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***In-vitro* cell culture study on Organosilane coated Mg discs**

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INTRODUCTION: Degradable Magnesium (Mg) and its alloys have a great potential as a material for resorbable implantable devices¹. Mg and its alloys have several advantages such as biocompatibility and their mechanical properties closely match to natural bone. However, rapid corrosion of Mg is hindering its use in clinical setting. One potential solution is to develop the coatings for highly corroding Mg surfaces. We used Organosilane (OS) coating to control the corrosion rate of Mg. The aim of this study was to test the hypothesis that OS multilayer self-assembled coating was cytocompatible and the surface modification of the coating will lead to higher rate of cells proliferation and decreased cells death.

METHODS: Self-assembled OS multilayer coating was formed on Mg discs using dip coating technique. The hybrid film formed via a simple sol-gel process based on the cohydrolysis and co-condensation of a mixture of alkyltriethoxysilanes and tetramethoxysilane. Furthermore, the OS coating was functionalized with 3-aminopropyltrimethoxysilane (APS) MC3T3-E1 pre-osteoblast cells were cultured on the OS coated and OS coated and aminated Mg disks for 15 days, to assess their cytocompatibility. Fluorescence dyes were used to visualize nuclei and actin cytoskeleton of the cells on the Mg discs. Cell viability was assessed using LIVE/DEAD assay.

RESULTS:

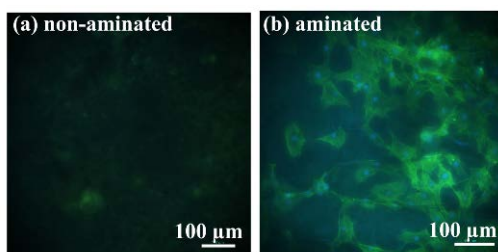


Fig. 1: Fluorescence imaging of the Mg discs exposed to MC3T3-E1 cells for 15 days: (a) non-aminated and (b) aminated OS coated Mg disc. The nuclei (blue) and actin filaments (green) indicate the presence of cells. Alexa Fluor® 488 dye for F-actin staining (green) and Hoechst 33342 dye for nuclei staining (blue) were used.

The fluorescence imaging results showed cell density on aminated OS discs was 28.40 ± 0.73

cells/10,000 μm^2 significantly higher ($p < 0.01$) than 17.83 ± 1.72 cells/10,000 μm^2 on OS coated discs after 15 days. These data indicate that amination of OS coating promoted cell attachment and/or cell proliferation due to the decrease in the surface hydrophobicity.

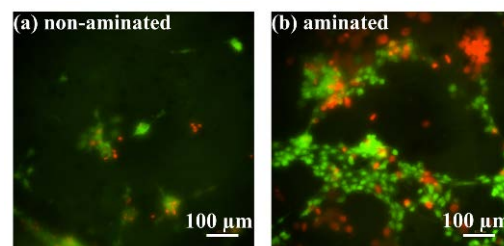


Fig. 2: Live/Dead cell assay after 11 days of culture on: (a) non-aminated and (b) aminated OS coated Mg disc.

The LIVE/DEAD cell viability assay confirmed that OS coated layer is cytocompatible. We observed 41% cell death on aminated OS coated discs compared to 87% cell death on non-aminated OS coated discs ($p < 0.029$).

DISCUSSION & CONCLUSIONS: With two-step coating process we have developed an anti-corrosive OS coating which is further functionalized with APS. We found that the cell viability and proliferation were highest when cultured on aminated OS compared to non-aminated OS. In summary, our study shows that organosilane self-assembled coating is cytocompatible and has a potential for surface functionalization of the Mg implantable devices with bioactive molecules. Our results suggest that surface functionalization can improve biocompatibility and histointegration of orthopedic degradable devices.

REFERENCES: ¹ Witte F. The history of biodegradable magnesium implants: a review. *Acta Biomater* . 2010;6(5):1680-92.

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