the tissues of rats given thyroxine were homogenized and subjected to fractionation into their subcellular components, triiodothyronine was more readily recovered than was the case where the whole homogenate was used. A possible explanation for this is discussed.

1. Gross, J., and LeBlond, C. P., *Endocrinology,* 1951, v48, 714.
2. ———, PROC. SOC. EXP. BIOL. AND MED., 1951, v76, 688.
3. Gross, J., and Pitt-Rivers, R., *Lancet,* 1952, v262, 439.
4. ———, *Biochem. J.,* 1953, v53, 645.
5. Roche, J., Lissitzky, S., and Michel, R., *Compt. rend. Acad. Sc.,* 1952, v234, 1228.
6. Gross, J., and Pitt-Rivers, R., *Lancet,* 1952, v262, 593.
7. ———, *Biochem. J.,* 1953, v53, 652.
8. Tomish, E. B., and Woollett, E. A., *Lancet,* 1953, v264, 726.
9. MacLagan, N. F., Sprott, W. E., and Wilkinson, J. H., *ibid.,* 1952, v263, 915.
10. Heming, A. E., and Holtkamp, D. E., *Fed. Proc.,* 1953, v12, 330.
11. Gross, J., Pitt-Rivers, R., and Trotter, W. R., *Lancet,* 1952, v262, 1044.
12. Albright, E. C., Larson, F. C., and Trust, R. H., PROC. SOC. EXP. BIOL. AND MED., 1954, v86, 137.
13. MacLagan, N. F., and Sprott, W. E., *Lancet,* 1954, v267, 368.
14. Roche, J., Michel, R., Michel, D., and Lissitzky, S., *Compt. rend. Soc. Biol.,* 1951, v145, 228.
15. Roche, J., and Michel, R., *Acta Endocrinologica,* 1954, v17, 385.
16. Kalant, H., Lee, R. B., and Sellers, E. A., *Endocrinology,* 1955, v56, 127.
17. Taurog, A., *The Thyroid,* Brookhaven Symposia in Biology, 1954, No. 7, 88.
18. Braasch, J. W., Flock, E. V., and Albert, A., *Endocrinology,* 1954, v55, 768.
19. Lee, N. D., and Williams, R. H., *ibid.,* 1954, v54, 5.
20. Thibault, O., and Pitt-Rivers, R., *Lancet,* 1955, v268, 285.

Received August 4, 1955. P.S.E.B.M., 1955, v90.

Intracellular Potassium Concentration in Normal and Diabetic Rat Liver.* (21950)

LOUIS TOBIAN,† WILLIAM I. MORSE,‡ AND A. BAIRD HASTINGS.
From the Department of Biological Chemistry, Harvard Medical School, Boston, Mass.

It has previously been shown that marked abnormalities of carbohydrate metabolism occur in liver slices from normal animals incubated under conditions leading to lowered intracellular potassium concentration(1). Thus, potassium depletion leads to decreased glycogen deposition from glucose and to decreased overall glucose utilization, while gluconeogenesis from pyruvate and other precursors is markedly increased. These metabolic abnormalities are qualitatively similar to those found in liver slices from diabetic rats even in the presence of normal intracellular potassium concentration(2,3). Furthermore, a number of observations have been made, both clinically and in experimental animals, suggesting that potassium depletion may lead to disturbances of carbohydrate metabolism qualitatively similar to those of diabetes mellitus(4,5). It seemed of interest, therefore, to determine whether lowered intracellular potassium concentration occurred in diabetic liver *in vivo*. Accordingly, electrolyte, water and glycogen analyses were made on diabetic and normal rat livers.

Material and methods. Male rats weighing 250 g were made diabetic by the intravenous injection of alloxan (40 mg per kilo). Three to 4 weeks later these animals, as well as normal controls, were fasted for 8 hours (water *ad lib.*), then sacrificed for blood and liver analysis. The animals were sacrificed under nembutal anesthesia by severing the carotid

* Supported by grants from the American Heart Assn. and the Eugene Higgins Trust through Harvard University.

† Established Investigator of the American Heart Assn.

‡ R. Samuel McLaughlin Traveling Fellow in Medicine.

TABLE I. Blood Analysis on Normal and Diabetic Rats.

Group	Blood sugar, mg/100 ml blood		Na	K
	Mean	Range	mEq/L serum	
Normal (11)	99	78-117	149.1 ± 6.4*	4.74 ± 1.5*
Diabetic (11)	292	121-470	144.7 ± 3.8	4.25 ± 1.8

* Mean values ± stand. error of mean.

arteries. The livers were removed, blotted free of blood, and 2 aliquots were taken. One was for glycogen analysis(6) and the other was used to determine water, sodium and potassium. This second aliquot was dried, defatted and extracted with 0.75 N HNO_3 as described by Lowry and Hastings(7). Sodium and potassium were determined by flame photometry. Whole blood was also analyzed for glucose(8) and blood serum for sodium and potassium.

Results. Table I shows the blood sugar and serum sodium and potassium values for 11 normal and 11 diabetic animals. The serum sodium and potassium concentrations were slightly lower in the diabetic than in the normal group. These data were used in calculating the intracellular potassium concentrations.

In Table II are recorded the analytical data obtained on the normal and diabetic livers, together with calculated values for extracellular water $(H_2O)_e$, intracellular water $(H_2O)_c$ and intracellular potassium concentration $[K]_c$. From the analytical data it may be seen that no statistically significant difference was observed in the concentrations of water, potassium or sodium between the normal and diabetic livers.

Using these data, and the assumption that the sodium of liver is entirely extracellular, the extracellular water $(H_2O)_e$ and intracellular water $(H_2O)_c$, in g per kilo of fat free tissue, were calculated according to methods previously described(7). No difference in either $(H_2O)_e$ or $(H_2O)_c$ was found between the livers of the normal and diabetic groups.

After making allowance for the amount of potassium in the extracellular water of the tissues, the concentration of potassium in the intracellular water $[K]_c$ was calculated. These values proved to be 187 mEq per kilo of intracellular water for the normal livers and 184 mEq per kilo of water for the diabetic livers. The difference is without significance.

Discussion. Since abnormalities in carbohydrate metabolism similar to those encountered in diabetic liver have been observed in normal liver where intracellular potassium concentration is experimentally reduced below normal, we have felt it important to determine whether the intracellular potassium concentration of liver is or is not reduced below normal in diabetes. From the data presented above it would appear that the intracellular potassium concentration is quite normal in diabetic liver. It would seem, therefore, that the metabolic abnormalities which have been described in hepatic tissue for diabetic animals are not attributable to a reduction in potassium concentration but rather it appears more likely that potassium depletion and insulin de-

TABLE II. Analytical Data on Normal and Diabetic Rat Livers. Mean values are expressed per kg of fat-free tissue (F.F.T.)—standard error of the mean.

Group	H_2O, g	K, mEq	Na, mEq	Glycogen, g
Normal (11)	715 ± 1.5	96.2 ± .6	31.4 ± 2.1	7.0 ± 1.2
Diabetic (11)	723 ± 2.8	95.5 ± 1.0	30.5 ± 1.2	19.4 ± 2.2
		Derived data		
	$(H_2O)_e$,* g/kg F.F.T.	$(H_2O)_c$,† g/kg F.F.T.	[K], mEq/kg (H_2O)	[K], mEq/100 g F.F. solids
Normal (11)	198 ± 15	517 ± 15	187 ± 8	33.8 ± .2
Diabetic (11)	198 ± 7	515 ± 7	184 ± 4	33.3 ± .2

* $(H_2O)_e$—g of extracellular H_2O per kg of F.F.T., calculated on the assumption that liver Na is extracellular.

† $(H_2O)_c$—g of intracellular H_2O per kg of F.F.T., calculated from total tissue H_2O and $(H_2O)_e$.

ficiency affect intracellular metabolism similarly but independently.

Summary. The intracellular potassium concentration was found to be normal in diabetic rat liver.

1. Hastings, A. B., Teng, C. T., Nesbett, F. B., and Sinex, F. M., *J. Biol. Chem.*, 1952, v194, 69.

2. Renold, A. E., Teng, C. T., Nesbett, F. B., and Hastings, A. B., *ibid.*, 1953, v204, 533.

3. Hastings, A. B., Renold, A. E., and Teng, C. T., *Trans. Assn. Am. Phys.*, 1953, v66, 129.

4. Gardner, L. I., Talbot, N. B., Cook, C. D., Bermenand, H., and Uribe, R., *J. Lab. and Clin. Med.*, 1950, v35, 592.

5. Eliel, L. P., Pearson, O. H., and White, F. C., *J. Clin. Invest.*, 1952, v31, 419.

6. Good, C. A., Kramer, H., and Somogyi, M., *J. Biol. Chem.*, 1933, v100, 485.

7. Lowry, O. H., and Hastings, A. B., *ibid.*, 1942, v143, 257.

8. Somogyi, M., *ibid.*, 1945, v160, 61.

Received August 10, 1955. P.S.E.B.M., 1955, v90.

Renin and Antirenin in Treatment of Long Term Experimental Renal Hypertension in the Dog.* (21951)

S. H. KREMEN AND G. E. WAKERLIN.
(With the technical assistance of L. Graham, B. Keith, and R. Speer.)
From the Department of Physiology, College of Medicine, University of Illinois, Chicago.

A number of reports have suggested that the pathogenesis of experimental renal hypertension changes with time. Thus it has been suggested that in acute experimental renal hypertension the pathogenesis is renal and in chronic experimental renal hypertension the pathogenesis is extrarenal or (according to one group of investigators) neurogenic. There have been no reported studies on the pathogenesis of long term experimental renal hypertension (*i.e.*, hypertension of 4 or more years duration) in the dog. Thus Ogden and colleagues(1,2) showed that certain sympatholytic and general anesthetic drugs decreased the blood pressure of chronic renal hypertensive rats but not of acute renal hypertensive rats. They also showed that the removal of a unilaterally constricted kidney in the rat caused a decrease in blood pressure in acute but not in chronic renal hypertensive rats. On the basis of these findings, Ogden suggested that the pathogenesis of experimental renal hypertension changes with time from a renal pressor mechanism to a neurogenic mechanism. Likewise, several groups of British investigators have reported that complete nephrectomy in the acute renal hypertensive rat produces a decrease in blood pressure, whereas there is no decrease during survival in the chronic hypertensive rat(3,4). These results are similar to those of earlier workers who reported that total nephrectomy in renal hypertension of one week's duration in the rabbit results in normotension but after 2 to 4 months of hypertension, total nephrectomy results in persistence of the hypertension during survival(5,6). Floyer and others have interpreted these findings as demonstrating an extrarenal mechanism for the pathogenesis of chronic experimental renal hypertension. Moss and Wakerlin(7) and Page(8) however, found no significant difference in the effects of sympatholytic and general anesthetic drugs in acute and chronic experimental renal hypertension in dogs. Moss and Wakerlin considered acute experimental renal hypertension in dogs to last 6 months following renal artery constriction and most of their acute hypertensive dogs were 3 to 6 months hypertensive. Since then, Haynes and Dexter(9) showed that an increase in plasma renin following renal artery constriction in the dog persists for some weeks and Shorr and colleagues(10) showed that increased plasma VEM is balanced by VDM 8 to 12 weeks following renal

* Aided by grants from the National Heart Institute, the Chicago Heart Assn.; and the American Heart Assn.