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Surface Modification of Ti-Nb-Zr Foams by Poly(3-Hydroxybutyrate)

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Abstract In this study, Ti-Nb-Zr superelastic foams were produced, characterized from the standpoint of their morphology and mechanical properties. To improve biocompatibility of these foams, they were subjected to surface modification by Poly(3-Hydroxybutyrate). The two-stage immersion of the Ti-Nb-Zr foams in the PHB-containing solution allows forming on their surface continuous polymer layers with incorporation of 6.4 % (w/w) of PHB.

Introduction

Over the past decades, intensive research has been carried out in the field of binary and multicomponent nickel-free superelastic titanium alloys, in particular of Ti-Nb, Ti-Nb-Zr alloy systems, as perspective materials for bone replacement [1-4]. These shape memory alloys (SMAs) demonstrate a unique combination of low Young's modulus (as low as 60–80 GPa), superelastic behavior, which is close to the behavior of bone, and contain only non-toxic elements, such as Ti, Nb, and Zr, in their chemical composition [5].

Even though the Young's modulus of bulk Ti-Nb-based SMAs is low as compared to other metallic biomaterials, it is still significantly higher than that of human bone [1,3]. This mechanical mismatch between the implant and bone leads to the "stress shielding" phenomenon, which is known to be the cause of bone resorption and implant loosening [1]. The idea to use metallic foams, the stiffness of which would be much lower than that of a bulk material is considered to be a promising solution to the problem of stiffness mismatch. In addition, porous structures allow bone ingrowth and, consequently, provide more efficient implant/bone fixation [6].

Recently, the space holder method has been applied to fabricate Ti-based foams for biomedical application [6-9]. This method allows manufacturing open-porosity foams with

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different porosity (up to 80 %) and the Young's modulus matching that of bones (1–30 GPa) [6]. Complex architecture of such foams makes extremely challenging the application of conventional surface modification techniques to create on their surface a "human body friendly" environment favorable for bone ingrowth. An exploratory study of this issue constitutes the main objective this work.

Poly(3-hydroxybutyrate), the basic polymer homologue of the polyhydroxyalkanoates' family (PHA), is the most common microbial polyester that has been used as a perspective biodegradable alternative to synthetic thermoplastics. Since PHB manifests simultaneously the biodegradable and biocompatible properties, it has received much attention as the base component for perspective medical devices and drug dosage formulations. PHB can also be used for the surface modification of metallic medical devices by coating and filling – to improve their biocompatibility and provide close integration of the device with a living tissue, e.g. bone tissue [10].

Experimental

Preparation of the Ti-Nb-Zr-foams. A 50 mm-diameter, 600 mm-long Ti-20.8at%Nb-5.5at%Zr (TNZ) ingot was manufactured by *Flowserve Corp.* (USA) and atomized by *TLS Technik Spezialpulver* (Germany). TNZ foams were fabricated using a powder metallurgy based technique called the "space holder process" described in detail in [9]. As a result, 15 mm-diameter, 15 mm-high cylindrical samples of ~50 % ($P\approx0.5$) porosity foams were obtained. For the surface modification study, these samples were EDM-cut into 1 mm-thick disks, with their centre-line axis either parallel or perpendicular to the compaction direction.

Production of PHB. PHB was produced by bacterial biosynthesis [11]. A PHA producer *Azotobacter chroococcum* strain 7B, a non-symbiotic nitrogen-fixing bacterium able to overproduce PHB (to 80 % of cell dry weight) was used. The strain was isolated from the wheat rhizosphere (sod-podzolic soil) and maintained on Ashby's medium. For PHB synthesis in cells, the culture was grown in shaker flasks (containing 100 ml of the medium) at 30 °C in Burk's medium, containing sucrose as the primary carbon source. Strain growth and polymer accumulation was controlled by nephelometry and light microscopy. The polymer isolation and purification from *A. chroococcum* comprised the following stages: (1) polymer extraction with chloroform in a shaker for 12 hours at 37 °C; (2) separation of polymer solutions from cell debris by filtration; (3) polymer precipitation from chloroform solution with isopropanol; (4) subsequent repeated cycles of dissolution in chloroform and precipitation with isopropanol for 4–5 times to remove any additives and contaminants, and (5) drying at 60°C. Details of PHB and its copolymers biotechnological production have been published in [11].

Modification of metallic foams with polymer. The technique of multiple impregnation of a porous metallic sample in a 1 % polymer solution of PHB in chloroform for several days was used. Soaking was carried out in one stage and in two stages, when the polymer impregnated sample was dried and then again placed in a polymer solution. After soaking, the samples were incubated in distilled water for 2 hours to determine the effectiveness of polymer deposition on the metallic substrate. The entry of PHB into the metallic foams was measured by weighing.

Characterization. The foams' porosity (P), pore size and distribution were characterized using two techniques: a combination of metallography (*NMM-800TRF* optical microscope) and image processing (*ImageJ* software) technique [9], and the independent Archimedes porosity measurement technique [12]. Six images at different locations of each specimen were processed by *Image J* software one by one. The pore sizes range from micro-pores (<10 μm) to macro-pores (from 100 to 1000 μm). The effect of the pore size on the sample architecture is analysed in terms of the ratio "volume of an individual pore to the total volume of pores", which is also

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termed as "point impact". The cumulative impact is calculated by summarizing the volume of pores in different diameter ranges, divided by the total pore volume in the sample.

The cyclic compression tests with incrementally increased engineering strain up to either ε=50 % or specimen failure (ε at each cycle corresponds to 0.02 of the initial sample height) were performed using an *MTS' Alliance RF/200* (ξ=0.002 s^{-1}).

Polymer coating of metallic foams was studied by scanning electron microscopy (SEM). For the SEM investigation, the samples (uncoated Ti-Nb-Zr-foams, metallic foams coated by one stage and two stages technique in polymer solution soaking) were mounted on aluminium stumps, coated with gold in a sputtering device for 15 min at 15 mA (*IB-3,Giko*) and examined under a scanning electron microscope (*JSM-6380LA, JEOL*).

Results and discussion

Typical image of pore structure of Ti-Nb-Zr sample (*P*=0.49) is presented in Fig. 1a. It can be seen that even though the number of pores smaller than 90 μm is high (Fig. 1b), their collective impact is low. High collective impact of the pores bigger than 90 μm was observed (Fig. 1b).

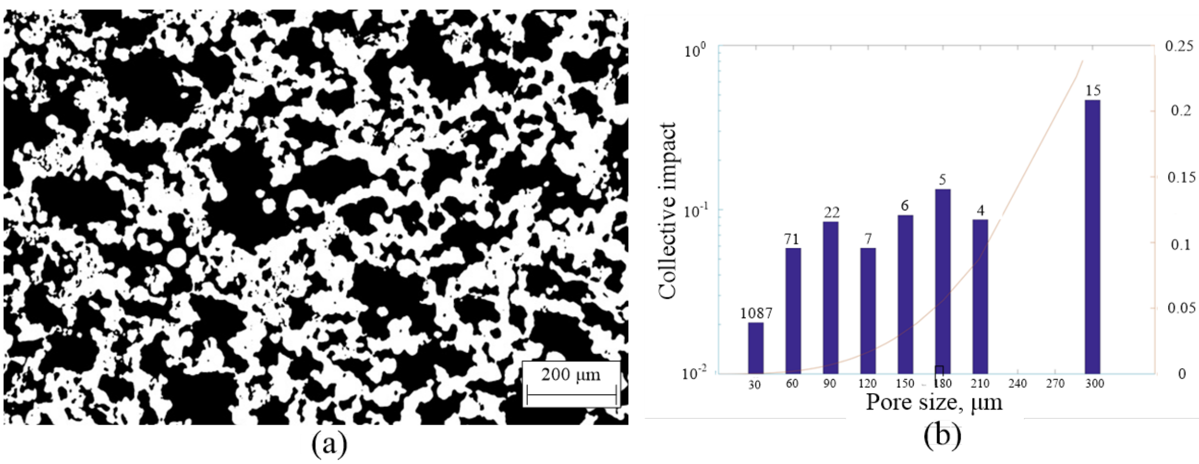

Figure 1. Typical binarized image of pore structure (a) and pore size distribution diagram (b) of Ti-Nb-Zr sample (P=0.49).

Mechanical characterization of fabricated foams was carried out by cyclic compressing tests. A typical stress-strain diagram is presented in Fig. 2. The following parameters were extracted from this diagram: the yield stress (σ_y^*), the engineering stress at ε=40 % (σ_{20}^*), and the Young's modulus at ε=20 % (E^*). The yield stress σ_y^* corresponds to the intersection of the tangent lines to the elastic and the plateau regions [13]. The apparent Young's modulus is determined for the 10th testing cycle (ε=20 %) from the tangent to the point of maximum stress (σ_{20}^*) on the unloading portion of the stress–strain diagram (Fig. 2). The Young's modulus is assessed at an intermediate level of strain to avoid both the influence of the specimen geometry (small strains) and the foam compaction (large strains) on the measured values.

The manufactured TNZ foams (*P*=0.49) demonstrate an interesting combination of low Young's modulus (E^*=5.7 GPa), which is close to that of bone, with high yield stress (σ_y^*=214 MPa), which is more than twice as high as that of bone (Fig. 2) [3].

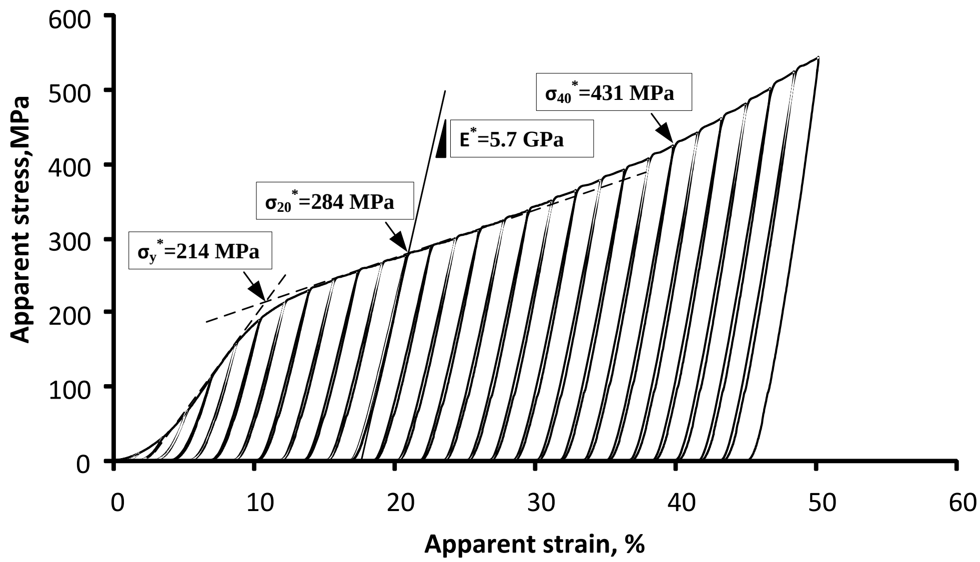

Figure 2. Typical stress-strain diagram of Ti-Nb-Zr foam (P=0.49).

PHB with a molecular weight of 3.55×10^5 Da was produced by bacterial biosynthesis. The coating of Ti-Nb-Zr foams with PHB by one stage and two stages technique of metal foams soaking in polymer solution resulted in the formation of the PHB coating on the surface and inside of pores of the Ti-Nb-Zr foam samples (Fig. 3).

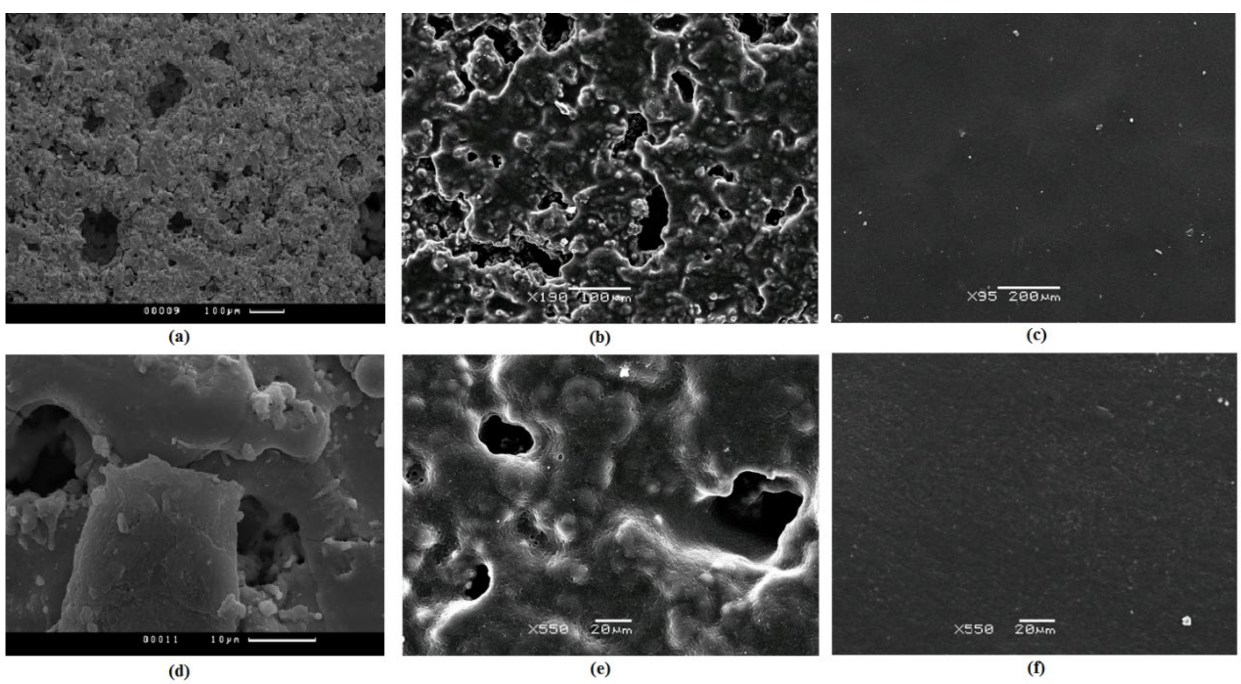

Figure 3. Ti-Nb-Zr foams (a, d) coated with PHB by one stage (b, e) and two stages (c, f) technique of multiple impregnation in polymer solution of Ti-Nb-Zr foam samples; SEM, ×95–190 (a,b,c), ×550–2000 (d, e, f).

Use of the one stage technique of soaking the Ti-Nb-Zr foams in polymer solution led to the formation of a thin and discontinuous polymer layer with a 4.6 % (w/w) content of PHB. The use of the two stage technique allows forming a continuous polymer layer on the surface of Ti-Nb-Zr foam with incorporation of 6.4 % (w/w) of PHB. The incubation of the Ti-Nb-Zr foams coated with PHB in water did not cause detachment of the polymer from the metal substrate, the weight of coated devices did not change.

Conclusion

Thus, this study showed some preliminary results of the successful surface modification of Ti-Nb-Zr foams via the two-stage immersion of these foams in a PHB-containing solution. These results constitute a starting point for the deeper investigation of the applicability of novel surface modification techniques to superelastic Ni-free foams for bone replacement.

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References

[1] M. Geetha, A.K. Singh, R. Asokamani, A.K. Gogia, Ti based biomaterials, the ultimate choice for orthopaedic implants – A review. Prog. in Mater. Sci. 54(3) (2009) 397-425. https://doi.org/10.1016/j.pmatsci.2008.06.004

[2] S. Miyazaki, H.Y. Kim, H. Hosoda, Development and characterization of Ni-free Ti-base shape memory and superelastic alloys, Mater. Sci. and Eng.: A. 438 (2006) 18-24. https://doi.org/10.1016/j.msea.2006.02.054

[3] V. Brailovski, S. Prokoshkin, M. Gauthier, K. Inaekyan, S. Dubinskiy, M. Petrzhik, M. Filonov. Bulk and porous metastable beta Ti–Nb–Zr(Ta) alloys for biomedical applications, Mater. Sci. and Eng.: C. 31 (2011) 643-657. https://doi.org/10.1016/j.msec.2010.12.008

[4] V. Sheremetyev, V. Brailovski, S.Prokoshkin, K. Inaekyan, S. Dubinskiy, Functional fatigue behavior of superelastic beta Ti-22Nb-6Zr(at%) alloy for load-bearing biomedical applications, Mater. Sci. and Eng.: C. 58 (2016) 935-944. https://doi.org/10.1016/j.msec.2015.09.060

[5] M. Niinomi, Recent titanium R&D for biomedical applications in Japan, JOM. 51 (1999) 32-34. https://doi.org/10.1007/s11837-999-0091-x

[6] G. Lewis, Properties of open-cell porous metals and alloys for orthopaedic applications, J. Mater. Sci.: Mater. in Med. 24 (2013) 2293-2325. https://doi.org/10.1007/s10856-013-4998-y

[7] X. Wang, Y. Li, J. Xiong, P. D. Hodgson, C. Wen. Porous TiNbZr alloy scaffolds for biomedical application, Acta Biomater. 5(9) (2009) 3616-3624. https://doi.org/10.1016/j.actbio.2009.06.002

[8] W. Niu, C. Bai, G. Qiu, Q. Wang. Processing and properties of porous titanium using space holder technique, Mater. Sci. and Eng.: A 506 (2009) 148-151. https://doi.org/10.1016/j.msea.2008.11.022

[9] J. Rivard, V. Brailovski, S. Dubinskiy, S. Prokoshkin, Fabrication, morphology and mechanical properties of Ti and metastable Ti-based alloy foams for biomedical applications, Mater. Sci. and Eng.: C. 45 (2014) 421-433. https://doi.org/10.1016/j.msec.2014.09.033

[10] A.P. Bonartsev, S.G. Yakovlev, E.V. Filatova, G.M. Soboleva, T.K. Makhina, G.A. Bonartseva, K.V. Shaitan, V.O. Popov, M.P. Kirpichnikov, Sustained release of the antitumor

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drug paclitaxel from poly(3-hydroxybutyrate)-based microspheres. Bioch. (Moscow) Suppl. Ser. B: Biomed. Chem. 6 (2012) 42-47.

[11] A.P. Bonartsev, I.I. Zharkova, S.G. Yakovlev, V.L. Myshkina, T.K. Mahina, V.V. Voinova, A.L. Zernov, V.A. Zhuikov, E.A. Akoulina, E.V. Ivanova, E.S. Kuznetsova, K.V. Shaitan, G.A. Bonartseva, Biosynthesis of poly(3-hydroxybutyrate) copolymers by Azotobacter chroococcum 7B: A precursor feeding strategy, Prep. Biochem. and Biotech. 47 (2017) 173-184. https://doi.org/10.1080/10826068.2016.1188317

[12] Standard Test Methods for Apparent Porosity, Water Absorption, Apparent Specific Gravity, and Bulk Density of Burned Refractory Brick and Shapes by Boiling Water, ASTM International, West Conshohocken (PA) (2010), p. 3

[13] L. Peroni, M. Avalle, M. Peroni, The mechanical behaviour of aluminium foam structures in different loading conditions, Inter. J. Imp. Eng. 35 (2008) 644-658. https://doi.org/10.1016/j.ijimpeng.2007.02.007