IN Vitro Generation Of Dendritic Cells From Cd34+
Cells Using Gm-Csf And Ifn-γ Versus Gst-Ii4
(Gm-Csf, Scf, Tnf-α, Il-4)

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Background Dendritic cells (DCs) are now recognized as the powerful antigen presenting cells which play a crucial role in generating immune responses against specific antigens. DCs can be differentiated from monocytes or CD34+ hematopoietic stem cells (HSCs). Stem cell factor (SCF) and FLT-3L are necessary for proliferation of DCs, whereas TNF-α, GM-CSF, IL-4 are necessary for differentiation of DCs. Formerly, DCs subsets were divided by lineage specific manner such as myeloid or lymphoid DCs. But recently, in mice system, some study suggested that environmental conditions can change the myeloid DCs to lymphoid DCs, and vice versa. Here, we investigated whether the human DCs can be differentiated from CD34+ HSCs and whether IFN-γ has certain roles adding GM-CSF compared with GM-CSF, SCF, TNF-α and IL-4 (GST/IL-4) in differentiation of DCs from CD34+ human HSCs.

Methods In GST/IL-4 group, CD34+ cells of patients with malignancies were collected and cultured in X-vivo 20 media with GST/IL-4 for 2 weeks. In GM-CSF/IFN-γ group, CD34+ HSCs were cultured in X-vivo 20 media with GM-CSF for first week, and with IFN-γ for second week. Both groups of DCs were maturated with ionomycin for 24 hours prior to harvest and were estimated with light microscopy, immunophenotype, dextran-FITC uptake, mixed lymphocyte reaction (MLR) and cytokine secretion such as IL-12 and IFN for evaluation of their morphologic, immunophenotypic and functional characteristics.

Results On the 14th day of culture, cells of both groups showed common feature of DCs in light and electronic microscopy. In GST/IL-4 group, DCs were CD1a+ strongly whereas the DCs of GM-CSF/IFN-γ group were CD1a+, CD8+. HLA-DR and another co-stimulatory molecules and ligand molecules (CD40, CD80, CD83 and CD86) were co-expressed in both groups of DCs. MLR assay revealed that DCs from both groups were similarly efficient to activating T cells. IL-12 production of both DCs were not different but the production of IFN-γ was uniquely detected in DCs of GM-CSF/IFN-γ group.

Conclusion We can suggest that 1) GM-CSF/IFN-γ can be used for DC differentiation from human CD34+ cells and that 2) in view of IFN-γ production and expression of CD8, GM-CSF/IFN-γ generated human DCs maybe the in vitro form of lymphoid DCs, as formerly known.