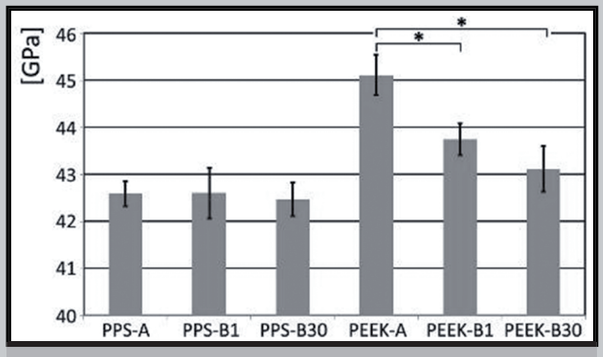

FIG. 2. The modulus of elasticity in bending (*denotes statistically significant differences, Mann-Whitney post-hoc test, α = 0.05).

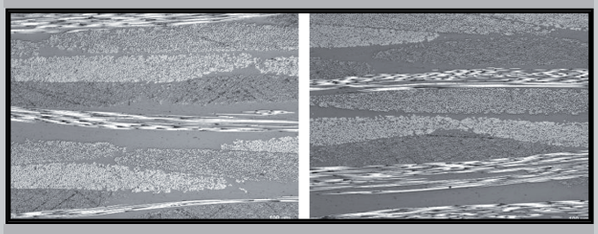

FIG. 3. Micrographs of polished sections of T300/PPS composites (from left: A and B30).

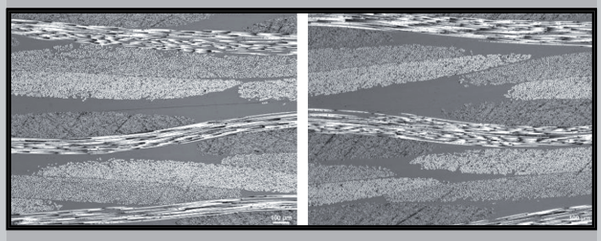

FIG. 4. Micrographs of polished sections of T300/PEEK composites (from left: A and B30).

Conclusions

On the basis of our analyses, we can state that both PEEK and PPS composites are good candidates for application as radiolucent materials providing resistance against sterilization decomposition. Presently, increased sterilization processes periods are applied and further analyses of physical properties are performed.

Acknowledgements

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ADHESION AND GROWTH OF HUMAN OSTEOBLAST-LIKE MG 63 CELLS ON TITANIUM AND STAINLESS STEEL SAMPLES DEVELOPED FOR CONSTRUCTING BONE IMPLANTS

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Titanium and stainless steel are strong, corrosion-resistant and biocompatible metals. Thanks to their remarkable properties, they have been in use for a long time in clinical medicine, mainly for constructing and replacing large joints, in particular the bone-anchoring parts, e.g. cups and stems, and also for fabricating orthopaedic screws and splints. In the Czech Republic, these devices are produced by Beznoska Ltd., and are clinically applied in the Orthopaedic Clinic, Bulovka Faculty Hospital in Prague.

This study has investigated the biocompatibility of samples made of pure titanium (according to quality standard ISO 5832-2) and corrosion-resistant steel (quality standards ISO 5832-1 and AISI 316L), obtained from Beznoska. In addition to Fe, the steel samples contained C (max. 0.025 wt.%), Si (0.6 wt.%), Mn (1.7 wt.%), P (max. 0.025 wt.%), S (max. 0.003 wt.%), Cr (17.5 wt.%), Ni (13.5 wt.%), Mo (2.8 wt.%), and Cu (max. 0.1 wt.%). The materials were used in the form of square samples (9x9 mm or 30x30 mm, thickness 1 mm). Both Ti and steel samples were grinded with SiO_2. The surface of the steel samples was then treated by polishing with Al_2O_3 paste (grain size up to 1 μm), while the surface of the Ti samples, i.e. a material not suitable for polishing, was finished by brushing using another type of Al_2O_3 paste with slightly larger grains. Thus, the surface of the steel samples was finally smoother and glossy, while the Ti surface was rougher and matted.

For the in vitro biocompatibility tests, human osteoblast-like MG 63 cells (European Collection of Cell Cultures, Salisbury, UK) were used. The smaller samples (9 x 9 mm) were inserted into polystyrene 24-well cell culture plates (TPP, Trasadingen, Switzerland; well diameter 1.5 cm). Each well contained 25 000 cells (approx. 14150 cells/cm^2) and 1.5 ml of Dulbecco's Modified Eagle Minimum Essential Medium (DMEM; Sigma, USA, Cat. No. 10270-106) supplemented with 10% foetal bovine serum (FBS; Gibco, Cat. No. 10270-106) and gentamicin (40 μg/ml, LEK, Slovenia). These samples were used for evaluating the size of the cell spreading area (day 1), and for evaluating cell shape and cell viability (days 1, 4 and 7 after seeding). The size of the cell spreading area was measured using Atlas Software (Tescan Ltd., Brno, Czech Republic). The viability of the cells was determined by the LIVE/DEAD viability/

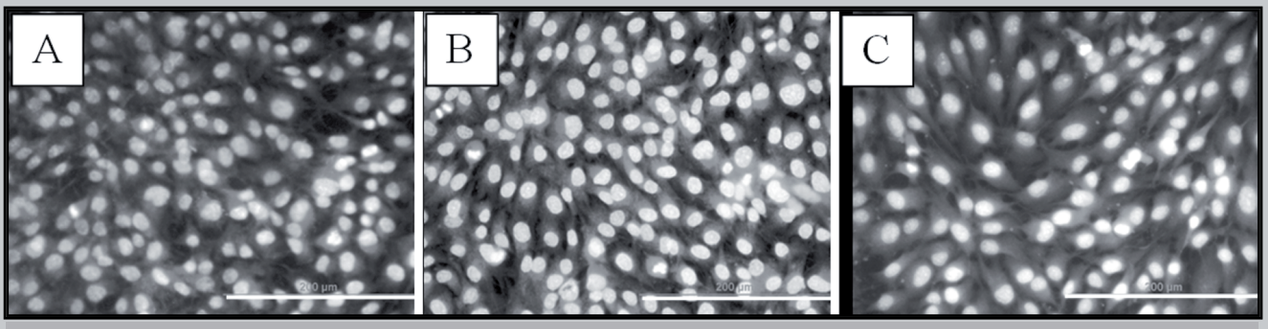

FIG. 1. Morphology of human osteoblast-like MG 63 on day 4 after seeding on titanium (A), on stainless steel (B), and on the control polystyrene culture dish (C). Cells stained with Texas Red C2-Maleimide and Hoechst #33342. Olympus IX 51 microscope, obj. 20, DP 70 digital camera, bar = 200 µm.

cytotoxicity kit for mammalian cells (Invitrogen, Molecular Probes, USA).

The larger samples (30x30 mm) were inserted into GAMA polystyrene dishes (diameter 5 cm; GAMA Group Joint-Stock Company, Ceske Budejovice, Czech Republic) and seeded with 300000 cells/dish (approx. 15300 cells/cm^2) suspended in 9 ml of the above mentioned culture medium. These samples were used for evaluating the cell number on days 1, 4 and 7 after seeding, using a Beckman Vi-CELL XR Cell Analyser automatic cell counter.

The results indicated that the number of initially adhering cells on day 1 after seeding was significantly lower on the titanium (5320±390 cells/cm^2) and on the stainless steel (4110±370 cells/cm^2) than on the control polystyrene culture dishes (7740±350 cells/cm^2). However, on day 4 after seeding, the cell population density on both metallic materials studied here became significantly higher than on the control polystyrene dishes (75200±2 890 cells/cm^2 on Ti and 90870±2 350 cells/cm^2 on steel vs. 56440±1180 cells/cm^2 on polystyrene). This suggests faster cell proliferation on both metallic materials than on polystyrene. At the same time, the cell number on the stainless steel samples was significantly higher than on the Ti samples. On day 7, the differences in number of adhered cells on both studied metals and on the control polystyrene substrate was on an average similar (from 328780±680 cells/cm^2 to 362 200±760 cells/cm^2). The cell viability on all tested materials was almost 100% in all culture intervals. The morphology of the cells on the studied materials was similar to the morphology of the adhered cells on the control polystyrene dishes, i.e. the cells were mostly flat and polygonal, and the size of their cell spreading areas was similar on all tested materials. The cells were distributed homogeneously on the entire material surface, and on day 4 they started to form confluent cell layers (FIG.1).

It can be concluded that the tests of biocompatibility confirmed that the titanium and the stainless steel promoted the adhesion and growth of bone-derived cells, and thus these materials are promising for construction of bone implants and for their good integration with the surrounding bone tissue. Further studies on osteogenic cell differentiation, potential immune activation and the response of the bone cells to growth factors, including bone morphogenetic protein, are in progress.

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BIOAPATITE MADE FROM CHICKEN FEMUR BONE

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Introduction

The inorganic part of the human bone has chemical and structural similarities with hydroxyapatite (HA) in the form of plate-shaped nanocrystals that are 2–3 nm in thickness and tens of nanometers in length and width [1]. Bones do not have a pure or a stoichiometric HA but incorporate many elements; some of them at the ppm level [2]. Ionic substitution can affect the crystal structure, crystallinity, surface charge, solubility etc., leading to major changes in the biological performance upon implantation. Nano HA was successfully synthesized from biowaste chicken eggshells and from different types of sea creatures. Another possible source for preparation of bioapatite (BAP) can be warm-blooded animal bones (e.g. bovine bone) [3]. In this study we report a preparation and characterization of bioapatite from chicken femur, such as another alternative source of bone apatite.