

The effect of surface topography on human bone marrow cells

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INTRODUCTION: Bone marrow cells contain among others mesenchymal stem cells that have the potential of self-renewal and can differentiate into various cell types such as osteoblasts, chondrocytes, myoblasts, fibroblasts and adipocytes [1]. These cells are among the first cells contacting the bone implant surface. This study investigates the effect of different defined surface topographies on the adhesion, morphology, migration and differentiation of these cells.

METHODS:

The structured culture dishes were produced by injection moulding. Copies of the following topographies were fabricated; plane, a surface with hemispheres of 30 μm in diameter and a spacing of 20 μm (30/20), a surface with hemispheres of 50 μm and no spacing (50/00), a polished and etched surface (p/e) and a surface with hemispheres of 30 μm and 20 μm spacing with a additional secondary etched structure (30/20e). The resulted dishes were titanium coated (70 nm) by physical vapour deposition, respectively sputtering.

Adult human bone marrow cells (HBMCs) were independently isolated from marrow of 3 patients obtaining a total hip replacement surgery. Adherent and expanded cells of the first passage were seeded out at a density of 5000/cm² and analysed after 7 days of cultivation. The cell viability was assessed by measuring the conversion of MTT to MTT-formazan per DNA as an index. Adherent cells were fluorescently stained for filamentous actin, the focal adhesion protein vinculin and the nucleus in order to enable a statement on the cytoskeleton architecture.

RESULTS: HBMCs on the 50/00 structure showed high MTT conversion per DNA values in comparison to all other structures whereas for p/e very low values were measured (Fig.1A) and was confirmed by another cell viability test (lysosomal activity, data not shown). CLSM analysis of the cultures showed variable cell densities and morphologies on the different structures (Fig.1B). On 50/00 surfaces the cultures were as dense as on the reference plane surface, whereas on all the other surfaces less cells were observed. On the 50/00 the cells span from one hemisphere to the other in contrast to the 30/20 where the cells were

located around the hemispheres. On both etched surface copies (p/e and 30/20e) the cells often formed star-shaped clusters.

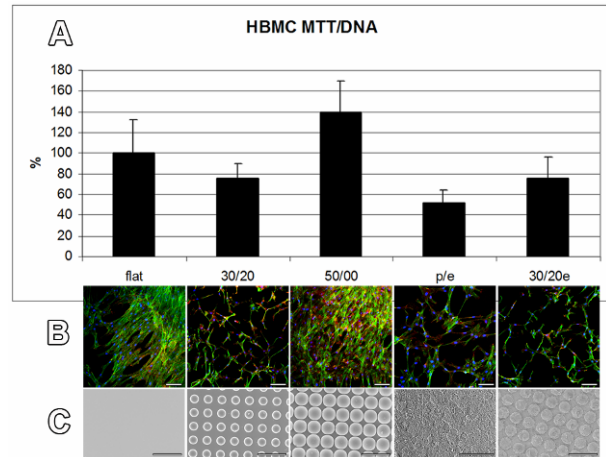


Fig. 1: HBMCs on different surface topographies. (A) MTT conversion per DNA, (B), HBMCs stained for F-actin (green), vinculin (red) and nucleus (blue), (C) raster electron microscopy images of the different topographies. Bar: 100 μm .

DISCUSSION & CONCLUSIONS: These results show that the topography influences the cell function and morphology. The 50/00 structure seems to promote cell adhesion, proliferation and has a positive impact on the metabolism of the cell. Furthermore, it can be assumed that HBMCs on copies of both etched surfaces behave differently from those kept on the polished 50/00. The comparison of the 50/00 with the 30/20 suggests that not the hemispheres themselves are the stimulating parameter, but rather the spacing between the hemispheres. The results of this study show how surface topography influences the functionality of the cells. Further investigations will be dedicated to the characterisation of the differentiation behaviour of HBMCs as function of surface topography.

REFERENCES: ¹ Pittenger M.F. et al. (1999). *Multilineage potential of adult human mesenchymal stem cells, Science 284, 143-147.*