

## **Improved responsiveness of Langerhans/dendritic cells to allergens**

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Langerhans cells (LC) are immature dendritic cells (DC) that are highly specialized antigen-presenting cells (APC) located in the skin, mucosa, and lymphoid tissues. LC play a key role in the induction phase of contact allergenicity, and hence it may be possible to develop an in vitro LC-based assay for contact sensitization. The difficulty in harvesting and the short survival time of LC in culture has prevented researchers from widespread use of LC. Although improvements have been made, generating large number of cells remains a limiting factor and the functionality and cytokine production capacity of cells in response to different stimuli is not consistent. Here we report a new method of generating LC from CD34+ progenitor cells harvested from umbilical cord blood. The generated LC were expanded 200 fold and they expressed CD1a and HLA-DR, characteristic of LC. LC were cultured for up to 41 days with no significant changes in CD1a and HLA-DR expression. Transmission electron microscopy showed the presence of Birbeck granules, a key ultrastructural marker of LC. Upon stimulation with lipopolysaccharide and phorbol 12-myristate 13-acetate, the LC showed a reproducible (n = 4), high level of gene and protein responsiveness in terms of IL-12, MIP-1 $\alpha$ , MIP-3 $\alpha$ , IL-6, and TNF- $\alpha$  expression. The generated cells were infectable with HIV and they were able to stimulate allogeneic T cells. In conclusion, we have developed a method to harvest and culture functional LC expressing key LC markers. These cells have longer life span in culture and can be used in: 1) allergenicity, 2) viral infection, 3) antigen presentation, 4) immuno-therapeutic, and numerous other studies.

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