11018 P

Effect of Insulin on Rate of Appearance of Free Sugar in Liver Brei.

M. TAUBENHAUS, R. LEVINE AND S. SOSKIN.

From the Department of Metabolism and Endocrinology, Michael Reese Hospital, and the Department of Physiology, University of Chicago.

Jensen[1] and Soskin[2] have recently summarized the literature regarding the action of insulin on the liver. The indirect evidence available favors the view that insulin inhibits glycogenolysis. More direct evidence has been reported by Issekutz and Szende,[3] who demonstrated that perfused livers removed from frogs which had previously received insulin, produced less sugar than did the livers of untreated frogs. Similar, but less well controlled results, were obtained by Siegel,[4] Molitor and Pollak,[5] and Popper[6] by different methods. On the other hand, Lundsgaard, *et al.*,[7, 8] were unable to show that insulin had any action on glycogen breakdown or deposition in the perfused livers of cats and dogs.

In a preceding communication we have shown a quantitative inhibition exerted by added glucose on the rate of appearance of free sugar in glycogenolyzing liver brei. This offered the opportunity for the testing of insulin action on the liver.

Normal, fed dogs were anesthetized with nembutal and a lobe of liver was removed after tying a broad linen band around its base. There was no bleeding and no detectable symptoms of shock. Insulin (1 unit per kg body weight) was then administered subcutaneously or intravenously as desired. Thirty to 45 minutes after the insulin administration, the remainder of the liver was removed. Total carbohydrate and free sugar were determined at varying intervals under constant conditions, as detailed in the preceding paper.[9]

Results and Discussion. The possible influence of the operative

1 Jensen, H., *Insulin*, Commonwealth Fund, 1938, New York.

2 Soskin, S., *Physiol. Rev.*, Jan., 1940, in press.

3 Issekutz, B., and Szende, J., *Biochem. Z.*, 1934, **272**, 412.

4 Siegel, R., *Klin. Wchnschr.*, 1929, **8**, 1069.

5 Molitor, H., and Pollak, L., *Arch. f. exp. Path. u. Pharmakol.*, 1930, **154**, 280.

6 Popper, H., and Wozasek, O., *Z. f. d. ges. exp. Med.*, 1931, **77**, 414.

7 Lundsgaard, E., *Johns Hopkins Hosp. Bull.*, 1938, **63**.

8 Lundsgaard, E., Nielsen, N. E., and Ørskov, S. L., *Skand. Arch. Physiol.*, 1936, **73**, 296.

9 Soskin, S., Levine, R., and Taubenhaus, M., PROC. SOC. EXP. BIOL. AND MED., 1939, **42**, 689.

TABLE I.
Inhibition of Glycogenolysis in Liver Brei by Dextrose, and by Dextrose + Insulin.

	Time, min	Total* Cho.	Amt* of dextrose added	Appearance of free sugar*		% inhibition	
				without insulin	with insulin	with dextrose alone	with dextrose + insulin
I.	0	2778	0	141	—	—	—
	60	—	0	299	—	—	—
	60	—	94	319	165	0	45
	60	2897	186	267	145	11	52
II.	0	3313	0	254	—	—	—
	60	—	0	1997	—	—	—
	60	—	100	2027	1180	0	42
	60	—	208	1565	1073	22	46
	60	—	418	1229	805	39	60
	60	3410	836	1260	662	37	67
III.	0	4184	0	105	217	—	—
	60	—	0	1317	1269	—	—
	60	—	100	1196	1066	9	19
	60	—	200	1080	892	18	32
	60	—	450	1019	690	23	48
	60	4000	900	446	222	66	83

* All values are mg per 100 g of liver, calculated as for glucose.

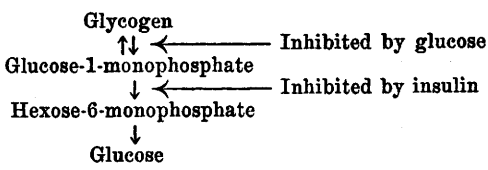
procedure on our results was ascertained by running control experiments in which no insulin was given. There was no difference between the rates of glycogenolysis in the first and second liver samples.

The data summarized in Table I show that in the liver samples removed after insulin administration, there was a significantly lower rate of appearance of free sugar than in the samples removed before insulin was given. When glucose was added *in vitro* to both sets of liver samples, the rate of glycogenolysis was inhibited to a greater extent and by smaller amounts of added glucose in the "insulinized" samples than in the controls.

It was also attempted to reproduce the above effect by adding the insulin *in vitro,* but without success.

The recent work of Gill and Lehmann[10] when related to what is known of the course of glycogen breakdown in the liver, suggests a tentative explanation of the mechanism of the above inhibition. They have shown that insulin inhibits the *in vitro* transformation of the glucose—1-ester to the 6-ester in muscle extract. If this action can be demonstrated to occur in liver, the inhibiting influence of both glucose and insulin could be indicated as follows:

10 Gill, P. M., and Lehmann, H., *Biochem. J.*, 1939, **33**, 1151.



Summary. Insulin inhibits glycogenolysis in the liver, and reinforces the inhibitory effect of added dextrose.

11019

Influence of Vitamin A upon Urea Clearance in the Rat.*

R. C. HERRIN. (Introduced by W. J. Meek.)

From the Department of Physiology, University of Wisconsin Medical School.

The study of the effect of vitamin A upon renal excretion was extended to the rat because the diet could be made of simpler materials and avitaminosis A of a more severe degree could be studied economically.

Procedure. A high protein diet was used in order to make the physiological strain on the kidneys greater. It was composed of casein 56, lard 20, starch 16, yeast 5 and Wesson's salt mixture 3. For the first 100 days of the experiment the casein was extracted with ether but as this left appreciable amounts of vitamin A the casein thereafter was dry heated for 2 weeks in the autoclave by passing steam at 15 pounds pressure through the outer jacket of the autoclave. This prolonged heating has been used for many years to free casein of vitamin A. The rats were exposed to light from a quartz Hg vapor lamp to furnish vitamin D. The rats weighed 30 to 40 g when placed on the experimental diet.

The technic of Farr and Smadel[1] with modifications was used in making the urea clearance determinations. The rats were handled for training before being used for clearance study. They were placed in the urine collection cages without a preliminary withdrawal of food. After a urine collection period of 350 to over 600 minutes, blood was drawn from the tail, then the urine was collected for another period of approximately the same length. The same blood

* This study received financial support by grants from the Committee on Scientific Research of the American Medical Association and the Wisconsin Alumni Research Foundation.

1 Farr, Lee E., and Smadel, Joseph E., *Am. J. Physiol.*, 1936, **116**, 349.