

between 2.2 and 3.5 μg per gram of fresh weight with an average value of 2.9 μg per gram. The muscle riboflavin values for 30 pellagrins lay within the same range with an average value of 2.8 μg per gram of fresh muscle.

3. From these results we have concluded that a determination of blood and muscle riboflavin values is of little significance in the evaluation of a state of riboflavin deficiency in man.

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Assay of Secretin.

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Recently a secretin preparation designed for human use was developed and tested clinically in normal individuals and in patients with pancreatic and biliary tract disease.^{1, 2, 3} Prepared by an unpublished method, this product is now manufactured and sold under the name *Pancreotest* by the Astra Laboratories, Sodertalje, Sweden. Through the courtesy of Messrs. Astra, a sample of Pancreotest was made available to us. This particular material had been produced recently (May 1, 1940) and was standardized by the manufacturer against the crystalline secretin of Hammarsten, *et al.*⁴ Having this standardized product we could evaluate its potency in relation to that of our own secretin preparations and investigate an apparent discrepancy in the expression of secretin units. Assays were conducted on 5 dogs by the technic previously described.⁵

Results. Pancreotest was found to be free of vasodilator substances when injected intravenously in amounts up to 10 mg. Weight for weight it was about half as potent as our standard SI preparation in stimulating the pancreas to secrete.⁵ The data on the 5 dogs are given in Table I.

Discussion. As is evident from the table, a given amount of Pan-

¹ Agren and Lagerlof, *Acta med. Skand.*, 1936, **90**, 1.

² Agren and Lagerlof, *Acta med. Skand.*, 1937, **92**, 359.

³ Lagerlof, *Quarterly J. Med.*, 1939, **8**, 115.

⁴ Hammarsten, Agren, Hammarsten, and Wilander, *Biochem. Z.*, 1933, **264**, 272.

⁵ Greengard and Ivy, *Am. J. Physiol.*, 1938, **124**, 427.

TABLE I.
Secretory Response to Weighed Amounts of SI and of Pancreotest.

Dog No.	SI injected (mg)	Pancreatic juice (drops)	Pancreotest injected (mg)	Pancreatic juice (drops)
1	1.0	17	1.0	10
	0.5	7	0.5	2
	0.5	8	1.0	7
2	0.5	36	0.5	20
	0.1	7	0.1	0
	0.4	15	0.8	17
3	0.5	19	1.0	20
	0.25	7	0.5	7
4	0.5	3	0.5	0
	1.0	8	1.0	2
	2.0	21	2.0	9
5	0.25	7	0.5	8
	0.5	16	1.0	17
	1.0	38	2.0	36

creotest is equivalent to half the same amount of SI in stimulating the pancreas to secrete. The corollaries to this finding are several: (a) The sample of Pancreotest was stated by the manufacturer to contain 2 "clinical units" per milligram which in turn is equivalent to 32 "cat units" as defined by Wilander and Agren.⁶ Our assays indicate the material to contain 2 threshold doses or "dog units" per milligram. Hence one "dog threshold dose" equals 16 cat units. (b) In a previous communication⁵ we reported that one "dog unit" is equal to 20 Wilander-Agren "cat units". Our Pancreotest assays show this ratio to be smaller, or about 1:16. The discrepancy is probably explained as follows: Our⁵ comparisons were made on the basis of cat's pancreatic juice obtained by direct cannulation. The Swedish investigators collected mixed secretions from the lumen of the duodenum. (c) The crystalline secretin of Hammarsten, *et al.*, has a reported potency of 253 cat units per milligram. On the basis of 16 cat units per "dog threshold dose", this is equivalent to 15 or 16 dog threshold doses per milligram, and the "dog threshold dose" of the Hammarsten preparation is therefore about 0.065 mg. This is approximately the same potency as our own crystalline secretin picrolonate, and these observations check well with our⁵ previously reported potency of 0.0036 mg (secretin picrolonate) according to our modification of the Wilander-Agren cat assay method. It should be emphasized again that our crystalline secretin picrolonate contains

⁶ Wilander and Agren, *Biochem. Z.*, 1932, **250**, 489.

about 80% picrolonic acid, whereas this constituent comprises only a small portion of the Hammarsten preparation. The "dog threshold dose" for our secretin base is 14 gamma.⁵ (d) The preparation, Pancreotest, is about half as potent as our standard SI material, which we have considered to be too imperfect for use in humans. The secretin used by us⁷ on human subjects is 10 times as potent as our SI and 20 times as potent as Pancreotest.

Conclusions. The commercial secretin preparation, Pancreotest, has been assayed on dogs and found to be half as potent as our standard material. An apparent discrepancy in the expression of units of secretin has been explained.

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Electrolytic Splitting of Liver Albumin.

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The albumin used in these studies was prepared from dog liver perfused *in situ* until free from blood. In all cases the animals had been well fed. The organ was excised, frozen in liquid air, powdered, and stored at -13°C . Preparation of the liver albumin followed upon extraction of the powdered liver with 0.5 *M* $(\text{NH}_4)_2\text{SO}_4$, and salting out of the globulins in 2.1 *M* $(\text{NH}_4)_2\text{SO}_4$. The albumin was salted out by increasing the concentration of ammonium sulphate to 3.5 *M* and was rendered free from any detectable amount of globulin by reprecipitation with ammonium sulphate. After 3 or 4 reprecipitations, the amount of material salted out in 2.1 *M* $(\text{NH}_4)_2\text{SO}_4$ was reduced to the vanishing point. The pH was maintained at 6.5 to 7.0 throughout this treatment. The amount of albumin obtained from 100 g of liver was 0.9 to 1.5 g.

Electrophoretic studies were carried out in the Tiselius apparatus, using for observation of the boundaries the Toepler "Schlieren" method as modified by Longworth. In a medium consisting of 0.1 *M* NaCl and 0.02 *M* $\text{KH}_2\text{PO}_4/\text{K}_2\text{HPO}_4$, following upon dialysis against a fluid of the same composition, electrophoretic analysis (pH 5.8 to 7.3) revealed 2 fractions; both moved towards the positive electrode. One fraction was considerably larger than the other and

⁷ Voegtlin, Greengard and Ivy, *Am. J. Physiol.*, 1934, **110**, 198.