produces an increase in the Mg_e, accompanied by an accelerated uptake of Mg^{28} in muscle, appendix, and heart(8). Digitalis, in the present study, produced similar changes in magnesium metabolism. Since it is difficult to explain these findings solely on the basis of the cardiotonic effects of digitalis, the results of the present study furnish additional evidence that this drug also has an extracardiac action.

Summary. In a test group of 8 rabbits, administration of digitoxin by intravenous injections of 0.1 mg/kg, given daily for 8 days, produced no significant changes in the external balance of magnesium. On the day following the last injection the exchangeable body content of magnesium, measured with Mg^{28}, was significantly increased. The results suggest that digitoxin produces subtle changes in the dynamics of magnesium metabolism which are not reflected in the external balance data.

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Use of Radioiodinated Antipyrine to Measure Liver Function in the Rat.* (27035)

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Radioiodinated antipyrine (RIAP), when injected in the rat, releases significant quantities of its I^{131} tag within one hour. The I^{131} tag appears in the peripheral blood in an increasingly diffusible form. This phenomenon can be measured by the rate of dialysis of radioactivity from plasma through a cellophane membrane(1). Brodie and Axelrod(2) have shown that in man, 30% of a dose of antipyrine is hydroxylated by the liver at the 4′ position, immediately conjugated with glucuronic or sulfuric acid and excreted in the bile. It has been suggested that since the I^{131} tag of RIAP is located at the 4′ position, its separation from the antipyrine molecule was a function of the liver(1). In the experiments reported here, liver function was altered by 3 different methods in rats. RIAP was then administered to these and to control animals. A plasma sample was obtained after one hour, and the amount of radioactivity remaining in the sample after a one hour dialysis was determined. The results indicate that impairment of liver function in the rat greatly reduces the loss of I^{131} tag from RIAP.

Methods. Young adult Sprague-Dawley rats, weighing 150-200 g were used in all experiments, male rats in Exp. I and II, females in Exp. III. 20 μc of RIAP (Abbott) in 0.5 ml 0.9% saline was administered intravenously to each animal. In all animals, anesthesia was induced with 40 mg/Kg sodium pentobarbital, given intraperitoneally.

In Exp. I, male rats were divided into experimental and control groups and allowed food and water *ad libitum* until anesthetized. The experimental animals were then eviscerated after ligating and severing the celiac axis and portal vein. The liver was left *in situ*, after

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the technique described by Russel(3). RIAP was then injected into the inferior vena cava and the incision closed by wound clips. After one hour, blood samples were obtained by aortic catheterization. The blood was centrifuged and 0.5 ml plasma samples placed into counting tubes. One ml of plasma was then placed in a cellophane bag (80 μ thick, pore size 30 Å) suspended in 20 ml of distilled water and the dialysis allowed to proceed for one hour(1). At the end of this time, 0.5 ml samples were taken from inside the dialysis bag. All samples were then counted in a well-type scintillation counter and the percent of radioactivity remaining inside the dialysis bag calculated.

In Exp. II, male rats were separated into experimental and control groups and fasted for 12 hours with free access to water. The experimental animals were then given an intraperitoneal injection of 0.3 ml/100 g of a 1:10 mixture of carbon tetrachloride USP and mineral oil, according to the method of Koch-Weser, Farber and Popper(4). Twenty-four hours after this injection, the animals were anesthetized, laparotomies performed and RIAP injected. One hour later blood samples were obtained and treated as in Exp. I.

In Exp. III, female rats were divided into 3 groups and fasted for 12 hours. The rats comprising Group 1 were then given an intraperitoneal injection of 75 mg/100 g of an aqueous solution of dl-ethionine in 3 divided doses(4). Animals Group 2 were given carbon tetrachloride as described above. The animals in Group 3 served as controls. After 48 hours, the animals were anesthetized and treated as described in the preceding experiments.

Results. The data from the 3 experiments are given in Table I and shown graphically in Fig. 1. After dialysis, the percent of radioactivity remaining in plasma samples from control rats in Exp. I and II, with unimpaired liver function, remained within a narrow range (5.6-10.6%). In Exp III, after 60 hours of fasting, a higher percentage of radioactivity remained ($P<.01$, Fig. 1). All methods utilized to impair liver function resulted in a significantly higher percentage of radioactivity remaining in the plasma of experimental animals after dialysis. The least loss of I^{131} from RIAP was seen in those animals with ligation of the celiac axis and portal vein. Acute poisoning with carbon tetrachloride or with dl-ethionine resulted in percentages lower than the experimental group of Exp. I, but significantly higher than in control groups. Significantly higher percentages were observed 24 hours after carbon tetrachloride poisoning than after 48 hours.

TABLE I. Results of dialysis of radioactivity from rat plasma after injection of radioiodinated antipyrine.

Exp.	No. Rats	% Radioactivity remaining in plasma after 1 hr dialysis ± S.D. (range)
I A Evisceration	10	26.5 ± 5.8 (17.7 – 32.9)
I B Control	10	7.8 ± .5 (5.0 – 10.6)
II A CCl_4 24 hr	6	18.5 ± 1.7 (16.5 – 21.3)
II B Control	6	8.2 ± 1.0 (6.8 – 9.2)
III A Ethionine	7	16.5 ± 3.9 (9.8 – 21.8)
III B CCl_4 48 hr	6	14.3 ± 2.6 (11.1 – 18.6)
III C Control	8	9.1 ± 1.5 (7.5 – 12.4)

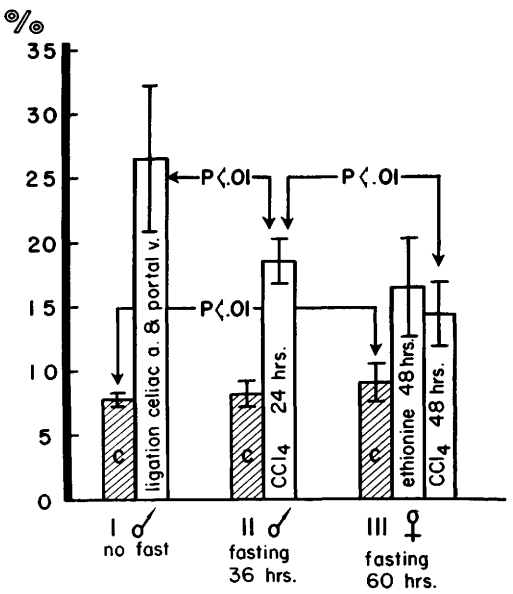

FIG. 1. Chart showing percent radioactivity remaining inside the cellophane membrane after 1 hr. dialysis of plasma from experimental and control (c) rats. Vertical bars represent S.D.

Discussion. This study shows that impairment of liver function results in a decrease in diffusibility of radioactivity through cellophane in the plasma of the rat following injection of RIAP. This decrease can be detected by a relatively simple procedure since it is correlated with the percent radioactivity remaining in a plasma sample after dialysis.

The comparatively narrow ranges of values observed in the control animals of Exp. I and II indicated that the amount of freely diffusible radioactivity appearing in the plasma of rats one hour after administration of RIAP is relatively constant. This range was taken to indicate normal liver function. The less diffusible form of radioactivity observed in the control animals of Exp. III differed significantly from this range, indicating impairment of liver function after a 60 hour fast. It has been shown by Addis *et al.*(5) that the fasting rat loses 20% of its liver protein after 48 hours, and 40% after 168 hours. Miller(6) has demonstrated a loss of liver catalase, alkaline phosphatase, xanthine dehydrogenase and cathepsin activity in the fasting rat, which parallels or exceeds the loss of liver protein. A decrease in deaminating, transaminating and glycogen forming functions in protein-depleted rats has been described by Kaplansky et al(7). The present study indicates that along with the above impairment of enzymatic activity, there is an impairment of ability of the fasting rat liver to release I^{131} tag from the RIAP molecule.

It was noted that more severe forms of liver damage, such as that produced by ligation of its blood supply, produced the greatest decrease in cellophane diffusibility of the I^{131} tag, although the lesser degree of liver damage produced by carbon tetrachloride and dl-ethionine poisoning also resulted in a marked decrease in diffusibility.

Russel has reported that ligation of the celiac axis and portal vein results in total impairment of liver function in the rat(3). The results obtained in these eviscerated rats differed significantly from those rats poisoned with carbon tetrachloride or dl-ethionine. A statistically significant difference was also noted between the results in those animals studied 24 hours after carbon tetrachloride poisoning and those studied at 48 hours, reflecting the greater degree of liver function that has been reported to take place at this time(4). These data demonstrate that varying degrees of liver damage can be quantitated by this method.

Koch-Weser, Farber and Popper(4) measured liver function at several intervals following carbon tetrachloride and dl-ethionine poisoning, using the doses described above. At 48 hours they reported the following: control rats; BSP retention in mg % of serum, 0.12 ± 0.130; bilirubin 2.10 ± 0.64 mg %. Two days after ethionine poisoning; BSP 4.05 ± 0.980 mg % (80% retention).

It is concluded that impairment of liver function can be detected and quantitated by measuring the cellophane diffusibility of the I^{131} tag one hour after intravenous injection of RIAP in the rat. It is possible that this procedure can be adapted as an accurate measure of liver function in human subjects.

Summary. To determine whether impairment of liver function interferes with release of I^{131} tag from radioiodinated antipyrine, groups of rats were functionally hepatectomized, poisoned with carbon tetrachloride or poisoned with dl-ethionine. RIAP was then injected intravenously and the diffusibility of the I^{131} tag through a cellophane membrane measured after one hour. Functional hepatectomy resulted in a marked decrease in diffusibility, while carbon tetrachloride and dl-ethionine poisoning produced a significant decrease in diffusibility. Return of liver function after carbon tetrachloride poisoning resulted in an increase of diffusibility. The data support the conclusion that the release of I^{131} tag from RIAP is a function of the liver and can be used to measure liver function.

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