. And the compressive modulus increased accordingly. The compressive modulus and strength of the Zn7Al-Ti2AlN composites were shown in figure 5(b), (c). EDS results of the feature region marked in figure 5(a).

Figure 2. (a) Line scanning analysis result of the atomic concentration (at%) of Al element, indicating that the Al accumulated at the interface of the matrix and Ti2AlN, (c), (d) EDS results of the feature region marked in figure 5(a).

Figure 3. FE-EMPA micrograph and WDS results (at%) of phase.

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transfer ability of Ti$_2$AlN, and the higher strength was achieved in the Zn7Al-Ti$_2$AlN composites. This means the strengthening effect of the Ti$_2$AlN reinforcement was outstanding. However, Ti$_2$AlN aggregation also appeared in the composites with an increase in content of Ti$_2$AlN. Particularly, when Ti$_2$AlN reached to 15 wt%, some Ti$_2$AlN aggregates were observed in Zn7Al-Ti$_2$AlN (figure 4(c)) and a decrease of modulus and strength of Zn7Al-Ti$_2$AlN occurred. The hardness of Zn7Al-Ti$_2$AlN composite was shown in figure 5(d). It was obvious that the hardness enhanced significantly as the Ti$_2$AlN content increased. This was because the hardness of Ti$_2$AlN (408 HV) was much higher than that of Zn7Al (74 HV).

The fracture surfaces of Zn7Al alloy and Zn7Al-10Ti$_2$AlN composite were shown in figures 6(a), (b). The fracture surface of Zn7Al-10Ti$_2$AlN composite was composed of two distinct phases. One was Ti$_2$AlN particle and the other was Zn7Al matrix. It was seen that the Ti$_2$AlN particle was broken and in tight compaction by Zn7Al matrix, which demonstrated that a strong interfacial bonding between Zn7Al matrix and Ti$_2$AlN reinforcement caused them to fracture together. The fracture mechanism ahead of the crack tips in the Zn7Al-Ti$_2$AlN was shown in figure 6(c). In the process of compression, crack was generated in the matrix, and with the further increase of the experimental force, the crack continued to expand in the matrix. As shown in figure 6(c), the crack proceeded linearly and smoothly in the Zn7Al matrix before striking the Ti$_2$AlN particles.
and then continued by winding around or passing through them via the Ti$_2$AlN/matrix interface. Due to the strong interfacial bonding and high strength of Ti$_2$AlN, crack propagation was effectively blocked. Thereby, the mechanical properties of Zn$_7$Al-Ti$_2$AlN composites were improved.

3.4. Degradation behavior
The results acquired from the electrochemical experiments with respect to Tafel polarization curves, corrosion potential ($E_{corr}$) and current density ($I_{corr}$) were shown in figures 7(a), (b). It was noticeable that the $I_{corr}$ of composites increased significantly in contrast to Zn$_7$Al alloy. The $I_{corr}$ of Zn$_7$Al-10Ti$_2$AlN was almost 3 times as that of Zn$_7$Al alloy ($7.79 \mu$A cm$^{-2}$ versus 2.76 $\mu$A cm$^{-2}$), which might be owing to the presence of galvanic micro-cells between the matrix and Ti$_2$AlN. As to the $E_{corr}$, it decreased with the addition of Ti$_2$AlN as well. The corrosion rates calculated by the $I_{corr}$ were shown in figure 7(c). Zn$_7$Al alloy showed the lowest corrosion rate, followed by Zn$_7$Al-5Ti$_2$AlN, Zn$_7$Al-15Ti$_2$AlN and Zn$_7$Al-10Ti$_2$AlN composites. The degradation rate of the Zn$_7$Al alloy and Zn$_7$Al-Ti$_2$AlN composites for 21 days was presented in figure 7(d). The sequence of degradation rates after immersion in SBF solution for 21 days was:

Zn$_7$Al-15Ti$_2$AlN > Zn$_7$Al-10Ti$_2$AlN > Zn$_7$Al-5Ti$_2$AlN > Zn$_7$Al. A possible reason was that the galvanic microcells accelerated corrosion of the matrix.

The surface morphologies and elements composition of Zn$_7$Al alloy and Zn$_7$Al-Ti$_2$AlN composites immersed in SBF solution for 21 d were shown in figure 8. Some white clusters/particles deposition could be observed, which formed degradation products precipitated on sample surface. In several areas accumulation of massive degradation products was apparent, but other regions were present where little amount of degradation products were found. All the degradation surfaces did not appear any severe local corrosion pits. The high multiple corrosion morphology was shown in the figures 8(e)–(h). In the surface of the Zn$_7$Al alloy (figure 8(c)), there were almost all particle degradation products. When addition of Ti$_2$AlN, cluster corrosion products appeared and with the Ti$_2$AlN content increasing, more degradation products formed. This was because the galvanic micro-cells between the matrix and Ti$_2$AlN accelerated degradation rate. EDS analyses of degradation products of Zn$_7$Al alloy and Zn$_7$Al-Ti$_2$AlN composites were listed in figures 8(i)–(l). According to the EDS results, the degradation products were mainly composed of Zn, Al, Ti, Ca, P, Cl and O. What’s more, with the Ti$_2$AlN content increasing, more Ca elements were detected. The reason was that the Ti$_2$AlN had a stronger affinity to the Ca ions, which was confirmed by the previous study.

3.5. Cytocompatibility
The cell viability evaluation results with extracts of Zn$_7$Al alloy and Zn$_7$Al-Ti$_2$AlN composites were shown in figures 9(a), (b). The Cell viability larger than 75% could be regarded as acceptable cytocompatibility according to ISO standards Part 5. It was clearly that all the cell viability met the cytocompatibility standards and the cell
viability was improved with the addition of Ti$_2$AlN. The fluorescence photographs of the cells cultured in the extracts of Zn$_7$Al and Zn$_7$Al–Ti$_2$AlN composites for 1 d and 5 d were shown in figure 9(c). The MG-63 cells showed benign feedback to Zn$_7$Al alloy and Zn$_7$Al–Ti$_2$AlN composites and had good spreading morphologies. What’s more, with the Ti$_2$AlN content increasing, the number of cells gradually increased. According to the previous study, the strong affinity between the Ca$^{2+}$ and Ti$_2$AlN promoted cell vitality.

Figure 7. Electrochemical measurement results of Zn$_7$Al and Zn$_7$Al–Ti$_2$AlN composites. (a) Tafel polarization curves, (b) $E_{corr}$ and $I_{corr}$, (c) corrosion rates calculated by the corrosion current density, (d) degradation rates for 21 days.

Figure 8. Surface morphologies of Zn$_7$Al alloy and Zn$_7$Al–Ti$_2$AlN composites immersed for 21 days. (a)–(d) low magnification of the surface morphologies, (e), (f) high magnification of the surface morphologies, (i)–(l) EDS results marked in (e)–(h).
4. Conclusions

In present study, in situ decomposition of Ti$_2$AlN was used to promote the interfacial bonding in the Zn7Al-Ti$_2$AlN composites prepared via laser melting. The results showed that a strong interfacial bonding was obtained according to the reaction equation: Ti$_2$AlN $\rightarrow$ Al$_{0.64}$Ti$_{0.36}$ + TiN + Al at the interface, which contributed to improved load transfer of Ti$_2$AlN. The compressive strength and hardness of Zn7Al-Ti$_2$AlN composite were enhanced from 155 MPa and 74 HV to 205 MPa and 80 HV, respectively, when the Ti$_2$AlN content was 10 wt%. Its degradation rate increased to 0.14 mm y$^{-1}$ in SBF solution, which was close to bone grow rate of 0.5 mm y$^{-1}$. Cytocompatibility evaluation showed that the Zn7Al-Ti$_2$AlN composites presented good cytocompatibility. In consideration of the high compressive strength, and acceptable biocompatibility, the Zn7Al–10Ti$_2$AlN composite could be a suitable material for biomedical application.

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