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Corrosion behavior, biocompatibility and biomechanical stability of a prototype magnesium-based biodegradable intramedullary nailing system

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ABSTRACT

Implants made of degradable magnesium alloys are a potential alternative to conventional orthopaedic implant materials, e.g. stainless steel or titanium. Intramedullary nails made of the magnesium alloy LAE442 were subjected to cyclic fatigue tests in both distilled water and Hank's Balanced Salt Solution (HBSS) at 37.5 °C until implant failure or a limit of 500,000 cycles was reached. In distilled water, four of the five nails were still intact after the end of the biomechanical test. In HBSS, a breakage within the first 70,000 bending cycles was observed. Additionally, the degradation rate of this alloy was determined in HBSS according to the weight loss method (0.24 ± 0.12 mm year^{-1}) and based on gas release (0.21 ± 0.03 mm year^{-1}) with a standard eudiometer. A cytotoxicity test with L929 cells was carried out in accordance with EN ISO 10993-5/12. This test demonstrated sufficient cell viability of the diluted extracts (50%, 25% and 12.5%). The relative metabolic activity of the 100% extract was reduced slightly below 70%, which is classified as a threshold value for cytotoxicity. In conclusion, this *in vitro* study indicates that intramedullary nails made of LAE442 may not have the required fatigue resistance for load-bearing applications and the development of a corrosion-protective coating may be necessary to prevent early failure of the implant.

1. Introduction

Orthopaedic devices, such as intramedullary nails, are commonly used for diaphyseal fracture fixation of long bones. Commercially available intramedullary nail devices were successfully introduced by Küntscher in 1939 [1]. Since that time, these implants have been amongst the most successful implants for fracture treatment. Ordinarily, these devices are made of stainless steel or titanium. Transversely disposed holes near the proximal and distal ends of the nails enable the nail to be screwed to the bone. Despite continuous enhancements of these implants to meet patients' needs, the nails have to be removed in a second operation after successful healing of the fracture, typically within two years of the first operation. In addition to causing extra stress to the patient, these repeat operations increase the danger of bone refracture or morbidity, as well as the risk of infection, and add a further burden to the healthcare system. Therefore, biodegradable materials, such as magnesium or polylactic acids, have recently been intensively investigated as an alternative for medical applications to avoid follow-up surgery [2]. Of all these biodegradable materials, magnesium alloys possess the biggest strength and offer mechanical properties closest to those of natural bone. With an elastic modulus of 41–46 GPa, magnesium is more than 2.6 and 4.6 times less stiff than titanium and steel, respectively, and may therewith counter stress-shielding problems during fracture treatments with steel and titanium implants [2,3]. Thus, magnesium alloys seem to be promising bioabsorbable materials as substitutes for steel or titanium in high-load implant applications, such as intramedullary fracture fixation. Several studies have investigated the behaviour of different magnesium alloys, *in vitro* and *in vivo*, to obtain a better understanding of this material and its biocompatibility, corrosion characteristics, degradation rates or cytotoxicity [4–8]. Referring to this, the magnesium alloy LAE442 used in the present study showed an acceptable host response as well as an applicable slow corrosion rate in an *in vivo* rabbit model [8]. Furthermore, little morphological change in efferent lymph nodes could be observed, combined with an absence of critical bone response in the bone after implantation of

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LAE422 rods in rabbits could be observed [9,10]. However, for orthopaedic applications with load-bearing function, mechanical capability under cyclic loading of corrosive, magnesium-based implants is also a major issue in assessing their applicability. In the past few years, the corrosion fatigue behaviour of magnesium-based alloys has mainly been studied for engineering applications [11–14]. Until now, only very few publications describe the fatigue behaviour of magnesium alloys or magnesium-based implants in simulated body fluid. Increased corrosion rates of two different magnesium alloys (AZ91D and WE43) were reported under cyclic tensile loading, compared with a static immersion test. Furthermore, these studies recorded a much lower fatigue strength of AZ91D probes in simulated body fluid than in air [15]. The tensile corrosion fatigue behaviour of AZ91D in simulated body fluid was also investigated. Additionally, the role of electrochemical polarization on corrosion fatigue cracking was studied [16]. It was concluded that pits were the initiation sites for corrosion fatigue and that nucleation as well as growth of pits was influenced by electrochemical polarization. Furthermore, the study recorded a significantly lower fatigue life for probes tested in simulated body fluid than for those tested in air [16]. However, most existing studies investigated fatigue behaviour solely under tensile loading using probes with simplified geometry which does not represent the *in vivo* loading conditions of most orthopaedic implants such as intramedullary nails. Those are predominantly loaded in bending, torsion and compression [17]. No studies are yet available investigating the performance of magnesium-based implants considering an environment similar to body fluids and loading conditions which are comparable to those acting in the human body. Furthermore, no study has yet described the cytotoxicity of LAE442 to evaluate the biocompatibility of the material, consequently, (i) the performance of intramedullary nails machined from a prototype magnesium alloy (LAE442) were investigated by evaluating the corrosion fatigue of LAE442 nails under cyclic bending in simulated body fluid; (ii) the corrosion rate was determined using the mass loss method and the gas evolution method with a standard eudiometer; and (iii) the *in vitro* cytotoxicity of LAE442 extracts according to EN ISO 10993-5/12 was identified.

2. Materials and methods

2.1. Materials and preparation

The magnesium alloy LAE442 (with a nominal composition of 90 wt.% magnesium, 4 wt.% lithium, 4 wt.% aluminium and 2 wt.% rare earth metals) was produced using pure magnesium (Magnesium Electron Limited, Manchester, UK, 99.94%) by gravity die casting and subsequent hot extrusion. The extruded material was analyzed by inductively coupled plasma atomic emission spectroscopy (ICP-OES; Spectro Ciros Vision EOP, SPECTRO Analytical Instruments GmbH, Kleve, Germany). The final magnesium alloy material consisted of 3.7 wt.% lithium, 3.62 wt.% aluminium, 0.73 wt.% cerium, 0.38 wt.% lanthanum, 0.16 wt.% neodymium, and 0.03 wt.% praseodymium. The alloy's microstructure was homogeneous and did not show any signs of a distinct texture. The average grain size was 16.59 ± 1.87 μm. The mechanical properties of the magnesium alloy LAE442 were determined in previous investigations according to ISO 6892–1:2009. The tests were performed with the universal testing machine Model Z 250 (Zwick GmbH & Co. KG, Ulm, Germany). The alloy exhibited a yield strength of 140 MPa, tensile strength of 239.3 MPa, and an elongation to failure of 15.4%.

Ten intramedullary nails, and six cylindrical and fourteen cuboidal samples were manufactured from the extruded bar stock (10 mm) on a computer numerically controlled (CNC) lathe and a CNC milling machine, respectively. Each intramedullary nail had a total length of 130 mm and a shaft diameter of 9 mm. The end was equipped with an M5 thread, allowing the use of a target device, and the front part was tapered off. Two screw holes with a diameter of 4 mm were drilled perpendicularly to each other on the proximal as well as the distal end of the nail (Fig. 1). Cylindrical samples were used to determine the corrosion rate and had a length of 10 mm and a diameter of 8 mm. Cuboidal blocks (5 × 4 × 4 mm) with a surface area of 1.12 cm^2 were used to evaluate *in vitro* cytotoxicity.

The surfaces of the magnesium samples were pickled prior to testing with a solution of 5 ml 65% nitric acid, 20 ml 85% glycerol and 5 ml 100% acetic acid (all sourced from Merck, Darmstadt, Germany) for 30 s. In the next step, the samples were ultrasonically cleaned for 2 min with isopropanol. Subsequently, the samples were dried in a vacuum drying oven (Heraeus Vacuum oven, Thermo Scientific, Bonn, Germany) at 50 °C for about 20 min. The samples for the cytotoxicity test of the extracts were gamma sterilized by 25 kGy of ^{60}Co gamma radiation (BBF, Sterilisationsservice, Kernen, Germany) prior to testing.

2.2. Immersion test

The immersion test was carried out in Hanks' Balanced Salt Solution (HBSS; Biochrom, Berlin, Germany) to simulate normal ion concentrations under physiological conditions. The corrosion media contained 8.00 g l^{-1} NaCl, 0.4 g l^{-1} KCl, 0.04 g l^{-1} Na_2HPO_4, 0.06 g l^{-1} KH_2PO_4, 0.20 g l^{-1} $MgSO_4x7H_2O$, 0.14 g l^{-1} $CaCl_2$, 1.00 g l^{-1} glucose and 0.35 g l^{-1} $NaHCO_3$. Cylindrical samples were used to determine the corrosion rate. A three-fold determination was performed, and the samples were suspended from a non-resorbable wire (Ethicon Prolene, 4–0, Johnson & Johnson, New Brunswick, NJ, USA) into 500 ml HBSS at 37.5 °C for 14 days. The pH of the solution was monitored during the immersion test using a pH meter (Schott Instruments Lab850, SI Analytics, Mainz, Germany). After immersion, the samples were dried in a vacuum drying oven. The corrosion layer was removed with 20% chromic acid, and the degradation rate was evaluated according to the weight loss method by the use of a precision weighing balance (ALC-110.4, Acculab, Göttingen, Germany) [18]:

$$\text{rate} = \Delta W \frac{W_{\text{before}} - W_{\text{after}}}{At},$$

where ΔW is the weight loss, W_{before} (mg) is the weight of the sample before corrosion, W_{after} (mg) is the weight after corrosion, A is the sample surface area (cm^2) and t is the immersion time (days). This weight loss rate was converted into an average corrosion rate P_W (mm $year^{-1}$) using the formula:

$$P_W = 3.65\Delta W/\rho,$$

where ρ is the density of magnesium (1.74 g cm^{-3}). This results in the following equation [18,19]:

$$P_W = 2.1\Delta W.$$

Fig. 1. Intramedullary nail made of magnesium-based alloy LAE442. This test implant had a length of 130 mm and two holes for intramedullary screw fixation at the proximal and distal ends.