[haematologica reports]
2006;2(3):16-18

Reciprocal activating interaction between dendritic cells and aminobiphosphonates-stimulated T cells: role of CD86 and inflammatory cytokines

CONTI L[1]
CASETTI R[2]
CARDONE M[1]
VARANO B[1]
MARTINO A[2]
BELARDELLI F[1]
POCCIA F[2]
GESSANI S[1]

[1]Department of Cell Biology and Neurosciences, Istituto Superiore di Sanità, Rome, Italy; [2]Laboratory of Cellular Immunology, National Institute for Infectious Diseases "L.Spallanzani", Rome, Italy

Cognate interactions are a common feature of the acquired immune response, where intimate cell contacts govern critical events such as Ag presentation and delivery of T cell help to CTLs and B cells, while the coordinated control of innate immunity is mostly mediated by cytokine loops. However, a number of studies recently highlighted the importance of DC interactions with NK cells, an important effector cell type of innate immunity, in the regulation of DC maturation.[1, 2, 3] Moreover, Gerosa *et al.*[3] reported the existence of a cross-talk between immature DCs (iDCs) and resting NK cells, leading to cell activation on microbial stimulus encounter. Finally, few studies have also reported some functional interactions between DC and T cell subsets[4, 5, 6] but none of these studies has investigated the mechanisms of action of different classes of phosphoantigens or described functional reciprocal interactions between T cells and DCs. Thus we investigated the reciprocal interactions between human monocyte-derived DCs and antigen-stimulated circulating γδ T lymphocytes, bearing the Vδ2 TCR (Vδ2). We demonstrated that co-culture of iDCs with peripheral blood Vδ2 T cells stimulated with either pyrophosphomonoesters (IPP) or aminobiphosphonates (es. pamidronate, PAM) leads to a significant up-modulation of CD86 and MHC class I molecules and to the acquisition of functional features typical of activated DCs. DC activation induced by both IPP- and PAM-stimulated γδ T cells was mostly mediated by TNF-α and IFN-γ secreted by activated lymphocytes. However, the effect of PAM-activated γδ T cells, but not that of IPP-activated cells, required cell-to-cell contact. In fact, as shown in Figure 1, neither phenotypic (i.e. CD86 induction) nor functional changes (i.e. Dextran-FITC uptake) were observed when PAM-stimulated γδ T cells and DCs were separated by a semipermeable membrane.

Reciprocally, activation of Vδ2 T cells by PAM, but not by IPP, was dependent on cell contact with iDCs. In fact, we observed that induction of activation markers (Figure 2A) and secretion of inflammatory cytokines (Figure 2C) by PAM-stimulated Vδ2 T cells occurred only in the presence of DCs. Moreover, when PAM-stimulated DC/γδ T cell co-cultures were separated by a semipermeable membrane or treated with blocking anti-CD86 antibodies, induction of CD25 and CD69 in Vδ2 T cells was strongly reduced (Figure 2B). In contrast, Vδ2 T cell activation by IPP was not affected by DCs.

These results demonstrate for the first time a bi-directional activating interaction between iDCs and aminobiphosphonates-stimulated T lymphocytes, thus suggesting a potential adjuvant role of this early cross-talk in the therapeutic activity of aminobiphosphonate drugs. It has already been hypothesized that aminobiphosphonates exert their antitumor activity either directly, by inducing cell cycle arrest in tumor cells,[7] or through activation of T cells.[8] Our finding that aminobiphosphonate-stimulated T cells induce DC activation, would argue in favor of a wider spectrum of action for these compounds, unraveling their potential capacity of inducing specific immune responses against malignancies. In fact, DCs activated by contact with PAM-stimulated T cells, produce IL-12 and might in turn stimulate T cell activation and expansion as well as the recruitment of other T cells, thus contributing to the amplification of the immune response. Moreover, Lopez *et al.*[9] recently demonstrated a role for IL-12 in protecting T cells from programmed cell death induced by mitogenic stimulation, thus favoring their expansion and antitumor activity. The broad cross-reactivity of circulating T cells and the possibility to improve their expansion in response to phosphorylated metabolites might provide a useful tool to manipulate the activation state of this cell population for therapeutic and/or vaccination exploitation in both

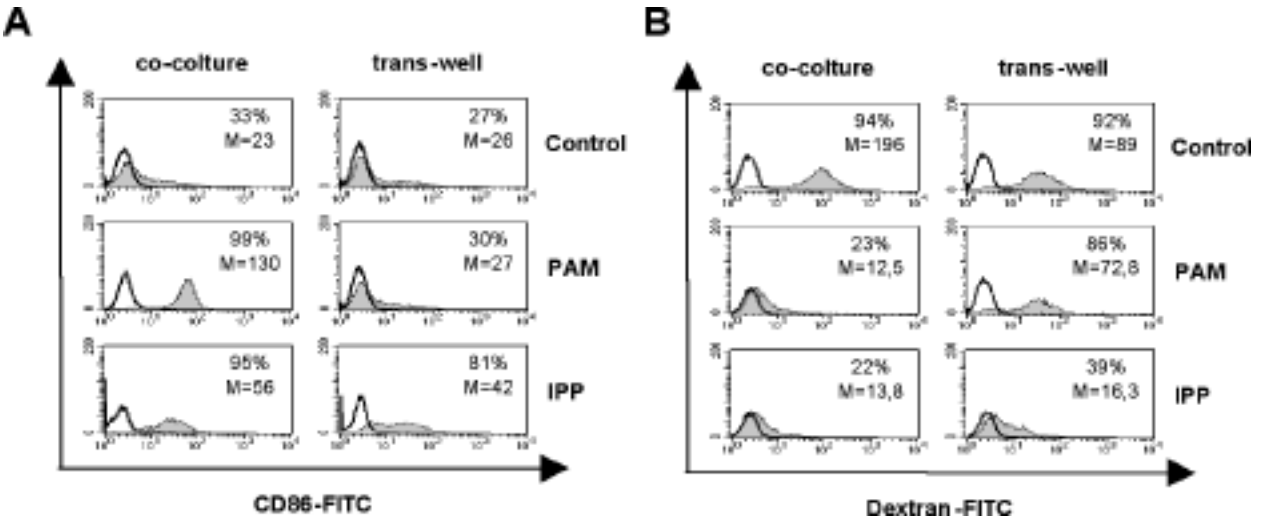

Figure 1. Role of cell-to-cell contact on the DC phenotypic and functional changes induced by PAM- and IPP-activated γδ T lymphocytes. iDCs and purified γδ T lymphocytes were co-cultured or separated by a semipermeable membrane in trans-well chambers. 48 h after PAM or IPP stimulation, DCs were subject to (A) phenotypic analysis and (B) dextran-FITC uptake.

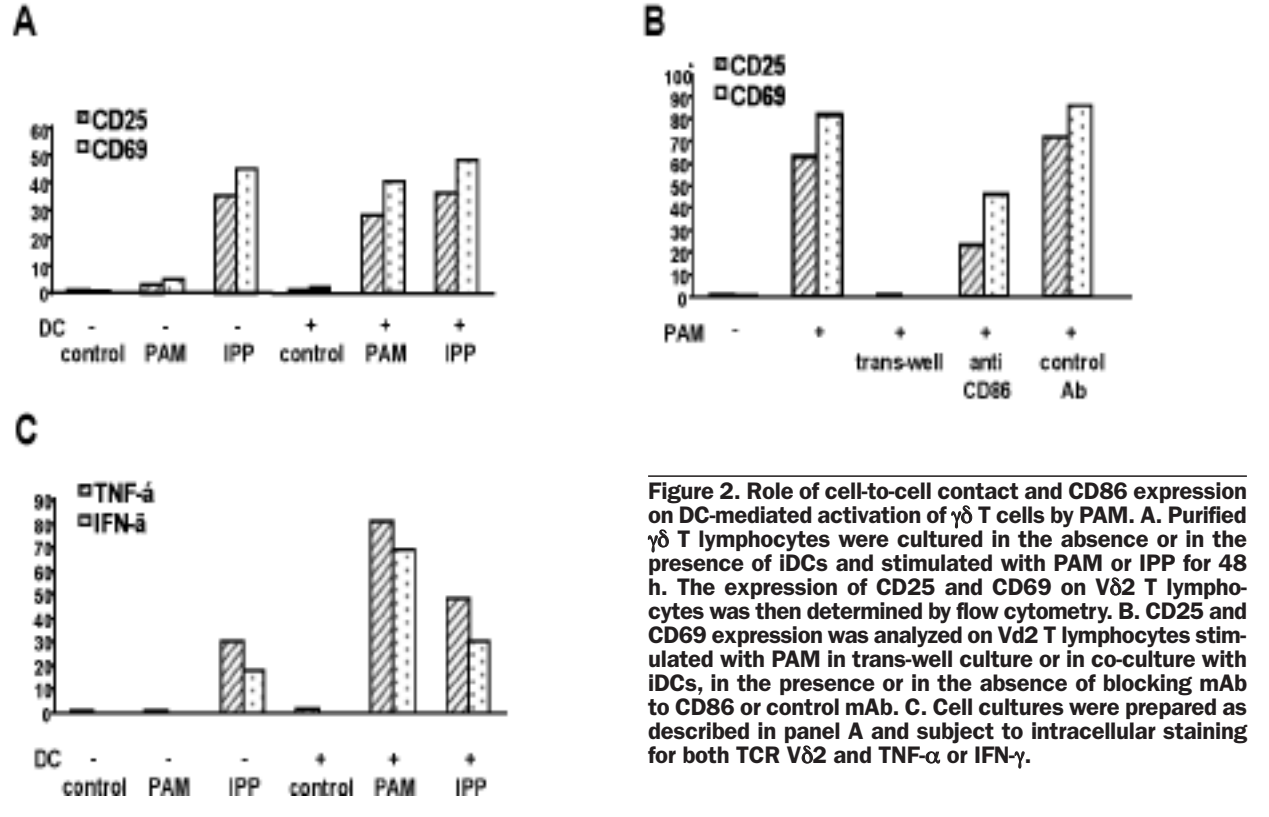

Figure 2. Role of cell-to-cell contact and CD86 expression on DC-mediated activation of γδ T cells by PAM. A. Purified γδ T lymphocytes were cultured in the absence or in the presence of iDCs and stimulated with PAM or IPP for 48 h. The expression of CD25 and CD69 on Vδ2 T lymphocytes was then determined by flow cytometry. B. CD25 and CD69 expression was analyzed on Vd2 T lymphocytes stimulated with PAM in trans-well culture or in co-culture with iDCs, in the presence or in the absence of blocking mAb to CD86 or control mAb. C. Cell cultures were prepared as described in panel A and subject to intracellular staining for both TCR Vδ2 and TNF-α or IFN-γ.

malignancies and infectious diseases. The use of aminobiphosphonates as activating Ag would limit the activation of T cells at specific districts, where iDCs and resting T cells coexist, thus avoiding uncontrolled immune activation and cytotoxic responses. Therefore, aminobiphosphonates could act as useful vaccine adjuvants capable of linking innate to adaptive immunity by rapidly activating T cells in the presence of DCs and potentially inducing T cell-mediated protective immunity.

References

1. Ferlazzo G, Tsang ML, Moretta L, Melioli G, Steinman RM, Munz C. Human dendritic cells activate resting natural killer (NK) cells and are recognized via the NKp30 receptor by activated NK cells. J Exp Med 2002;195:343.
2. Piccioli D, Sbrana S, Melandri E, Valiante NM. Contact-dependent stimulation and inhibition of dendritic cells by natural killer cells. J Exp Med 2002;195:335.
3. Gerosa F, Baldani-Guerra B, Nisii C, Marchesini V, Carra G, Trinchieri G. Reciprocal activating interactions between natural killer cells and dendritic cells. J Exp Med 2002;195:327.
4. Leslie DS, Vincent MS, Spada FM, Das H, Sugit M, Morita CT. CD1-mediated/T cell maturation of dendritic cells. J. Exp Med 2002;196:1575.
5. Ismaili J, Olislagers V, Poupot R, Fourniè JJ, Goldman M. Human T cells induce dendritic cell maturation. Clin Immunol 2002;103: 296.
6. Ye Z, Haley S, Gee AP, Henslee-Downey PJ, Lamb LS. *In vitro* interactions between T cells, DC, and CD4+ T cells; implications for the immunotherapy of leukemia. Cytotherapy 2002;4:293.
7. Raje N, Anderson KC. The evolving role of biphosphonate therapyin multiple myeloma. Blood 2000;96:381.
8. Kunzmann V, Bauer E, Feurle J, Weissinger F, Tony HP, Wilhelm M. Stimulation of T cells by aminobiphosphonates and induction of anti plasma cell activity in multiple myeloma. Blood 2000;96:384.
9. Lopez RD, Xu S, Guo B, Negrin RS, Waller EK. CD2-mediated IL-12-dependent signals render human -T cells resistant to mitogen-induced apoptosis, permitting the large-scale ex vivo expansion of functionally distinct lymphocytes: implications for the development of adoptive immunotherapy strategies. Blood 2000;96: 3827.