CHANGES IN PROTEIN METABOLISM IN EXPERIMENTAL BURNS AGAINST A BACKGROUND OF AMINAZINE ACTION

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Following burns, the protein metabolism of the organism undergoes considerable changes of which the most typical is the intensification of protein breakdown [6]. Attempts to directly affect the protein metabolism of burned subjects do not always lead to the desired results, as the regulatory mechanisms of protein metabolism in the intact organism have not been adequately studied.

The question of the participation of the nervous system in the tissue metabolism of protein is extremely unclear. The study of this problem advances with the use in research practice of methods and substances which permit local changes in the functional state of specific sections of the nervous system. One of these substances is aminazine, in a fairly broad spectrum of the action of which it is possible to distinguish its selective inhibition of the adrenergic components of the reticular formation of the brain stem [1,2,4,5,12]. Apparently, aminazine blocks not only the ascending but also the descending effects of the reticular formation [3], with which are probably connected the various metabolic disorders caused by it. Most work on this question is concerned with the effect of aminazine on the metabolism of lipids [11,15,17-19], glucose and glycogen in the organism [14] and on mineral metabolism [22]. Findings on the effect of aminazine on protein metabolism are less specific. A number of authors indicate some change in the enzyme activity of blood and tissues [5,7,16]. No changes were observed in the amino acids and protein fractions of blood [20]. It was established [10,13] that aminazine did not change the intensity of incorporation of S^{35}-methionine into the tissue proteins of animals.

Considering the relative stability of protein metabolism during the action of aminazine in normal conditions, it was of interest to study whether the aminazine blockade of reticular formation affects the protein metabolism of the organism exposed to burns. The present work was devoted to a study of this question.

EXPERIMENTAL METHODS

The trials were with male rabbits 2.5-3 kg bodyweight. There were 4 series of trials: I) on healthy animals; II) on healthy animals given aminazine (7 mg/kg) twice daily for 5 days; III) on rabbits with burns caused by immersion of the rear extremities in boiling water for 20 sec; IV) on rabbits subjected to standard burning and given aminazine 40 min before burning, and then according to the scheme indicated for series II.

The investigation was carried out on the 5th day after burning. The intensity of tissue autolysis was studied in all animals by Folin's method after incubation for 2 h at pH 3.8, the intensity of incorporation of S^{35}-methionine into proteins after injection into rabbits 18 h before slaughter at the rate of 10,000 imp/min per 1 g bodyweight, the content in some tissues of RNA and DNA separated by the method of Schmidt and Tannhauser and estimated on a spectrophotometer.

EXPERIMENTAL RESULTS AND DISCUSSION

From the data in the table, it follows that, on the 5th day after burning in a number of organs and tissues of rabbits (liver, intact muscles and muscles in the zone of burning, intestine), the processes of autolysis were intensified

Effect of Aminazine on Autolysis of Tissues, Incorporation of S^{35}-Methionine into Tissue Proteins, and Content of Nucleic Acids Following Burning

	Control rabbits	Control rabbits given aminazine	P	Burned rabbits	Burned rabbits given aminazine	P
	M+m			M+m		
Autolysis of tissues (in μg tyrosine per 1 mg protein)						
Liver	7,5 ± 0,25	8,4 ± 0,8	>0,2	9,9 ± 0,6	6,1 ± 0,1	<0,01
Muscles:						
intact	0,38 ± 0,16	0,36 ± 0,06	>0,5	0,94 ± 0.09	0,52 ± 0,04	<0,001
in zone of burning	0,34 ± 0,02	0,36 ± 0,06	>0,5	1,8 ± 0,25	1,76 ± 0,02	>0,5
Kidney	10,9 ± 0,5	11,8 ± 1,2	>0,2	11,9 ± 1,0	9,4 ± 0,6	<0,05
Adrenals	5,8 ± 0,8	4,1 ± 0,65	>0,1	5,4 ± 0,8	5,4 ± 0,7	—
Intestine	4,6 ± 0,1	5,6 ± 0,9	>0,2	8,0 ± 0,5	5,3 ± 0,8	<0,02
Brain	2,2 ± 0,2	1,6 ± 0,6	>0,2	2,2 ± 0,2	1,9 ± 0,2	>0,2
Incorporation of S^{35}-methionine (in impulses/min/5 mg protein)						
Serum	208 ± 27	211 ± 43	>0,5	467 ± 60	586 ± 93	>0,2
Liver	284 ± 22	238 ± 31	>0,2	296 ± 28	324 ± 23	>0,5
Muscles:						
intact	48 ± 1,8	45 ± 11	>0,5	30 ± 9	23 ± 4,6	>0,5
in zone of burning	—	—	—	40 ± 13	44 ± 13	>0,5
Kidney	628 ± 81	629 ± 44	>0,5	538 ± 78	593 ± 87	>0,5
Adrenals	451 ± 22	425 ± 24	>0,2	275 ± 63	373 ± 56	>0,2
Intestine	671 ± 20	625 ± 15	>0,05	479 ± 90	368 ± 78	>0,2
Brain	94 ± 12	76 ± 12	>0,5	73 + 9	56 ± 10	>0,2
Content P DNA (in mg %)						
Liver	19,5 ± 1,2	15,5 ± 1,5	>0,05	18,5	17,3	
Intact muscles	3,6	3,6		4,2	4,0	
Content P DNA (in mg %)						
Liver	41,9	41,9		56,0	54,7 ± 0,7	
Intact muscles	6,1	5,6 ± 0,25		7,2 ± 0,71	6,8 ± 0,33	
Number of trials	7	6		10	10	

as we have already reported in previous work [6]. Thus, if in the liver of a healthy animal the autolytic activity of the tissue was 7.5 μg tyrosine per 1 mg protein, after burning it was 9.9 μg. The difference was statistically significant ($P < 0.01$). The differences in autolytic activity in the intact musculature of the healthy and burned rabbit were respectively 0.34 and 0.94 μg tyrosine per 1 mg protein ($P < 0.01$). In the burned muscles, autolysis increased even more sharply. A considerable increase in autolytic activity in these experiments was noted in the intestine: 4.6 μg tyrosine per 1 mg protein in normal and 8 μg tyrosine per 1 mg protein in burned rabbits ($P < 0.001$). In the burned rabbits injected with aminazine, the autolytic processes were not intensified at all or autolysis was reduced in comparison with that in burned rabbits not given aminazine. The table also shows the action of aminazine on healthy animals. It was shown that, in this case, aminazine had no effect on the intensity of the autolytic processes.

To show that the autolysis-inhibitory action of aminazine following burning is not connected with the direct effect of aminazine on the cell and the enzymic processes in it, the autolysis of some tissues was studied in the presence of different amounts of aminazine (1, 10, and 100 μg per 1 ml) in the incubation medium.

It was found that the addition of aminazine in the trials with tissues of healthy and burned rabbits did not reduce the extent of autolysis. Thus, if the autolytic activity of the liver of a healthy rabbit was 6.7 μg tyrosine per 1 mg protein, in the presence of 1, 10, and 100 μg aminazine in the incubation medium, it ranged from 6.2 to 8 μg tyrosine per 1 mg protein. In the burned rabbit, the extent of autolysis of 9.5 μg tyrosine per 1 mg protein in the presence of aminazine ranged from 9.3 to 10 μg tyrosine per 1 mg protein. The same relationship was observed also in other organs studied (muscles, kidney, intestine).

Parallel with the studies of autolysis processes to some extent characterizing the activity of proteolytic systems in the live animal, the synthesis of tissue proteins was studied in the same trials from the incorporation into them of S^{35}-methionine, as was the content of nucleic acids in some tissues. The table shows that, on the 5th day after the

rabbit was subjected to burning, the incorporation of S^{35}-methionine into the proteins of intact muscles, kidney, adrenals and the walls of the small intestine was inhibited by 25-30%. The amount of RNA was increased in the liver and that of RNA and DNA in muscles. No effect of aminazine was noted in these experiments. It can be concluded from the trials that injection of aminazine had no effect on the processes of protein synthesis or on the content of nucleic acids in tissues in healthy or burned animals. As aminazine, in any concentration studied, did not inhibit autolytic processes in vitro, we assume that the inhibition of autolysis in burned animals observed in the trial is due not to its direct action on tissue enzymes, but is mediated by the nervous system. The change in the intensity of the breakdown processes of tissue protein following the change in the functional state of reticular formation by aminazine indicates the relation of the fine mechanisms of the intermediate metabolism of proteins to the regulatory influence of the nervous system.

In connection with the differing effect of aminazine action on autolytic processes in healthy and burned rabbits, two basic aspects are admissable. It may be thought that the action of aminazine is poor when the condition of the nervous system is normal and has a considerably stronger effect on reticular formation when it is stimulated [2], which can occur following profuse pathological afferent impulsation, characteristic of burns. On the other hand, it can be suggested that, in the healthy organism, regulation of the normal level of breakdown of tissue proteins is provided for also following limitation of impulsation from reticular formation, which was observed in the trials when healthy rabbits were injected with aminazine. But, apparently, the intensified breakdown of tissue proteins after burning was attended by supplementary impulsation from reticular formation which may be reflected by the protein metabolism as a result of the stimulation of other sections of the nervous system, the activation or the occurrence of neuro-endocrine reactions. Independently of the possible concrete mechanisms connecting protein metabolism with the functional state of reticular formation, the injection of aminazine in that case may normalize the breakdown processes of the tissue proteins following burning, as was the case in our studies. As the injection of aminazine in normal conditions or with burning did not change the nucleic acid content of tissues and the intensity of protein synthesis, it may be thought that the regulation of those processes occurs without the participation of reticular formation.

The aim of the experiments was to study some stages of the regulatory processes of protein metabolism following burns, but the question of the administration of aminazine to burned subjects has by no means been cleared up. In view of the many contradictory findings on the clinical effectiveness of injecting aminazine following burning, we cannot consider one isolated finding of a reduction in the intensity of the autolytic processes as an indication for the use of the preparation.

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