were well developed and actively secretory. There were decidua-like cells in the stroma.

Our results show that both the effects previously found in rodents and those reported in primates, can be obtained in the same species (rats) by varying the dosage of testosterone propionate. This explains the apparently contradictory results which have been obtained with this hormone, and obviates the necessity for postulating a species difference in its action. The estrus-suppressing effects in our rats were obtained with much smaller doses of testosterone propionate than have hitherto been used. It seems likely that the similar results which others have obtained in primates, were due to the arbitrary choice of a relatively small dosage for those experiments.

The different effects of the large and small doses of the androgen may be explained by the different actions of these doses on the anterior hypophysis, and by the direct effects of the amount of androgen injected. Both large and small doses depress the elaboration of follicle-stimulating hormone by the pituitary, as judged by the cessation of estrous cycles. Large doses, however, also cause a release of luteinizing hormone, as indicated by the numerous large corpora lutea in the ovaries. This difference in the effects of large and small doses on the pituitary is similar to that which is known to occur with the estrogens.[9] Thus a small dose of testosterone results in cessation of the estrous cycles, ovarian atrophy, diminished estrin and involution of the uterus. A large dose results in cessation of the estrous cycles, persistent corpora lutea and diminished estrin. But uterine involution does not occur because the larger amount of androgen present acts directly on the uterus to prevent the atrophy and to produce the progestation-like changes.[5, 6]

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Utilization of Carbohydrate by the Phlorhizinized Dog.*

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The glycosuria produced by the administration of phlorhizin has served as an experimental tool in the study of carbohydrate metabo-

9 Hohling, W., *Klin. Wchnschr.*, 1934, **13**, 97.

* Aided by the Max Pam Fund for Metabolic Research.

lism. The scientific literature on the subject, however, presents an anomalous situation as regards the ability of the phlorhizinized animal to utilize sugar. On the basis of characteristic R.Q. values and urinary D:N ratios, Lusk and others[1] concluded that so-called phlorhizin diabetes is essentially similar to pancreatic diabetes, in that no carbohydrate is utilized by either type of animal. However, later work has shown that the phlorhizinized dog exhibits every conventionally accepted criterion of considerable ability to utilize carbohydrate. Thus administration of glucose raises the R.Q., diminishes ketosis,[2, 3] spares protein breakdown,[4] and leads to a synthesis of glycogen.[3] Isolated tissues from phlorhizinized animals, react to a glucose medium with a rise in R.Q. similar to that exhibited by tissues from normal animals.[5]

A parallel situation has existed in regard to the question of the ability of depancreatized animals to utilize sugar. Here too, the characteristic R.Q. of 0.7 and the D:N ratio of about 2.8 were put forward as proof that the depancreatized dog does not utilize carbohydrate. But, as in the case of phlorhizin diabetes, evidence is accumulating which shows that the depancreatized animal is well able to make use of sugar. Removal of the liver from a depancreatized dog leads to a similar fall in the blood sugar level as occurs after hepatectomy in the normal animal. Glucose has to be given to prevent hypoglycemia and maintain life.[6] It has been shown that completely depancreatized dogs maintained without insulin, react to glucose administration with a rise in R.Q., a diminution of ketosis and a sparing of protein.[7, 8] More recently we have shown that carbohydrate utilization is directly related to the height of the blood sugar level, in the depancreatized as well as in the normal dog. At his usual hyperglycemic blood sugar level the diabetic dog uses as much or even more carbohydrate than does the normal dog at his normal blood level.[9]

It is obvious that phlorhizination and pancreatectomy produce metabolic disturbances which are in many ways similar to each other, but which differ in one important respect: pancreatectomy is followed

1 Lusk, G., *The Science of Nutrition,* F. Lippincott Co., Philadelphia, 1925.

2 (a) Wierzuchowski, M., *J. Biol. Chem.,* 1926, **68**, 385; (b) Deuel, H. J., Wilson, H. E. C., and Milhorat, A. T., *J. Biol. Chem.,* 1927, **74**, 265.

3 Nash, T. P., *Physiol. Rev.,* 1927, **7**, 385.

4 Mirsky, I. A., Heiman, J. D., and Swadesh, S., *Am. J. Physiol.,* 1937, **120**, 681.

5 Shorr, E., Loebel, R. O., and Richardson, H. B., *J. Biol. Chem.,* 1930, **86**, 529.

6 Mann, F. C., and Magath, T. B., *Arch. Int. Med.,* 1923, **31**, 797.

7 Soskin, S., *J. Nutrition,* 1930, **3**, 99.

8 Ring, G. C., *Am. J. Physiol.,* 1934, **109**, 88.

9 Soskin, S., and Levine, R., *Am. J. Physiol.,* 1937, **120**, 761.

by hyperglycemia, phlorhizin administration by hypoglycemia. In view of our findings that carbohydrate utilization is directly related to the height of the blood sugar level, it seemed important to study the use of sugar by the phlorhizinized animal, taking this relationship into account.

Dogs were fasted for 3 days and eviscerated on the morning of the fourth day. During this period, 4 daily doses of 1 g of phlorhizin in oil were administered; the last dose being given on the morning of the operation. The effectiveness of the phlorhizination was checked by determining the blood sugar level, and in some animals by calculating their D:N ratios. The details of sugar administration and calculation of utilization will be found in our previous publication.[9]

There were 13 successful experiments on 13 dogs (Fig. 1). All except 2 of the 13 dogs showed a rate of utilization similar to that of normal animals at the same blood sugar levels. The 2 exceptions which showed a smaller rate of utilization, also exhibited a glycogen breakdown far below the average found in the normal, depancreatized and phlorhizinized animals of the present and previous[10] series. Although we have included the results from these 2 animals in Fig. 1, it seems justifiable to exclude them from our discussion for the time being.

Table I compares the average rates of glycogen breakdown and lactic acid increase in normal, depancreatized and phlorhizinized dogs. If we take into account the lower initial glycogen values of the phlorhizinized animals, it can be seen that their rate of muscle glycogen breakdown compares to that of normal rather than to that of depancreatized animals. The rate of lactic acid accumulation shows similar relationships.

It is evident from our results that as compared with normal dogs, there is essentially no diminution in the utilization of carbohydrates by phlorhizinized animals, at blood sugar levels ranging from 9 to 800 mg %. This agrees with other evidence to the same effect, quoted in our introductory remarks. Drury[11] in a recent study of eviscerated phlorhizinized dogs, concluded that they use only one-third the normal amount of carbohydrate. However, he did not properly take into account the carbohydrate supplied by glycogen breakdown, and only measured the disappearance of sugar from the blood stream.

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10 Soskin, S., Levine, R., and Heller, R. E., PROC. SOC. EXP. BIOL. AND MED., 1938, **38**, 6.

11 Drury, D. R., Bergman, H. C., and Greely, P. O., *Am. J. Physiol.*, 1936, **117**, 323.

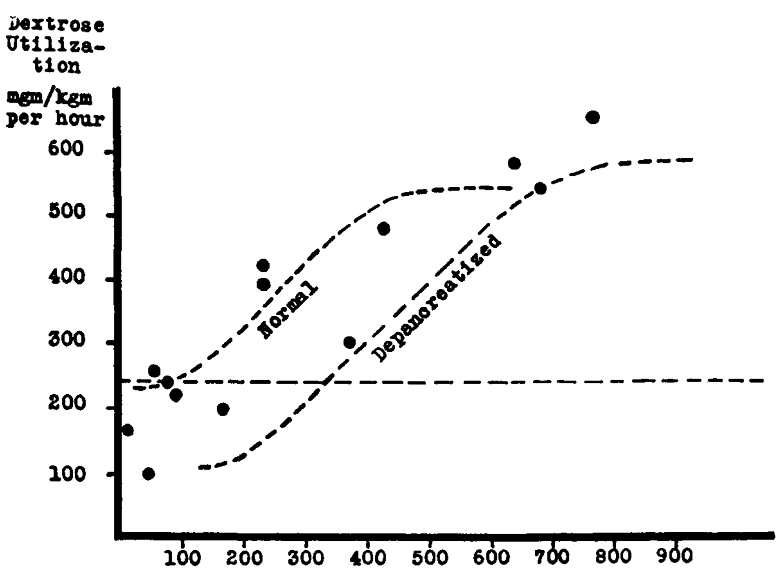

FIG. 1.

Each black dot represents an experimental result on a different phlorhizinized dog. The smooth curves for the normal and depancreatized animals, which are included for comparison, are derived from previously reported experiments.[9] The horizontal line indicates the rate of dextrose utilization of normal dogs within the normal glycemic range.

TABLE I.

Condition	Avg muscle glycogen Initial mg%	Avg muscle glycogen Final mg%	Avg blood lactic acid Initial mg%	Avg blood lactic acid Final mg%	Avg decrease in glycogen mg/%/hr	Avg rise in lactic acid mg/%/hr
Normal	511	355	50.5	106.7	43.1	15.2
Depancreatized	337	217	118.1	183.2	38.4	21.0
Phlorhizinized	431	314	99.4	152.2	32.5	14.5

Completely phlorhizinized dogs excrete large quantities of sugar in the urine. At the same time they use normal amounts of carbohydrate. There must, therefore, be an overproduction of sugar in these animals. Hence, the low R.Q. values in these animals must be related to the overproduction rather than to an underutilization of sugar. These low R.Q.'s may result from a preponderance of reactions in which oxygen is used up without a corresponding production of carbon dioxide, such as the formation of sugar from proteins and fatty acids. That the liver of the phlorhizinized animal contributes a very low R.Q. which reduces the composite R.Q. of the whole animal, has been shown by direct observation of the perfused liver from phlorhizinized animals.[12]

In view of the fact that the utilization of carbohydrate is not inhibited by phlorhizin, the amount of sugar in the urine cannot represent all the sugar formed by the liver. It is, therefore, impossible to consider the D:N ratio of the urine as an index of the amount of sugar formed from protein or other precursors. Direct determinations on perfused livers of normal as well as phlorhizinized animals show that the D:N ratios range between 5.0 and 14.0.[12, 13] These figures present a truer picture of the derivation of sugar than the urinary D:N ratios, which are lowered by the utilization of a considerable portion of the carbohydrate formed.

We have concluded previously that the glycosuria and hyperglycemia of pancreatic diabetes are also due to an overproduction of sugar from non-carbohydrate sources. However, the causative mechanisms of overproduction are not the same in the phlorhizinized as in the depancreatized animal. The removal of the pancreas disturbs the normal regulation of hepatic glycogenesis and glycogenolysis which is dependent upon the height of the blood sugar and the presence of insulin.[14] This disturbance leads to uninhibited glycogenolysis and excessive gluconeogenesis. However, in the phlorhizinized dog, insulin is not lacking.[15] The increased hepatic glycogenolysis and gluconeogenesis are due to the persistently low blood sugar level resulting from the lowered renal threshold for sugar. This lowered threshold which is responsible for the continual glycosuria is due to the inhibition of the mechanism for the reabsorption of glucose in the kidney tubules. Recently Lundsgaard and others have shown that the transfer of sugar from the kidney tubules back to the blood stream probably occurs by way of the formation of hexose-phosphate. This phosphorylation of glucose is inhibited by phlorhizin.[16] Although all tissues are subject to this same action of phlorhizin, the kidney shows the greatest effects because it has a limited ability to destroy phlorhizin and also because the drug accumulates in the kidney in a larger concentration. Muscle tissue destroys phlorhizin very quickly and thus the phosphorylation mechanisms in muscle are not disturbed to any great extent.[17] Our results

12 Blixenkrone-Møller, N., *Ztschr. f. physiol. Chemie,* 1938, **252**, 117.

13 Heller, H., *Acta Med. Scand.,* 1936, **90**, 489.

14 Soskin, S., Mirsky, I. A., Zimmerman, L. M., and Heller, R. E., *Am. J. Physiol.,* 1936, **114**, 648.

15 Nash, T. P., and Benedict, S. R., *J. Biol. Chem.,* 1924, **61**, 423.

16 (a) Lundsgaard, E., *Biochem. Ztschr.,* 1933, **264**, 209; (b) Kalckar, H., *Enzymologia,* 1937, **2**, 47.

17 Lambrechts, A., *Compt. Rend. Soc. Biol.,* 1935, **118**, 1248.

tend to confirm that this is the general rule since, except for the 2 instances mentioned above, there was no inhibition of glycogen breakdown in our animals.

The excessive gluconeogenesis in both phlorhizin and pancreatic diabetes, although caused by different mechanisms, nevertheless, leads to the same consequences, namely, continued glycosuria, low liver glycogen values, the accumulation of fat in the liver, ketosis and lowered R.Q. To this extent the two conditions are similar. The presence of insulin in the phlorhizinized animal, however, enables it to maintain the normal relationship between the height of the blood sugar level and the rate of entry of blood sugar into the tissues.[9, 18] This differs from the depancreatized animal, which requires a higher blood sugar level to allow a normal rate of entry.

Summary. At similar blood sugar levels, the phlorhizinized dog utilizes dextrose at the same rates as does the normal animal. The similarities and differences between phlorhizin diabetes and pancreatic diabetes are briefly discussed.

10235 P

Effect of Vitamin A Deficiency on the Rate of Apposition of Dentin.*

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Sixty albino rats placed on complete Vitamin-A-deficiency diet were given 2 intraperitoneal injections of 0.5 cc of a 2% solution of alizarine Red S (Table I). Alizarine is deposited as a red line in the dentin growing and calcifying at the time of each injection.[1] By measuring, in ground sections of incisors, the distance between the 2 injection effects and dividing this by the time interval daily rates of apposition were obtained.

The daily rate of normal dentin apposition averages 16 μ in both the labial and lingual portions of the tooth. In the experimental animals the rate of dentin apposition increased on the labial and

18 Soskin, S., and Levine, R., *Am. J. Physiol.*, 1938, **123**, 192.

* Aided by a grant from Mead Johnson and Co.

1 Schour, I., and Hoffman, M. M., PROC. SOC. EXP. BIOL. AND MED., 1938, **37**, 710.