Microstructure and characteristics of the metal–ceramic composite (MgCa-HA/TCP) fabricated by liquid metal infiltration

X. N. Gu,1 X. Wang,2 N. Li,1 L. Li,2 Y. F. Zheng,1,2 Xigeng Miao3

1State Key Laboratory for Turbulence and Complex System and Department of Advanced Materials and Nanotechnology, College of Engineering, Peking University, Beijing 100871, China
2Center for Biomedical Materials and Engineering, Harbin Engineering University, Harbin 150001, China
3Institute of Health and Biomedical Innovation, Queensland University of Technology, Kelvin Grove, QLD 4059, Australia

Received 23 December 2010; revised 13 April 2011; accepted 20 April 2011
Published online 23 August 2011 in Wiley Online Library (wileyonlinelibrary.com). DOI: 10.1002/jbm.b.31879

Abstract: In this article, a novel MgCa alloy-hydroxyapatite-tricalcium phosphate (HA/TCP) composite was fabricated using the liquid alloy infiltration technique. The feasibility of the composite for biomedical applications was studied through mechanical testing, electrochemical testing, immersion testing, and cell culture evaluation. It was shown that the composite had a strength about 200-fold higher than that of the original porous HA/TCP scaffold but retained half of the strength of the bulk MgCa alloy. The corrosion test indicated that the resulting composite exhibited an average corrosion rate of 0.029 mL cm⁻² h⁻¹ in the Hank’s solution at 37°C, which was slower than that of the bulk MgCa alloy alone. The indirect cytotoxicity evaluation revealed that 100% concentrated (i.e., undiluted or as-collected) extract of the MgCa-HA/TCP composite showed significant toxicity to L-929 and MG63 cells (p < 0.05). In contrast, the diluted extracts with 50 and 10% concentrations of the MgCa-HA/TCP composite exhibited a similar degree of cell viability (p > 0.05), equivalent to the grade I cytotoxicity of the standard ISO 10993-5: 1999.

Key Words: MgCa-HA/TCP composite, scaffold, mechanical property, corrosion, cytotoxicity


INTRODUCTION
Magnesium alloys are attractive candidates for degradable implant applications due to their good mechanical properties and biocompatibility, in comparison with biodegradable polymers.1–5 However, their fast degradation/corrosion rates challenge the practical use of magnesium alloys due to the mismatch of the bone healing rate and the gas cavity generated around the implant during its degradation.9,10 Magnesium alloys also do not have the bone-bonding ability shown in calcium phosphate bioceramics.

Recently, metal matrix composites (MMC), consisting of an adequate ceramic reinforcement phase, are found to be effective for simultaneously increasing the strength, slowing down the corrosion rate, and improving the bioactivity of magnesium alloys.6–8 Several processing techniques, including powder metallurgy (P/M),6,7 the traditional casting method,8 and so forth, have been developed. The MMCs made of magnesium alloy AZ91D as a matrix and 20 wt % hydroxyapatite (HA) particles as a reinforcement through a P/M route, exhibited higher strength, more stable corrosion rate and improved cytocompatibility than the AZ91D matrix.6 According to Ye et al.,8 the introduction of the 1 wt. % gelatin coated nano-sized HA particles into the Mg-2.9Zn-0.7Zr alloy under magnetic agitation also indicated more favorable corrosion resistance and cytocompatibility than the Mg-Zn-Zr matrix alloy.

Among the techniques currently available for the processing of MMCs, infiltration of a molten metal into a porous ceramic preform represents a technique more suitable for achieving a high volume fraction of reinforcements (>50%) in MMCs.9–12 First, this process shows its superiority in the extreme uniform distribution of the ceramic reinforcements, eliminating residual porosities and interfacial reactions between the reinforcements and the matrix.9,10 Second, this process also shows the advantageous ability of fabricating the final, or near-final shaped composite components, minimizing the generally difficult machining of MMCs.11 Third, when suitable barriers are used to prevent the infiltration of a molten metal, it is also possible to produce selectively reinforced components, within which the metal is reinforced only where needed.9,11

This technique inspires us to adopt ceramic scaffolds for tissue engineering as the porous ceramic preforms for metal...
infiltration because of their excellent bio-compatibility and mature preparation technologies. In addition, the resulting properties of these composites can be adjusted by free selection of different porous scaffolds as well as the molten metals. The research reported here was a preliminary study about the composite fabricated by molten metal infiltration technique, adopting the HA/TCP scaffolds as the ceramic preform and the Mg-1Ca alloy as the metal phase, both of which showed good biocompatibility in our previous studies. The microstructure, mechanical properties, and in vitro corrosion behavior in simulated body fluid were investigated in this study. The cell response to the composite extract was also evaluated using the indirect cytotoxicity assay.

MATERIALS AND METHODS
Materials preparation
The Mg-1wt%Ca alloy (expressed as MgCa alloy in the following) was casted by using commercial pure Mg (99.98%) and Ca (99.95%) in a crucible under a mixed gas atmosphere of SF₆ and CO₂. To prepare the porous HA/TCP platforms, polyurethane (PU) foams were dipped into the ceramic slurry (a mixture of 160 g HA and 40 g β-TCP) and gently squeezed to allow the slurry penetrating the foams before drying at room temperature for at least 24 h. The ceramic slurry-coated PU foams were sintered in a furnace and the final HA/TCP scaffolds were removed from the furnace after it had cooled down. The average pore size of the scaffold was 500 μm and the porosity was 87%. The specific fabrication route of the HA/TCP scaffold was described in our previous work.

MgCa-HA/TCP composites were fabricated by infiltrating the molten MgCa alloy into the porous scaffold using a vacuum suction casting machine. The porous ceramic was preheated at 150°C and positioned in the vacuum suction casting machine, which was pre-pumped to 0.02–0.03 MPa. The matrix MgCa alloy was re-melted in the furnace under a mixed gas atmosphere of SF₆ and CO₂ and the temperature was kept at 700–720°C. Then the infiltration of the molten MgCa alloy was driven by the vacuum and held for 2 min while the melt was solidified. For the corrosion and extracting tests, the composite samples (with the size of 10 × 10 × 2 mm³) were mechanically polished up to 2,000 grit, ultrasonically cleaned in acetone, absolute ethanol and distilled water and then dried in air.

Microstructural characterization
The microstructure of the composite was observed by an optical microscope (Olympus BX51 M) and an environmental scanning electron microscope (ESEM, Quanta 200FEG), equipped with an energy-dispersive spectroscopy (EDS) attachment. An X-ray diffractometer (XRD, Rigaku DMAX 2400) was used to characterize the phases of the composite using the Cu Kα radiation at the step size of 0.02° with a scanning speed of 4° min⁻¹.

Mechanical testing
Uniaxial compression testing was conducted on an Instron 8562 testing machine at a constant nominal strain rate of 2 × 10⁻⁴ s⁻¹ at room temperature. The test samples with the size of 3 × 3 × 6 mm³ were prepared according to the ASTM-E9-09. Five identical samples were used for the compressive tests.

Electrochemical test
A conventional three electrode cell with the composite sample (the exposed area of 1 cm²) as a working electrode, the saturated calomel electrode (SCE) as a reference electrode, and a platinum electrode as the counter electrode was used. The electrochemical tests were carried out in the Hank’s solution (NaCl 8.00 g L⁻¹, KCl 0.40 g L⁻¹, CaCl₂ 0.14 g L⁻¹, NaHCO₃ 0.35 g L⁻¹, MgSO₄·7H₂O 0.20 g L⁻¹, Na₂HPO₄·12H₂O 0.12 g L⁻¹, KH₂PO₄ 0.06 g L⁻¹) at 37°C ± 0.5°C using an electrochemical workstation (CHI660C). The open circuit potential (EOC) was measured as a function of time. The potentiodynamic polarization test was measured from 300 mV below the OCP value at a scanning rate of 1 mV s⁻¹. The electrochemical impedance spectroscopy (EIS) measurements were carried out from 100 mHz to 100 kHz at OCP values.

Immersion test
The immersion test was carried out in Hank’s solution at 37±0.5°C according to ASTM-G31-72. After 1, 10, and 20 days’ immersion, the samples were removed out of the solution, rinsed with distilled water, and dried in air. The change of pH value of the Hank’s solution as well as the amount of hydrogen generated from the samples was monitored using a calibrated burette according to the method described in Ref. throughout the 20 days’ immersion. An average of three measurements was taken for each group.

Indirect cytotoxicity evaluation
The experimental composite samples were sterilized by ultraviolet-radiation for at least 2 h. The samples were then immersed in DMEM in a humidified atmosphere with 5% CO₂ at 37°C, with the surface area/extraction medium ratio of 3 cm² mL⁻¹. After 72 h of incubation, the supernatant fluid was withdrawn and centrifuged, and then was serially diluted to 50 and 10% concentrations. On the other hand, Murine fibroblast (L-929) cells and Human osteosarcoma cells (MG63) were cultured in the Dulbecco’s modified Eagle’s medium (DMEM), with 10% fetal bovine serum (FBS), 100 U mL⁻¹ penicillin and 100 μg mL⁻¹ streptomycin at 37°C in a humidified atmosphere of 5% CO₂. The cells were incubated in 96-well cell culture plates at 3 × 10⁴ cells/100 μL of medium in each well for 24 h to allow cell attachment. In order to evaluate the cytotoxicity of the extracts from the composite samples, the medium in the wells was then replaced with 100 μL of 100, 50, and 10% extracts and incubated for 1, 3, and 5 days. For spectrophotometric measurements, 10 μL MTT was added to each well for 4 h of incubation and then 100 μL formazan

MICROSTRUCTURE AND CHARACTERISTICS OF THE METAL–CERAMIC COMPOSITE
solubilization solution was added to each well for overnight. The measurements were carried out at 570 nm with a reference wavelength of 630 nm by a microplate reader (Bio-RAD680). The background of MTT results cultured in extracts without cells was subtracted. Meanwhile, the Mg concentration of the extracts was measured by inductively coupled plasma atomic emission spectrometry (Leeman, Profile ICP-AES). The pH value of the extracts was also measured. Statistical analysis was conducted to evaluate the differences in cell viability by the analysis of variance (ANOVA). The statistical significance was defined as 0.05.

RESULTS

Microstructure and mechanical property

The infiltration of the molten MgCa alloy into the HA/TCP scaffold, with the original total porosity 87%, was successfully achieved and a typical optical micrograph of the resulting composite was shown in Figure 1(a). The magnified SEM image and the EDS line-scan of the interface between the HA/TCP scaffold and the MgCa alloy matrix [as shown in Figure 1(b)] indicated sharp transitions of the element intensity profiles. Figure 2 presents the XRD analysis of the MgCa-HA/TCP composite. The XRD pattern exhibits only the reflection peaks of the α-Mg, Mg₂Ca, HA, and TCP phases.

Figure 3 showed the representative compression stress-strain curves of the MgCa-HA/TCP composites. The average compressive strength and the elongation for the resulting composites were (128.7 ± 15.1) MPa and (13.5 ± 0.7)%, which were approximately half of those properties of the MgCa alloy but about 200-fold higher than those of the original porous HA/TCP scaffolds (inset in Figure 3).

Electrochemical measurements

Figure 4 showed the potentiodynamic polarization curves and the Nyquist plots obtained for the MgCa-HA/TCP composite in Hank’s solution at 37°C. The experimental composite clearly indicated much more positive corrosion potential value (-1.49 V), as well as the decreased kinetics of anodic and cathodic reactions, than the bulk MgCa matrix alloy (-1.88 V). The corrosion current density of the experimental composite was much lower (15.23 μA cm⁻²) than the bulk MgCa matrix alloy (178.58 μA cm⁻²), indicating the better corrosion resistance.
of the experimental composite. From Figure 4(b), the EIS spectrum of the composite and the matrix alloy showed one semi-circle indicating an electrochemical process dominated by the mechanism of charge transfer across the interface involved in the dissolution of the MgCa alloy substrate. The polarization resistance of the samples can be obtained from the diameter of the near semi-circular spectra which was related to the corrosion rate. The MgCa-HA/TCP composite exhibited a higher polarization resistance ($\sim 1200$ $\text{cm}^2$) than the bulk MgCa matrix alloy ($\sim 400$ $\Omega$ $\text{cm}^2$).

**Immersion tests**

Figure 5(a) showed the hydrogen evolution over the immersion period for the MgCa-HA/TCP composite and the bulk MgCa matrix alloy in the Hank’s solution at $37^\circ\text{C}$. The MgCa-HA/TCP composite revealed an amount of hydrogen within the same immersion period that was less than one-third of that of the bulk MgCa matrix alloy. In contrast, the pH value of the Hank’s solution incubating the MgCa-HA/TCP composite and the bulk MgCa matrix alloy revealed a similar changing trend, as shown in Figure 5(b).

Figure 6 illustrated the morphologies of the MgCa-HA/TCP composite after 1, 10, and 20 days’ immersion in Hank’s solution at $37^\circ\text{C}$. The composite exhibited a flat and smooth surface with the scaffold structure being visible after 1 day immersion and some shallow pits could be seen after removing the corrosion product on the sample surface [Figure 6(a,b)]. After 10 days of immersion, a corrosion product layer was clearly observed on the surface of the MgCa-HA/TCP composite without any sign of the scaffold structure [Figure 6(c)]. However, the scaffold structure and the polishing scratches on the surface of the infiltrated MgCa alloy could be seen after removing the corrosion product [Figure 6(d)]. The corrosion product layer became compact after 20 days of immersion with some nano-sized holes on the surface [inset in Figure 6(e)]. Some deep corrosion pits were observed on the surface of the infiltrated MgCa alloy, revealing the dissolution of the MgCa alloy and the scaffold structure was still clear after cleaning the corrosion product [Figure 6(f)]. The EDS results of the composite before and after the CrO3 cleaning were shown in Figure 7. It indicated that the corrosion product contained C, O, Mg, P,
and Ca elements [Figure 7(a)]. After removing the corrosion product, the composition of the MgCa-HA/TCP composite [Figure 7(b)] was similar to the uncorroded one. The much higher contents of the elements O, P and Ca on the surface of the MgCa-HA/TCP composite immersed for 20 days [Figure 7(a)] suggested the deposition of a compound containing Ca and P elements. XRD results revealed that the corrosion products were mainly Mg(OH)\(_2\) and HA (as shown in Figure 8). In addition, the peak from the \(\alpha\)-Mg was still high indicating the good corrosion resistance of the infiltrated MgCa matrix alloy.

**Cytotoxicity**

The pH value of the 100% MgCa-HA/TCP composite extract was 8.87 ± 0.34 and the Mg ion concentration was 10.06 ± 0.69 mM. Figure 9 showed the cytotoxicity results of the different concentrated extracts of the MgCa-HA/TCP composite using the L-929 and MG63 cells. It was clearly seen that the 100% composite extract indicated the Grade II cytotoxicity to L-929 and MG63 cells. After 50 and 10% degrees of dilution, the cell viability was significantly improved (\(p < 0.05\)) with Grade I toxicity to L-929 and MG63 cells.

**DISCUSSION**

The MgCa-HA/TCP composite shows an extreme improvement in the strength and the elongation in comparison with the original porous HA/TCP ceramic scaffold.\(^{17}\) According to the micro-mechanical model of Gibson and Ashby, the crushing of ceramic foams is based on bending of the struts.\(^{19}\) The interpenetration of the MgCa alloy phase stabilizes the ceramic struts and partially prevents strut bending, resulting

![Figure 6](image-url)

**FIGURE 6.** Surface morphologies of the MgCa-HA/TCP alloy composite after (a)–(b) 1 day, (c)–(d) 10 days, and (e)–(f) 20 days of immersion in the Hank’s solution before [(a), (c), and (e)] and after [(b), (d), and (f)] the CrO\(_3\) cleaning.

![Figure 7](image-url)

**FIGURE 7.** EDS results of the MgCa-HA/TCP alloy composite for the 20-day immersion in the Hank’s solution (a) before and (b) after the CrO\(_3\) cleaning. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]
in a major improvement of the mechanical properties of the ceramic scaffold. However, the resulting MgCa-HA/TCP composite indicates inferior values of the mechanical property compared to the bulk MgCa alloy probably due to the poor interfacial bonding between the HA/TCP scaffold and the alloy matrix. Load transfer from the metal matrix to the ceramic skeleton is, therefore, supposed to be reduced and the improved bonding is needed to increase the stiffness and delay the onset of ceramic/metal de-cohesion. Zeschky et al. also reported the reduced mechanical property of AZ91 infiltrated SiOAC ceramic foams compared to the AZ91 bulk alloy due to the crack opening at the ceramic/metal interface, which can be improved by further oxidation. Additionally, the pore size and the porosity of the scaffold also influences the resulting mechanical property of the composite since the debonding depends on the length of the dislocation pile up at the interface, and, therefore, on the pore size of scaffold.

In general, the MgCa-HA/TCP composite exhibits a slower corrosion rate than the matrix MgCa alloy. The formation of the protective layer on the sample surface is responsible for the improved corrosion resistance of the composite. The HA/TCP scaffold in the composite acts as the hydroxyapatite nuclei and the hydroxyapatite will grow spontaneously consuming the Ca2+ and PO43-. The multiple protection effects offered by corrosion product Mg(OH)2 and hydroxyapatite may be the reason for the slower corrosion rate observed for the composite. Witte et al. and Ye et al. also reported the protective layers, composed of multiple corrosion products, on the AZ91D/HA and Mg-Zn-Zr/HA composites, exhibited better corrosion resistance than the single Mg(OH)2 layer. However, severe local corrosion seems to be irritated by the ceramic scaffold for the composite. The ceramic/metal interface has the capability of forming a galvanic couple in the electrolyte and the electrically conducting scaffold becomes the noble member. The corrosion initiated along the interface allowing the electrolyte deep penetration. As the corrosion proceeds, the MgCa alloy matrix dissolves gradually with an intact HA/TCP scaffold structure left, as shown in Figure 6(d,f). The
and eventually becomes specific tissue. From the above extracts indicate similar L-929 and MG63 cell viability (adjusting the pH value of the extracts. 28 Given the body aration techniques.

compatibility can be expected with the combined advan-

ious Mg material extracts, such as pure Mg,27 Mg-Nd-Zn-

Zr28 and Mg-Zn-Y,29 and the cell viability increased after

fluid exchange in vivo. Furthermore, as the cor-

rosion of the MgCa-HA/TCP composite proceeds, good bio-

compatibility can be expected with the combined advan-

tages of the HA/TCP scaffold and the MgCa matrix alloy. On

the one hand, the dissolving magnesium ion will activate

bone cells1,2 and thus accelerate the bone restoration pro-

cess. On the other hand, as the MgCa alloy matrix dissolves, the

HA/TCP scaffold structure will be left which shows good osteoconductivity17 and allows the invasion of cells and eventually becomes specific tissue. From the above facts, the infiltration method shows promising potential in preparing Mg alloy based composite for orthopedic application.

CONCLUSIONS

The MgCa-HA/TCP composite was fabricated by the molten metal infiltration technique for orthopedic applications. Compared to the single MgCa alloy matrix, the MgCa-HA/ TCP composite exhibited the inferior mechanical property (decreased by 50%) but with superior corrosion resistance (improved by 68%). The average corrosion rate of the resulted composite was 0.029 mL cm⁻² h⁻¹ after the immersion in Hank’s solution for 20 days. The immersion test indicated that the MgCa alloy matrix gradually dissolved and induced the apatite deposition. The bioactive HA/TCP scaffold was left behind due to the much slower degradation rate of the ceramic scaffold. In vitro cytotoxicity tests of the composite revealed that the cell viability was related to the extract concentration, with Grade 1 cytotoxicity for 50 and 10% concentrated extracts.

REFERENCES


9. Peng LM, Cao JW, Noda K, Han KS. Mechanical properties of ce-

ramic-metal composites by pressure infiltration of metal into po-


10. Mattern A, Huchler B, Staudenecker D, Oberacker R, Nagel A, Hoffmann MJ. Preparation of interpenetrating ceramic-metal com-


12. Zeschky J, Lo J, Höfler T, Greipl P. Mg alloy infiltrated Si-C cer-


15. Hutmacher DW. Scaffolds in tissue engineering bone and carti-


16. Gu T-MG, Orton DG, Hollister SJ, Feinberg SE, Halloran JW. The me-


18. Song G, Atrens A. Understanding magnesium corrosion. A frame-


20. Contreras A, López VH, Bedolla E. Mg/TiC composites manufac-


24. Ferrando WA. Review of corrosion and corrosion control of mag-


25. Feng A, Han Y. The microstructure, mechanical and corrosion properties of calcium polyphosphate reinforced ZK60A magne-