

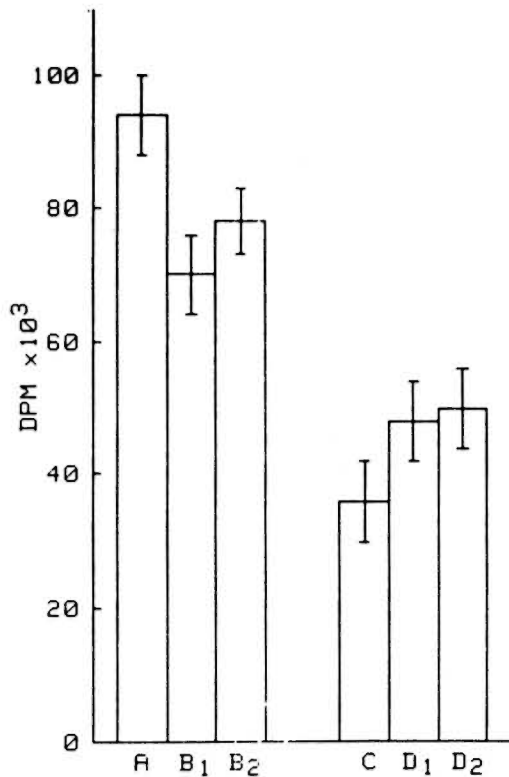
Adherent Suppressor Cells in Polar Lepromatous Leprosy

TO THE EDITOR:

Patients with polar lepromatous leprosy exhibit tolerance to *M. leprae* specific antigens in a variety of systems to test for delayed-type hypersensitivity and cell-mediated immunity. We wish to report results of preliminary investigations into the type of cells responsible for this immunological tolerance.

Mononuclear cells from three patients with lepromatous leprosy were isolated from whole blood by conventional methodology (1). The patients had been treated with dapsone (DDS) for one to two years and had not experienced complications dur-

ing the period of treatment. Adherent cells were removed from the mononuclear cell suspension in a two-step procedure. In brief, the mononuclear cells were cultured on plastic petri dishes in RPMI 1640 containing 20% heat inactivated fetal calf serum but without antibiotics for 2 hr at 37°C in 5% CO₂ and 98% humidity. Nonadherent cells were removed by decantation and incubated overnight under the same conditions. After overnight incubation, nonadherent cells were removed and set up in culture. The procedure was, in our hands, highly effective and resulted in a depletion of adherent cells, which are activated mac-



A = whole cells plus concanavalin A, mean \pm S.D., N = 3.

B₁ = whole cells plus concanavalin A plus Dharmendra, mean \pm S.D., N = 3, $p < 0.005$ between B₁ and A.

B₂ = whole cells plus concanavalin A plus *M. leprae*, mean \pm S.D., N = 3, $p < 0.010$ between B₂ and A.

C = cells depleted of adherent cells plus concanavalin A, mean \pm S.D., N = 3.

D₁ = cells depleted of adherent cells plus concanavalin A plus Dharmendra, mean \pm S.D., N = 3, $p < 0.005$ between D₁ and C.

D₂ = cells depleted of adherent cells plus concanavalin A plus *M. leprae*, mean \pm S.D., N = 3, $p < 0.005$ between D₂ and C.

THE FIGURE. The effect of Dharmendra antigen and *M. leprae* on the lymphocyte transformation of mononuclear cells from polar lepromatous leprosy patients in response to concanavalin A. Statistical tests were performed using the Student's *t* test.

rophages, and permitted us to retain for cultures a percentage of nonadherent cells showing monocyte characteristics by stain-

ing with peroxidase and nonspecific esterase. This percentage of $5.0 \pm 0.9\%$ is compulsory for lymphocyte blast transformation tests. Three day cultures were set up with concanavalin A using either unfractionated mononuclear cells or mononuclear cell suspensions depleted of adherent cells. Cells exposed to *M. leprae* specific antigens (either Dharmendra antigen or armadillo-derived integral *M. leprae*) were handled in an identical fashion except that they were exposed to the antigens for a period of three days before the addition of concanavalin A. The antigens remained in the cultures during the final three days' incubation with concanavalin A. The cultures were pulsed with ³H-thymidine and counted.

We observed (The Figure) that unfractionated mononuclear cells from these lepromatous cases are suppressed in their response to concanavalin A by either Dharmendra antigen or integral *M. leprae*; cells which have been depleted of adherent cells show enhanced ³H-thymidine incorporation in the presence of Dharmendra antigen or integral *M. leprae*. Thus, the cells responsible for suppression appear to be adherent cells which are activated macrophages. These macrophages or supernatants from their culture are able to suppress blast transformation in mixed leukocyte cultures with cells from the blood of healthy individuals (data not shown). This suggests the production of a "monokine" by these macrophages, but the mechanisms of this inhibitory effect are not clear and require further investigation.

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