A105 25-HYDROXYVITAMIN D₃ CONVERSION BY DENDRITIC CELLS AND T CELLS DRIVES 1,25-DIHYDROXYVITAMIN D₃ MEDIATED ANTI-INFLAMMATORY CD4⁺ T CELL RESPONSES

L Jeffery, K Raza, A Filer, D Sansom *School of Immunity and Infection, University of Birmingham, Birmingham, UK*

10.1136/ard.2010.148981.8

Background Low vitamin D status is associated with an increased risk of autoimmune diseases, including rheumatoid arthritis. Thus, understanding how vitamin D modifies immune reactions holds therapeutic potential. The authors have shown that 1,25(OH)₂D₃, acts directly upon human CD4 T cells, suppressing inflammatory cytokines (interleukin (IL)-17, IL-21, IFN γ and IL-22) while enhancing regulatory-markers (CTLA-4, CD25, FoxP3 and IL-10). However, the short half life of 1,25(OH)₂D₃ and its low serum level imply that local conversion of 25(OH)D₃, the major circulating form of vitamin D, by 1 α -hydroxylase (CYP27B1), is necessary for immune regulation in vivo. Thus, the authors have studied the effect of 25(OH)D₃ upon T cell responses. In view of the critical role of CTLA-4 in immune-regulation and its strong sensitivity to 1,25(OH)₂D₃, the authors have also investigated CTLA-4 function in vitamin D-modified immune responses.

Methods CD4⁺CD25⁻ T cells were purified from human peripheral blood (PB) and stimulated with LPS-matured allo-genic dendritic cells (DCs) plus anti-CD3 or with anti-CD3/CD28 beads in the presence and absence of 25(OH)D₃. FoxP3, CTLA-4 and cytokines were measured by flow cytometry and CYP27B1 by qPCR. In function assays, CTLA-4 was blocked with anti-CTLA-4 and T cell division monitored. DC expression of CD80 and CD86 was assessed by FACS. PB and synovial fluid (SF) mononuclear cells, from patients with synovitis, were stimulated with antiCD3 in the presence and absence of 1,25(OH)₂D₃ and cytokines examined by FACS.

Results Stimulation of T cells by DCs in the presence of 25(OH)D₃ led to strong suppression of IL-17 and IFN γ but upregulation of CTLA-4 and CTLA-4⁺FoxP3⁺ frequencies. By contrast, in the absence of DCs, 25(OH)D₃ caused modest CTLA-4 induction and IFN γ suppression ($p < 0.05$ for all). These differences corresponded with CYP27B1 levels, as maturation induced much higher CYP27B1 in DCs than arose in T cells ($p < 0.05$). CTLA-4 blockade overcame 1,25(OH)₂D₃-mediated suppression of T cell division in DC stimulations and prevented downregulation of co-stimulatory CD80 and CD86 on DCs. PB T cells from synovitis patients responded normally to 1,25(OH)₂D₃ with suppressed IL-17 and IFN γ ($p < 0.0001$). 1,25(OH)₂D₃ also reduced IL-17 ($p < 0.05$) but not IFN γ in SF T cells.

Conclusions The authors have shown that DCs can efficiently convert 25(OH)D₃ to drive 1,25(OH)₂D₃-modified T cell responses and that upregulation of CTLA-4 is mechanistically important in immune suppression by vitamin D. Thus, 25(OH)D₃ supplement could be useful in the treatment of conditions such as RA. This is supported by the authors' finding that T cells from synovitis patients can respond to 1,25(OH)₂D₃.