



ENRICHMENT OF NK CELLS BY PANNING ON *ERYTHRINA CRISTAGALLI* PLATES

This procedure is a modification of the method for NK cell isolation described in the following reference:

Koren, H.S. and Argov, S. "Novel Approaches for Enrichment of NK Cells" in *Macrophage Biology* (Progress in Leukocyte Biology, Volume 4, 1985) pg. 695-705, (Reichard, Sherwood, and Kojima, M. ed) Alan R. Liss, New York.

A variety of methods are used to obtain enriched fractions of NK cells such as column adherence techniques, Percoll discontinuous gradients and monoclonal antibodies. The following procedure utilizes the lectin isolated from *Erythrina cristagalli* (Cat. No. L-1140), taking advantage of its ability to discriminate between NK cells and other mononuclear cells in a negative selection technique.

PROCEDURE:

- 1) Bacteriological plastic plates (60 mm) are coated with 2 ml of *Erythrina cristagalli* lectin in PBS, pH 7.8, at a concentration of 250 ug/ml for one hour at 24 °C.
- 2) Decant the unbound *Erythrina cristagalli* lectin and wash the plate thoroughly with PBS. (The decanted lectin solution may be stored at 4°C with 0.05% sodium azide and reused for further plating.)
- 3) Add 1×10^7 lymphocytes in 4 ml of RPMI-1640 or PBS for one hour at 4 °C.
- 4) After collecting the unbound cells (enriched NK fraction), the bound cells can be removed by the addition of 200 mM lactose, 0.5 mM EDTA.
- 5) Both the unbound and bound cells can be washed several times with RPMI or PBS containing 200 mM lactose to remove any trace amounts of lectin which might be present.