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GAMMA INTERFERON PRODUCTION INDUCED BY ANTIGENS IN PATIENTS WITH LEPROSY AND AMERICAN CUTANEOUS LEISHMANIASIS

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Abstract. In this study, we measured gamma interferon production in mononuclear cell cultures from patients with diverse forms of leprosy and American cutaneous leishmaniasis. We studied patients with lepromatous, borderline lepromatous, borderline, and borderline tuberculoid forms of leprosy, as well as a Mitsuda-negative contact. In leishmaniasis we studied patients with localized cutaneous, mucocutaneous, and diffuse cutaneous forms of the disease. High correlation was observed between gamma interferon production and lymphocyte proliferation assays in both diseases. Resistant forms of both diseases showed significant reactivity, while the severe progressive forms were characterized by insignificant responses in both assays. Localized cutaneous leishmaniasis is characterized by variability in gamma interferon production, which may be of prognostic value in longitudinal studies.

Immune gamma interferon (IFN-γ) is produced during an immune response by antigen-specific T cells and probably also by natural killer (NK) cells recruited by the T cell product interleukin-2 (IL-2). IFN-γ was discovered and originally measured on the basis of its antiviral activity.[1] IFN-γ may act on macrophages to induce or "switch on" interleukin-1 (IL-1) release and secretion. This released IL-1 could then augment the proliferation of stimulated T cells.[2]

Leprosy and leishmaniasis are characterized by a broad spectrum of clinical manifestations, which depend in part upon the host's immune response.[3] While the immune mechanisms and antigen-specific immunological defects which characterize different clinical forms of disease are not fully understood, macrophage activation by IFN-γ probably represents a fundamental prerequisite for the elimination of these intracellular pathogens. Production of this lymphokine has been studied in leprosy[4, 5] but to our knowledge has not been measured systematically in American cutaneous leishmaniasis.

This preliminary study evaluates IFN-γ production in mononuclear cell cultures of patients with different clinical forms of these two diseases upon exposure to antigen.

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MATERIALS AND METHODS

Isolation of mononuclear cells

Mononuclear cells were isolated from heparinized peripheral blood by flotation over Ficoll-Hypaque gradients[6] and cultivated at a density of 2×10^5 viable cells/0.2 ml in microtiter plates. The cells were cultured in RPMI 1640 medium containing 10 U/ml penicillin, 100 μg/ml streptomycin, and 10% heat-inactivated pooled normal human AB serum.

Stimulation of lymphocytes

Leprosy patients. The antigens used were a) 20 μl of soluble extract of *Mycobacterium leprae*, 25 μg protein/ml; b) 20 μl purified *M. leprae*, 60×10^6 bac/ml; and c) 20 μl heat-killed bacillus Calmette-Guerin (BCG) (Connaught Laboratories Ltd., Willowdale, Ontario, Canada), 0.18 mg/ml.

M. leprae was purified from experimentally infected armadillo tissues by the Draper protocol.[7] Soluble antigen was prepared from purified bacilli by partial disintegration (8 passes through an Aminco French pressure cell at 10,000 lb/in²), elimination of bacillary debris by centrifugation at 39,000 × *g*, 4°C, 1 hr, and filtration of the supernate with a Millipore membrane, pore size 0.45 μ. Protein content was determined by the Lowry method. This antigen was facilitated

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