8 VITAMIN D MODULATES AROMATASE EXPRESSION IN HUMAN MACROPHAGES AND DOWNREGULATES PROINFLAMMATORY CYTOKINE PRODUCTION VIA ERK/MAPK SIGNALING.

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Background and objectives Vitamin D_3 (calcitriol) was found to downregulate innate immune response in human macrophages and showing immune-modulatory effects[1]. Gender-linked differences in serum levels of sex hormones and their peripheral metabolites play an important role in the neuroendocrine immune/inflammatory response also in monocyte/macrophage cells[2].

This in vitro study evaluated the effects of calcitriol on the expression and the activity of the aromatase involved in estrogen production in cultures of human macrophages and its possible ability to modulate the production of the proinflammatory cytokines (IL-1β, IL-6 and tumour necrosis factor α (TNFα)) involved in the innate immunity response.

Materials and methods Cultures of human THP-1 were activated to macrophages by interferon γ (IFNγ) (500 U/ml) and treated for 24 h with calcitriol (10^{-8}M) and 17β-estradiol (E_2, 10^{-8}M) alone and in combination. Untreated macrophages (THP-1-activated) were used as controls (cnt). P450-aromatase synthesis was evaluated by immunocytochemistry (ICC) and western blotting (WB), whereas the expression of the related CYP19A1 gene was quantified by Real-Time PCR (RT-PCR). IL-1β, IL-6 and TNFα synthesis was analysed by enzyme immunoassay (ELISA) and WB. The signal transduction pathway of mitogen activated proteins ERK1/2 was evaluated by WB.

Results. P450-aromatase synthesis and related CYP19A1 mRNA expression were found significantly downregulated by calcitriol in presence of E_2 ($p<0.001$ vs E_2-treated cells). The IL-1β, IL-6 and TNFα production decreased in the THP-1-activated macrophages treated with calcitriol with and without E_2 ($p<0.001$ in calcitriol alone vs cnt and in calcitriol/E_2 vs E_2 for all cytokines). The data were obtained by ELISA and confirmed by WB. Interestingly, calcitriol decreased ERK1/2 phosphorilation both alone (vs cnt) and in presence of E2 (vs E2-treated cells) reducing the ERK pathway activation, as observed by WB.

Conclusions Our data suggest that calcitriol downregulated the aromatase expression, involved in the production of estrogens, and decreased proinflammatory cytokine production by human macrophages. These downregulatory effects of calcitriol were found to correlate with the reduction in phosphorilation of ERK pathway. These results suggest a possible role of calcitriol on innate immunity of human macrophage that drive

the inflammatory processes in RA and its anti-inflammatory and immunosuppressive intracrine activity.

REFERENCES

1. **Do J E**. Effects of vitamin D on expression of Toll-like receptors of monocytes from patients with Becet's disease. *Rheumatology* 2008;**47**:840–48.
2. **Cutolo M**, Straub RH, Bijlsma JW. Neuroendocrine-immune interactions in synovitis. *Nat Clin Pract Rheumatol* 2007;**3**:627–34.