

Implant osseointegration: *in vitro* analysis of titanium microtopography.

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INTRODUCTION: Internal fixation devices are often removed to avoid: growth disturbances in paediatrics¹; delayed infection; implant migration/breakage; allergic reactions; soft tissue irritation (e.g gliding tendons); implant protrusion / intrusion (e.g. into a joint); build up of fretting particles in unrelated organs (from loose multi component implants), as well as being cosmetically disturbing (protrusion under skin). Advocates of life long retention maintain that difficulty in removing a device due to extraosseous formation warrant their preservation to avoid complications such as increased operative time, blood loss & debris contamination. Problems associated with excessive bony over-growth account for ~7% of all complications encountered. Thus, we investigated the potential of surface polishing of titanium & its alloys to reduce excessive osseointegration, as surface micro-topography is known to directly influence tissue integration².

METHODS: Commercially pure titanium (cpTi), Titanium6%Niobium7%Aluminium (TAN), Titanium15%Molybdenum (Ti15Mo) & Stainless steel (Ss, negative control) were studied. Micro-rough standard surfaces of the cpTi (TS), TAN (NS) & Ti15Mo (MS) were included as positive controls. Variants were electropolished (TE, NE, ME) or pastepolished (TP, NP, MP). Surfaces were characterized by AFM, non-contact profilometry, contact angle, XPS & SEM. Response of 6 day old rat calvaria (RC) cells for osteocalcin (OC) gene expression, Alizarin red-S(AR-S), & tritiated thymidine (³H-Tdr). Confocal fluorescent microscopy of the cell cytoskeleton (actin, tubulin & vinculin) were assessed at 48hr.

RESULTS: Surface analysis reveals that polishing successfully reduced the micro-roughness of all materials (Fig.1) without significantly affecting hydrophilicity; or the chemical properties due to their anodisation following treatment, increasing their oxide layer thickness. OC gene expression on polished samples was significantly reduced (Fig.2; ANOVA; p<0.05) compared to positive controls however due to the superior bone bonding ability of titanium, this was higher

than the negative control. AR-S quantification supported this. ³H-Tdr incorporation results showed distinct proliferative outcomes for all samples. Generally, a peak was seen at 14 days culture. Interestingly, paste polished TAN showed an obvious stunt in proliferation compared to other samples (data not shown). Ongoing viability & morphological analysis will help elucidate this finding. Fluorescent microscopy results indicate that surface polishing allowed cells to adopt cytoskeletal morphology similar to that seen on the negative control & not the idiosyncratic osteoblast morphology documented for micro-rough standard surface.

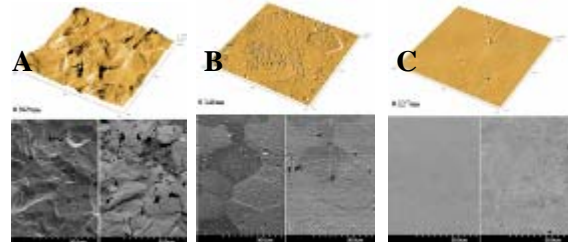


Fig.1. Reduction of surface micro-topography of cp.Ti with electropolishing (B). (A) Positive control (TS). (C) Negative control (Ss).

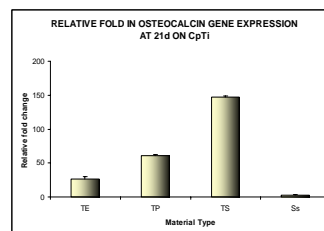


Fig.2. Representative Data of OC expression on cp.Ti & Ss negative control.

DISCUSSION & CONCLUSIONS: Surface polishing appears to exert its affect on reducing extraosseous formation from a cellular level by inhibiting osteoblast ability to mineralise & produce mature matrix. We believe surface polishing suspends RC cells in a proliferative state & prevents subsequent bone formation. *In vivo* work from our laboratory corroborates these findings.

REFERENCES:¹ Peterson.(2005) J.Paediatric. Orthop. 25: 107-115. ² Meredith et al., (2007) J.Mater.Sci. Med 18: 405-413.

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