STIMULATING ACTION OF CARNOSINE ON HEMATOPOIETIC STEM CELLS

E. G. Kurella, V. V. Mal'tseva, L. S. Seslavina, and S. L. Stvolinskii

UDC 577.336.616-017.11

KEY WORDS: carnosine; colony formation index; survival rate; radioprotector

Among the many biological effects of the natural histidine-containing dipeptide, carnosine [12, 7], its ability to protect the body against the action of radiation is noteworthy [13, 5]. The radioprotective effect of carnosine may have a number of different causes. One of them, put forward by Goncharenko and Kudryashov [1], might be connected with the ability of carnosine to influence background levels of endogenous radioprotectors serotonin and histamine. However, it has been found that although carnosine modifies biogenic amine levels in the mouse spleen after irradiation, this is not the cause of its radioprotective action, for injection of carnosine before irradiation reduces the mortality of the animals but does not affect serotonin and histamine concentrations [5].

The basis of the effect of carnosine may perhaps be its antioxidative action, manifested as ability to interact with active forms of oxygen and with hydroxyl radicals [7, 8, 9]. However, the possibility of a membrane-stabilizing effect likewise cannot be ruled out, as has been demonstrated on blood cells [3]. In order to obtain a wider picture of the mechanism of action of carnosine as a radioprotector, we studied the effect of this substance on hematopoietic stem cells, when administered in different ways.

EXPERIMENTAL METHOD

In experiments to determine the effect of carnosine on the survival rate of irradiated animals, adult noninbred albino mice (weighing 20 g) were used; carnosine was given with the drinking water at different stages, before and after irradiation (dose 5.0-5.5 Gy). Each series consisted of 10 animals.

To discover the effect of carnosine on hematopoietic stem cells the method of endogenous splenic colonies [10] was used. Adult (CBA × C57BL)F_1 mice weighing 24-26 g were used in the experiments. Carnosine was injected intraperitoneally in a dose of 1 mg per mouse, 30 min, 1 h, and 24 h after irradiation. The animals were irradiated in a single dose of 6 Gy ($LD_{50/30}$) with ^{60}Co γ-rays. On the 8th day after irradiation the animals were killed and the number of readily visible colonies formed on account of colony-forming units which remained viable, was counted on the surface of the spleen.

The carnosine used in the experiments was obtained preparatively from beef [12].

EXPERIMENTAL RESULTS

After a single dose of x-ray irradiation of noninbred mice with a dose of 5.0-5.5 Gy the mean length of survival of the animals was 12-17 days, and their survival rate by the 30th day was 11% after a dose of 5.5 Gy and 30% after 5.0 Gy. Administration of carnosine with the drinking water in a dose of 50 or 100 mg/kg body weight increased both these

Department of Biochemistry, Faculty of Biology, M. V. Lomonosov Moscow University. (Presented by Academician of the Academy of Medical Sciences of the USSR S. E. Severin.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 112, No. 7, pp. 52-53, July, 1991. Original article submitted November 25, 1990.

 0007-4888/91/0007-0966$12.50

TABLE 1. Effect of Carnosine on Survival Rate of Animals after X-Ray Irradiation

Group of animals	Dose of irradiation, Gy	Dose of carnosine, mg/kg body weight	Number of series	Mean life span, days	Survival rate, %
Irradiation without protection	5,0	—	1	17	30
	5,5	—	4	12±2	11±5
Carnosine 1 day before irradiation	5,0	50	1	27	60
	5,5	100	1	26	80
	5,0	50	4	20±3	42±10
Carnosine 2 days before irradiation	5,0	50	2	15±1	26±5
Carnosine 1 day after irradiation	5,0	50	1	30	100
	5,0	100	1	26	70
	5,5	50	4	16±4	25±8
Carnosine 2 days after irradiation	5,5	50	2	18±4	58±5

TABLE 2. Effect of Carnosine (50 mg/kg body weight) on Activity of Colony Formation by Hematopoietic Stem Cells of (CBA × C57BL)F_1 Mice

Group of animals	No. of animal 1 2 3 4 5 6 7 8 9 10 number of colonies in spleen 8 days after irradiation	Mean value	Index of activation of colony formation
Control (irradiated)	1 3 1 3 0 4 4 1 3 2	2,0±1,3	—
Carnosine 30 min before irradiat	4 7 3 2 4 2 2 5 9 13	5,1±3,4	2,6
Carnosine 1 h after irradiation	2 5 3 2 3 10 3 1 0 4	3,3±2,6	1,7
Carnosine 24 h after irradiation	3 1 3 4 2 3 5 3 4 2	3,0±1,1	1,5

parameters, and the larger dose was less effective. This also was observed when the anticarcinogenic action of carnosine was studied [10, 11].

Comparison of the effects of carnosine depending on the time of its administration showed that its action is best if the interval between irradiation and administration of carnosine is shortened. For instance, if carnosine was given after 24 h it was more effective than after 48 h, and if carnosine was given 2 days before irradiation, hardly any effect was found (Table 1).

We concluded from analysis of these results that for the optimal protective action to be exhibited, carnosine should be given either immediately before irradiation or soon after. This policy was adopted when carnosine was given in the experiments on inbred animals.

The results of the second series of experiments are given in Table 2. Whatever the mode of its administration to the animals carnosine had a stimulating effect on endocolony formation. Carnosine acted most effectively on stem cells when given immediately before irradiation (30 min before), when the number of colonies in the spleen of the experimental mice was increased by more than 2.5 times. However, a significant effect also continued to be observed when carnosine was given 1 h or 24 h after irradiation (the index of activation of colony formation was 1.7 and 1.5 respectively). Whereas the positive effect in the first case can be linked with some degree of probability with the direct action of carnosine as a quencher of free radicals, the postradiation protective effect must perhaps be explained by the direct action of carnosine on the hematopoietic system and, consequently, on the immune system.

Japanese workers have found a number of effects of carnosine on both T and B systems of immunity [11, 13]. It has also been shown that carnosine and related compounds (β-alanine, homocarnosine) accelerate granulation and stimulate repair processes, which has been demonstrated both experimentally and clinically [7, 13, 4]. The stimulating action of carnosine on hematopoiesis, which we have demonstrated, accounts for the marked increase in the survival rate after sublethal irradiation of animals receiving carnosine. Different methods of administration of carnosine will perhaps enable it to be used for both prophylactic and therapeutic purposes.

LITERATURE CITED

1. E. N. Goncharenko and Yu. B. Kudryashov, Moscow (1980), p. 176.
2. O. P. Lyskova, A. M. Dupin, et al., Claim for Invention No. 4695347 (1989).

3. V. V. Mal'tseva, L. S. Seslavina, N. K. Matveeva, and G. T. Sukhikh, Biol. Nauki, No. 6, 148 (1990).
4. S. A. Silaeva, N. V. Gulyaeva, et al., Byull. Éksp. Biol. Med., No. 2, 180 (1990).
5. S. E. Severin, A. A. Boldyrev, et al., Radiobiologiya, **30** (1990).
6. O. Aruoma, M. Laughton, and B. Halliwell, Biochem. J., **264**, 863 (1989).
7. A. A. Boldyrev, Int. J. Biochem., **22**, 129 (1990).
8. A. A. Boldyrev, A. M. Dupin, E. V. Pindel, and S. E. Severin, Comp. Biochem. Physiol., **89B**, 245 (1988).
9. R. Kohen, Y. Yamamoto, K. Cundy, and B. Ames, Proc. Nat. Acad. Sci. USA, **85**, 3175 (1988).
10. K. Nagai and T. Suda, Surgery, **100**, 815 (1986).
11. K. Nagai and T. Suda, Meth. Find. Exp. Clin. Pharmacol., **10**, 497 (1988).
12. S. E. Severin, International Congress of Biochemistry, No. 1, 45 (1964).
13. S. Shimanaka, 31st International Congress of Physiology, Helsinki (1989), p. 3306.
14. J. E. Till, E. A. McCulloch, and Z. A. Siminovitch, Radiol. Res., **18**, 95 (1963).

CHANGES IN ACTIVITY OF ADENOSINE DEAMINASE AND ANTIOXIDATIVE ENZYMES IN PATIENTS WITH DUST DISEASES OF THE LUNGS

I. B. Nemechek, B. L. Lur'e, and B. T. Velichkovskii

UDC 616.24-003-07:[616.153.1:577.152.191].193]-074

KEY WORDS: adenosine deaminase; superoxide dismutase; catalase; pneumoconiosis; chronic dust bronchitis

Morphological, biochemical, and clinical manifestations of the pneumoconioses give indirect proof that immune mechanisms are involved in the development of this disease. Despite many attempts to discover antigenic changes characteristic of pneumoconioses in the lungs and other changes which can be regarded as probable specific manifestations of immune mechanisms in the development of this pathology, we have as yet no integral picture of the disturbances of the immune system [1]. Further investigations into this problem are accordingly indicated.

Enzymic disturbances of purine metabolism in immunodeficiency states were first discovered in 1972 [11]. One of the key enzymes of purine metabolism is adenosine deaminase (ADA), which is widely distributed in human and animal tissues [5]. Its principal substrates are adenosine and deoxyadenosine. Normally ADA decomposes purines and is involved in the formation of hypoxanthine, a source for purine resynthesis. It is now known that a decrease in ADA activity accompanies many primary and secondary immunodeficiency states. Highest ADA activity in man is observed in lymphoid tissue, erythrocytes, and tissues of the gastrointestinal tract, where ADA activity is necessary for the utilization of alimentary purines, taken in with the food. Maximal ADA activity is found in cortisone-sensitive thymocytes. Some increase in ADA activity is observed in the lymphocytes of persons "developing" an immune response [10].

Recently the important role of active forms of oxygen (AFO) in the development of dust diseases of the lungs has been discovered. The main sources of AFO in the body are phagocytic cells, platelets, eosinophils, and endotheliocytes. AFO play a dual role in the function of aerobic organisms. They are responsible for realization of the mechanism of the bactericidal effect, and for the formation of biologically active substances (prostaglandins, leukotrienes); they are also

N. I. Pirogov Second Moscow Medical Institute. Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 112, No. 7, pp. 53-56, July, 1991. Original article submitted October 7, 1990.

 0007-4888/91/0007-0968$12.50