ments, urinary secretion was similar to that following hemorrhage. During this stage, considerable hemoconcentration (75% in one of the experiments) was present, yet the infusion stimulated considerable kidney secretion. It seems evident from these 2 groups of experiments that acidosis and hemoconcentration are not the important factors in complete anuria of shock. We may therefore assume, that reduced blood volume, hemoconcentration, low blood pressure, and acidosis do not lead to a permanent anuria, *i.e.*, one which is not abolished by infusions.

The second type of anuria may be called burn anuria. This type did not always respond to an infusion fluid. In experiment B1 the kidneys secreted urine only after the second infusion. In experiment B2 a total amount of 2,430 cc of saline had no effect on kidney secretion. The difference in response might be attributed to different amounts of hemoglobin in the plasma. In B1 the plasma hemoglobin was 1385 mg %, whereas in experiment B2 it was 3680 mg %. The severity of the anuria in all burn experiments frequently could be associated with the degree of hemoglobinemia.

The third type of anuria may be called crush anuria. It should not be mistaken for the first type, *i.e.*, the anuria of low blood pressure and diminished blood volume. In crush anuria, the kidneys always secreted well after the first infusion as in the first type, but the urine was deeply pigmented, presumably with myohemoglobin.[4] This was a sign of impending damage to the kidneys. Unlike following burns, the anuria developed very slowly and only after 18 hours or longer the kidneys might stop secreting, even after infusion. It was important to give infusions in order to determine the extent of damage to the kidneys. It seemed possible to prevent the crush anuria by giving certain infusion fluids early. Saline and plasma did not prevent it in all cases. In many cases complete anuria did not occur either in burns or in crush injuries, but all experiments exhibited some degree of oliguria.

Summary and Conclusions. I. Three different types of anuria were observed in these experiments.

1. Anuria following a severe hemorrhage or a reduced blood volume.
2. Anuria following a severe burn.
3. Anuria following a crush injury: this type developed much later, 18 to 36 hours, after injury.

II. The possible factors for the appearance of anuria are: low blood pressure, a reduced blood volume, the hemoglobinemia present in burns, and an unknown substance. This substance may be myohemoglobin or a toxic agent released from the damaged limb.

III. Permanent damage to the kidneys and anuria may develop following crushing or burning, because renal function seems to be most impaired following these two traumatic agents.

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14596

Absorption of Galactose by Renal Tubules of the Dog.

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In previous papers from this laboratory evidence was presented to show that in the rat and in man, thyroid hormone increases the rate of the intestinal absorption of substances that are transferred actively by a mechanism generally considered to involve phosphorylation; the hormone was without influence on the rate of absorption of those substances that

are considered not to undergo phosphorylation.[1,2] Our[3] interest in the manner by which thyroid hormone exerts an influence on the mechanism involved in the active transfer of solutes across membranous barriers has led to a study of the effect of this hormone on renal tubular transfers. In a recent publication[4] we reported that thyroid hormone accelerates the active transfer (absorption) of glucose by the renal tubules of the dog. The observation that thyroxin also accelerates the active tubular transfer of diodrast, together with other considerations, led us to postulate that thyroid hormone may exert its characteristic effect on metabolism and on transfer mechanisms by increasing the effective concentration of adenosinetriphosphatase. Additional data supporting this hypothesis is now available.[5] In this report the comparison of intestinal and renal transfer mechanisms, and the influence of thyroid hormone thereupon, has been extended to include galactose. It is well known that the absorption of glucose and galactose by the intestine involves an active transfer mechanism;[6] it is likewise known that such a mechanism is involved in the renal tubular absorption of glucose.[7] The behavior of the renal tubules toward galactose is less clear.[8]

Experimental. I. The renal tubular absorption of galactose has been studied by the use of the method of Shannon and Fisher[9] in 2 female dogs in the normal state and in one animal after the administration of 9 daily doses (40 mg) of thyroxin. The details of our experimental procedure were as described previously,[4] with the exception that each urine collection period was extended to about 25 minutes (exactly timed). An interval of one-half hour was allowed for the establishment of equilibrium of plasma levels in those experiments involving changes in the concentration of galactose in the plasma. Reducing values were determined according to the method of Hagedorn and Jensen (1923). Filtrates (plasma and urine) were cleared of glucose by yeast fermentation. The non-sugar reducing substances were, in the case of urine, removed by precipitation with mercury[10] and, in the case of plasma, determined on suitably prepared blanks.

Results. It is evident from the data presented in Table I that within the limits of a four-fold change in the concentration of galactose in the plasma, a relatively constant fraction (about 40%) of the galactose filtered through the glomerulus is absorbed by the tubules. It is also apparent that within the limits studied (Creatinine U/P ratios, 8-67), the fraction absorbed is independent of the degree of concentration of the glomerular filtrate. The data for the animal in the hyperthyroid state suggests that thyroid hormone is without influence on the renal tubular transfer of galactose. It is unfortunate that circumstances prevented the accumulation of a greater amount of data on the animals in the hyperthyroid state. However, the data is more significant than at first appears, for the animals that were employed in this investigation were the same as those used in our previous study of the influence of thyroxin on the glucose Tm and the diodrast Tm.[4] Since these 2 studies were carried on simultaneously, and since Dog C showed a marked increase in the tubular transfer of glucose as a result of the administration of thyroxin only a few days prior to the galactose study, there can be little doubt that the galactose data was typical of the hyperthyroid animal.

Discussion. A comparison of the behavior

1 Althausen, T. L., and Stockholm, M., *Am. J. Physiol.*, 1938, **123**, 577.

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3 Eiler, John J., Stockholm, M., and Althausen, T. L., *J. Biol. Chem.*, 1940, **134**, 283.

4 Eiler, John J., Althausen, T. L., and Stockholm, Mabel, *Am. J. Physiol.*, 1944, **140**, 699.

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7 Smith, Homer W., *Lectures on the Kidney*, University Extension Division, University of Kansas, 1943.

8 Harding, V. J., and Grant, G. A., *J. Biol. Chem.*, 1933, **99**, 629.

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10 West, E. S., and Peterson, V. L., *Biochem. J.*, 1932, **26**, 1720.

TABLE I.
Renal Tubular Absorption of Galactose.

			Creatinine			Galactose			
Dog	Date	State of animal	Plasma mg %	U/P ratio	Clearance cc/min	Plasma mg %	Filtered mg/min	Absorbed mg/min	Ratio Absorbed / Filtered
B	3/5/42	Normal	32.0	12.7	44.6	229	102	44	.43
			29.3	7.9	47.4	229	109	40	.37
			28.7	7.8	46.5	229	107	44	.41
	3/10		28.6	9.8	53.7	113	61	25	.41
			28.2	9.2	47.0	107	50	18	.36
			28.1	13.1	47.0	102	48	18	.38
			31.3	12.3	47.9	229	110	39	.36
			31.4	10.2	46.8	292	137	52	.38
								Avg	.39
C	7/7	"	31.7	12.8	67.6	179	122	70	.51
			27.8	23.1	64.7	175	113	71	.63
			26.5	40.4	68.6	165	113	66	.58
	7/9		32.6	9.8	62.0	371	230	92	.40
			28.2	16.5	66.0	415	274	120	.44
			27.8	13.6	65.5	446	292	116	.40
								Avg	.49
	5/29	Hyperthyroid	28.2	43.2	73.4	158	116	48	.41
			25.2	67.1	80.5	146	118	59	.50
			24.1	49.8	89.6	197	176	79	.45
			23.5	—	92.8	241	224	100	.45
								Avg	.45

of the intestine and the renal tubules toward glucose and galactose shows that while the intestine handles these 2 sugars in a similar fashion, there is a notable difference in the treatment they are accorded by the renal tubules. The question arises whether or not the differences in renal tubular absorption of these sugars is due to the absence of an active mechanism for the transfer of galactose. The present data is compatible with passive diffusion providing it is assumed, as has been shown to be the case for glucose, that the distal portions of the tubules are relatively, or actually, impermeable to galactose. The constant fraction of filtered galactose that is absorbed would then be occasioned by the diffusion gradient effected by the obligate[11] absorption of water in the proximal tubules, which in the range of U/P ratios studied (8-67) represents a constant fraction of the glomerular filtrate.[12] However, such an interpretation necessitates the conclusion that under a given diffusion gradient the membranes of the proximal tubules are equally permeable to galactose and urea, for Shannon[13] has estimated that about 40% of the filtered urea is absorbed by virtue of diffusion in the proximal tubules. Unless it be assumed that phlorhizin alters the permeability of the proximal tubular membranes toward monosaccharides, in addition to inhibiting a transfer mechanism, studies involving the use of this drug militate against a high rate for the penetration of sugars. Additional evidence may be cited against passive diffusion. A comparison of the data for galactose with that obtained from a study

[11] Smith, Homer W., *The Physiology of the Kidney*, Oxford University Press.

[12] Walker, Arthur M., and Bott, Phyllis A., *Am. J. Physiol.*, 1941, **134**, 580.

[13] Shannon, J. A., *Am. J. Physiol.*, 1938, **122**, 782.

of the tubular transfer of xylose[14] would indicate that the differences in the tubular absorption for glucose and galactose is not to be attributed to the absence of an active mechanism for the transfer of galactose. The tubular transfer of galactose and xylose is quite similar in several respects; the fraction of the filtered sugar that is absorbed is independent, in both cases, of the concentration of the sugar in the plasma, and of moderate changes in the degree of concentration of the glomerular filtrate. A demonstrated difference lies in the magnitude of the fraction absorbed. In contrast to 40% for galactose only 27% of the filtered xylose is absorbed. The observation that elevation of the plasma glucose level completely and reversibly blocks the tubular absorption of xylose, together with conclusions drawn from a sound, critical analysis of the kinetics of transfer mechanisms, has led Shannon[14] to conclude that xylose is absorbed by an active tubular process which is identical with that responsible for the absorption of glucose. Studies involving the use of phlorhizin would appear to support such a conclusion. Since galactose is absorbed by the renal tubules at a greater rate than xylose, it seems reasonable to conclude that the tubular absorption of the galactose also involves an active transfer mechanism. The difference in behavior of glucose and galactose, including the differences in their response to thyroxin, may then be attributed to a difference in the rate determining step in the sequence of reactions responsible for the active transfer.

Summary. The renal tubules of the dog absorb about 40% of the galactose that appears in the glomerular filtrate; within rather wide limits the fraction that is absorbed is independent of the concentration of plasma galactose and of the degree of concentration of the glomerular filtrate. In contrast to glucose, the renal tubular transfer of galactose is not altered by the administration of thyroxin. It is likely that a transfer mechanism is involved in the tubular absorption of this sugar.

[14] Shannon, J. A., *Am. J. Physiol.*, 1938, **122**, 755.

14597 P

Electrophoresis of Purified Prothrombin.

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Prothrombin has not been identified as a separate component in the electrophoresis patterns of oxalated or citrated plasma, because it is not present in sufficient concentration to permit detection. For this reason extensive electrophoretic data can be obtained only on concentrated preparations of prothrombin. In previous purification work[1,2] impurities have been removed in progressively greater proportion. In recent work we have obtained preparations of still higher purity. This offered an opportunity to study the electrophoretic properties of prothrombin for the first time.

The Tiselius apparatus used for this work is equipped with a Schlieren type lens and a Svensson diaphragm. The mobilities were calculated from observations of the descending boundary, in accordance with the recommendations of Longsworth and MacInnes.[3] The values for any given pH represent the average of several determinations. A new prothrombin preparation was made for each run. It is planned to offer a comprehensive presentation

[1] Seegers, W. H., Smith, H. P., Warner, E. D., and Brinkhous, K. M., *J. Biol. Chem.*, 1938, **123**, 751.

[2] Seegers, W. H., *J. Biol. Chem.*, 1940, **136**, 103.

[3] Longsworth, L. G., and MacInnes, D. A., *J. Am. Chem. Soc.*, 1940, **62**, 705.