EFFECT OF IMMUNODEPRESSION ON VIRUS MULTIPLICATION AND INTERFERON AND ANTIBODY FORMATION IN ANIMALS

V. D. Solov'ev,* A. M. Sorokin, and N. R. Gutman

UDC 616.988-092.9-06:612.017.1-064]-092

The use of massive doses of azathioprine under the experimental conditions used had no significant effect on multiplication of type A/PR8 influenza virus and Coxsackie A6 virus. Meanwhile, serum interferon and antibody production was totally suppressed. The blood leukocytes of mice receiving azathioprine did not produce interferon *in vitro*. The results indicate that a leading role in the pathogenesis of virus infections developing after organ transplantation when massive doses of immunodepressants are used for "rejection crises" is played by disturbance of the formation of the factors of antiviral immunity. The results may be of practical importance for the development of optimal immunodepression programs for organ transplantation.

KEY WORDS: immunodepression; virus infections; leukocytes; interferon; organ transplantation.

Evidence is now accumulating of an increase in the frequency of development of viral, fungal, and bacterial infections and the development of malignant tumors in patients undergoing transplantation of certain organs [5-8, 10, 11]. The widespread use of immunodepressants, although justifiable from a clinical point of view, also has disadvantages. One disadvantage of every immunodepressant is that it lacks selectivity of action. All not only depress the ability of the host to reject the graft, i.e., transplantation immunity, but they also give rise to a more or less marked response of the immunity system as a whole. The most characteristic example of this is the damage done to the immunological mechanisms for protection of the host against various infections. Depending on the type of model, the scheme of administration of the immunodepressant, and the substances used, the formation of cellular and humoral factors of resistance to infections is variously inhibited. The writers showed previously that interferon (IF) formation and antibody (AB) production differ in their sensitivity to the action of identical immunodepressants [3, 4].

This paper gives the results of a study of the multiplication of certain viruses and also of IF and AB formation in animals with depressed immunoreactivity.

EXPERIMENTAL METHOD

Noninbred mice weighing 20-25 g were used in the experiments to induce IF and AB formation.

Newcastle disease virus (NDV), strains EM2 and PR8 of type A influenza virus, and Coxsackie A6 virus were used as inducers. The titers of the viruses were: influenza type A/EM2 10^7 EID_{50}, 320 ha.u., A/PR8 10^7 EID_{50}, 1280 ha.u., and NDV 10^8 CPD_{50}, 640 ha.u.

*Academician of the Academy of Medical Sciences of the USSR.

N. F. Gamelaya Institute of Epidemiology and Microbiology, Academy of Medical Sciences of the USSR, Moscow. Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 80, No. 12, pp. 50-53, December, 1975. Original article submitted March 26, 1975.

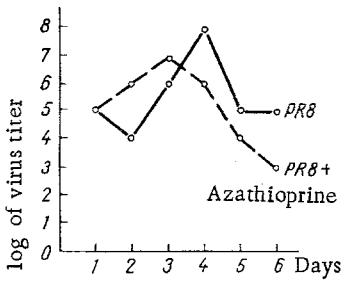

Fig. 1

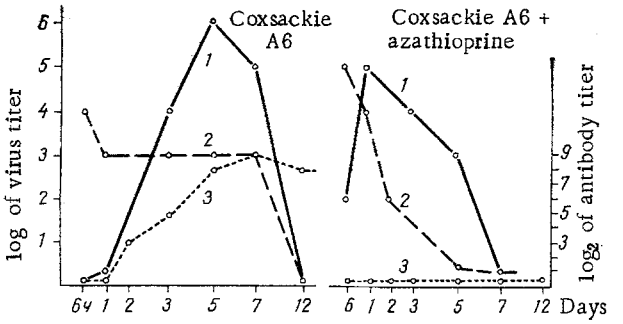

Fig. 2

Fig. 1. Effect of azathioprine on multiplication of type A/PR8 influenza virus in lungs of mice weighing 5-6 g. Virus injected intranasally in a dose of 1 LD_{50} (0.03 ml), azathioprine given by mouth in a dose of 0.1 ml daily for 7 days, starting from day of infection with virus.

Fig. 2. Effect of azathioprine on multiplication of Coxsackie A6 virus and antibody production. Coxsackie A6 virus injected intravenously in a dose of 1 ml (10^{10} LD_{50}), azathioprine given by mouth in a dose of 0.1 ml daily for 7 days starting from day of infection with virus: 1) multiplication of virus in heart; 2) in blood; 3) antibody production.

The methods of induction and determination of IF and AB, of administration of azathioprine, and of determining the multiplication of type A/PR8 influenza and Coxsackie A6 viruses were described earlier [1-4].

EXPERIMENTAL RESULTS AND DISCUSSION

The writers showed previously that azathioprine, in a dose of 1250 mg/kg, completely suppresses anti-NDV AB formation in mice and lowers the serum IF level considerably [4]. It will be clear from Table 1 that a similar pattern was found when various strains of type A influenza virus were used as inducers.

Blood leukocytes of mice receiving azathioprine did not produce IF in response to induction by NDV (less than 1:4 compared with 1:128 in the control), in confirmation of earlier observations. When type A/PR8 influenza virus, adapted to mice, was used it was found that multiplication of the virus in the lungs receiving azathioprine did not differ significantly from its multiplication in the control group (Fig. 1).

Similar results indicating absence of any marked effect of azathioprine on virus multiplication were obtained in experiments with Coxsackie A6 virus (Fig. 2).

The results thus show that intensive immunodepressive therapy even with massive doses of azathioprine (lethal or close to lethal for mice) had no significant effect under the experimental conditions used on multiplication of the various viruses. Meanwhile IF and AT formation were disturbed to some degree or other. The results obtained with these experimental models thus suggest that a leading role in the pathogenesis of infections in patients with depressed immunoreactivity is played by the disturbance of the immunological mechanisms of resistance to infections. In particular, as the writers also showed previously, IF production by blood leukocytes *in vitro* is affected. Similar results, indicating depression of the IF-producing ability of the blood leukocytes of persons undergoing kidney transplantation followed by immunodepressive therapy have recently been obtained by Rytel and Balay [12]. Meanwhile, in "rejection crises" azathioprine is used in very high and near-toxic doses, although for only a short time. As the results of the present experiments show, these doses not only inhibit transplantation immunity, but also inhibit IF formation. The possibility cannot be ruled out that the high frequency of provocation of cytomegalovirus infection after transplantation could be connected with the inhibition of IF formation, for IF is known to restrict multiplication of cytomegalovirus [9]. The comparative importance of cellular and humoral factors of protection in the pathogenesis of virus infections varies very considerably. In acute infections, despite its very slight or even lack of effect on virus multiplication, azathioprine depresses the formation of antivirus im-

TABLE 1. Effect of Azathioprine on IF and AB Formation

Inducer virus	IF titer (in units)	AB titer (in HIT)*
Influenza A/EM2	32	160
« A/EM2+azathioprine	8	0
« A/PR8	16	640
« A/PR8+azathioprine	<4	0
NDV	4096	160
NDV+azathioprine	512	0

*Hemagglutination inhibition test.

munity. In some infections multiplication of viruses in animals with depressed immunoreactivity is, on the contrary, intensified.

During prolonged administration of immunodepressants their effect may be cumulative, so that antivirus immunity is inhibited even more. Azathioprine affects the cells of the lymphoid system much more than dividing cells of any other type, so that the cellular substrate of immunogenesis and of virus multiplication can also be differentiated in different virus infections.

The results described above thus point to the need for a more detailed study of IF and AB formation after organ transplantation and during proliferation of viruses in patients treated with immunodepressants, with the object of establishing optical schemes of administration of immunodepressants alone or in various combinations. Considering that IF production by blood leukocytes *in vitro* correlates with its formation *in vivo* [2], this process should also be studied in connection with different schemes of immunodepression.

Since immunodepressants can activate latent infectious (cytomegalovirus, viruses of the herpes group), a promising method of prevention of virus infections complicating the post-transplantation period could be by the timely administration of IF, because of its broad and nonspecific spectrum of antiviral action; no other single preparation can give such an effect.

LITERATURE CITED

1. N. R. Gutman and V. D. Solov'ev, Vopr. Virusol., No. 3, 281 (1969).
2. V. D. Solov'ev, T. A. Bektemirov, and A. E. Gumennik, Vopr. Virusol., No. 5, 531 (1967).
3. A. M. Sorokin, in: Problems in Radiation Immunology and Microbiology [in Russian], Moscow (1969), pp. 41-42.
4. A. M. Sorokin, Vopr. Virusol., No. 4, 443 (1973).
5. H. K. Anderson and E. S. Spencer, Acta Med. Scand., 186, 7 (1969).
6. J. E. Craighead, J. B. Hanshaw, and C. B. Carpenter, J. Am. Med. Assn., 201, 725 (1967).
7. T. C. Eickhoff, D. B. Olin, R. J. Anderson, et al., Transplant Proc., 4, 693 (1972).
8. R. B. Hill, B. E. Dahrling, T. E. Starzl, et al., Am. J. Med., 42, 327 (1967).
9. D. J. Land, M. T. Thomas, and J. Gresser, C. R. Acad. Sci. (Paris), 268, 3137 (1969).
10. M. H. Lessof, Postgrad. Med. J., 118, 327 (1972).
11. I. Penn, Malignant Tumors in Organ Recipients, Springer-Verlag, New York (1970).
12. M. W. Rytel and J. Balay, J. Infect. Dis., 127, 445 (1973).