Summary and Conclusions. Toxic material, which causes death of mice on intravenous injection, can be extracted from progestational endometrium and from placenta of several mammalian species. Small amounts of material with a similar effect are present in extracts of skeletal muscle. Quantitative estimation of the toxicity of a variety of tissue extracts has been made. Addition of blood or serum rapidly diminishes this toxic property.

Special appreciation is due Miss Carol Spaulding for assistance in conducting this investigation.

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Liver Necrosis in Mice Following Injection of Toxic Extracts of Progestational Endometrium and of Placenta.

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During a study of the action of toxic extracts of endometrium and of placenta[1] a number of treated mice were found to have focal necrosis of the liver. Liver lesions have appeared in 174 of the animals; this represents an incidence of about 30% in mice which received sublethal amounts of toxic extract injected intravenously. The animals ranged from 1½ to 2 months of age. The lesions have been studied further by examining the livers of treated animals at different intervals following the injections.

Results. Within the first hour after injection, irregular areas of discoloration could be seen through the capsule of the liver. At first they were dark red, but after 24 hours they were usually paler than the adjacent liver tissue, and became sharply defined. Large lesions often presented a mottled appearance, with darker spots on a pale background (Fig. 1). The lesions were usually near the periphery of the lobes, and their size and number were variable. Usually the altered zones were small, and they occurred most frequently in the large left lobe, but all lobes were involved when the lesions were extensive. Occasionally, with massive lesions, there were fibrinous adhesions of the involved parts to adjacent structures. Small lesions were invariably covered by a smooth surface, although they were usually situated just beneath the liver capsule.

Microscopically there was dilatation of the hepatic sinusoids in the earliest lesions recognized, and erythrocytes were sometimes seen outside of vessels. Within 3 hours vacuolation of hepatic cells could be seen (Fig. 2) in the altered regions, and the nuclei were sometimes small and deeply stained. After 17 to 20 hours much of the cell structure had been destroyed (Fig. 3) in sharply demarcated zones, bordering which the adjacent hepatic cells were sometimes unusually deeply stained. At this stage there were fairly numerous polymorphonuclear leucocytes, particularly near the margin of the lesions.

The lesions had no constant position in the liver lobules. When the lesions were minimal, involving only small groups of cells, they were mid-zonal distribution. Sometimes entire lobules were necrotic, and the lesions commonly extended throughout several lobules or parts of several lobules without apparent predilection for any part.

In several animals there were tiny pedunculated lobes of liver tissue protruding caudally near the right kidney. These all showed necrosis in treated animals, even in the absence of lesions elsewhere. Several recent thrombi in vessels within the narrow stalk which attached these structures to the

[1] Schneider, C. L., PROC. SOC. EXP. BIOL. AND MED., 1946, **62**, 322.

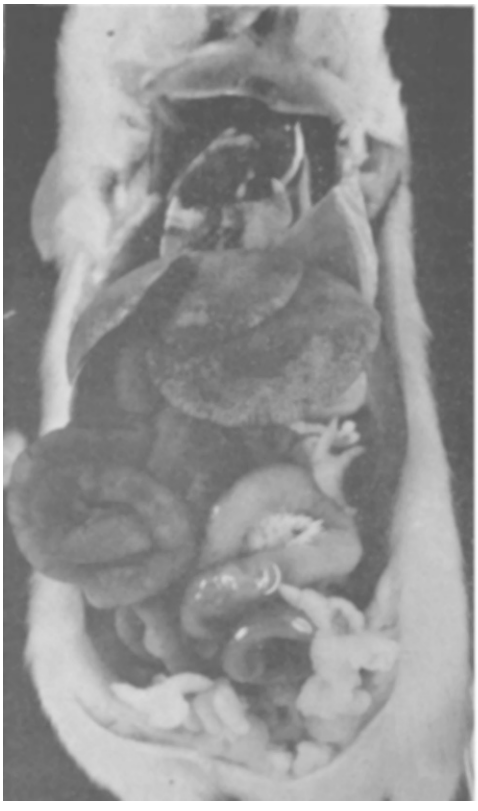

Fig. 1.
Abdominal organs of a mouse killed 17 hours after intravenous injection of an extract of rabbit uterus at estrus. The mottled portions of the liver indicate the necrotic zones.

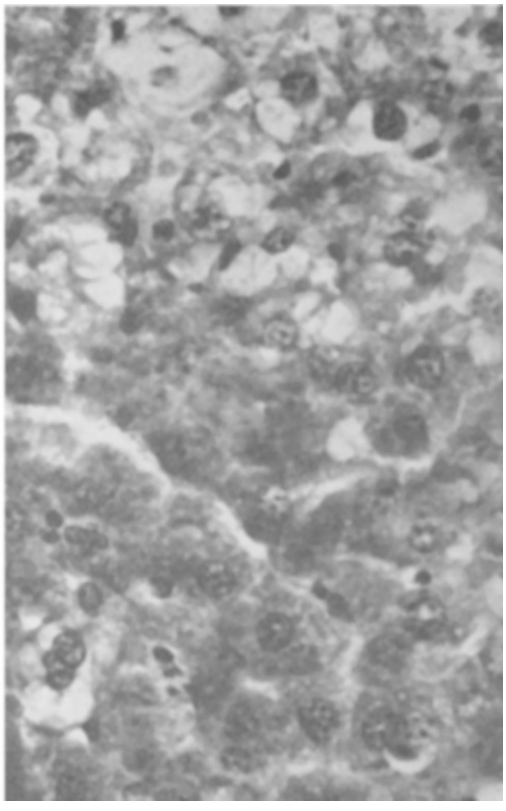

Fig. 2.
Histological section of a mouse liver removed 1½ hours after injection of a toxic extract of rabbit uterus. In the upper half of the photograph are many abnormal vacuolated cells.

liver suggested a possible mechanism for the formation of these lesions. Thrombi were also found in small veins adjacent to other areas of liver necrosis, although they were usually not demonstrable in association with small lesions.

The time required for absorption of the necrotic liver was dependent upon the size of the lesion. Necrotic cells sometimes persisted for 4 days or longer. Mitotic figures could usually be seen in the surrounding hepatic tissue after 2-4 days. There was relatively little growth of fibrous tissue, which was pronounced only at the periphery of the large lesions.

The sex of the mice, a preliminary period of starvation or deprivation of water, a meat diet for 2 days, and variations in the character of the anesthetic at the time of injection, had no recognizable influence upon the development of the liver lesions.

Injections of only slightly less than the fatal quantity of toxic extract were most frequently followed by liver damage, and under these conditions the lesions were often large. However, liver necrosis did not occur invariably in such animals, and lesions developed in some mice after injection of much less than a lethal quantity of an active extract. The liver necrosis, like the acute death of mice, occurred regularly only after intravenous injection of the toxin.

When toxic extracts were mixed with serum *in vitro* there was rapid diminution of the toxic property as measured by the increased amount of the extract necessary to produce death of mice.[1] Injection of sublethal quantities of this partially detoxified extract

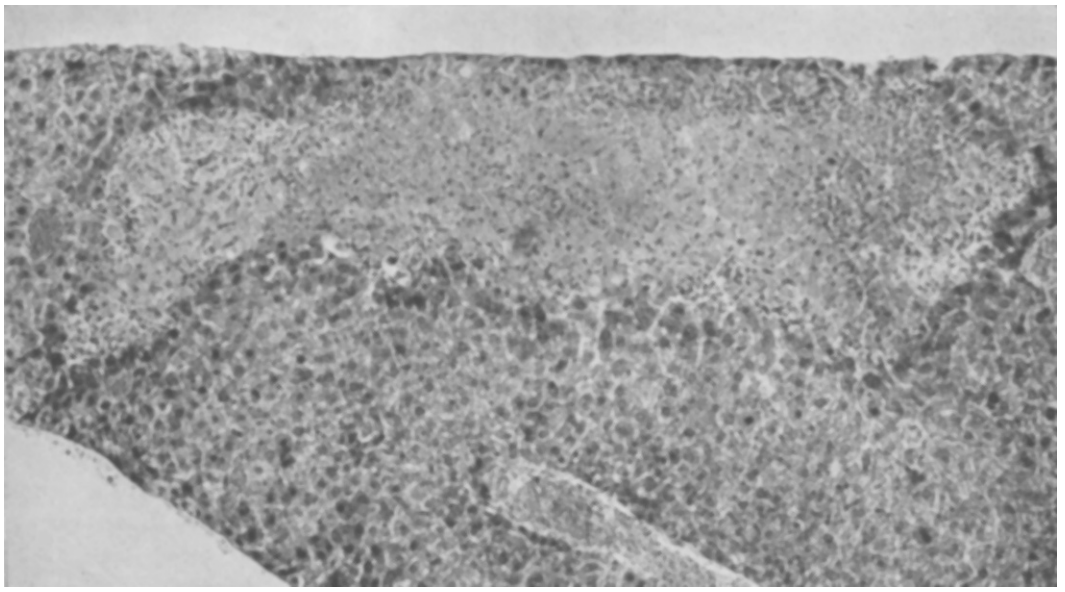

Fig. 3.
Histological section of a mouse liver removed 20 hours after injection of a toxic uterine extract, showing a zone of necrosis adjacent to the liver surface.

did not produce liver lesions more frequently than did much smaller amounts of untreated extract. Similarly, slow injection of several times the quantity which was lethal on rapid injection did not increase the incidence of lesions. These observations suggest that the production of liver lesions is related to the ability of the extract to kill animals, and that serum or plasma influences both properties.

In 203 control mice, most of which had been given quickly fatal injections of toxic uterine extracts, grossly visible liver lesions similar to those in the experimental animals were found in 3 instances. None of these had received injected extract long enough prior to death to account for the lesions. An occasional animal injected with extracts of other types of tissue also showed liver lesions, although the only tissue to elicit the change with a frequency distinctly greater than that of the control group was skeletal muscle. Of 69 mice injected with muscle extracts, 14 developed liver lesions.

Most of the mice with liver lesions showed no other gross tissue changes. Likewise, histological sections of kidney, spleen, heart, lung, thyroid, adrenals and brain from several of the animals disclosed no abnormalities. In a few instances there was hemorrhage in one or more tissues, including the intestine, mesenteric lymph nodes, lungs and eye. No underlying lesion was detected. Several thrombi were found on the atrial endocardium.

Five rabbits and 4 dogs which survived intravenous injections of toxic extracts showed no liver necrosis. Of 51 rats surviving intravenous injections only 2 had liver necrosis similar to that of the mice.

Discussion. Krichesky and co-workers[2,3] did not mention liver necrosis in their rabbits injected with toxic extracts of pregnant or pseudopregnant uteri, but focal liver necrosis has been reported by various investigators using placental extract in an effort to produce experimental toxemia of pregnancy. Young[4] with suspensions of dried, autolysed, powdered placenta injected hypodermically, obtained liver necrosis in 40% of 15 guinea pigs. Others were reported to have "degenerative lesions."

2 Krichesky, B., and Pollock, W., *Am. J. Physiol.*, 1940, **130**, 319.

3 Krichesky, B., and Mahler, J., *Endocrinology*, 1942, **30**, 616.

4 Young, J., *J. Obs. and Gyn. Brit. Emp.*, 1914, **26**, 1.

Obata[5] observed lesions in 3 of 12 rabbits injected intravenously with saline extracts of fresh placenta. He mentioned hemorrhagic liver lesions in his injected mice. Oden[6] upon repeating the methods of Young with 3 guinea pigs and 7 rabbits, declared that the liver lesions were diffuse, with "no lesions in any manner resembling those of eclampsia." No mention was made of liver lesions in his mice that received injections of extract. Bartholomew and Kracke[7] reported liver and kidney lesions in rabbits and guinea pigs.

From these reported observations and from the circumstances leading to the development of the liver lesions here reported, it is apparent that there is a relationship between the development of focal liver necrosis and the injection of toxic extracts of endometrium and of placenta. However, the appearance of a few similar foci of necrosis in animals which received injections of extracts of a variety of tissues, or occasionally too soon after injection to be a result of the treatment, suggests that the experimental lesions may not result directly from the action of the extract, but may follow activation of some latent process. Since most of the changes in the liver developed after the animals that received injections of extract had suffered a temporary collapse or shock-like state, this non-specific influence may have been a factor in the pathogenesis of the lesions. The possibility that the lesions are identical with the spontaneous focal liver necrosis in mice reported by Olitsky and Casals[8] has been considered, but the lesions described here were grossly visible and involved larger zones than did the spontaneous lesions. No inclusion bodies have been found in any of the sections of liver from animals used in this study.

Summary. Focal liver necrosis occurred in 30% of 500 mice which survived the intravenous injection of single sublethal quantities of toxic extracts of endometrium or placenta. Occasional similar lesions were seen in mice which were injected with other tissue extracts. Although related to the effect of the injected toxic material from endometrium or placenta, the lesions may be indirect or nonspecific effects of the injections.

Special appreciation is due Miss Carol Spaulding for assistance in conducting this investigation.

[5] Obata, I., *J. Immunology*, 1919, **4**, 111.

[6] Oden, C. L. A., *J. Mich. Med. Soc.*, 1925, **24**, 110.

[7] Bartholomew, R. A., and Kracke, R. R., *Am. J. Obs. and Gyn.*, 1932, **24**, 797.

[8] Olitsky, P. K., and Casals, J., PROC. SOC. EXP. BIOL. AND MED., 1945, **60**, 48.

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Influence of Dietary Iodine on Susceptibility of Rats to Alpha Naphthylthiourea Poisoning.*

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Richter[1] proposed the use of α-naphthylthiourea (ANTU) as a rodenticide for the control of Norway rats because of its high toxicity to carnivorous species and its relatively low toxicity to omnivorous and herbivorous species.

While it has not yet been reported that any

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† The experimental data in this paper are taken from a thesis submitted *in absentia* in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Biochemistry in the Graduate School of the University of Illinois.

[1] Richter, C. P., *J. A. M. A.*, 1945, **129**, 927.