Effect of Metal Ions and Particles on Osteoblast Mineralisation on Grit-blasted, Titanium-coated and Hydroxyapatite-coated Prosthesis Surfaces

Karan Shah¹, Edward R. Draper², Alison Gartland¹, J. Mark Wilkinson¹.
¹University of Sheffield, Sheffield, United Kingdom, ²JRI Orthopaedics Ltd., Sheffield, United Kingdom.

Disclosures:

Introduction: Local joint tissue and systemic concentrations of cobalt and chromium are persistently elevated after metal-on-metal hip resurfacing (MoMHR) versus normal physiological levels. Successful osseo-integration is critical for survival of hip resurfacing acetabular components. However, failure of osseo-integration is a common mechanism of prosthesis failure. We have previously shown that cobalt (Co) and chromium (Cr) ions (Co²⁺, Cr³⁺ and Cr⁶⁺) impair osteoblast survival and function at clinically relevant concentrations¹, which may explain the observed poor integration of these implants. By studying the effect of different surface coating treatments on osteoblast function and mineralisation in presence of Co and Cr nanoparticles and ions, we provide evidence that prosthesis surface coating modification modulates osteoblast responses to metal debris and may improve survivorship of these prostheses.

Methods: Human osteoblast cell line (SaOS-2) were cultured on CoCr alloy disks (JRI Orthopaedics Ltd, Sheffield) with grit-blasted (GB), titanium-coated (TI) and hydroxyapatite-coated (HA) surfaces with mean surface roughness (Sz) of 57µm, 79µm and 66µm respectively. The cells were treated with Co and Cr nanoparticles (100 particles each of Co and Cr per cell) or metal ions (1000µg/L of Co²⁺ and Cr³⁺). Mineralisation by osteoblasts was induced by supplementing the feeding media with 10nM dexamethasone and 50µg/mL ascorbic acid. On day 17 of 21 day culture, the media was supplemented with 5mM inorganic phosphates and 20µM xylenol orange, a fluorescent label that incorporates into newly mineralized matrix. The percentage mineralized area was measured by analyzing fluorescence images by ImageJ. Osteoblast alkaline phosphatase (ALP) activity, a marker for cell differentiation was assessed by measuring pNPP substrate hydrolysis by cell lysate after 7 day exposure to metal ions and particles. All experiments were repeated 3 times with 3-5 replicates in each experiment and data analysed using One-way ANOVA with Kruskal-Wallis post-hoc test in Prism5®, GraphPad Software, Inc. San Diego, USA.

Results: Osteoblast mineralisation was 92% of HA surfaces versus 0.8% of GB and 0.2% of TI (p<0.0001). Osteoblasts treated with nanoparticles showed no mineralisation on GB or TI surfaces, but 62% of the HA surface was mineralized. Osteoblasts treated with metal ions on HA surfaces had significantly higher mineralisation (75%) compared to those on GB (0.4%) and TI surfaces (0.1%). ALP activity of osteoblasts on HA was 48% lower compared to GB (p<0.0001) and 52% lower than TI (p<0.0001). ALP activity on HA was also lower by 30% compared to TI when treated with nanoparticles and by 22% with metal ions (p<0.05).

Discussion: We found an increased rate of cellular differentiation, with an early decrease in ALP activity and subsequently higher mineralisation on HA coatings. HA-coated surfaces also showed better resistance to impaired mineralisation with metal exposure compared to GB and TI coatings, with nanoparticles being more detrimental to metal ions for all surfaces.

Significance: Uncemented prostheses that rely on osseointegration for survival are more commonly used than cemented. Although there is a decline in the use of metal-on-metal bearings, many prostheses are modular with CoCr junctions that release metal debris. This study highlights the benefit HA coatings offer by protecting osteoblasts from the negative effects of metal debris which will be of significant benefit for osseointegration of hip prosthesis.

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Fig 1 A) Osteoblast mineralisation on implant surfaces measured after 21 days by alizarin orange staining. B) Percentage response for mineralisation with metal ions and nanoparticles relative to control (*p<0.05, ***p<0.001).