Acta Derm Venereol (Stockh) 1983; 63: 384–387

Basic and Interferon-augmented Natural Killer (NK) Cell Activity in Psoriasis

CHRISTER T. JANSÉN and MARKKU VIANDER

Departments of Dermatology and Medical Microbiology, University of Turku, Finland

Jansén, C T, Viander M. Basic and interferon-augmented natural killer (NK) cell activity in psoriasis. Acta Derm Venereol (Stockh) 1983; 63: 384–387.

The natural killer (NK) cell activity of lymphocytes from 24 psoriatic patients was compared with that of 34 normal controls and 19 patients with non-psoriatic dermatoses. In the range of effector/target cell ratios used (6.25 to 100), the basal NK cell activity level was similar in the three groups. Moreover, both interferon alpha (IFN-α) and interferon gamma (IFN-γ) augmented the NK cell activity to the same extent in psoriasis as in the two control groups. We conclude that this important natural cancer surveillance mechanism is functioning properly in psoriatic patients. *Key words: Psoriasis; NK cells; Interferon; Cancer immunity.* (Received December 22, 1982.)

C. T. Jansén, Department of Dermatology, University of Turku, SF-20520 Turku 52 (TYKS), Finland

Psoriasis is a chronically persisting disease needing prolonged treatment, often continuing for decades. Recently much attention has been paid to the possible carcinogenicity of psoriasis treatment modalities, and considerable effort is presently being expended on defining the inherent risks of different psoriasis treatments, such as photochemotherapy. There is, in parallel, an increasing awareness of the fact that host factors may be most important in determining the outcome of exposures to tumorigenic environmental factors. There is now ample evidence from animal experiments that immunological defence mechanisms may, in particular, determine the susceptibility to development of skin cancers (9).

Although host immune factors presumably play a similar role in the case of skin cancer development in man (10), the existence and strength of such immune mechanisms are difficult to evaluate. One exception is the recently characterized natural killer (NK) cell system, which is probably one of the major cancer surveillance systems in mammals, including man (5). This NK cell activity resides in a small subpopulation of spleen and peripheral blood lymphocytes, and is activated by the presence of interferon (IFN) (6).

To acquire a biological measure of the effectiveness of cancer surveillance mechanisms in psoriasis patients, we have assayed the NK cell activity level and its IFN augmentation in psoriatics and compared it with that of patients with other skin disorders and normal controls.

MATERIAL AND METHODS

Patients and controls

The study group consisted of 24 patients suffering from plaque-type psoriasis of long-standing duration (4–64 years, mean 18 years). The patients were neither receiving any systemic antipsoriatic remedies nor had they recently (≤3 months) undergone phototherapy or photochemotherapy. The ages of the psoriatics varied from 18 to 73 years, mean 44.1 years. Of the 23 patients, 15 were male.

The normal controls consisted of 34 healthy volunteers from among the hospital and laboratory personnel. The ages of the normal controls, 15 of which were male, varied between 24 and 42 years, mean 31 years. In addition, a control group of 19 patients suffering from skin disorders unrelated to psoriasis was analysed. The control patients included 4 cases of photodermatoses, 3 cases each of leg

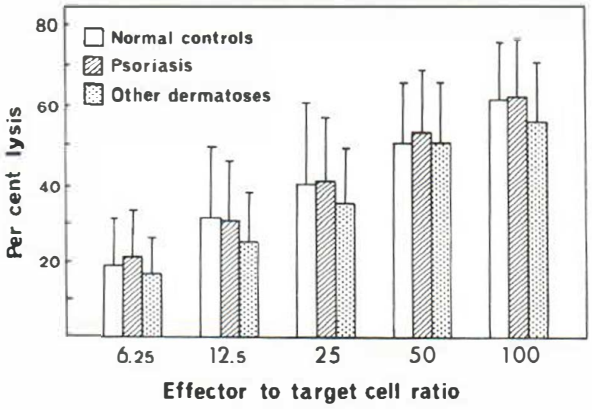

Fig. 1. NK cell activity levels, expressed as a percentage of maximal lysis. Mean values and standard deviations shown for the three population groups, at five different effector/target ratios.

ulcer and neurodermatitis, 2 cases each of atopic eczema, lichen planus and warts, and 1 case each of acne, pityriasis lichenoides, and bee allergy. In the 19 control patients,13 of whom were male, the ages ranged from 9 to 67 years, with a mean of 45.6 years.

NK cell determinations

Effector cells. Peripheral blood mononuclear cells were separated from 15 ml of venous blood by means of Ficoll-Isopaque (Pharmacia Fine Chemicals AB, Uppsala, Sweden) gradient centrifugation (1). The mononuclear cells were depleted of adherent cells by incubation for 60 min at 37°C in 5% CO_2 atmosphere on plastic tissue culture dishes (Sterilin Ltd., Teddington, England) in RPMI 1640 medium (Grand Island Biological Company, Grand Island, New York) supplemented with 10% fetal calf serum (FCS, Gibco Europe Ltd., Glasgow, Scotland) and gentamycin (1 μg/ml). The nonadherent cells were harvested and resuspended in fresh RPMI 1640 medium with 10%.

Cytotoxicity assay. The cell line K-562 was used as target in a 4-hour microcytotoxicity assay, as described earlier (12, 13). In brief, 2×10^6 target cells in 0.5 ml were labelled with 400 μCi ^{51}Cr isotope (The Radiochemical Centre, Amersham, England) for 1 hour at 37°C and then washed twice with the medium and resuspended in RPMI 1640 with 10% FCS at a concentration of 5×10^4/ml. The cytotoxicity assay was performed in round-bottom microplates (Nunc, Roskilde, Denmark). 5×10^3 ^{51}Cr-labelled target cells in 0.1 ml were mixed with 0.1 ml of effector cells in different effector/target (ET) ratios ranging from 6.25 : 1. The assays were set up in triplicate, and control wells containing only target cells were included in every plate to determine the maximum and spontaneous release. After incubation for 4 hours at 37°C in 5% CO_2 atmosphere the cell-free supernatants were collected using the Titertek Supernatant Collection System (Flow Laboratories, Irvine, Scotland) and counted in a well-type gamma counter (Wallac, Turku, Finland). The specific ^{51}Cr release was determined according to the following formula: (experimental release—spontaneous release/ maximal release—spontaneous release)×100. Maximal release was determined by incubating labelled K-562 cells in 5% sodium dodecyl sulphate (SDS).

Interferon. Sendai virus induced, partially purified human leukocyte interferon (IFN-α) was kindly provided by Dr. Kari Cantell, Central Public Health Laboratory, Helsinki, Finland. The production and purification procedures have been described previously (2). Similarly, a preparation of immune interferon (IFN-γ), produced in lectin-stimulated human peripheral blood mononuclear cell cultures, was provided by Dr Cantell. For use in the NK-assays, interferon was preincubated with the effector cells 10 min before adding target cells for the 4-hour cytotoxicity assay. The final concentration of IFN-alpha was 1000 U/ml and that of IFN-gamma, 100 U/ml in each well.

RESULTS

Basic NK cell activity

Fig. 1 gives the basic NK cell activities in the three study groups, at the five different effector to target (ET) cell ratios tested. In all study groups, the lytic response increased proportionally to the increase in effector cell concentration, indicating that the assay

system functioned properly. When the magnitude of the responses in the three study groups was compared, no significant differences were found.

Response to interferon

The effects of IFN-α and IFN-γ on the NK cell activities are recorded in Table I. In general, the NK cell activities in all study groups were augmented by interferon, although in a few individual cases non-responsiveness or slight inhibition was seen (data not shown). On the whole, the response in the three study groups were of equal magnitude (Table I), and IFN-alpha and IFN-gamma were equally effective. Although there was a slight trend for the IFN responses in psoriatic patients to run lower than those for the normal controls, this difference was not statistically significant.

DISCUSSION

Animal studies have implied that the NK cell system may function as an important "first line" cancer defence mechanism by detecting and destroying early clones of mutated cells (5). By showing that a normal level of basic NK cell activity is found in the peripheral blood of psoriatics, our study implies that this cancer surveillance system does operate properly in psoriasis suffers. This finding is in agreement with data recently presented by Hunyadi et al (7). In that study, as well as in ours, K-562 erythroblastoid target cells were used. In our study, however, five different effector/target cell ratios were used, ranging from 6.25 to 100, in contrast to the three ratios of 10 to 40 used by Hunyadi et al. (7).

Our study was extended to measure the augmentation of the NK cell activity by interferon, using both IFN-alpha (leukocyte IFN) and IFN-gamma (immune IFN). The IFN-induced augmentation is considered to depend on both a recruitment of active NK cells from inactive precursor cells, and a direct augmenting effect on the lytic activity of the mature NK cells (11). We found that this activation capacity was of the same magnitude in psoriatics as in the controls, which adds further support to the concept of an intact NK cell system in psoriatics.

The cellular IFN responsiveness in psoriasis may also be regarded in a larger biological context, since interferons are known to be potent inhibitors of cell proliferation (3), acting through specific cell surface receptors (4). Theoretically, interferon responsiveness might be altered, e.g. in rapidly proliferating psoriatic epidermal cells. Interestingly, IFN-α did not influence the DNA synthesis rate of human psoriatic skin explants cultured in intraperitoneal diffusion chambers in mice (8). While our present study includes no data on epidermal cells, the normal IFN responsiveness of the lymphocytic cells may be interpreted as evidence against any general impairment in IFN response mechanisms in psoriasis.

Table I. *Augmentation of NK cell activities by interferon-alpha (IFN-α, 1000 U/ml) and interferon-gamma (IFN-γ, 100 U/ml)*

Results are expressed as augmentation indices ± standard deviation at an effector/target cell ratio 25 : 1. Figures in parentheses indicate numbers of persons tested

Population	IFN-α	IFN-γ
Normal controls	1.50±0.50 (34)	1.46±0.39 (20)
Psoriatics	1.43±0.44 (24)	1.32±0.35 (15)
Other patients	1.46±0.31 (19)	1.37±0.52 (13)

REFERENCES

1. Bøyum A. Separation of leucocytes from blood and bone marrow. Scand J Clin Lab Invest 1968; 21: Suppl. 97: 1–109.
2. Cantell K, Hirvonen S. Large-scale production of human leucocyte interferon containing 10^8 units per ml. J Gen Virol 1978; 39: 541–543.
3. Creasey AA, Bartholomew JC, Merigan TC. Role of G_0–G_1 arrest in the inhibition of tumour cell growth by interferon. Proc Natl Acad Sci USA 1980; 77: 471–1475.
4. Grollman EF, Lee G, Ramos F, Lazo PS, Kaback HR, Friedman RM, Kohn LD. Relationships of the structure and function of the interferon receptor to hormone receptors and establishment of the antiviral state. Cancer Res 1978; 38: 4172–4185.
5. Herbermann RB, Holden HT. Natural killer cells as antitumor effector cells. J Natl Cancer Inst 1979; 62: 441–445.
6. Herbermann RB, Djeu JY, Kay HD, Oraldo JR, Riccardi C, Bonnard GD, Holden HT, Fagnani R, Santoni A, Puccetti P. Natural killer cells. Characteristics and regulation of activity. Immunol Rev 1979; 44: 43–70.
7. Hunyadi J, Szegedi G, Nigra TP. Natural killer cell activity in psoriasis vulgaris. Arch Dermatol Res 1981; 271: 107–109.
8. Kariniemi A. Effect of human leucocyte interferon on DNA synthesis in human psoriatic skin cultured in diffusion chambers. Acta Pathol Microbiol Scand Sect A 1977; 85: 270–272.
9. Kripke M. Immunology of UV-induced skin cancer. Photochem Photobiol 1981; 32: 837–839.
10. Marshall V. Premalignant and malignant skin tumours in immunosuppressed patients. Transplantation 1974; 17: 272–275.
11. Targan S, Dorey F. Interferon activation of "prespontaneous-killer" (pre-SK) cells and alteration in kinetics of lysis of both "pre-SK" and active SK cells. J Immunol 1980; 124: 2157–2161.
12. Viander M., Uksila J, Lassila O, Jansén C.T. Natural killer cell activity in atopic dermatitis. Arch Dermatol Res 1982; 274: 283–288.
13. West WH, Cannon GB, Kay HD, Bonnard GD, Herbermann RB. Natural cytotoxic reactivity of human lymphocytes against a myeloid cell line: characterization of effector cells. J Immunol 1977; 118: 355–361.