NATURE OF THE TOXICITY OF THE SERUM AND ORGANS OF RATS AFTER THERMAL BURNS

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The serum, liver, and kidneys of a rat 24 h after a thermal burn of degree IIIb acquire marked toxic properties, which increase progressively on the 5th and 8th days. The toxic properties of the liver and kidneys are independent of the blood contained in them and are manifested in organs washed free from blood. Activity of cathepsins in the serum, liver, and kidneys rises later than the toxicity appears: it is increased 5 days, and further increased 8 days after burning.

The work of Fedorov [5] has shown that a "burn toxin" is present in the serum of burned persons and animals. Later investigations [1, 6, 9, 11] have confirmed that blood serum after thermal burns and extracts from burned skin possess toxic properties, and if injected into a healthy animal they give rise to severe toxic manifestations terminating in the recipient's death. The origin and properties of the "burn toxin" have been studied intensively and are at the present time the subject of lively discussion. The discussion has centered in particular on the connection between the burn toxin and proteolytic enzymes [12], whose activity is increased in burned tissues and in intact organs after burns [3].

The objects of the investigation described below were as follows: to discover whether not only the serum and burned skin but also the internal organs of burned animals possess toxic properties, and to study the dynamics of the appearance of toxic properties and the increase in activity of the proteolytic enzymes in the serum, liver, and kidneys.

EXPERIMENTAL METHOD

A flame burn of degree IIIb covering 15-20% of the body surface (exposure 50 sec) was inflicted on albino rats weighing 150-200 g. Burned and control (intact) rats were killed 1, 5, and 8 days after burning. Under sterile conditions the organs were removed and blood collected in a sterile vessel. To exclude the effect of admixture of blood during the subsequent determination of the toxicity and proteolytic activity in the organs, the organs were perfused with sterile Ringer's solution. The method of perfusion developed by the writers enabled this to be done without a period of anoxia of the organs. The perfusion was carried out through arteries and veins, including the portal vein. The toxicity and proteolytic activity was thus determined first before perfusion in the left lobe of the liver, and later after perfusion in the remainder of the liver. In the kidneys the tests were carried out before and after perfusion on rats of different groups. The tissue was homogenized in a sterile glass homogenizer, and the toxicity of the homogenates and serum was determined by the blood culture method [4]. The toxic effect was determined from the inhibition of migration of leukocytes of experimental cultures on the addition of the test material to them, relative to control cultures, in which Ringer's solution was added to the nutrient medium. An increase in migration of the experimental cultures relative to the control was expressed as a positive percentage, and inhibition (toxic effect) as neg-

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TABLE 1. Toxicity and Proteolytic Enzyme Activity of Serum and Organs of Burned Rats (M ± m)*

Time after burning (in days)	Number of animals	Serum		Liver				Kidneys			
		toxicity	activity of proteolytic enzymes	toxicity		activity of proteolytic enzymes		toxicity		activity of proteolytic enzymes	
				in unperfused organ	in perfused organ	in unperfused organ	in perfused organ	in unperfused organ	in perfused organ	in unperfused organ	in perfused organ
Control (intact rats)	20	14±4	10±3	11±4	8±2,4	5,7±0,4	5,7±0,4	12±3	11±2,4	8,7±0,7	12±1
1	8	—22±4,3	12±3	—9±3	—9±3,3	7,0±1,5	7,6±2	—12±2	—14±4	11,4±1,5	12,9±2,0
5	8	—28±0,3	26±1	—15±3	—16±1	8,1±0,3	8,3±1,5	—12±2	—12±4	10,6±0,5	9,5±0,5
8	10	—28±0,3	30±4	—25±3	—26±4	9,1±1,0	10,0±2	—20±2	—20±3	11,6±1,0	11,6±0,8

*Toxicity expressed in conventional units, cathepsin activity in g tyrosine/mg protein.

ative. Activity of the proteolytic enzymes (cathepsins) was determined in glycerol extracts of the organs relative to hemoglobin [8].

EXPERIMENTAL RESULTS

The results are given in Table 1. It will be noted that neither toxicity nor cathepsin activity was changed after perfusion of the organs and complete removal of the blood. Consequently, all the results reflect the toxicity and cathepsin activity of the organs themselves and not of the blood serum. A well-marked toxicity appeared in the serum, liver, and kidneys 24 h after burning. No significant increase in the activity of "acid" proteolytic enzymes could be found at this period.

By the end of the first day the normal enzyme activity in the serum was evidently restored after its initial increase during the first minutes and hours after burning [2, 10]. On the 6th-7th day a marked and prolonged rise of activity of proteolytic enzymes usually began in the organs. In the present experiments 5 days after burning the activity of proteolytic enzymes increased significantly in the serum and liver, while in the kidneys a tendency for cathepsin activity to increase was observed. By this time the toxicity of the serum and liver had increased considerably.

The toxic properties of the serum, liver, and kidneys of the burned rats were increased still further after 8 days, and the proteolytic enzyme activity in these organs and the serum also was increased.

Since toxic properties of the serum and internal organs appeared much earlier than the increase in activity of the enzymes catalyzing protein breakdown, the burn toxin cannot be identified with the tissue acid proteinases.

However, the subsequent parallel increase in toxicity and cathepsin activity may indicate some connection between activation of protein breakdown and the toxic properties of the tissues of the burned organism.

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