

## Decreased Release of Interferon- $\gamma$ by Peripheral Blood Mononuclear Cells of Patients with Chronic Dermatophytosis in Response to Stimulation with Trichophytin

Sir,

One of the key functional parameters determining the immune response to an infecting organism is the nature of cytokines produced by T cells. Our present study was focused on interferon- $\gamma$  (IFN- $\gamma$ ) release by peripheral blood mononuclear cells (PBMC) in patients with chronic dermatophytosis, and the pathogenesis of chronic dermatophytosis is discussed in particular in relation to the association with a possible deficiency of this immunoregulatory cytokine.

We investigated 5 patients with dermatophyte infection (tinea pedis). All patients had *T. rubrum* infection, as demonstrated by a positive KOH examination and isolation of the causative fungi on Sabouraud's dextrose agar. Three patients, with a duration of infection of more than a year, were considered chronic cases. Peripheral venous blood was drawn from the patients and PBMC were isolated from the blood by density centrifugation. Trichophytin was prepared with *T. mentagrophytes* SM 0111 = RV 27961 (*Arthroderma vanbreuseghemii*) as reported previously (1).

PBMC ( $1 \times 10^6$ /ml), suspended in RPMI-1640 medium (GIBCO, Grand Island, New York, U.S.A) supplemented with 100 U/ml penicillin, 100  $\mu$ g/ml streptomycin, and 10% fetal calf serum, were cultured with and without trichophytin (50  $\mu$ g/ml) for 72 h at 37°C in a humidified atmosphere containing 5% CO<sub>2</sub>. Cell-free supernatants were collected and stored frozen at -70°C until needed. IFN- $\gamma$  activity in the culture supernatants was determined with a RIA test kit (Centocor, Melferne, PA, U.S.A).

When PBMC were incubated with trichophytin, high levels of IFN- $\gamma$  were detected in the culture supernatants of PBMC from the patients with non-chronic infection, as reported previously (2). In contrast, markedly lower levels were found in the chronically infected patients. When PBMC were incubated without trichophytin, there were no differences in the release of IFN- $\gamma$  (Fig. 1).

Our results indicate that the production of IFN- $\gamma$  by PBMC from the patients with chronic dermatophyte infection in response to stimulation with trichophytin was impaired in contrast to that of non-chronically infected patients. IFN- $\gamma$  plays an important role in the effector phase of the DTH reaction (3). In a previous study (2), we showed that IFN- $\gamma$  was produced in peripheral lymphocytes obtained from patients with non-chronic dermatophyte infection, suggesting that patients with non-chronic dermatophyte infections have circulating trichophytin-specific T lymphocytes capable of producing IFN- $\gamma$ , which is known to play a role in the development of the DTH reaction in the skin.

The DTH response is known to play a critical role in host resistance to dermatophyte infection, mainly because resolution of the disease is usually accompanied by the development of DTH (4). The effector mechanism of the DTH response involved in the eradication of dermatophytes from the skin is poorly understood. DTH-mediating T lymphocytes may act by

secreting cytokines which attract and/or activate macrophages and neutrophils (2, 5). These cells were found to be able to kill the dermatophyte. IFN- $\gamma$  was previously identified as a macrophage-activating factor (6). Activated macrophages may destroy the dermatophyte more efficiently. The alteration in the epidermis induced by the DTH reaction may allow serum inhibitory factors to come in contact with the dermatophyte existing in the stratum corneum. This contact may lead to the inactivation of the dermatophyte. DTH may accelerate epidermal proliferation, which facilitates the elimination of the dermatophyte from the stratum corneum with resultant desquamation. IFN- $\gamma$  was re-

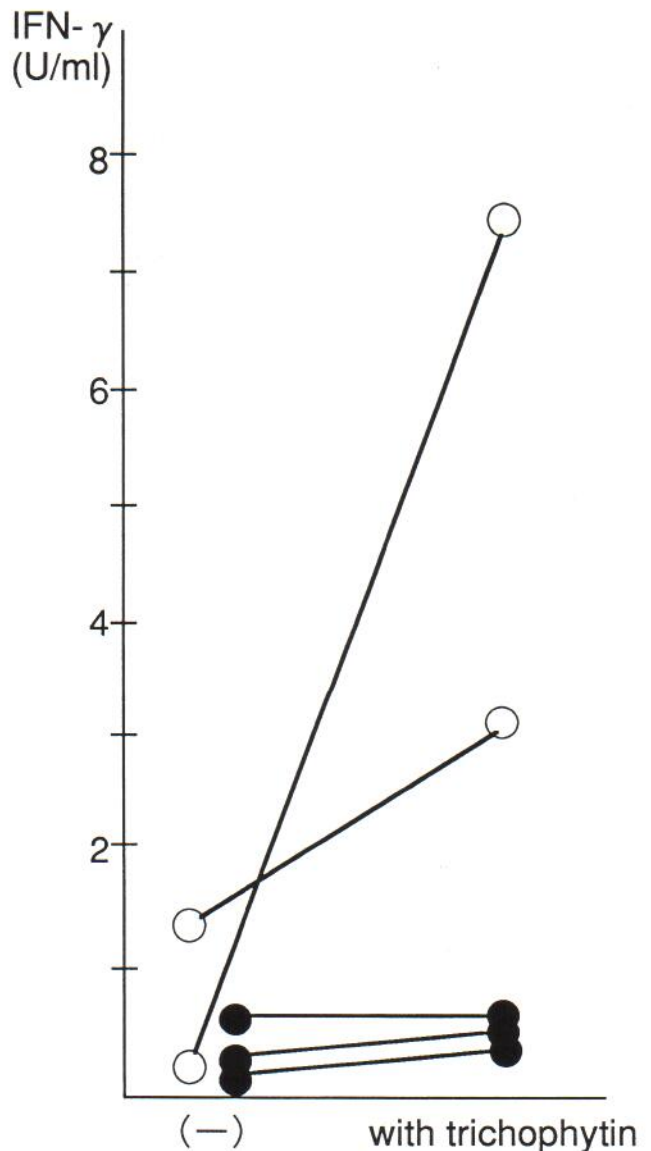


Fig. 1. IFN- $\gamma$  release by peripheral blood mononuclear cells (PBMC) from patients with chronic (●) and non-chronic (○) dermatophytosis. PBMC were cultured with and without trichophytin.

ported to contribute to keratinocyte hyperproliferation *in vivo* (7). In this way, the DTH response would act as an effector mechanism.

The chronicity in dermatophytosis may depend on a number of factors, such as immunogenicity and invasiveness of the infecting organism, the site of infection and the patient's immune response (8–10). Studies on the immune response in chronic dermatophytosis have been carried out. A defect of DTH would be important in determining the chronicity of dermatophyte infection, since persistence of infection appears to correlate with impaired or absent DTH. It has been reported that chronically infected patients show negative skin test and poor lymphocyte transformation test results. Thus, the association between T lymphocyte hyporeactivity and chronic dermatophytosis is well recognized. However, its significance is not clearly understood.

One of the key functional parameters determining the immune responses to infectious microorganisms is the nature of the cytokines produced by T lymphocytes. In the present study, *in vitro* T lymphocyte hyposensitivity to dermatophyte antigen was shown by measuring the release of the T cell-derived cytokine, IFN- $\gamma$ , which plays an important role in the effector phase of the DTH reaction. Our data support the hypothesis that individuals predisposed to chronic dermatophyte infection exhibit depressed DTH to dermatophyte antigen. It is possible that a decreased release of IFN- $\gamma$  at the site of infection might explain the inability of chronically infected patients to eradicate dermatophytes from the skin.

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