

Production of Lymphokines and Chemotactic Activity of Malaria Patient Monocytes

Principal Investigators : Robert A. Wells, MAJ, MSC
Katchrinnee Pavanand, M.D.
Sandor Zolyomi, SFC, USA
Barnyen Permpnich

Associate Investigators : Somchit Tulyayon
Prasit Sookto
Niphon Chuanak

OBJECTIVE : To develop assays for the production of lymphokines by malaria patient lymphocytes and for the measurement of chemotaxis of patient monocytes induced by such lymphokines.

BACKGROUND : The protective reaction to infection with malaria is notorious for its inefficiency. The immune response is characterized by excessive synthesis of immunoglobulin - most of which has no demonstrable anti-malarial activity. Although few investigations have been directed toward the cellular immunology of malaria, it has been reported that a concomitant state of immunosuppression stems from defective macrophage function (1).

It was reasoned that such defects might stem from defective responsiveness to monocyte chemotactic factors normally synthesized by lymphocytes. Such defects had been reported in other disease states such as the Wiskott-Aldrich syndrome (2) and chronic mucocutaneous candidiasis (3). As an extension of this rationale a preliminary battery of assays was conducted to test the migratory competence of monocytes across a membrane in response to lymphocyte associated chemotactic factors.

METHODS : The chemotactic test substances utilized were as follow : *E. coli* supernatant or factors (of lymphocyte origin) synthesized against human and simian malaria antigen or factors for the plant mitogens concanavalin A, phytohemagglutinin and purified protein derivative. For the chemotactic assays against *E. coli* supernatant 2×10^6 /ml mononuclear cells from either normal Thai donors or malaria patients were incubated at 37°C in 5% CO₂. Controls for these assays were normal media. For the chemotactic assays against the malarial antigens and the mitogens 2×10^6 /ml mononuclear cells from normal donors were incubated as above with the respective material over a wide range of concentrations. Controls for this series of assays incorporated the short term (non-incubation) exposure of mononuclear cells to each material prior to second-stage decanting of the supernatants for the chemotactic assay.

RESULTS : The results of chemotactic assays of normal and patient monocytes against *E. coli* supernatant and of normal monocytes against the above indicated materials are indicated in Table 1. The data suggests that there is demonstrable chemotactic activity by each parameter employed. It is equally clear that the chemotactic index in each case is much lower than had been hoped. While

these values might be enhanced by purification and concentration techniques it is felt that these requirements vrs time and manpower are disproportionate to the relative value of such work. It is thus our intention to make our data available to other in-house investigators and to abort further pursuit of this work in favor of research with higher priority. This project should therefore be considered complete.

REFERENCES :

1. Loose, L.D., Cook, J.A., and Diluzio, N.R. : Malarial Immunosuppression - A Macrophage Mediated Defect. in Basic Research in Malaria, E.H. Sadun ed, Proc. Helminth. Soc. Wash., p. 484, Vol. 39, Nov. 72 (Special Issue).
2. Altman, L.C., Snyderman, R., and Blaese, R.M. : Abnormalities of Chemotactic Lymphokine Synthesis and Mononuclear Leukocyte Chemotaxis in Wiskott-Aldrich Syndrome. J. Clin. Invest., 54:486-493, 1974.
3. Snyderman, R. et al. : Defective Mononuclear Leukocyte Chemotaxis : A Previously Unrecognized Immune Dysfunction Annls. Int. Med. 78:509-513, 1973.

Table 1. Monocyte Responsiveness to Chemotactic Factors

Supernatant origin	Monocyte origin	Total # of runs	Chemotactic index (C.I.) range mean
E. coli	Patient	3	1.91 - 3.26 2.4
E. coli	Normal	4	0.87 - 3.68 1.8
Concanavalin A	Normal	4	0.19 - 1.52 1.1
Phytohemagglutinin	Normal	6	0.8 - 2.2 1.5
Purified Protein Derivative	Normal	3	1.0 - 2.7 1.6
Malaria Antigen (human)	Normal	6	0.7 - 1.3 1.1
Malaria Antigen (simian)	Normal	2	1.0 - 1.2 1.1