28 days. A portion of the large intestinal wall, freed of feces, and a portion of liver were analyzed for emetine. The analysis was adapted from the general method of Brodie *et al.*[2]

Table I shows the emetine concentration in the liver and intestine. One hour after injection, the concentration of drug was much higher in the liver than in the intestine. At 12 hours, the liver concentration of drug was 41.6 mg per kg compared to the intestinal concentration of only 1.7 mg per kg. At the end of 4 days the low concentration in the intestine became almost undetectable whereas that of the liver was maintained for more than 4 weeks.

Summary. Injection of emetine into rabbits was followed by a relatively high drug concentration in the liver for more than a month, whereas the level in the intestine was always much lower and became undetectable after 4 days. The findings of a high concentration and prolonged presence of emetine in the liver and a low concentration and transient presence of the drug in the intestine are proposed as the explanation for the efficacy of emetine in amebic liver disease and the drug's failure in amebic intestinal disease in man.

[2] Brodie, B. B., Undenfriend, S., and Dill, W., *J. Biol. Chem.*, 1947, **168**, 335.

16488

Phospholipid Utilization in the Diabetic Dog.*

PAUL V. HARPER, JR., WILLIAM B. NEAL, JR., AND GEORGIANA R. ROGERS.
(Introduced by Lester R. Dragstedt.)
From the Department of Surgery, University of Chicago.

Depancreatized dogs maintained on insulin developed fatty livers, emaciation, and anemia.[1] Dragstedt maintains that this syndrome is due to the lack of a specific substance present in pancreas, which he has named lipocaic.[2] The syndrome is relieved or prevented by feeding choline, raw pancreas, and various choline-free pancreatic extracts.

It has been shown that the plasma phospholipid level is lowered in lipocaic deficiency.[3] Phospholipid is probably the chemically active form of fat and the significant lipid fraction in the transport of fat. There is evidence that choline deficiency in rats, which resembles lipocaic deficiency, results in an impairment of transportation of fat from the liver to the tissues.[4] In the present study, the plasma phospholipid turnover was measured in normal dogs and in diabetic dogs under a variety of conditions.

Numerous studies on phospholipid metabolism have been performed with the use of isotopic tracers. One conclusion from this work is that the plasma phospholipid is produced largely in the liver.[5]

In the present study, it is assumed that the plasma phospholipid is produced in the liver and withdrawn from the plasma for utilization by the tissues. Since the phospholipid levels remained constant during the period of measurement, the rate of production of new phos-

* This work has been aided by grants from the Rockefeller Foundation—Postwar Training of Medical Veterans Fund, Mr. Andrew E. Wigeland, and the Kenneth G. Smith and Douglas Smith Foundations for Medical Research of The University of Chicago.

[1] Allan, F. N., Bowie, J. J., MacLeod, J., Jr., and Robinson, W. L., *Brit. J. Exp. Path.*, 1924, **5**, 75.

[2] Dragstedt, L. R., Prohaska, J. V., and Harms, H. P., *Am. J. Physiol.*, 1936, **117**, 175.

[3] Chaikoff, I. L., and Kaplan, A., *J. Biol. Chem.*, 1934, **106**, 267.

[4] Stetten, D., Jr., and Salcedo, J., *J. Biol. Chem.*, 1944, **156**, 27.

[5] Fishler, M. C., Entenman, C., Montgomery, M. L., and Chaikoff, I. L., *J. Biol. Chem.*, 1943, **150**, 47.

TABLE I.
Fasting Normal Dogs.

Dog No.	Total plasma lipid (mg%)	Plasma lipid phosphorus (mg%)	Half-life of injected phospholipid (hr)	Utilization (mg% lipid phosphorus disappearing per hr)	Fraction of plasma phospholipid utilized per hr	Remarks
234	479	11.8	5.6	1.46	.124	
769	900	17.8	5.5	2.20	.126	
S-892	1020	21.1	6.8	2.16	.101	
965	450	8.4	5.0	1.17	.139	
913	—	17.5	7.2	1.67	.096	
913	—	12.2	7.0	1.21	.099	
913	—	17.1	5.7	2.08	.122	Fed
944	—	11.0	7.6	1.22	.091	
966	427	7.8	5.4	1.54	.128	Fed
1B	—	12.2	6.5	1.31	.107	Zilversmit's dogs[7]
2B	—	11.6	5.0	1.61	.139	
3B	—	17.9	5.4	2.29	.128	
Mean		13.8	6.0	1.66	.114	
Standard error of mean		±1.2	±.26	±.12	±.005	

pholipid equaled the rate of disappearance or utilization.

Methods. The rate of disappearance was measured by the method of Zilversmit *et al.*[6,7] with little modification. Donor dogs were injected subcutaneously with 2 to 3 millicuries of radioactive phosphorus as sodium phosphate, and 24 hours later the dogs were heparinized and bled and the plasma was separated. From 64 to 81% of the plasma radioactive phosphorus was in the phospholipid fraction and the remainder was inorganic phosphorus. The plasma containing the tagged phospholipid was injected intravenously into 7 normal and 12 diabetic dogs in the postabsorptive state. After allowing 20 to 30 minutes for complete mixing, blood samples were drawn at intervals of 1 to 2 hours, and the radioactivity and phosphorus content were determined on the trichloracetic acid precipitate of the plasma. All samples had activities of 3 to 20 times background activity and were counted for appropriate periods. Phosphorus was determined by the method of Fiske and Subbarow.[8] Plasma lipids were determined by the method previously reported from this laboratory.[9] All dogs were fasted before the determination for 18 to 20 hours. Half the daily insulin requirement plus 25 g of glucose in skimmed milk was given to all but the normal and control groups half an hour before the injection of the phospholipid. Two hundred g of raw pancreas a day for periods up to several weeks were administered to one group. All diabetic dogs were controlled with insulin on a regime previously reported from this laboratory.[9]

The fact that all the injected radioactive phosphorus was not in the form of phospholipid did not alter the findings measurably. The formation of new plasma phospholipid by the liver from injected inorganic radioactive phosphorus does not, in our experience, reach levels comparable to those observed in the present study (5 to 13 counts per second per 3 cc sample) until as much as 20 microcuries/kg are given, and even then not until 12 to 24 hours have elapsed. Since all the present experiments were terminated within 6 hours, and since the inorganic radioactive phosphorus injected in the plasma amounted to only .02 to .04 microcuries/kg it is evident that this factor may be neglected. The inorganic radioactive phosphorus in the plasma

[6] Zilversmit, D. B., Entenman, C., and Fishler, M. C., *J. Gen. Physiol.*, 1943, **26**, 325.

[7] Zilversmit, D. B., Entenman, C., Fishler, M. C., and Chaikoff, I. L., *J. Gen. Physiol.*, 1943, **26**, 333.

[8] Fiske, C. H., and Subbarow, Y., *J. Biol. Chem.*, 1925, **66**, 375.

[9] Dragstedt, L. R., Donovan, P. B., Clark, D. E., Goodpasture, W. C., and Vermeulen, C. W., *Am. J. Physiol.*, 1939, **127**, 755.

did not interfere with the determination of the radioactivity of the plasma phospholipid phosphorus as it was washed out with trichloracetic acid.

In all cases the disappearance of the injected phospholipid from the blood stream was found to follow a logarithmic curve. The time required for half the tagged phospholipid to disappear, or the half-life, was measured graphically from the plot of the logarithm of the radioactivity of the trichloracetic acid precipitate of the plasma against time. From this the utilization was calculated from the relation:

$$\text{Utilization} = \frac{\text{plasma lipid phosphorus(mg\%)}}{\text{half-life (hr)} \times 1.44} =$$

mg% lipid phosphorus disappearing per hour from the blood stream.

The fraction of the total plasma phospholipid disappearing per hour equals utilization / plasma lipid phosphorus (mg%) or

$$\frac{1}{\text{half-life (hr)} \times 1.44}$$

The standard error of the mean for the various groups was calculated from the formula for small samples:

$$S_x = \frac{\sigma}{\sqrt{N-1}}$$

Results. The results for normal dogs are summarized in Table I. The striking finding in this group was the relative constancy of the fraction of plasma phospholipid utilized per hour in spite of wide variations in the plasma phospholipid concentration among the various dogs. This suggests that utilization of phospholipids is roughly proportional to its concentration. Tables II and III show the results for diabetic dogs fasting and given insulin. It is seen that no essential difference exists between the two groups. Comparing these two groups to the normal group, the usual changes of lipocaic deficiency are present in varying degrees. On the average, there is a decreased plasma lipid and plasma lipid phosphorus. The injected phospholipid disappears at a significantly greater rate in the diabetic groups, the half-life averaging 4.7 and 4.8 hours against 6.0 hours for the normal dogs. However, because of the lower phospholipid level in the plasma of the diabetic dogs, the

TABLE II.
Fasting Diabetic Dogs.

Dog No.	Type of preparation	Daily insulin requirement (units)	Total plasma lipid (mg%)	Plasma lipid phosphorus (mg%)	Half-life of injected phospholipid (hr)	Utilization (mg lipid phosphorus disappearing per hr)	Fraction of plasma phospholipid utilized per hr	Remarks
930	Total pancreatectomy	10	500	8.8	3.7	1.66	.188	10 wks postop. No suppl. 5 wks
934	" "	34	570	11.5	4.8	1.66	.145	3 wks postop.
801	Partial pancreatectomy with ligation of duct	32	450	11.2	5.0	1.56	.139	136 wks postop. No supp. 9 wks
892	Same	4	290	7.9	5.4	1.02	.129	38 wks postop. No suppl. 20 wks
922	Pancreatic duct ligation	4	213	8.1	5.0	1.12	.139	19 wks postop.
	Mean	17	406	9.7	4.8	1.40	.148	
	Standard error of mean		±66	±.2	±.3	±.14	±.010	

TABLE III.
Diabetic Dogs Given Insulin and Sugar.

Dog No.	Type of preparation	Daily insulin requirement (units)	Total plasma lipid (mg%)	Plasma lipid phosphorus (mg%)	Half life of injected phospholipid (hr)	Utilization (mg% lipid phosphorus disappearing per hr)	Fraction of plasma phospholipid utilized per hr	Remarks
919	Total pancreatectomy	64	770	9.1	3.6	1.74	.192	30 wks postop. No suppl. 1 wk
933	" "	38	467	9.0	4.9	1.27	.141	10 wks postop. No suppl.
933	" "	28	325	5.4	6.0	.63	.116	14 wks postop. No suppl.
934	" "	24	490	11.5	4.7	1.70	.147	8 wks postop. No suppl.
890	" "	68	650	11.4	5.9	1.33	.118	40 wks postop. No suppl. 18 wks
489	Partial pancreatectomy with ligation of duct	44	830	15.3	5.2	2.04	.133	102 wks postop. No suppl. 15 wks
892	Same	4	240	4.7	3.5	.93	.198	44 wks postop. No suppl. 26 wks
895	Same	4	310	12.4	4.0	2.16	.173	40 wks postop. No suppl. 5 wks
923	Pancreatic duct ligation	4	455	11.4	4.4	1.81	.158	25 wks postop. No suppl.
	Mean	35	505	10.0	4.7	1.50	.153	
	Standard error of mean		±66	±1.1	±.3	±.16	±.011	

absolute utilization of plasma phospholipid is slightly less in the diabetic groups. The fraction of plasma phospholipid utilized per hour, however, is increased in the diabetic dog, indicating that for a given plasma level, phospholipid is utilized more rapidly in the diabetic dog, regardless of whether he is getting insulin; and conversely, for a given rate of utilization, the diabetic dog has a lower plasma lipid phosphorus. Again, as in the normal dog, there is a relatively wide fluctuation in the absolute utilization which parallels the phosphorus level, while the fraction of plasma phospholipid utilized per hour is relatively constant.

Upon administration of raw pancreas to the diabetic dogs, the plasma lipid and lipid phosphorus level rose to normal, as shown in Table IV, but the utilization of phospholipid rose well above the normal level, and the fraction utilized per hour was higher in this group. The half-life of the injected phospholipid was shorter.

Table V shows the results in one dog with a subtotal pancreatectomy with the remnant of pancreas remaining attached to a duct. In this preparation, the very high utilization of phospholipid parallels the high lipid phosphorus level. However, the fraction utilized per hour is in the same range as the other diabetic dogs. The effect of insulin and sugar on the fraction of the phospholipid utilized per hour is negligible.

Preliminary results on 3 diabetic dogs given 2 g of choline chloride a day for 3 days showed effects similar to the group fed raw pancreas. Plasma lipids averaged 451 mg%; plasma lipid phosphorus averaged 9.2 mg%; half-life of injected phospholipid averaged 4.0 hours; utilization averaged 1.70 mg% per hour; and the fraction of phospholipid utilized per hour averaged .177, the highest of all groups. No significant effect was observed in two fasting normal dogs given similar amounts of choline.

It is apparent from the data that in both the normal and the diabetic dog the rate of utilization of phospholipid is proportional to the concentration of phospholipid in the plasma. Two dogs on which determinations were made under a variety of conditions illus-

TABLE IV.
Diabetic Dogs Given 200 Grams per Day Raw Pancreas, Insulin, and Sugar.

Dog No.	Type of preparation	Daily insulin requirement (units)	Total plasma lipid (mg%)	Plasma lipid phosphorus (mg%)	Half-life of injected phospholipid (hr)	Utilization (mg% lipid phosphorus disappearing per hr)	Fraction of plasma phospholipid utilized per hr	Time on raw pancreas (wks)	Remarks
933	Total Pancreatectomy	62	960	11.3	3.6	2.17	.192	1	15 wks postop.
933	,,	92	1300	16.6	3.3	3.50	.210	5	19 ,, ,,
919	,, ,,	58	930	16.7	5.0	2.32	.139	2	35 ,, ,,
919	,, ,,	96	1100	12.8	4.4	2.02	.158	3	28 ,, ,,
934	,, ,,	42	1300	18.7	4.6	2.82	.151	5	15 ,, ,,
801	Partial pancreatectomy with ligation of duct	40	1470	18.4	5.7	2.24	.122	5	145 ,, ,,
892	Same	12	480	8.8	3.7	1.65	.187	2	46 ,, ,,
892	Same	10	747	13.8	3.8	2.52	.182	5	50 ,, ,,
	Mean	65	1030	14.7	4.2	2.42	.168		
	Standard error of mean		±84	±.4	±.3	±.22	±.011		

TABLE V.
Subtotal Pancreatectomy with Remnant on Duct—No Supplement.

Daily insulin requirement (units)	Total plasma lipid (mg%)	Plasma lipid phosphorus (mg%)	Half-life of injected phospholipid (hr)	Utilization (mg% lipid phosphorus disappearing per hr)	Fraction of plasma phospholipid utilized per hr	Remarks
50	1450	23.7	5.0	3.30	.139	Insulin and sugar. 3 wks postop.
94	1850	30.1	4.8	4.35	.145	Fasted 6 wks postop.

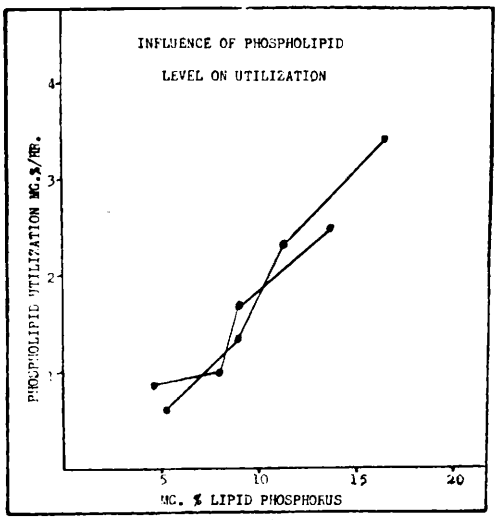

FIG. 1.

trate this point particularly well (Fig. 1). This confirms the conclusion arrived at by Balfour[10] from less direct evidence, and is entirely consistent with the observations of Flock and Bollman[11] on chickens. This concept permits a broader interpretation of the significance of the plasma lipid phosphorus level.

It is also apparent that the utilization of plasma phospholipid is altered after production of diabetes by pancreatectomy in such a manner that phospholipid disappears from the plasma more rapidly in the diabetic dog than in the normal; that is to say, the fraction of the circulating plasma phospholipid utilized per hour is significantly increased in the diabetic animals. This effect seems to be unchanged whether the animal is fasted or given insulin and sugar and may be slightly increased when the animal is fed raw pancreas. The significance of this alteration in the phospholipid metabolism is not clear, especially as it seems to be aggravated instead of corrected by feeding raw pancreas and by choline.

Summary and conclusions. Phospholipid tagged with radioactive phosphorus was injected intravenously into normal and diabetic dogs. From the rate of disappearance of the

[10] Balfour, W. M., *Gastroenterology*, 1947, **9**, 686.

[11] Flock, E. V., and Bollman, J. L., *J. Biol. Chem.*, 1944, **156**, 151.

tagged phospholipid from the blood stream and the plasma lipid phosphorus level, the rate of utilization of phospholipid was calculated. It was observed in both normal and diabetic groups that the rate of utilization was roughly proportional to the plasma lipid phosphorus concentration. The absolute utilization of plasma phospholipid was slightly lower in the diabetic group, but the proportion of plasma phospholipid utilized per hour was significantly higher in this group. Administration of insulin and sugar to diabetic dogs caused no significant change. Administration of raw pancreas to diabetic dogs increased the absolute utilization very markedly and the proportion utilized per hour only slightly.

16489

Effect of Celiac Ganglionectomy upon Experimental Peptic Ulcer Formation.*

C. W. LILLEHEI† AND OWEN H. WANGENSTEEN.

From the Department of Surgery, University of Minnesota Hospitals, Minneapolis, Minn.

In a recent report,[1] we have investigated the effect on the gastrointestinal tract of complete excision of the celiac, superior, and inferior mesenteric ganglia. Extirpation of these ganglia removes essentially the postganglionic sympathetic fibers to the gastrointestinal tract from the stomach through the colon and results in a severe bloody diarrhea associated with typical appearing peptic ulcers in somewhat less than 50% of the dogs. These ulcers are located for the most part in the stomach or duodenum, but occasionally as far down as the terminal ileum or colon. Likewise, these ulcers often progress to hemorrhage or perforation. The question has been raised by ourselves as well as others as to whether these ulcers are an effect of the removal of the postganglionic sympathetic innervation or whether they develop as the consequence of a state of protein deficiency and emaciation which several investigators[2,3] have shown to be often associated with peptic ulcer in dogs.

This investigation was undertaken to appraise what specific effects, if any, complete excision of the postganglionic sympathetic innervation to the stomach and duodenum has upon susceptibility to gastroduodenal ulceration.

Experiment. Male and female dogs of medium size, previously dewormed and between the ages of 1½ to 5 years were used in these experiments. The animals were divided into 3 series as follows: Series I, dogs in which complete excision of the celiac ganglia was performed leaving undisturbed the vagi nerves and the superior and inferior mesenteric ganglia. This operation removes the postganglionic sympathetic innervation of the stomach, duodenum, and also that of the liver, spleen, and pancreas as well. Series II, consists of dogs in which in addition to complete excision of the celiac ganglia, the superior mesenteric ganglia were removed also. This operation removes the postganglionic sympathetic fibers to the stomach, duodenum, and small intestine. Series III consists of a control series of normal dogs of comparable age and size.

Operative procedure. For ganglionectomy intravenous nembutal anesthesia was used, sterile technic, and a midline abdominal in-

* Supported by a grant from the Surgical Study Section of the United States Public Health Service, the John and Mary R. Markle Foundation, and the Graduate School of the University of Minnesota.

† National Cancer Trainee.

1 Lillehei, C. W., Dixon, J. L., Friesen, S. R., and Wangensteen, O. H., PROC. SOC. EXP. BIOL. AND MED., 1948, **68**.

2 Weech, A. A., and Paige, B. H., *Am. J. Path.*, 1937, **13**, 249.

3 Keller, A. D., *Arch. Path.*, 1936, **21**, 127.