



## DENDRITIC CELL DIFFERENTIATION IN A PERIPHERAL BLOOD MONONUCLEATED CELL CULTURE TREATED WITH INTERLEUKIN-2

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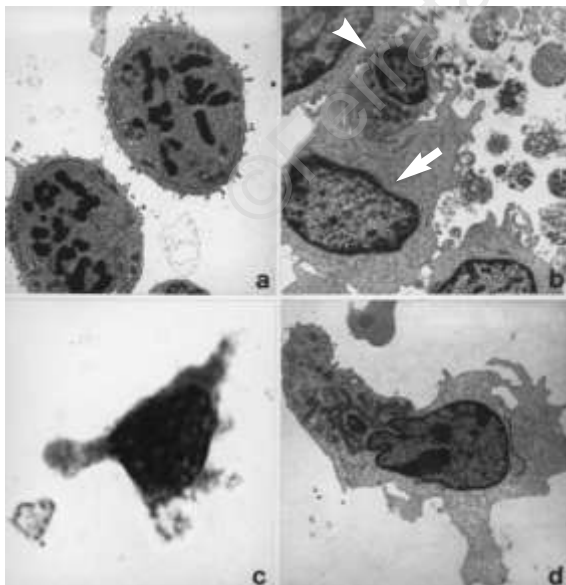
Peripheral blood mononucleated cells (PBMC) isolated by a 1077-density gradient (Histopaque, Gibco) from a healthy subject were cultured in an RPMI 1640 medium added to 10% fetal calf serum in the presence of recombinant interleukin-2 (rIL-2, Eurocetus, 100 pg/mL) for 8 days. Fresh, complete medium containing rIL-2 was replaced every two days.<sup>1</sup> Part of the culture samples were stained with May-Grünwald-Giemsa for light microscopy and part were processed for electron microscopy analysis with a Zeiss TEM at 50 KV.

An intense proliferation (high mitotic index) (Figure 1a) was observed. An unusual appearance of enlarged and irregularly-shaped cells with cytoplasmic projections typical of dendritic cells was detected. Ultrastructural analysis revealed some of the morphological peculiarities of these cells: nuclear invaginations, mitochondria cluster in the

juxta-nuclear region, lysosomes, cytoplasmic projections and frequent association to small leukocytes. We were not able to reproduce the same results with other cultures derived from different subjects; in these cases, dendritic cells were only sporadically detected. Recently, dendritic cell colony-forming units (CFU-DC) induced *in vitro* from bone marrow cells, myeloid precursors and human peripheral blood monocytes have been shown.<sup>2,3</sup> Various cytokines (IL-1, IL-3, IL-4, IL-6, TNF $\alpha$ , GMC-SF, IFN $\gamma$ ) are described to be dendritic cell inducers.<sup>4</sup> We obtained dendritic cells starting from PBMC cultures treated with interleukin-2. Although we cannot exclude the production of various other cytokines in our system, the proliferation of dendritic cells must be considered an unexpected result in this case.

### References

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**Figure 1. Light (c) and electron microscopy (a, b, d) morphology of dendritic blood cells cultured for 8 days with recombinant interleukin-2 (rIL-2).**

**a = undifferentiated cells during the proliferation ( $\times 4.000$ );**

**b = profile of direct contact between a dendritic cell (arrow) and a small leukocyte (arrowhead) ( $\times 5.500$ );**

**c, d = possible terminal stage of dendritic cell differentiation: enlarged size and irregular shape are detectable through electron microscopy; (d) numerous mitochondria are localized in the juxta-nuclear region (c =  $\times 3.800$ ; d =  $\times 6.000$ ).**