



## Polyethylene particles stimulate expression of ITAM-related molecules in peri-implant tissues and when stimulating osteoclastogenesis in vitro

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### ABSTRACT

Wear particle-induced orthopaedic prosthesis loosening is associated with elevated osteoclast activity. The immunoreceptor tyrosine-based activation motif (ITAM)-related molecules OSCAR, Fcγ, TREM2 and DAP12 are important for osteoclast formation. The aim of this study was to determine if these molecules are involved in peri-implant loosening by investigating their expression in peri-implant tissues obtained at revision of joint replacement components containing polyethylene (PE) wear particles, and in osteoclasts formed in vitro in the presence of PE particles. The results showed that there was a marked and statistically significant increase in protein levels of the ITAM-related molecules in the revision tissues. The levels of OSCAR, Fcγ, TREM2 and DAP12 mRNA in the revision tissues were also increased. In vitro PE particles stimulated osteoclast resorption in the presence of 50 ng ml<sup>-1</sup> receptor activator NFκB (RANKL) and significantly elevated the expression of OSCAR, Fcγ, TREM2 and DAP12 during osteoclast formation. These findings suggest that the ITAM signalling molecules and their co-receptors have a role in pathogenic bone loss associated with implant PE wear.

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### 1. Introduction

Aseptic loosening associated with peri-implant bone loss is the most common reason for joint revision surgery [1]. This peri-implant osteolysis (PO) is associated with wear particles liberated from the bearing surfaces of the prosthesis, stimulating a chronic inflammatory response dominated by macrophages and multinucleated foreign body giant cells [2–4]. There is strong evidence showing that polyethylene (PE) particles produced by wear of the articulating surface, particularly in hip prostheses, are important in peri-prosthetic bone loss around articulations with PE linings [5,6]. Increased remodelling of peri-implant bone and immature bone formation has been demonstrated around loosened prosthetic hip implants, indicating that there is increased osteoclast activity that is partially compensated for by high osteoblast activity in peri-implant osteolysis [7]. PE wear particles liberated from bearing surfaces of orthopaedic implants are major determinants of PO and have a multitude of cellular effects [8]. Cells containing PE wear particles in human peri-implant tissues express osteoclast

phenotypic markers, such as tartrate-resistant acid phosphatase (TRAP), cathepsin K (CatK) and calcitonin receptor (CTR), and these genes are further induced by interaction between osteoclasts and the bone matrix [4]. Consistent with these findings cells isolated from revision tissue readily form bone-resorbing osteoclasts in vitro [3].

Receptor activator of NFκB ligand (RANKL) and its receptor RANK are key molecules regulating osteoclasts in health and disease. High levels of RANK and RANKL are present in peri-implant tissues obtained at revision surgery [3,9]. RANKL binding to RANK on osteoclasts results in an intracellular signalling cascade leading to stimulation of the key osteoclast transcriptional factor, nuclear factor of activated T cells 1 (NFATc1) [10,11]. NFATc1 then initiates the terminal stages of osteoclast formation by directly inducing genes such as CTR, cathepsin K, matrix metalloproteinase 9 (MMP-9), TRACP, β3-integrin and osteoclast-associated receptor (OSCAR) [9,12–18].

Recent reports indicate that NFATc1 may also be stimulated by immunoreceptor tyrosine-based activation motif (ITAM) signalling pathways in addition to RANK/RANKL signalling [19]. Osteoclast associated receptor (OSCAR) and triggering receptor expressed on myeloid cells 2 (TREM2) are recently identified transmembrane receptor proteins that associate with the adaptor proteins Fc

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