These results are in part contradictory to those obtained by Ikeda,[2] who found that glycogen from mammalian tissues induced organ-specific antibodies when injected into rabbits.

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Rôle of Thyroid in Increased Protein Metabolism of Phlorhizin Diabetes.*

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The mechanism of phlorhizin diabetes has been the subject of much investigation and controversy. Two distinct views exist, one postulating that the action of phlorhizin is exclusively renal[1, 2] and the other that some primary extrarenal factor also is involved.[3, 4] These views arise from the fact that although no apparent effect can be observed when phlorhizin is administered to dogs without renal function,[2] a definite hyperglycemia,[3] and abnormalities in the glucose tolerance curve[4] may occur in such animals if the phlorhizination is completed before the kidneys are incapacitated.

The observations of Dann, Chambers and Lusk[5] indicate the involvement of extrarenal factors in the genesis of phlorhizin diabetes. They demonstrated that the administration of phlorhizin to the thyroidectomized dog does not result in the increased basal metabolic rate and the increased nitrogen excretion which occur in the intact phlorhizinized dog. During the course of a series of studies on nitrogen metabolism, we had occasion to make some observations which confirm and extend those of Dann, Chambers, and Lusk.

Three groups of animals were studied. The first group consisted of normal dogs which had been fasted for 3 to 4 days; the second, of normal dogs which had received a daily subcutaneous injection

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[2] Ikeda, G., *Jap. J. Exp. Med. Sci. Trans.*, 1932, **7**, 231.

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[1] Minkowski, O., *Arch. f. exp. Path. u. Pharm.*, 1892, **31**, 85.

[2] Deuel, H. J., Jr., Wilson, H. E. C., and Milhorat, A. T., *J. Biol. Chem.*, 1927, **74**, 265.

[3] Underhill, F. P., *J. Biol. Chem.*, 1912, **13**, 15.

[4] Goldstein, L. A., Tatelbaum, A. J., Ehre, S., and Murlin, J. R., *Am. J. Physiol.*, 1932, **101**, 166.

[5] Dann, M., Chambers, W. H., and Lusk, G., *J. Biol. Chem.*, 1931, **94**, 511.

of a suspension of phlorhizin in oil for 3 to 4 days, during which time all food was withheld; the third, of dogs subjected to a complete thyroidectomy and subsequently phlorhizinized.

On the fourth day of fasting and of phlorhizination, a bilateral nephrectomy was performed under sodium amytal anesthesia which was maintained throughout the experimental period. Immediately after the completion of this operation, an arterial blood sample was drawn from an exposed femoral artery for determination of non-protein-nitrogen by a micro-Kjeldahl method (digestion according to the Koch-McMeekin and distillation by the Bock-Benedict method), sugar by the Somogyi modification of the Shaffer-Hartmann method and total solids by drying a weighed quantity of blood to constant weight. Arterial blood samples were drawn again at 2- to 4-hourly intervals for 12 hours. In a number of instances, the experimental period was extended an additional 12 hours in order to note any late changes in the accumulation of blood non-protein-nitrogen.

During the early stages of these studies, all non-protein-nitrogen and sugar values of the blood were corrected for changes in the water content, but this was subsequently abandoned because of the negligible changes observed.

In another communication[6] we have presented evidence to show that the rate of non-protein-nitrogen accumulation in the blood of nephrectomized dogs is as accurate a gauge of protein metabolism as is the determination of urinary nitrogen excretion. Accordingly, the data summarized in Table I reveal that even in the absence of kidneys, the protein metabolism of the phlorhizinized dog is about 30% higher than that of the normal dog. This is true even if the experimental period is extended over 24 hours instead of 12 as herein reported. Thus, it is obvious that the removal of the kidneys does not result in a complete cessation of phlorhizin diabetes. However, when phlorhizin is administered to the previously thyroidectomized dog, the rate of protein metabolism is the same or slightly lower than that of the normal dog after the kidneys are removed. This indicates that the process of phlorhizination probably results in stimulation of the thyroid gland which in turn is responsible for part of the increased protein metabolism observed in phlorhizin diabetes.

In spite of the fact that the rate of protein metabolism is greater in the normal phlorhizinized-nephrectomized dogs than in the previously thyroidectomized, phlorhizinized-nephrectomized dogs, the

[6] Mirsky, I. A., Heiman, J. D., and Swadesh, S., (in press).

TABLE I.

The Blood Non-Protein Nitrogen of Nephrectomized Normal, Phlorhizinized and Thyroidectomized-Phlorhizinized Dogs.

Experiments	No. of Exp.	Blood Sugar							Blood Non-Protein Nitrogen						
		0	4 hr.		8 hr.		12 hr.		0	4 hr.		8 hr.		12 hr.	
		Actual	Actual	Increase	Actual	Increase	Actual	Increase	Actual	Actual	Increase	Actual	Increase	Actual	Increase
		mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%	mg.%
Normal Controls	5	66	66	0	66	0	70	4	29.7	40.4	10.7	52.3	22.6	66.1	36.4
Phlorhizinized Controls	5	50	106	56	97	47	104	54	45.6	59.2	13.6	74.7	29.1	92.8	47.2
Thyroidectomized-Phlorhizinized	3	31	75	44	95	64	85	84	31.5	43.3	11.5	53.8	20.0	62.5	31.0

blood sugar levels are approximately the same. Thus, both sets of animals have a low initial blood sugar level before nephrectomy, and rise to a level somewhat higher than that observed in normal animals after nephrectomy. This post-nephrectomy blood sugar level is maintained constant over the period of time studied. Hence, it is probable that the blood sugar *per se* is not the factor responsible for the stimulation of the thyroid gland.

Summary and Conclusions. The administration of phlorhizin results in a stimulation of the thyroid gland which in turn is responsible for part of the increased protein metabolism observed in phlorhizin diabetes. Since this increased protein metabolism of the phlorhizinized animal continues even after the removal of the kidneys, but does not occur when phlorhizin is administered to previously nephrectomized animals,[2] it is probable that this drug exerts some specific effect on the kidney, which is in turn responsible for the thyroid stimulation. Our observations do not offer evidence for the possibility that the blood sugar level is the responsible factor in the increased activity of the thyroid but suggest some primary renal factor.

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Deleterious Effects of Insulin Shock.

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The effects of a hypoglycemia of long duration on the various tissues and the organism as a whole are of fundamental importance. On the practical side the possibility of the use of excessive doses of the new slow acting protamine insulin by untrained individuals, because of the lack of immediate results or the accidental administration of the concentrated precipitate from a vial which has not been agitated, make essential a knowledge of the effects of hypoglycemia due to insulin shock.

In experiments on dogs we have found that when a state of insulin shock is maintained for 24 hours or longer with protamine zinc insulin it is apparently impossible to resuscitate the animal and death always ensues. Often this result follows when the period is shorter. A typical protocol follows: A male dog weighing 29 kg., last fed the day before the experiment started, was given 40 units (1 cc.)