ACTIVE VITAMIN D ANALOGUES DO NOT DIRECTLY AFFECT VASCULAR SMOOTH MUSCLE CELL CALCIFICATION, BUT INDUCE A PROCALCIFIC TO ANTICALCIFIC PARACRINE SWITCH IN CO-CULTURED HUMAN MACROPHAGES.

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Purpose: Vascular calcification is highly correlated with morbidity mortality in end stage renal disease (ESRD) patients, and is commonly associated with inflammation. Clinical studies indicate a survival benefit of Vitamin D treatment in ESRD patients. Thus, we examined the effects of the active Vitamin D analogues, calcitriol and paricalcitol, on the ability of macrophages to regulate vascular smooth muscle cell (SMC) calcification *in vitro* using a macrophage/SMC co-culture system. **Methods:** Macrophages were obtained by phorbol-ester induced differentiation of THP-1 cells. Vitamin D receptor expression was confirmed on both macrophage and SMC by RT-PCR. Macrophage/SMC co-cultures were established using transwell inserts. Mineralization in SMC co-cultures was induced by treatment with 2.6 mM phosphate for 10 days. **Results**: Macrophages induced a three-fold increase in SMC calcification in the co-culture system compared with SMC cultured alone (SMC versus Macrophage/SMC: 18.65 $\pm$ 3.8 versus 66.97 $\pm$ 11.19 μg/mg protein, P=0.009). In contrast, treatment of macrophage/SMC co-cultures with various concentrations (0.5, 5 and 50 nM) of calcitriol and paricalcitol significantly inhibited SMC calcification in a dose-dependent manner, with maximal ~5-fold inhibition observed with 50 nM paricalcitol. Neither calcitriol nor paricalcitol inhibited calcification of SMC culture alone, indicating a paracrine effect. Mechanistically, real-time PCR revealed the induction of osteopontin and inhibition of bone morphogenetic protein 2 mRNA by both calcitriol and paricalcitol in macrophages. Levels of alkaline phosphatase and oncostatin M mRNAs were induced preferentially by calcitriol, and TNFα mRNA levels were preferentially inhibited by paricalcitol treatment. **Conclusions** The ability of active Vitamin D analogues to direct a switch in the paracrine activities of macrophages from procalcific to anticalcific may contribute to their observed survival benefits in CKD patient