

fection. The femoral vein is very small in rats. We were not able to locate it without cutting the skin. The jugular vein is by far the largest of the peripheral vessels of the rat. Blood may be drawn from it without much difficulty because of the ease in locating it by its pronounced pulsation. The jugular vein of the rat runs anteriorly *over* the clavicle about 10 mm. from the middle of the sternum.

If a rat is given a little ether, placed on its back on a board and tied so that the head is approximately one inch lower than the rest of the body, the hair clipped over the clavicle, it is easy to locate the jugular by the pulsation and to feel the clavicle. With the clavicle to support the vessel and the pulsation to direct us, it is not difficult to place the needle into the vessel. This same route has been used by us for injecting various substances and also for transfusing rats.

It has also been found possible to collect blood from rats according to the above technique without the use of an anesthetic.

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Studies on Calcium and Phosphorus in Bile-fistula Dogs.

J. D. GREAVES AND CARL L. A. SCHMIDT.

From the Division of Biochemistry, University of California Medical School, Berkeley.

The work of Pavlov,¹ Looser,² Wisner and Whipple,³ Düttman,⁴ Dietrich,⁵ Gilbert,⁶ and Buchbinder and Kern⁷ has shown that absence of bile from the intestinal tract of animals leads to osteoporosis. The gross manifestations of the condition are softening of the bones and spontaneous fracture. The condition is not brought about when only small amounts of bile are permitted to enter the intestinal tract. Results in accord with the above were obtained by Seidel⁸ in 2 human cases. Two explanations to account for this phenomenon have been advanced: (a) Klinke⁹ postulates that in the

¹ Pavlov, I. P., *Verein. Ges. russ. Aertate*, 1904, **72**, 314.

² Looser, E., *Verhandl. deut. Path. Ges.*, 1907, **11**, 291.

³ Wisner, F. P., and Whipple, G. H., *Am. J. Physiol.*, 1922, **60**, 119.

⁴ Düttmann, G., *Beitr. klin. Chir.*, 1923, **139**, 720.

⁵ Dietrich, H., *Beitr. klin. Chir.*, 1925, **134**, 530.

⁶ Gilbert, E., *Z. Ges. exp. Med.*, 1924, **43**, 539.

⁷ Buchbinder, W. C., and Kern, R., *Arch. Int. Med.*, 1927, **40**, 900; *Am. J. Physiol.*, 1927, **80**, 273.

⁸ Seidel, H., *Münch med. Wochschr.*, 1910, **57**, 2034.

⁹ Klinke, K., *Klin. Wochschr.*, 1928, **1**, 385; *Ergeb. d. Physiol.*, 1928, **26**, 279.

absence of bile there is lack of calcium absorption, (b) Seifert¹⁰ suggests that in the absence of bile, vitamin D is not absorbed. Support for the latter hypothesis is furnished by Tammann¹¹ who showed that parenteral administration of vitamin D alleviated the condition.

There has been no systematic work dealing with calcium and phosphorus balances in bile fistula dogs. King, Bigelow and Pearce¹² reported negative calcium balances over a period of a week and Emerson¹³ reported decreased fecal calcium output over several 3-day periods. However, from the standpoint of a calcium balance Emerson's results have little significance.

In the present work, calcium and phosphorus balances were carried out on 2 female dogs in which bile was excluded from the intestinal tract by cholecystonephrostomy. This operation was

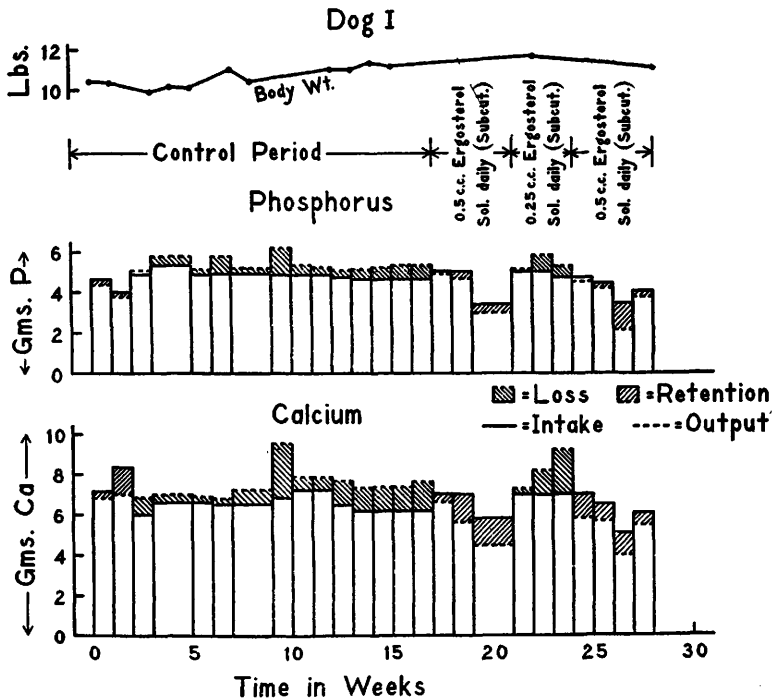


FIG. 1.

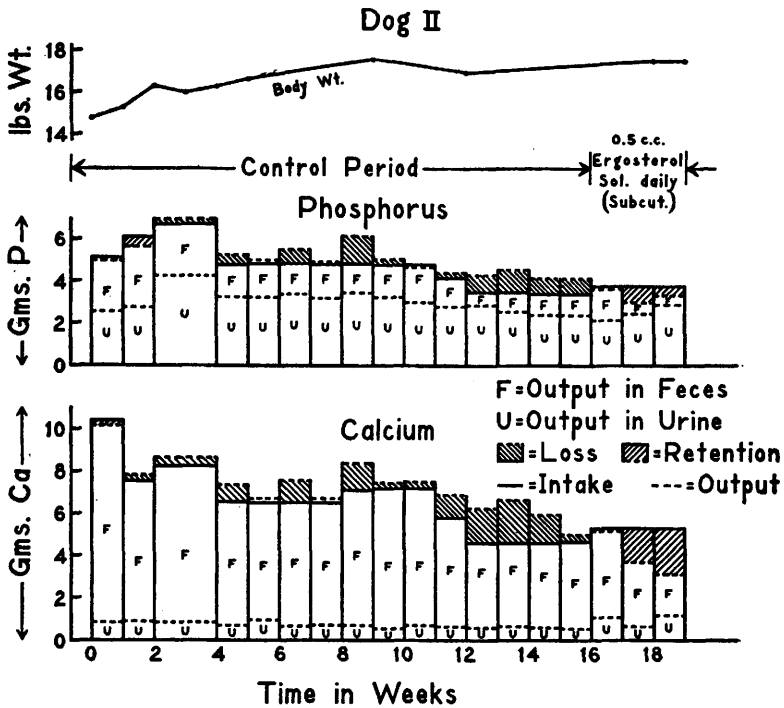
¹⁰ Seifert, E., *Beitr. klin. Chir.*, 1926, **136**, 496.

¹¹ Tammann, H., *Beitr. klin. Chir.*, 1928, **142**, 83.

¹² King, J. H., Bigelow, J. E., and Pearce, L., *J. Exp. Med.*, 1911, **14**, 159.

¹³ Emerson, W. C., *J. Lab. Clin. Med.*, 1928, **14**, 122.

kindly performed for us by Dr. C. M. Johnson according to the technique described by Kapsinow, Engle and Harvey.¹⁴ These animals were fed a miscellaneous diet, comparatively low in fat, for periods of 128 and 189 days respectively, after which they were placed on a diet consisting of casein 25 parts, cracker meal 48 parts, sucrose 5 parts, vitavose 18 parts, agar 2 parts, and salt mixture (calcium carbonate 1 part, sodium chloride 2 parts, magnesium carbonate 0.2 part) 2 parts. An adjustment period of one to 2 weeks was allowed before calcium and phosphorus balances were begun. For dog II, urine and feces were analyzed separately, while for dog I, composite analyses only were made.



The balance data are presented in Figs. 1 and 2. It will be noted that dog I was in negative calcium and phosphorus balance during a period of 17 weeks. On administration of 0.5 cc. Viosterol (Squibb 100 D) solution daily for a period of 4 weeks both the calcium and phosphorus balances became positive. When this dose was decreased to one-half of this amount, both the calcium and the

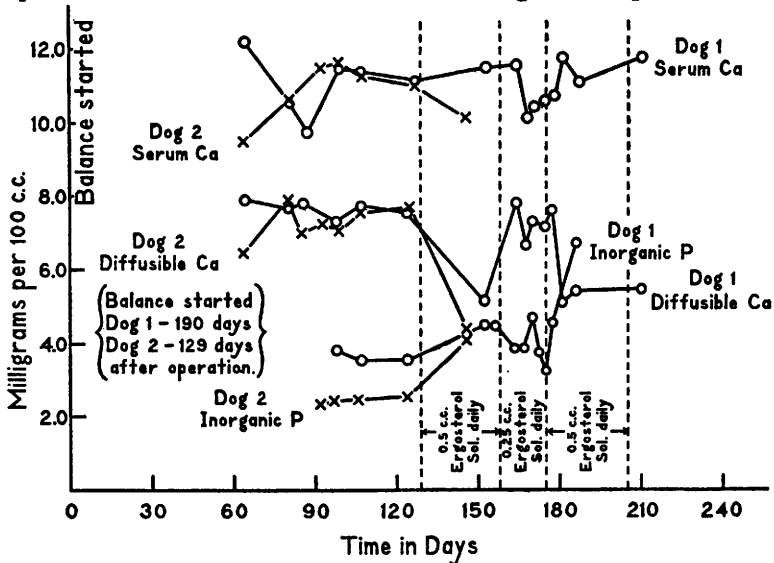
¹⁴ Kapsinow, R., Engle, L. P., and Harvey, S. C., *Surg. Gynec. and Obs.*, 1924, 39, 62.

phosphorus balances became negative. On increasing the dose of viosterol solution to 0.5 cc., both the calcium and the phosphorus balances again became positive. Similar results were obtained with dog II. Administration of 0.5 cc. solution of viosterol subcutaneously shifted a 16 weeks' negative calcium and phosphorus balance to a positive balance.

We are also reporting some data dealing with the distribution of blood calcium in these dogs. Sekitoo¹⁵ has reported low blood calcium in bile fistula rabbits. Emerson¹⁸ found a gradual rise in diffusible and blood calcium in his internal bile fistula dogs over an 84-day period. However, the low values for diffusible calcium which Emerson reports without apparently any indications of tetany cast considerable doubt on his results.

Our results are given graphically in Fig. 3. In both dogs during the periods of negative calcium balance the diffusible calcium was higher, while during the period of positive calcium balance, the values reached a lower and more nearly normal value. During the periods of negative phosphorus balance the serum phosphorus in both animals was low. This value was increased to a more nearly normal value when the animal was in positive phosphorus balance.

It was necessary to terminate the experiments for uncontrollable reasons. The results while incomplete nevertheless point strongly to a depletion of vitamin D in the bile fistula dogs as the possible cause



¹⁵ Sekitoo, T., *J. Biochem.* (Tokio), 1930, 11, 391.

for the osteoporosis. It is possible that in normal animals, deoxycholic acid acts as a carrier for the absorption of vitamin D across the intestines. If so, it follows that in the absence of bile, little or no absorption of vitamin D would take place.¹⁶

It is hoped to resume experimental work in this field at an early date.

¹⁶ Schmidt, W., and Schmidt, C. L. A., *Univ. Calif. Pub. Physiol.*, 1930, **17**, 211.