time of the organism's exposure to the preparations be prolonged.

REFERENCES

1. G. A. Belokrylov, *Zh. Mikrobiol.*, № 4, 23-28 (1976).
2. G. A. Belokrylov, I. V. Molchanova, and O. Ya. Popova, *Byull. Eksp. Biol. Med.*, **108**, № 11, 584-587 (1989).
3. E. D. Gol'dberg, G. V. Karpova, V. V. Zhdanov, *et al.*, *Pat. Fiziol.*, № 1, 10-11 (1990).
4. V. N. Zakharov, A. V. Karaulov, V. V. Sokolov, and V. N. Frash, in: *Changes in the Blood System for Exposure to Radiation and Benzene* [in Russian], Novosibirsk (1990).
5. R. V. Petrov, V. N. Shvets, and V. M. Man'ko, *Dokl. Akad. Nauk SSSR*, **204**, № 2, 489-492 (1972).
6. V. A. Tutel'yan and L. V. Kravchenko, *Mycotoxins* [in Russian], Moscow (1985).
7. I. Ya. Uchitel', *Macrophages in Immunity* [in Russian], Moscow (1978).
8. T. G. Shemerovskaya, T. D. Zorina, N. K. Artemenko, *et al.*, *Immunologiya*, № 1, 44-46 (1987).
9. A. V. Shtelzner, in: *Immunologische Arbeitsmethoden* (Ed. by H. Friemel *et al.*), Jena (1976).

ONCOLOGY

Increased Resistance to P3-X63-Ag8.653-MOPC Plasmacytoma after Exposure to Repeated Stress

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The antitumor effect of multiply repeated stress is demonstrated. The maximal inhibitory effect on tumor growth is achieved by a moderate physical load (swimming), the efficacy of immobilization stress is somewhat lower, while intensive physical load is considerably less effective.

Key Words: *chronic stress; tumor growth*

Reports that exposure to stress before transplantation or induction of a tumor may have an antitumor effect are of great interest [1-5,7]. The inhibition of tumor growth has been achieved by electrical shock [3], cold stress [4], handling [7], and periodic separation of newborn mice from the mother [5]. The effect has been demonstrated on various models of experimental tumor process: MSV virus inoculation [3], transplantation of Gross virus-induced lymphoma [4], Walker 256 carcinoma [7], and Ehrlich ascitic carcinoma [5]. However, there are also some reports that stress stimulates [8,9] or has no effect [6] on tumor growth.

Bearing in mind that the present data on this topic are contradictory and limited, in the present study we attempted to develop an experimental model for studying the effect of different kinds of stress in the pretransplantation period on antitumor resistance.

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MATERIALS AND METHODS

The experiments were carried out on 164 female BALB/c mice aged 6 to 8 months. The P3-X63-

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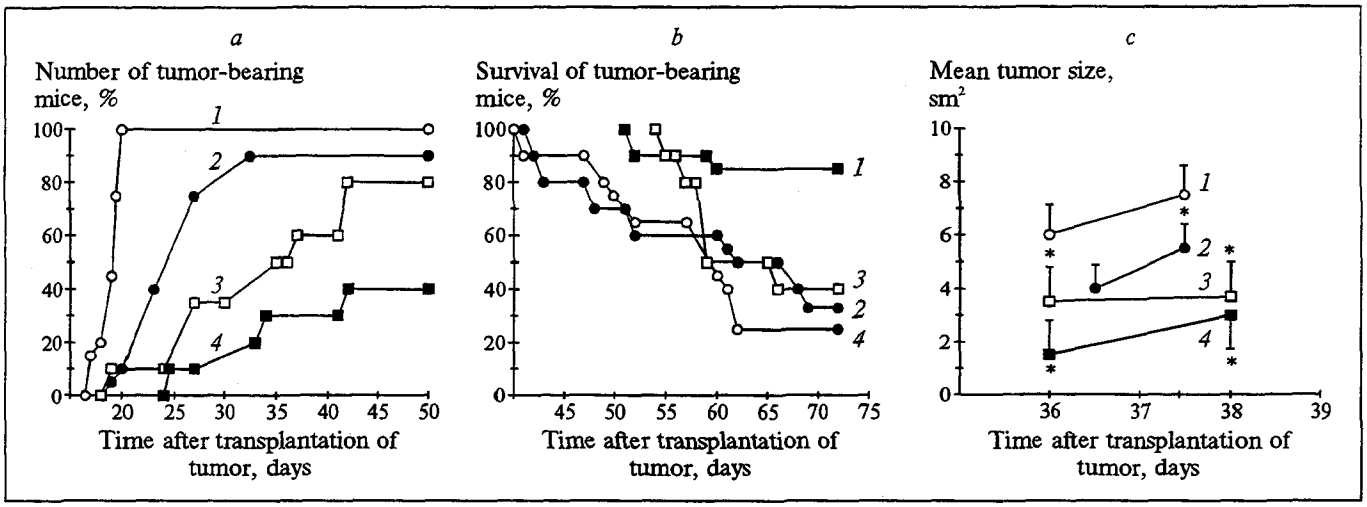

Fig. 1. Effect of intensive and moderate physical loads (swimming) and immobilization stress on growth of P3-X63-Ag8.653-MOPC plasmacytoma in mice (*a*), survival of tumor-bearing mice (*b*), and tumor size (*c*). *1*) control group; *2*) group S1, intensive physical load (swimming); *3*) group S2, immobilization stress; *4*) group S4, moderate physical load. *: $p<0.05$

Ag8.653-MOPC syngeneic plasmacytoma strain was obtained from the Research Institute of Human Morphology, Russian Academy of Medical Sciences. Tumor cells for transplantation were isolated from ascitic fluid, diluted to a concentration of 8×10^3 cells/ml in medium 199, and injected subcutaneously in a volume of 0.5 ml to the right femur. Tumor size was determined by the formula: A×B, where A and B are two perpendicular dimensions. The effect of stress on the development of the experimental tumor process was evaluated from the following parameters: the dynamics of tumor appearance, the death of tumor-bearing mice, and the size of the tumor. The following groups of mice (8-19 mice per group) were used: C (control), intact female BALB/c mice; S1, intensive physical load (swimming): the mice were forced to swim for 20 min at a water temperature of 37°C 3 times a day over 4 days; S2, immobilization stress: the mice were placed in plastic tubes with food and water ad libitum for 6 hours over 4 days; S3, moderate physical load: the mice were made to swim during 6 min (3 min at 28°C, after which they were promptly transferred to 38°C water) 3 times a day over 4 days.

The mice of the S1, S2, and S3 groups were subjected to stress according to the above scheme over 2 weeks, and after 15-16 days the tumor cells were injected. The reproducibility and reliability of the results were evaluated in 3 independent experiments performed in spring (experiment № 1), in summer (experiment № 2), and in winter (experiment № 3). Statistical processing of the results was carried out using the Wilcoxon-Mann-Whitney test.

TABLE 1. Parameters Characterizing Tumor Process in Female BALB/c Mice in Control and after Intensive (S1) and Moderate (S3) Physical Loads and Immobilization Stress (from Three Independent Experiments)

Parameter	№ of experiment	Group of mice			
		C	S1	S2	S3
Total of tumor-bearing mice, %	1	92.2	83.3	73.7	36.4
	2	100.0	93.3	80.0	87.5
	3	47.3	20.2	22.2	13.3
Time of tumor emergence in 50% of mice, days	1	19.5	26.0	37.0	80.0
	2	20.0	24.5	25.5	21.0
	3	>40.0	>80.0	>80.0	>80.0
Median of latent period, days	1	20.0	27.0	27.0	33.5
	2	21.0	25.0	25.5	21.0
	3	22.0	44.0	19.5	30.0
Total fraction of tumor-bearing survivors after 45 days, %	1	92.3	66.6	100.0	100.0
	2	70.0	86.6	90.0	72.5
	3	100.0	100.0	100.0	100.0
Life-span of 50% of tumor-bearing mice, days	1	58.0	60.5	58.0	60.0

RESULTS

The results of the first experiment, presented in Fig. 1, show that multiply repeated stress exposures (moderate and intensive physical loads and immobilization stress) before transplantation of P3-X63-Ag8.653-MOPC syngeneic plasmacytoma cells lead to an inhibition of tumor growth. This manifests itself in a prolonged latent period and in a decreased number of tumor-bearing mice in comparison with the control group. Reliable differences in tumor size between the experimental and control groups were also found. The effect of exposures to stress on the life-span of the tumor-bearing mice was negligible. The revealed effect of post-stress inhibition of tumor growth was examined in analogous experiments № 2 and 3.

Table 1 summarizes the results of all three experiments. The data confirm the reliability and reproducibility of the antitumor effect of all kinds of stress used. Analysis of the results of all three experiments revealed that the number of tumor-bearing mice after exposure to the intensive physical load (group S1) was lower by 24.9% ($p \leqslant 0.05$), after immobilization stress (group S2) by 31.3% ($p \leqslant 0.05$), and after the moderate physical load (group S3) by 48.4% ($p \leqslant 0.05$) than that in the control group (taken as 100%). All kinds of stress used reliably delayed the time of tumor emergence in 50% of the mice: 1.51-fold in group S1, 1.72-fold in group S2, and 2.38-fold in group S3. The intensive physical load (group S1) resulted in a reliable increase of the median of the latent period. The post-stress changes in the number of tumor-bearing mice still alive 45 days after tumor transplantation (experiments Nos. 1, 2, and 3) and the data on the life-span of 50% of the tumor-bearing mice suggest a negligible effect of stress on the late stages of the tumor process.

The reproducibility and reliability of the positive effect of stress in the form of intensive and moderate physical loads (swimming) and immobilization during the early stages of experimental tumor processes were confirmed in three independent experiments. The most pronounced inhibitory effect on tumor growth was achieved by the moderate physical load (swimming); immobilization stress was less effective, while the antitumor effect of the intensive physical load (swimming) was considerably lower. The experimental model developed allows for comparing different kinds of stress in terms of the efficacy of tumor growth inhibition.

REFERENCES

1. L. V. Volkova, in: *Stress: Adaptation and Dysfunction*, IV All-Union Symposium, Abstract, Kishinev (1991), p. 147.
2. R. Adler and S. B. Freidman, *J. Comp. Physiol.*, **59**, 361 (1965).
3. A. Amkraut and G. F. Solomon, *Cancer Res.*, **32**, 1428 (1972).
4. S. R. Burchfield, S. C. Woods, and M. S. Elich, *Physiol. Behav.*, **21**, 537 (1978).
5. R. C. La Barba and J. L. White, *Phychosom. Med.*, **33**, 458 (1971).
6. B. H. Hewberry, *J. Nat. Cancer Inst.*, **61**, 725 (1978).
7. G. Newton, C. G. Bly, and C. McCrary, *J. Nerv. Ment. Disease*, **134**, 522 (1962).
8. L. J. Peters and H. Kelly, *Cancer*, **39**, 1482 (1977).
9. V. Riley, *Science (Wash., D.C.)*, **189**, 465 (1975).