ON THE PATHOCHEMICAL CHARACTERISTICS OF ACUTE TOXIC FATTY DYSTROPHY OF THE LIVER

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Fatty infiltration of the liver, developing during toxic hepatitides, has been regarded until now as a result of mobilization of fat from the depots and its entrance into the liver due to depletion of the glycogen of that organ [2,3,6].

Along with the excessive inflow of fat, its inadequate removal from the liver is of major significance in the pathogenesis of fatty infiltration of this organ. According to the data of Recknagel et al. [10,11], the action of hepatotoxic poisons disrupts the so-called secretory functioning of the liver. The authors understand this latter to involve the capacity of the liver to release into the blood stream those triglycerides which have been formed in the organ from higher, free (non-esterified) fatty acids (NEFA), coming from the fat depots. They base their hypothesis on investigations with triton administration, which selectively inhibits the egress of triglycerides from the plasma, and causes an elevation of its level in the blood, at the same time not changing the triglyceride concentration in the liver. Subcutaneous injection of rats with carbon tetrachloride two hours before the administration of the triton prevented the elevation of the triglyceride level in the plasma, but also caused a rapid increase in its concentration in the liver. In subsequent investigation, Recknagel and coworkers showed that, with the administration of hepatotoxic poisons, the elevation of the concentration of triglycerides in the liver is accompanied by a reduction in its serum level. In the opinion of the authors, the toxic hepatitides are associated with a disturbance in the formation of lipoprotein complexes in the liver, as a result of which the triglycerides cannot leave this organ, and thus, they accumulate in it.

It should be noted that even in 1947, S. M. Leites [2], investigating the relation between the phospholipids and total lipids in the liver, indicated that not only was excessive entrance of fat important in the pathogenesis of toxic fatty infiltration of the liver, but also inhibition of its outflow, due to a relatively insufficient increase in the formation of phospholipids, which play an important role in the discharge of fat from the liver.

The following questions remain unelucidated in the pathogenesis of toxic fatty infiltration of the liver: 1) is the leading factor the mobilization of fat from the depots even at early stages in the development of liver damage, or is the primary factor the accumulation of fat in the liver as a result of disruption of its excretion?; 2) is the mobilization of fat from the fat depots accomplished in the form of triglycerides as such, or do NEFA exit from the fatty tissue after preliminary splitting of the triglycerides within the tissue? This specific mechanism of fat mobilization from the depots as a source of energy is accepted by a number of investigators [8,12,13], and also in our laboratory on the basis of experiments in which we investigated lipolysis in fatty tissue and the concentration of NEFA in the serum subsequent to the action of fat-mobilizing factors [4,5].

In addition, it remains an open question as to whether the changes in the lipolytic activity of the liver are important as a possible factor in the pathogenesis of its fatty dystrophy, since disruption of the splittting of triglycerides in this organ impedes the formation of phospholipids from the products of the splitting of fat and fatty acids.

We set out to study, at various stages in the development of toxic fatty infiltration of the liver, the lipolytic activity of the fat tissue, the concentration of NEFA and of total lipids in the serum, and also the lipolytic activity of the liver in comparison with a change in the concentration of total lipids in this organ.

Changes in the Concentration of Total Lipids in the Liver and its Lipolytic Activity, the Lipolytic Activity of the Fat Tissue, and also the Concentration of NEFA and Total Lipids in the Serum, in Control Experiments and after Single and Repeated Injections of CCl_4 (Mean Data: M ± m)

Dose of CCl_4	Duration after first injection	No. of animals	Total lipids in the liver (in g-%)*	Lipolytic activity of the fat tissue (in microequiv./ /ml/gram)†	Serum		Lipolytic activity of the liver (in microequiv./ml/ /gram†
					NEFA (in milliequiv./ /liter)	total lipids (in mg %)	
Control	—	12	19.87 ± 0.35	4.15 ± 0.12	1.03 ± 0.02	228.9 ± 10.98	18.69 ± 0.28
0.3/100 g	4 h	16	23.3 ± 0.41 p< 0.001	3.3 ± 0.4	1.3 ± 0.08 p< 0.01	274.0 ± 10.2 p< 0.01	20.11 ± 0.72 p< 0.05
Control	—	16	20.0 ± 0.17	3.75 ± 0.11	0.97 ± 0.05	234.5 ± 11.95	19.57 ± 0.49
0.15-0.3/100 g	48 h	40	27.8 ± 0.73 p< 0.001	5.23 ± 0.34 p< 0.001	1.13 ± 0.05 p< 0.02	264.0 ± 13.00	17.84 ± 0.33 p< 0.01
Control	—	14	20.4 ± 0.35	3.9 ± 0.16	0.95 ± 0.03	239.0 ± 6.24	20.08 ± 0.65
0.15-0.3/100 g (2 injections, with a day between)	fifth day	22	31.9 ± 1.05 p< 0.001	3.24 ± 0.3	0.88 ± 0.04	233.3 ± 11.13	19.13 ± 1.11
Control	—	6	19.6 ± 0.11	3.87 ± 0.12	1.03 ± 0.03	236.5 ± 9.59	20.36 ± 0.2
0.3/100 g (3 injections, every other day)	7th day	12	34.7 ± 1.81 p< 0.001	5.74 ± 0.71 p< 0.02	1.20 ± 0.1	215.4 ± 7.7	21.66 ± 0.42

* Calculation based on the dry weight of the liver tissue.

† The lipolytic activity of the fat tissue and the liver was determined from the difference between the concentrations of NEFA before and after incubation (150 min).

EXPERIMENTAL METHOD

As the subject of the investigations, we used male white rats, weighing 150-280 g, maintained on the normal laboratory diet.

Toxic fatty infiltration of the liver was caused by subcutaneous injection of carbon tetrachloride (CCl_4) in a dosage of 0.15-0.3 ml per 100 g of weight of the animal, with intervals between the injections of 1 day. The animals were sacrificed 4 and 48 h after the first injection, as well as 24 h after the 2nd, and 24 h after the 3rd, injections of CCl_4 (i.e., on the 5th and 7th days after initiation of the poisoning).

In both the experimental and control animals, we determined: 1) the concentration of total lipids in the liver by extraction of liver tissue, dried to a constant weight, with dichlorethane, in a Soxhlet apparatus; 2) the lipolytic activity of the epididimal fat tissue by the method of Gordon and Cherkes, with certain changes [4]; 3) the lipolytic activity of the liver, using tween-60 [4]; 4) the concentration of total lipids in the serum, according to the method of Kunkel [9]. The concentration of NEFA was determined by the method of Dale [7].

EXPERIMENTAL RESULTS

Only 4 h after the first injection of CCl_4, we noted an increase in the concentration of total lipids in the liver from an average of 19.87 to 23.3 g-% (see table). However, this increase in the level of total lipids was not connected with an increase in the lipolytic activity of the fat tissue. On the contrary, this index showed a tendency toward decrease. Thus, the increase in the concentration of total lipids in the liver 4 h after injection of CCl_4 is not caused by activation of fat mobilization from its depots.

At this stage of development of toxic fatty infiltration of the liver, the concentration of NEFA in the serum increased; the reason for this may be the observed increase in the lipolytic activity of the liver. The concentration of total lipids in the serum also increased.

Forty-eight hours after the first injection of CCl_4, the concentration of total lipids in the liver rose. In this case, there was an elevation in the lipolytic activity of the fat tissue, and in the serum concentration of NEFA. The lipolytic activity of the liver dropped. The concentration of total lipids in the serum remained unchanged. Thus, the alteration in the investigated indices suggests that, at this stage in the development of fatty dystrophy of the liver, the increase in its concentration of total lipids is caused by the activation of fat mobilization from the depots, and possibly, by inhibition of the lipolysis of triglycerides in the liver.

After the 2nd injection of CCl_4 (5th day after initiation of the poisoning), the concentration of fat in the liver continued to rise. However, this increase was not accompanied by either an increase in lipolysis in the fat tissue, or by an elevation in the concentration of NEFA or total lipids in the serum. Obviously, in this period of development of fatty dystrophy of the liver, the fat accumulation occurred as a result of the disruption of its egress from the organ.

Seven days after initiation of the poisoning, i.e., after the 3rd injection of CCl_4, the concentration of fat in the liver increased still further. We noted a parallel activation of lipolysis in the fat tissue, and a tendency toward elevation of the serum concentration of NEFA. The amount of total lipids in the serum and of lipolysis in the liver were in the normal range. Thus, at this stage of fatty dystrophy of the liver, we again observed activation of the fat mobilization from the depots.

In analyzing the obtained data, above all we must point out the regular and gradual increase in the amount of fat in the liver, subsequent to CCl_4 poisoning, which was observed even at 4 h after initiation of the poisoning.

Apparently, three factors play a role in the pathogenesis of this acute, toxic, fatty infiltration of the liver: activation of fat mobilization from the depots, inhibition of its egress from the liver (according to Recknagel and coworkers, a disruption of its "secretory" functioning), and inhibition of the lipolysis within the liver of esters from the higher fatty acids. At different stages in the development of fatty dystrophy of the liver, one of these factors becomes the predominant action. At 4 and 96 h after the initiation of the poisoning, the major factor is the disturbance in excretion of fat from the liver; at 48 h after the first injection and 24 h after the 3rd injection of CCl_4, the accumulation of fat in the liver runs parallel to the activation of its mobilization from the fat depots.

SUMMARY

Subjecting rats to either single or repeated injections of CCl_4 (0.15-0.3 ml-% per 100 g of body weight) leads to a gradual rise in the total lipid content of the liver; this is detectable as early as 4 h after the first injection. Changes in the lipolytic activity of the fat tissue are phasic in nature (there is a rise 48 h after the first and 24 h after the third CCl_4 injection, with a parallel rise in the serum non-esterified fatty acids). There is no increase in the lipolytic activity of the fat tissue 4 h after the first injection or 24 h after the second CCl_4 injection. The lipolytic activity of the liver increased 4 h, and decreased 48 h, after the first CCl_4 injection; this hepatic function showed no change at other stages in the development of toxic fatty dystrophy of the liver.

Evidently, three factors play a role in the pathogenesis of acute toxic fatty dystrophy of the liver, i.e., activation of fat mobilization from the depots, inhibition of fat excretion from the liver (according to Recknagel, a disturbance in "secretory" functioning of the liver), and inhibition of lipolysis of the higher fatty acid esters in this organ. The action of one of the mentioned factors predominates at different stages in the development of fatty dystrophy of the liver.

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
