REGENERATION OF THE LIVER IN MONKEYS

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Removal of 60-70% of the liver from rats [5, 8], mice [14], rabbits [3, 4], or dogs [7], is followed by rapid recovery to the original weight and size. Regrowth is caused by increased mitotic activity of the cells [1, 5] as well as by their hypertrophy [2]. Definite biochemical changes occurring in the remainder of the liver have been demonstrated [1, 10, 12]. The liver of monkeys has been less thoroughly studied. It is important to extend such investigations because Child and others [6] have shown that, anatomically, the structure of the portal system of macaque rhesus monkeys is similar to that of man. Mannix, Cornell, and O'Sullivan [9] have shown that, in the young macaque rhesus monkey, removal of an average of 53.4% of liver tissue was followed by regeneration within two months; however, the authors consider that, in the monkey, regenerative power is much less than in other experimental animals. B. P. Solopaev [3, 4] found no special features in the recovery process of macaque rhesus liver and maintains that, in monkeys, regeneration takes place as rapidly and as completely as it does in other animals.

Our task has been to make a cytological study of regenerating liver.

EXPERIMENTAL METHOD

The left front, posterior, and the right posterior lobe of the liver, comprising an average of 26% of the whole liver tissue, were removed from 14 macaque rhesus monkeys weighing 1900-3400 g. The operation was performed under local novocaine anesthesia. Before the operation, all the animals received 40% glucose solution intravenously, and morphine, caffeine, and camphor subcutaneously. A silk ligature was placed on each of the lobes to be removed, and the area was irrigated with penicillin. The wound was sewn up in layers. After the operation, for 2-4 days, all the animals received intravenous glucose twice per day and large doses of penicillin. On the second day after the operation, they were given white bread, fruit, sugar, tinned fruits, and a large amount of water. They were killed by decapitation. As controls, we used the portion of liver removed as well as the liver of two unoperated monkeys. The material was fixed in 12% formalin, in 80% ethyl alcohol, in Carnoy's fluid, or in cooled acetone. The methods of Brachet, Feulgen, and Gomori [11] were used to study the RNA and DNA content of the cytoplasm and nuclei of the hepatic cells of the control and regenerating livers; tests were also made for alkaline phosphatase activity. Also, we applied the method of Gomori [11] to test for reticular fibers; measurements of the nuclei were made on preparations stained for alkaline phosphatase. In every case, we measured 200 uninucleate hepatic cells and their nuclei by means of an ocular micrometer.

EXPERIMENTAL RESULTS

Out of the 14 monkeys, 7 died after the operation, one was kept for a long time, and the remaining 6 were killed 1, 2, 3, 6, 7, and 14 days after the operation. At the third day after the operation the residual lobes start to thicken, their edges become rounded off, and, at this stage, the color differs somewhat from the normal dark red hue; fatty infiltration is quite rapid in the first 1-2 days, it begins to fall off after the third, and ceases entirely by the fourteenth day; the relative and absolute weight of the regenerating liver has almost entirely recovered by this time (Tables 1 and 2).

Histological studies of the regenerating liver made on the first-second day after the operation (Fig. 1) showed no special features distinguishing regeneration of the liver in monkeys from that of other animals. All that could be found were occasional cells dividing mitotically or amitotically. Between the third and fourteenth days after the operation

TABLE 1. Weight of Liver of Operated Monkeys

No. of monkey	Duration of experiment (in days)	Body weight before operation (in g)	Body weight at death (in g)	Weight of portion of liver removed (in g)	Weight of regenerating liver (in g)	Relative weight of regenerating liver (in %)	Total wt. of removed and regenerating portion of the liver		Notes
							g	%	
2 259	1	2 400	2 400	19,0	55,0	2,3	74,0	3,1	Monkey died after operation
2 251	1	2 400	2 400	38,6	62,0	2,6	100,0	4,2	The same
2 309	1,5	1 900	1 900	14,5	43,1	2,3	57,6	3,0	The same
2 264	2	2 000	2 000	38,3	62,9	3,1	101,2	4,0	The same
2 469	2	3 400	3 500	24,0	90,2	2,6	114,2	3,3	Monkey killed
2 441	3	3 000	2 400	21,3	71,4	2,9	92,7	3,9	The same
2 404	5	3 100	2 700	22,0	77,3	2,8	99,3	3,7	The same
2 479	7	3 200	3 000	21,5	84,5	2,8	106,0	3,5	The same
2 300	14	2 000	1 900	16,0	60,5	3,2	76,5	4,0	The same
E		23 400	13 500	215,2	606,9	24,6	822,1	32,7	
M		2 600	2 700	23,9	67,4	2,7	98,0	3,6	

TABLE 2. Liver Weight of Control Monkeys

No. of Monkey	Body weight at death (in g)	Liver weight	
		Absolute wt. (in g.)	Relative wt. (as %)
1	3 130	113,4	3,6
2	3 118	118,0	3,7
3	3 402	114,8	3,3
4	3 000	82,3	2,7
M	3 162	107,4	3,4

TABLE 3. Area of Uninucleate Hepatic Cells and of the Nuclei (in μ^2)

No. of monkey	Unoperated liver		No. of monkey	Regenerating liver		Duration of experiment (in days)
	cell	nucleus		cell	nucleus	
2 267	238,3	18,8	2 267	259,4	30,1	2
2 469	213,5	25,4	2 469	225,7	27,2	3 1/2
2 479	249,3	25,4	2 479	428,6	43,0	7
2 300	234,5	25,4	2 300	261,3	34,2	14
M	233,6	23,7	*M*	293,9	33,6	

the reticulin framework of the regenerating liver differed from the preoperated condition as follows: the reticulin fibers were thickened and were more loosely distributed in the parenchyma. No appreciable differences in the amount of distribution of the nucleic acids was found. At all times, as in normal animals, the nuclei contained but few clumps of DNA, and these were distributed chiefly along the nuclear membrane and around the nucleolus. The cytoplasmic granules in the cells of the regenerating liver stained rather more intensely than normal with pyronine on the third, fifth, and seventh days after the operation.

The only definite differences concerned alkaline phosphatase. In the regenerating liver, between the third and seventh days its activity was much greater than it was in the control animals; it was found not only in the nuclear membrane, chromatin, and nucleolus which stained strongly a dark or dark brown color (as they did also in the control) but considerable amounts were also found in the cytoplasm. Normally, the the activity of this enzyme in the cytoplasm of hepatic cells is not appreciable (Figs. 2 and 3). The localization of the alkaline phosphatase in the liver parenchyma of the control animals was uneven: cells near the blood vessels always contained more of the enzyme than those near the center of the lobe; a similar distribution of alkaline phosphatase was maintained in the regenerating liver during the recovery process (between the third and fourteenth days after the operation).

Results of the measurement of the uninucleate hepatic cells and of their nuclei (Table 3) show that in regenerating monkey liver during recovery the area of the cytoplasm and nuclei increases. In the control group the average cell area was 236.1μ^2, and the nuclear area was 25μ^2; in the regenerating liver the corresponding figures were 268.7 and 29.9μ^2. After 14 days the amount of RNA and the alkaline phosphatase activity returned to their normal values, while the hypertrophy of the cells and nuclei was maintained throughout the whole period of study.

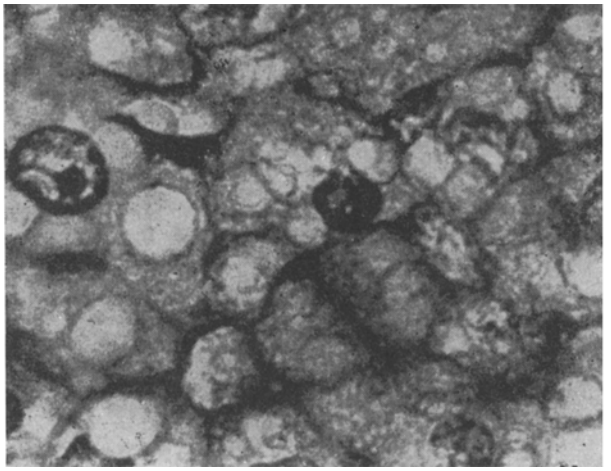

Fig. 1. Regenerating liver of monkey No. 2259 one day after operation. Vacuolization of the hepatic cells. Gomori's stain for alkaline phosphatase. Ocular 6×, objective 100/1,30, magnification 720.

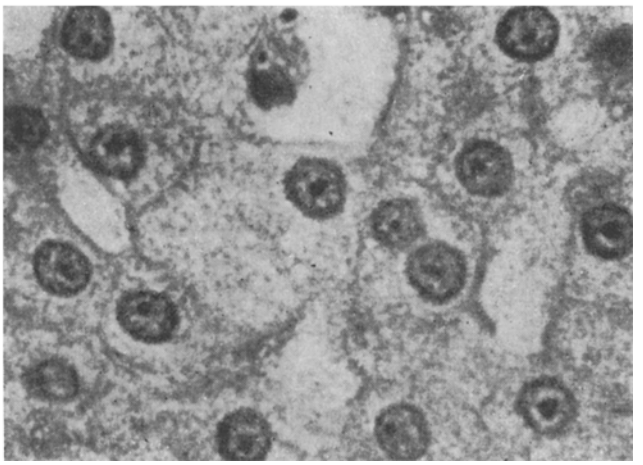

Fig. 2. Control liver of monkey No. 2479. Gomori's stain for alkaline phosphatase. Ocular 6×, objective 100/1, 30, magnification 720.

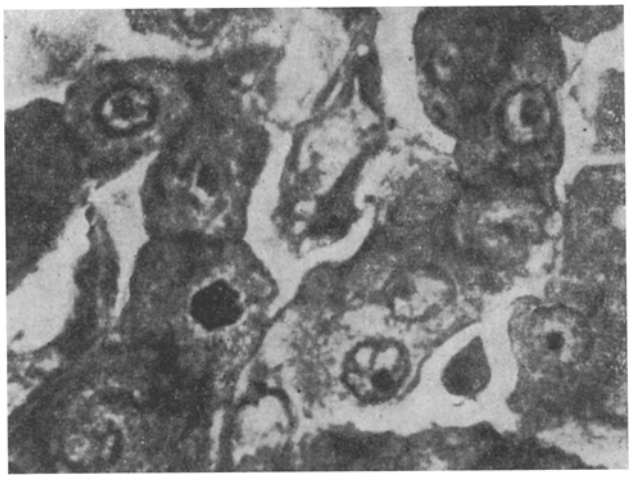

Fig. 3. Regenerating liver of monkey No. 2479, seven days after operation. Gomori's stain for alkaline phosphatase. Ocular 6×, objective 100/1, 30, magnification 720.

One subgroup consisted of animals which had died 1-2 days after the operation. Histological study of these monkeys showed that there was an abnormal arrangement of the trabeculae of the liver. Fatty infiltration had occurred: the hepatic cells were strongly vacuolized, and in many of them the nuclei were in various stages of disintegration and were pyknotic; the RNA in the hepatic cells of the monkeys that died was diffuse, and the clumps had disappeared. No dividing cells were found, and the area of the uninucleate hepatic cells and of the nuclei was reduced: in the control group, the cell area was 240.0μ^2, and, that of the nucleus, 27.7μ^2, while in the experimental group, the corresponding figures were 218.5 and 22.5μ^2. The features observed in the liver of the operated group consisted of small variations in the content of nucleic acids, hypertrophy of the hepatic cells, the small number of dividing cells, and the loose arrangement of the reticular fibers; we may therefore suppose that in the macaque rhesus monkey liver regeneration shows several special features and takes place less intensively than in other mammals.

SUMMARY

The posterior, left anterior, and right posterior lobes of the liver, representing 26.0% of the total liver tissue were removed from 14 macaque monkeys. Seven animals survived. Regeneration occurred through regenerative hypertrophy. Regrowth took place through the formation of new hepatic cells by mitosis and by amitosis, as well as by hypertrophy of these cells. The histochemical changes of the nucleic acids indicated that, in the monkey, regeneration is less intense than in the lower animals, and is due mainly to hepatic cell hypertrophy.

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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.
