# EXPERIMENTAL BIOLOGY

## THE EFFECT OF SERUM AND BLOOD PLASMA FROM ANIMALS UNDERGOING LIVER REGENERATION ON THE MITOTIC ACTIVITY OF LIVER CELLS IN INTACT ANIMALS

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Several authors have obtained the effect of increased mitotic activity in the regenerating livers of rats which have been injected with serum or plasma from donor animals from which part of the liver had previously been removed [2, 9]. A similar effect has been observed in the case of recipient rats with intact livers [3, 9]. These experiments suggest that during the process of regeneration the bloodstream contains some factor or other which has a stimulatory action on cell division. The opinion has also been expressed that during the process of regeneration the content of cell division inhibitors in the blood stream is reduced [4, 6].

Furthermore, whereas some suppression of the growth rate of the liver of recipient animals has been observed in certain cases following injection of serum from hepatectomized animals [8], in other cases no such effect on the number of mitoses has been observed [7].

From what has been said above, it is obvious that there are conflicting opinions as to the nature of the effect (if any) of humoral factors on the mitotic activity of the liver.

The aim of the present work was to study the effect of serum and plasma from animals undergoing liver regeneration on the mitotic activity of liver cells in intact animals.

### EXPERIMENTAL METHODS

Two series of experiments were carried out: one series on 2 month old gray hamsters (First series) and another (Second series) on nonpedigree male white rats weighing 170 g. Animals of the first series were divided into donors and recipients. The donor animals were in turn, divided into 3 subgroups: one of which underwent partial hepatectomy (about 65% of the liver was removed [5]; the second underwent a pseudo-operation (without damage to the organ), whilst the animals in the third group were left intact. Animals in the first 2 subgroups were used as sources of blood for the preparation of serum and plasma, the blood being removed from the abdominal artery, 48 h after the operation. Blood was obtained in the same way from the intact animals of the third subgroup. Serum was injected intraabdominally into the intact recipients of the corresponding subgroups. The injections were carried out over a period of 3 days at a rate of 3 injections per day, each injection consisting of 0.5 ml of serum diluted with twice its volume of sterile physiological saline solution.

On the morning following the last injection the animals were killed, and their livers fixed in preparation for subsequent histological investigation. Pieces of liver tissue were also removed from donor animals and fixed immediately after removal of their blood. In all cases the pieces of liver were taken from the right lobe.

From the preparations (sections 7 microns thick, stained with hematoxylin and eosin), counts were made of the number of mitoses per 12000-15000 cells in each animal; the early stages of mitoses were not counted. The mitotic coefficient (MC) was calculated as a percentage.

TABLE 1. Effect of Serum from Intact and Hepatectomized Hamsters on the Mitotic Activity of Livers in Intact Animals

| Group of animals | No. of animals in groups | MC (as %) |
|---|---|---|
| Intact donors | 10 | 0.24 ±0.04 |
| Intact recipients receiving serum from intact donors | 4 | 0.16 ±0.03 |
| Hepatectomized donors | 10 | 0.36 ±0.06 |
| Intact recipients receiving serum from hepatectomized donors | 11 | 0.35 ±0.06 |
| Donors, subjected to pseudo-operation | 10 | 0.12 ±0.02 |
| Intact recipients receiving blood from donors subjected to pseudo-operation | 6 | 0.13 ±0.05 |

TABLE 2. The Effect of Blood Plasma from Intact and Hepatectomized Rats on Mitotic Activity and the Number of Cells (per field of vision) in Livers of Intact Rats

| Group of animals | No. of animals in groups | MC (as %) | No. of cells in field of vision |
|---|---|---|---|
| Intact recipients, receiving plasma from hepatectomized donors | 16 | 0.29 ±0.05 | 28 ±2 |
| Intact recipients receiving plasma from intact donors | 15 | 0.10 ±0.05 | 36 ±1 |

The animals of the second series provided two subgroups of donors—one consisting of hepatectomized rats, the other of intact rats. Thirty two hours after hepatectomy, blood was obtained from the abdominal artery of rats in the first group, potassium citrate being added to prevent clotting. Blood was obtained similarly from the intact rats. Plasma was obtained from both sets of blood and this was injected into the 2 corresponding subgroups of intact recipients. Each recipient animal was given 2 injections: at 6 h (3 ml of plasma) and at 14 h (1 ml).

Forty eight hours after the first injection (i.e., at 7 h) the recipient rats were killed. Histological examination of their livers was carried out as for animals of the first series.

## EXPERIMENTAL RESULTS

As can be seen from Table 1, the mitotic activity in livers of intact hamsters receiving injections of serum from hepatectomized donors was equal to that in animals with regenerating livers.

The mitotic coefficient (MC) of regenerating livers was found to be higher than that of livers in hamsters subjected to pseudo-operation ($P = 0.003$) and than that of recipients of the second group which had received injections of normal serum ($P = 0.016$), or serum from donors subjected to pseudo-operation ($P = 0.012$). The number of mitoses in livers of intact hamsters receiving serum from hepatectomized animals was also higher than that of livers in the groups of animals enumerated in the last sentence (P being equal to 0.001, 0.018, 0.016 respectively). The mitotic coefficient (MC) of livers in normal (intact) hamsters was greater than that of animals subjected to pseudo-operation ($P = 0.03$), but did not differ essentially from that of regenerating livers ($P = 0.15$), or from livers of hamsters receiving serum from hepatectomized donors ($P = 0.13$). It is possible that it can be explained by the fact that normal hamsters were killed at a time when mitotic activity in the liver was at a maximum [1].

Nevertheless, it is reasonable to suggest that the injection of serum from hamsters subjected to partial liver resection produces an increase in the number of mitoses found in the livers of intact recipients, because the mitotic coefficient of regenerating livers and livers of intact recipients receiving serum from hepatectomized donors is almost equal.

The data obtained from the second series of experiments (using rats) is given in Table 2, from which it is evident that the injection of plasma from hepatectomized animals causes an increase in the number of mitoses in livers of intact rats ($P = 0.01$). In addition, it seems reasonable to suppose that the decrease in the number of liver cells per field of vision in experimental animals as compared with controls ($P < 0.01$) is due to an increase in the dimensions of the cells.

Thus, it can be said that removal of part of the liver and subsequent regeneration of the organ results in certain changes in the liquid component of the blood, such that when the latter is injected into intact animals, the mitotic activity of their liver cells is increased.

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