

ACTIVITY OF SERUM AND LIVER ENZYMES
AT VARIOUS STAGES OF DEVELOPMENT
OF EXPERIMENTAL CIRRHOSIS OF THE LIVER

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Liver damage was produced in rats by injection of CCl_4 . With the change from an acute parenchymatous lesion of the liver to a chronic lesion and cirrhosis, changes take place in the activity of various serum enzymes. Activity of transferases drops (alanine aminotransferase more rapidly than aspartate aminotransferase), activity of sorbitol dehydrogenase falls sharply, that of ornithine carbamoyltransferase falls slowly, and activity of alcohol dehydrogenase rises. The activity of all these enzymes in the liver falls.

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The character of changes in serum enzyme activity and the diagnostic importance of its determination in chronic forms of hepatitis and cirrhosis have not yet been adequately studied.

In this investigation we studied the principles governing changes in activity of various serum enzymes during the change from an acute to a chronic lesion and during development of cirrhosis of the liver.

EXPERIMENTAL METHOD

Experiments were carried out on 260 male albino rats weighing 250-300 g. Liver damage was produced by subcutaneous injection of carbon tetrachloride in a dose of 0.5 ml/100 g body weight in an equal volume of sunflower oil. Administration of CCl_4 to all animals began at the same time and the compound was injected every 3 days for 2.5 months, the animals then being sacrificed at various times after the first

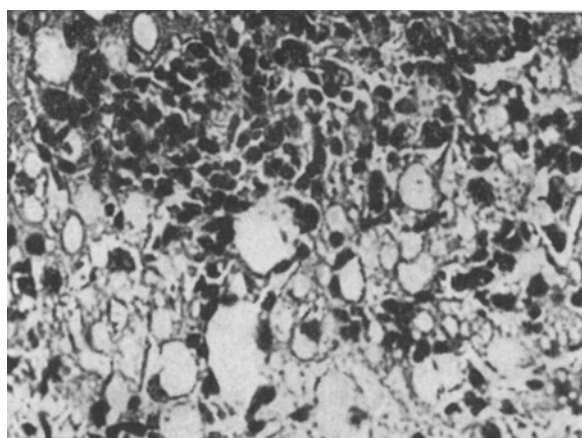


Fig. 1. Well-marked fatty infiltration of liver tissue, infiltration by numerous lymphocytes and histiocytes, and disappearance of the cytoplasm of single liver cells. 12 days after beginning of CCl_4 administration, hematoxylin-eosin, 200 \times .

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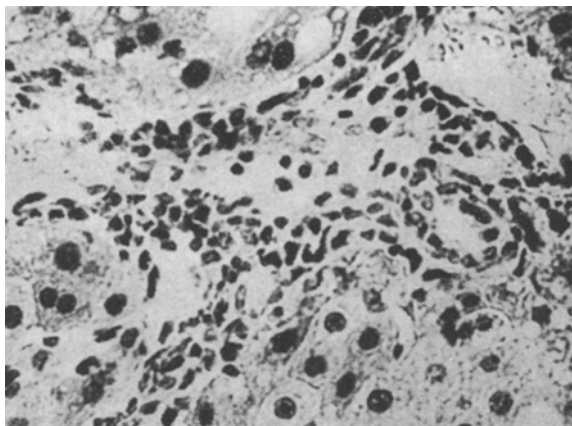


Fig. 2. Connective-tissue bands in liver tissue with small, newly formed blood vessels and inflammatory infiltration in these bands. Marked parenchymatous degeneration. Fatty infiltration and disappearance of cytoplasm of nearby liver cells. Solitary binuclear cells can be seen. 24th day. Hematoxylin-eosin, 200 \times .

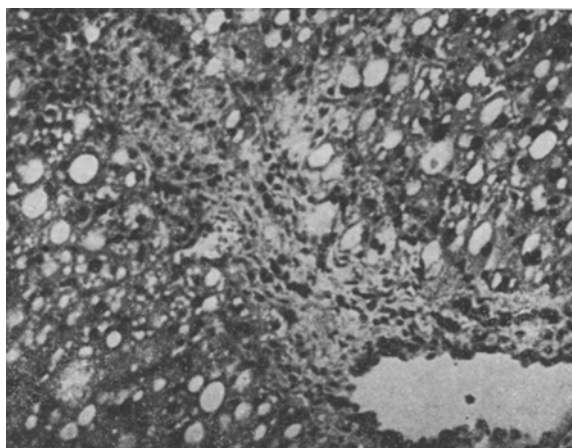


Fig. 3. Extensive connective-tissue fibrous band with subsiding inflammatory changes in it and fatty infiltration of surrounding liver tissue. 78th day. Hematoxylin-eosin, 200 \times .

injection (4, 8, 12, etc. up to 80 days). Animals of the control group were sacrificed along with the experimental animals.

Alcohol dehydrogenase activity was determined by the method of Roger and co-workers [3] and that of the other enzymes by methods described in previous papers [1, 2]. Activity of sorbitol dehydrogenase and aminotransferases was expressed in μ moles, that of ornithine carbamoyltransferase in μ g substrate/ml serum or g liver/h at 37°, and activity of alcohol dehydrogenase as the change in optical density readings after 10 min.

EXPERIMENTAL RESULTS

By injecting CCl_4 into rats at different times it was possible to examine the change from acute into chronic degeneration and the development of cirrhosis of the liver, in four main stages. Stages I and II (4-16 days from the beginning of CCl_4 administration) were characterized by the development of acute

TABLE 1. Serum Enzyme Activity in Animals at Different Stages of Development of Cirrhosis ($M \pm m$)

| Days from beginning of CCl_4 administration | Enzyme activity | | | | |
|---|--------------------------------------|--|---------------------------------------|--------------------------------------|--|
| | sorbitol dehydrogenase | ornithine carbamoyltransferase | alcohol dehydrogenase | alanine aminotransferase | aspartate aminotransferase |
| Control | (40) $0,22 \pm 0,05$ | (12) $6,1 \pm 0,5$ | (12) 0 | (40) $0,7 \pm 0,09$ | (40) $0,5 \pm 0,08$ |
| 8 | (8) $2,8 \pm 0,9$ $P < 0,001$ | (12) $19 \pm 1,1$ $P < 0,001$ | (12) $4,5 \pm 0,25$ $P < 0,001$ | (8) $2,4 \pm 0,5$ $P < 0,001$ | (8) $1,7 \pm 0,2$ $P < 0,001$ |
| 16 | (8) $3,3 \pm 0,86$ $P < 0,001$ | (10) $19,1 \pm 0,46$ $P < 0,001$ | (12) $6,9 \pm 1,0$ $P < 0,001$ | (8) $3,15 \pm 0,7$ $P < 0,001$ | (8) $2,4 \pm 0,34$ $P < 0,001$ |
| 48 | (8) $0,42 \pm 0,05$ $P > 0,2$ | (12) $16,1 \pm 1,3$ $P < 0,001$ | (12) $12,0 \pm 2,0$ $P < 0,001$ | (8) $0,96 \pm 0,12$ $P = 0,1$ | (8) $1,6 \pm 0,29$ $P < 0,001$ |
| 76-80 | (16) $0,17 \pm 0,015$ — | (12) $12,9 \pm 0,6$ $P < 0,001$ | (12) $13,3 \pm 1,5$ $P < 0,001$ | (16) $0,56 \pm 0,05$ — | (16) $1,65 \pm 0,26$ $P < 0,001$ |

Note. Number of animals (n) given in parentheses.

TABLE 2. Serum Enzyme Activity in the Liver at Different Stages of Development of Cirrhosis ($M \pm m$)

| Days from beginning of CCl_4 administration | Enzyme activity | | | | |
|---|--------------------------------|---------------------------------|-----------------------------|---------------------------------|----------------------------------|
| | sorbitol dehydrogenase | ornithine carbamoyltransferase | alcohol dehydrogenase | alanine aminotransferase | aspartate aminotransferase |
| Control | $86,5 \pm 4,8$ | $5\,300 \pm 180$ | $1\,190 \pm 80$ | $1\,839 \pm 27$ | $1\,587 \pm 18,5$ |
| 8 | $58,5 \pm 4,64$ $P < 0,001$ | $4\,300 \pm 100$ $P < 0,001$ | 925 ± 58 $P < 0,02$ | $1\,114 \pm 12,8$ $P < 0,01$ | $1\,069 \pm 15,2$ $P < 0,001$ |
| 16 | $56,5 \pm 3,9$ $P < 0,001$ | $3\,640 \pm 160$ $P < 0,001$ | 800 ± 48 $P < 0,001$ | $1\,044 \pm 11,7$ $P < 0,01$ | $1\,026 \pm 7,6$ $P < 0,001$ |
| 48 | $56,5 \pm 2,7$ $P < 0,001$ | $1\,610 \pm 200$ $P < 0,001$ | 610 ± 80 $P < 0,001$ | $570 \pm 8,9$ $P < 0,001$ | $688 \pm 8,4$ $P < 0,001$ |
| 76-80 | $39,8 \pm 3,1$ $P < 0,001$ | $1\,590 \pm 90$ $P < 0,001$ | 525 ± 45 $P < 0,001$ | $415 \pm 9,5$ $P < 0,001$ | $571 \pm 7,7$ $P < 0,001$ |

Note. Number of animals (n) given in parentheses.

degenerative and necrobiotic changes with the spread of fatty infiltration and diffuse inflammation in the stroma and parenchyma (Fig. 1), while stage III (24-48 days) was accompanied by proliferation of the adventitia of the blood vessel walls and proliferation of the connective tissues around the vessels and bile ducts and in the interalveolar septa (Fig. 2), and in stage IV (48-80 days) massive proliferation of the connective tissue and atrophy of the lobules were observed (Fig. 3).

Changes in enzyme activity in the blood serum and liver tissue (Tables 1 and 2) were dependent on the phase of development of the pathological process, and changes in the activity of different enzymes occurred in different directions.

During formation of chronic degeneration and cirrhosis the activity of all investigated enzymes in the liver decreased progressively. Dissociation between changes in activity of the transferases developed in the blood serum (a more rapid decrease in activity of alanine aminotransferase than on aspartate aminotransferase), while activity of sorbitol dehydrogenase fell sharply and that of ornithine carbamoyltransferase decreased slowly and activity of alcohol dehydrogenase increased.

The results given above indicate the absence of direct relationship between enzyme activity in the liver tissue and blood serum. They show that during development of cirrhosis of the liver the changes in activity of the various serum enzymes possess their own distinctive pattern.

LITERATURE CITED

1. L. L. Gromashevskaya, V. I. Demin, Z. P. Gette, et al., *Vopr. Med. Khimii*, No. 3, 246 (1964).
2. L. L. Gromashevskaya, V. N. Demchenko, and E. M. Mironova, *Ukr. Biokhim. Zh.*, No. 3, 262 (1967).
3. R. K. Bonnichsen and N. G. Brink, *Methods in Enzymology*, 1 (1955), p. 495.