89.4 ± 5.1% of the injected dose is in the liver and 1.5 ± 0.11% in the spleen, no difference between normal and irradiated mice being seen.

It may be concluded that the hepatic flow is slowed down immediately and up to 4 days after local irradiation. The difference of the liver flow between normal and irradiated mice is statistically significant (Table). The fact that the spleen uptake remained stable whether the liver region was irradiated or not, is proof that the delay clearance is caused by the slowed-down liver blood flow and not by irradiation damaged RES. When the reticuloendothelial cells of the liver are destroyed or blocked, the phagocytosis of the spleen increases compensatorily and the uptake is higher[6].

Our results are confirmed by PIOVELLA et al. who noticed under a transillumination microscope that the blood flow after γ-irradiation of the liver was delayed, with stasis and perivascular microhemorrhages. Conditions returned to normal after a small dose, whereas after a higher dose they were irreversible[7,8].

Zusammenfassung. Mit Hilfe der Clearance von kolloidalem Radiogold, der Leber- und Milzspeicherung wurde die Durchblutung der Leber nach lokaler Bestrahlung der Leberregion bestimmt. Im Anschluss an 500, 1000 oder 2000 R wurde eine Verminderung der Leberdurchblutung festgestellt. Eine Restitutio ad integrum wurde dosisunabhängig 3–4 Tage post irritationem beobachtet.

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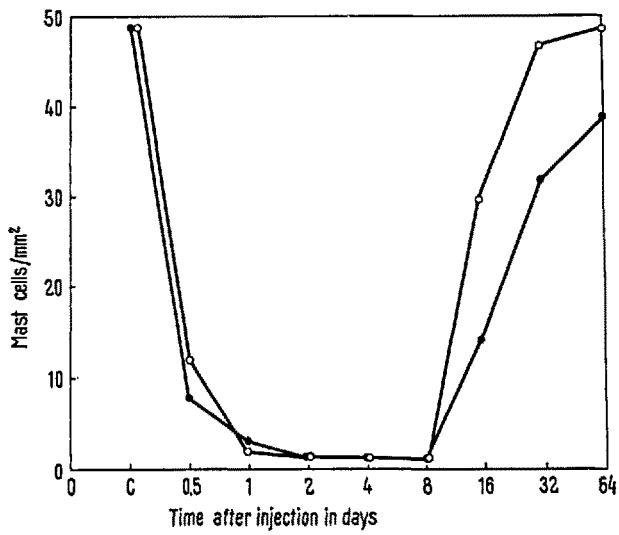
The Histamine and Heparin Content of the Rat's Mesenterial Mast Cells Regenerating after Application of Compound 48/80

The relationship between histamine and heparine in the mast cells is still obscure. The mast cells are a significant source of histamine and heparin, and the release of mast cell granules through experimental means is accompanied by the release of histamine and heparin. RILEY[1] supposes that, on rupture of the tissue mast cell, histamine first spreads rapidly through the tissues, and then a sluggish discharge of heparin follows. WERLE et al.[2] show that the histamine in the mast cells, and also the greatest part of the tissue histamine, forms a complex with heparin. In the complex the histamine acts as heparinate.

It is difficult to develop an experimental procedure making possible separate histamine or heparin release without disruption of the mast cells. SMITH[3], however, found that intraperitoneal administration of low concentrations of toluidine blue, protamine sulfate, or other histamine liberators brought about a significant release of histamine without any disruption of the mast cells in the tissues of the peritoneal cavity. It is, however, a well-known fact that in the majority of experimental conditions the mast cells are extremely sensitive to environmental changes, as well as to irritation or actual injury. Mast cells respond to injury of any kind by liberating substances normally held in their protoplasm. Thus the changes observed in the mast cells immediately after irritation of any kind must be noted with caution.

In this investigation, the appearance of histamine and heparin is examined in mast cells regenerating after previous and complete disruption caused by compound 48/80.

Material and methods. Adult male rats (250–300 g) were injected intraperitoneally with 200 μg compound 48/80 dissolved in 2cm³ of a 0.9% sodium chloride solution. The control animals were only injected with 2 cm³ sodium chloride solution. The animals were killed from half a day up to 64 days after the injection. From the mesentery of the lower small intestine, two specimens were taken. One specimen was immediately put into a solution of Reinecke-salt (saturated, filtered solution, diluted with water in the ratio 1:5) for 48 h (SCHAUER and WERLE[4]). The histamine of the mast cells precipitated as 'Reineckat', and, in the unstained translucent preparations of the mesenteries, the granules of the mast cells could clearly be seen. After fixation the mesenterial membrane was carefully – avoiding stretching – placed on a microscope slide. The other specimen was fixed in a 4% basic lead acetate, stained with a 1% aqueous solution of toluidine



Mast cell counts after application of compound 48/80. —●— Reinecke-salt, —○— toluidine blue.

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blue, and placed on a microscope slide. In both methods the mast cells were counted in an area of 10 mm², and the average mast cell count per 1 mm² was calculated.

Results and discussion. As the results obtained from the control material show, both the methods give the same result as far as untreated mast cells are concerned. It is also noted that, if there are differences in the mast cell counts obtained with both methods, these differences must be regarded as changes in the heparin or histamine content of the cells.

As expected, rapid disruption of the mast cells occurred, and only a few traces of mast cells could be found in the specimens up to 16 days. Thereafter new small mast cells began to appear in the specimens; that is, regeneration had begun. At 16 days the number of mast cells demonstrable with histamine stain was only about half the number of cells visible with metachromatic staining. Later the difference decreased. The count of the cells which became visible with toluidine blue reached the control level at 32 days. On the other hand, the number of the cells stained with Reinecke-salt was still only 80% of the control at 64 days.

The results point to the possibility that the bond between the histamine and heparin in the mast cells is not definitely fixed and that the amounts of the heparin and histamine in the mast cells may vary within certain limits. There are two explanations for the fact that the mast cell count obtained with histamine staining is smaller than the count obtained with toluidine blue staining. It may be possible that the effect of compound 48/80 lasts for a very long time. The second and more probable explanation is that the young and regenerating mast cells always have a lower concentration of histamine than full-grown mast cells.

The experimental conditions in this study were rather pathological. Whether the mast cells can vary their histamine and heparin content physiologically as well is a fundamental problem. This function is probable if we take into consideration their manifold role in the connective tissue.

Zusammenfassung. Der Histamin- und Heparingehalt in Mastzellen, die nach totaler Zerstörung mit Histaminliberator 48/80 regenerieren, wird untersucht. Die Ergebnisse zeigen, dass während des Regenerationsprozesses die Anzahl der Mastzellen, in denen sich das Histamin mit Reineckesalzlösung anfärbt, kleiner ist, als die Anzahl der Mastzellen, die mit Toluidinblau darzustellen sind.

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Catheptic Activity in the Cerebral Tissue of the Rabbit during Allergic Encephalomyelitis

The proteolytic enzymes of the cerebral tissue in normal and pathological conditions have so far been relatively little studied. In 1931 KREBS[1] found in the cerebral tissue a marked peptidasic and a reduced gelatinasic activity. EDLBACHER et al.[2], and later KIES et al.[3], confirmed the existence of proteinasic activity in the brain. The accurate researches of ANSELL and RICHTER[4] proved the existence of a proteinase with optimal pH 7.4, and of a cathepsine with optimal pH 3.5, in fresh cerebral tissue. As proved by several investigations[5,6], in demyelinizing processes, the state of equilibrium between the synthesis and the breakdown of proteins is modified. It may be presumed that these changes are also reflected by the enzymatic activity.

In our experiments, 30 rabbits were treated with an encephalitogenic emulsion, containing antipertussis vaccine, cattle brain homogenate, and Freund's adjuvant, inducing allergic encephalomyelitis[7]. The catheptic activity in the cerebral tissue homogenate (1:4 in isotonic sodium chloride solution) was determined at pH 5.0, with azocasein substrate, in the presence of 25% urea, and 0.05 *M* cysteine. The rate of proteolysis was expressed in μg of the azocasein, hydrolysed by 100 mg wet tissue, after 1 h incubation at 38°C. In the controls, an appreciable catheptic activity was observed, which remained practically unchanged after 10 days, i.e. after 2 injections of antigen. After 3 or 5 injections, i.e. after 20 or 32 days from the beginning of treatment, in the evolutive phase of the pathological process, the catheptic activity showed a significant increase, amounting to 88%, as compared to the controls (see Table).

The increase of the enzymatic activity might be in connection with the allergic inflammatory process of the brain. Recent investigations[8] demonstrated that, in anaphylactic and allergic reactions, the proteolytic enzymes

Mean values of the catheptic activity of the brain in the four groups studied (10 rabbits each)

Controls	Animals treated with encephalitogenic emulsion		
	After 10 days (2 injections of antigen)	After 20 days (3 injections of antigen)	After 32 days (5 injections of antigen)
280 ± 38.2	256 ± 25.8	488 ± 11.1	522 ± 20.5
	$p < 0.001$		

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