

Cytokine production by PBL and T cells of allergic patients stimulated with allergens measured with the Th1/Th2 cytometric bead array kit (CBA)

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Measuring cytokines in cell cultures are an important source of information describing dominant type(s) of reaction(s). The primary assays used are *in vitro* PBL cultures, allergen specific T-cell lines and clones which are tested against the relevant allergens. Even though a standard proliferation assay will provide some information we are also interested in whether the reaction is of a regulatory T-cell, Th1, Th2 or general inflammatory character. Cytokines detectable by the Th1/Th2 cytometric bead array kit from BD Bioscience can measure the following cytokines in ONE sample using less than 50 µl culture of supernatant: IFN-gamma (Th1), TNF-alfa (inflammatory), IL-10 (regulatory), IL-5 and IL-4 (Th2) and IL-2 (growth factor). This sounds too good to be true so we tested the kit against ELISAs, which did not show exactly the same results but the results were more consistent using the CBA kit. However, the CBA assay is very sensitive to BOTH light and heat (room temperature). Fortunately, by keeping samples on ice and in the dark the problem is considerably reduced. Speed and cost is reduced by using half the volume at all times during the procedure described by BD Bioscience and by counting 900 beads instead of the 1800 recommended.