in vit A was localized in parenchymal cells after the chyle injections as compared with the reticulo-endothelial cell localization following injection of the artificial emulsions, may have been a function of the smaller particle size of chyle, or perhaps it was due to the relative ease with which the chyle chylomicrons enter into the heparin clearing reaction(17).

Summary. Intravenous injection of vitamin A in the form of rat chyle into vit A-deficient assay rats produced a growth response twice that produced by the same dose given orally. The characteristic vit A autofluorescence was absent from the parenchymal cells of liver and adrenal cortex of assay rats depleted of vit A; the autofluorescence was restored to the distribution in these tissues characteristic of the normally fed rat only after intravenous injection of rat chyle which contained vit A. In contrast, intravenous injection of artificial emulsions which contained vit A resulted in a marked localization of vit A fluorescence in the reticulo-endothelial cells of spleen, lungs and liver and to a smaller extent in the liver and adrenal parenchymal cells. Most of the vit A content of rat plasma has been reported to be in the soluble fraction; however, we found most of the vit A content of post-absorptive rat chyle in the chylomicron fraction.

1. Moore, T., Vitamin A, Elsevier Publishing Co., Amsterdam, London, New York, and Princeton, 1957, p154.

2. Symposium on Intravenous Fat Emulsion, Metabolism, 1957, v6, 591.

3. Sherman, H. C., Todhunter, E. N., J. Nutrition, 1934, v8, 347.

4. Popper, H., Greenberg, R., Arch. Path., 1941, v31, 766.

5. Kimble, M. S., J. Lab. and Clin. Med., 1938-39, v24, 1055.

6. Bollman, J. L., Cain, J. C., Grindlay, J. H., ibid., 1948, v33, 1349.

7. Lindgren, F. T., Elliot, H. A., Gofman, J. W., J. Phys. and Colloid Chem., 1951, v55, 80.

8. Brown, W. D., Science, 1953, v118, 46.

9. Bateson, H. C., An Introduction to Statistics in the Medical Sciences, Burgess Publishing Co., Minneapolis, Minn., 1956.

10. Lease, J. G., Lease, E. J., Steenbock, H., Bauman, C. A., J. Lab. and Clin. Med., 1941-42, v27, 502.

11. Day, A. J., French, J. E., Quart. J. Exp. Physiol., 1959, v44, 239.

12. Byers, S. O., Mist-St. George, S., Friedman, M., Physiopathology of the Reticulo-Endothelial System, C. C Thomas, Springfield, Ill., 1957, p128.

13. Ganguly, J., Krinsky, N. I., Mehl, J. W., Deuel, H. J., Jr., Arch. Biochem. and Biophys., 1952, v38, 275.

14. Krinsky, H. I., Cornwell, D. G., Oncley, J. L., ibid., 1958, v73, 233.

15. Robinson, D. S., French, J. E., Quart. J. Exp. Physiol., 1957, v42, 1951.

16. Murray, R. G., Freeman, S., J. Lab. and Clin. Med., 1951, v38, 56.

17. Korn, E. D., J. Biol. Chem., 1955, v215, 15.

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Effect of Exclusion of Pancreatic Juice on Digestion and Mucosal Absorption of Fat in Dogs.* (29786)

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The well-established fact that pancreatic insufficiency results in defective lipid absorption has been based largely on the recovery and analysis of fecal lipids of animals lacking the external secretion of the pancreas(1-3). However, since the small intestine is probably the primary region in which such defects are manifested, it was felt that it would be fruitful to investigate in detail the intraluminar and mucosal lipid changes which occur in

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TABLE I. Weights of Lipid Recovered from Gastrointestinal Contents of Normal Dogs and Dogs Deprived of Pancreatic Juice.

		Fed (g)	Stomach (g)	Duodenum-Jejunum (mg)	Ileum (mg)
			A. Fat-free		
Normal	(15)*	0		91 ± 21†	120 ± 28
Depancreatized	(9)	0		109 ± 22	233 ± 59‡
			B. Cottonseed oil		
Normal	(7)	25	10.0 ± 1.1	1083 ± 270	490 ± 120
Depancreatized	(8)	25	8.0 ± .9	2576 ± 881	3080 ± 529§

* No. of samples given in parentheses.
† Mean ± S.E. of mean.
‡ $P < .05$.
§ $P < .01$.

the small intestine during deprivation of pancreatic juice. Thus, by characterizing the lipid of intestinal contents and mucosa following ingestion of a common dietary triglyceride, it was thought that it might be possible by comparing the amount and composition of these two fractions with those of normal dogs to make inferences regarding specific defects in the lipid digestion and absorption mechanisms which result from pancreatic insufficiency.

Methods. Normal dogs and dogs deprived of pancreatic juice were used in these studies. The data obtained on the normal dogs have been previously presented(4). Deprivation of pancreatic juice was achieved by removing the body of the pancreas leaving the uncinate process intact(5). The partially depancreatized dogs were allowed to recover from the surgical procedure for 7-10 days prior to use in an experiment. Subsequent necropsy revealed no connection between the duodenum and the remainder of the pancreas.

A fat-containing meal was prepared by mixing 25 g cottonseed oil triglycerides with 25 g casein, 25 g sucrose, and 5 g cellulose flour. This test meal was fed to 7 normal and 8 depancreatized dogs. In addition, a fat-free test meal consisting of 25 g casein, 25 g sucrose, and 5 g cellulose flour mixed with a little water was fed to 15 normal and 9 depancreatized dogs. This meal was fed in order to obtain an estimate of the amount and composition of the endogenous lipid which is present in the total lipid recovered from intestinal contents and mucosa after feeding fat.

The dogs were fed a test meal after a 24-hour fast and killed 4 hours later. The stomach and small intestine were removed separately and the small intestine divided into 2 segments. The proximal 40% of the small intestine was considered to represent duodenum-jejunum and the remainder, ileum. Collection of gastrointestinal contents and intestinal mucosa and the isolation and analyses of lipid recovered from these fractions have been described(4). Since only small amounts of intraluminar lipid were recovered after feeding the fat-free meal, pooling of these lipid samples from 3 dogs was necessary for the analyses.

The intraluminar and mucosal lipids were characterized by a number of determinations. Phospholipid was determined either by fractionation by silicic acid column chromatography(6) or by the method of Fiske and Subarrow(7). Free and esterified cholesterol were determined by the method of Sperry and Webb(8) and monoglycerides by the method of Pohle and Mehlenbacher(9). Free fatty acids were assayed under nitrogen by titration in 95% ethanol with alcoholic 0.02 N KOH to the phenolphthalein endpoint. That part of the total lipid not accounted for by assay was considered to consist of the combined fraction of diglycerides and triglycerides.

Results. The weights of lipid recovered from the contents of the stomach and small intestine of the dogs fed the test meals are given in Table I. Comparisons between normal and depancreatized dogs revealed that the amount of endogenous lipid recovered from the ileum of the depancreatized animals fed the fat-free meal was significantly greater than normal. This result can be compared with that of Pessoa *et al*(1), who reported

TABLE II. Composition* of Intestinal Intraluminar Lipids of Normal Dogs and Dogs Deprived of Pancreatic Juice.

Lipid component	Duodenum-jejunum				Ileum			
	Normal		Depancreatized		Normal		Depancreatized	
	(%)	(mg)	(%)	(mg)	(%)	(mg)	(%)	(mg)
A. Fat-free								
	(5)†		(3)		(5)		(3)	
PL	34.1 ± 7.7	31	45.9 ± 9.0	50	34.4 ± 7.2	41	54.1 ± 4.9	123
FCH	12.8 ± 2.2	12	7.6 ± 1.2	8	11.7 ± 1.7	14	5.8 ± .7	7
ECH	1.2 ± .4	1	1.7 ± 1.2	2	2.5 ± 1.3	3	2.6 ± .4	3
TCH	14.0 ± 2.5	13	9.3 ± 2.3	10	14.2 ± .6	17	8.4 ± .9	10
FFA	28.0 ± 4.8	25	15.4 ± 3.7	17	26.2 ± 4.2	31	21.6 ± 3.0	50
MG	8.8 ± 1.5	8	4.9 ± .8	5	7.3 ± 2.5	9	5.8 ± .6	14
DG + TG‡	15.1 ± 4.0	14	24.5 ± 5.0	27	17.9 ± 7.8	22	11.2 ± 2.3	26
B. Cottonseed oil								
	(7)		(8)		(7)		(8)	
PL	5.5 ± 2.0	60	4.1 ± .9	105	9.8 ± 2.9	48	4.4 ± 1.1	135
FCH	2.2 ± .5	24	.9 ± .2	23	7.1 ± .7	35	1.1 ± .3	34
ECH	.5 ± .1	5	.3 ± .1	8	.6 ± .1	3	.4 ± .1	12
TCH	2.7 ± .5	29	1.2 ± .2	31	7.7 ± .7	38	1.5 ± .4	46
FFA	30.6 ± 3.8	334	8.3 ± 1.3	214	50.5 ± 2.3	248	11.8 ± 2.3	364
MG	9.4 ± 1.7	94	4.4 ± .6	113	16.2 ± 1.7	79	6.1 ± .7	188
DG + TG	51.8 ± 5.1	566	82.0 ± 2.6	2113	15.8 ± 3.6	77	76.2 ± 3.3	2347

* Values are mean percentage composition by weight ± standard error of mean and mean weight in mg. Mean weights in mg of individual components were obtained by multiplying mean total weight of lipid recovered from a segment of intestine (Table I) by mean percentage by weight of a component. Abbreviations: PL, phospholipid; FCH, free cholesterol; ECH, esterified cholesterol; TCH, total cholesterol; FFA, free fatty acids; MG, monoglycerides; DG + TG, diglycerides and triglycerides.

† No. of samples in parentheses. Each sample for the fat-free meal represents lipid pooled from 3 dogs.

‡ Combined diglycerides and triglycerides were determined by difference between other fractions and 100%.

endogenous fecal lipid excretion to be about 3 times greater than normal in pancreatic duct-ligated dogs. As determined from the amount of lipid recovered from the stomach at the end of 4 hours, there was normal gastric evacuation of cottonseed oil by the depancreatized dogs. Thus, variations in gastric emptying time probably did not affect intestinal digestion and absorption of lipid by the operated animals. When the cottonseed oil meal was fed, significantly greater than normal quantities of lipid were found in the ileum of the depancreatized dogs. The presence of large amounts of unabsorbed lipid, either of endogenous or dietary origin, in the distal region of the intestine probably reflects the defective absorption of lipid which is known to accompany absence of the external secretion of the pancreas.

The composition of the intraluminar lipids recovered after feeding the test meals to normal and depancreatized dogs is given in Table II. If amounts of endogenous lipid comparable to those recovered after feeding the fat-free meal were present in corresponding intestinal segments of the dogs fed fat, nondietary lipid constituted an appreciable part of the total lipid recovered from some of the intestinal segments of the dogs fed cottonseed oil (Table I). Consequently, in order to obtain an estimate of the amount and composition of intraluminar lipids derived from dietary sources only, corrections were made as described previously(4) for the contribution of endogenous lipid to the various components making up the total lipid recovered after feeding fat. These corrections consisted of (1) excluding the weights of phospholipid and cholesterol recovered after feeding fat, since only trace amounts of these lipid components were present in the dietary fat and, (2) subtracting the weights of endogenous free fatty acids and glycerides recovered after ingestion of the fat-free meal from the weights of these same components recovered after the ingestion of fat. The corrected com-

TABLE III. Composition* of Gastric Lipids and Weights and Composition of Intestinal Intraluminar Lipids Corrected for Endogenous Lipid.

Lipid component	Normal			Depancreatized		
	Stomach	Duodenum-jejunum	Ileum	Stomach	Duodenum-jejunum	Ileum
Lipid wt, mg		947	342		2391	2809
FFA, %	2.9	32.6	63.4	1.4	8.2	11.2
MG, %	1.0	9.1	20.5	1.9	4.5	6.2
DG + TG, %	96.1	58.3	16.1	96.7	87.3	82.6

* Composition values are percentage by weight. Abbreviations as in Table II.

position values for the intestinal intraluminar lipids are given in Table III along with corresponding values for the gastric lipids.

As judged by the low concentration of free fatty acids and monoglycerides in the gastric lipids, cottonseed oil was only slightly hydrolyzed in the stomach of both groups of dogs. The corrected data on the intestinal lipids show that lipolysis was both progressive and extensive as cottonseed oil traversed the small intestine of the normal dogs. In this instance, the unabsorbed ileal lipid consisted mostly of free fatty acids and monoglycerides with only a small fraction of the total lipid being present as higher glycerides. On the other hand, although the absolute amounts of free fatty acids and monoglycerides recovered from corresponding intestinal segments of normal and depancreatized dogs were comparable (Table II), these lipid components recovered from both intestinal segments of the depancreatized dogs constituted only a relatively small fraction of the total lipid, which consisted mostly of di- and triglycerides.

The composition of the mucosal lipids of the dogs fed the 2 test meals (Table IV) is expressed as milligrams of lipid component per 100 g of wet mucosa. Exclusion of pancreatic juice did not alter from normal the composition of the mucosal endogenous lipid of the depancreatized dogs fed the fat-free meal. When fat was added to the meal, the amounts of mucosal phospholipids and cholesterol were not changed from those of dogs fed the fat-free meal. However, ingestion of fat was accompanied by significant increases in weight of the lipid fraction consisting of free fatty acids and glycerides (FFA + G) in both intestinal segments of the 2 groups of dogs. Increases in the weight of this lipid fraction are interpreted to mean that cottonseed oil was absorbed into the intestinal epithelium of all regions of the intestine of both groups of dogs. In addition, comparisons between normal and operated animals showed that the weight of free fatty acids plus glycerides was significantly greater in the duodenum-jejunum of the normal dogs ($P < .05$), whereas there was no difference for the ileum.

To evaluate the amount of mucosal free fatty acids and glycerides present as the result of absorption, the weights of the mucosal endogenous free fatty acids, monoglycerides, and di- and triglycerides recovered after feeding the fat-free meal were subtracted from those present after feeding fat. The sum of the corrected weights of these components is an estimate of the quantity of mucosal free fatty acids plus glycerides present in the mucosa as a result of absorption. The corrected values (Table V) show the same relationship between normal and depancreatized dogs as do the uncorrected weights. When corrected individual values are expressed as percentage composition by weight, it is apparent that the composition of the absorbed lipid is the same for corresponding intestinal segments of both groups of dogs. In each instance, most of the absorbed lipid was in the form of di- and triglycerides with a relatively small proportion being present as free fatty acids and monoglycerides.

Discussion. Conflicting results have been obtained from the few studies which have attempted to assess directly the intestinal digestion of triglycerides in the absence of pancreatic juice. Umber and Brugsch(10), using a single depancreatized dog, reported fat splitting to be 18% in the duodenum and 42% in the lower ileum. Some years later, Nothmann and Wendt(11) fed olive oil to dogs deprived of pancreatic juice and found

only 2-4% of the fat was hydrolyzed in the small intestine. Since gastric emptying was extremely rapid in these dogs, it was suggested(12) that lipolysis was limited because enhanced gastrointestinal motility did not allow sufficient time for digestion to occur. Douglas *et al*(13) subsequently reported that when different fat-containing mixed meals were fed to 5 depancreatized dogs, gastric emptying was normal and the average percentage hydrolysis of fat by these dogs was 33%, 50%, 60%, and 41% for the stomach, duodenum, jejunum, and ileum, respectively. These workers attributed the marked gastrointestinal digestion of fat mostly to the action of gastric lipase. However, the amount of free fatty acids in the dietary lipids was determined for one dog only (10.4%), making it difficult in most cases to accurately assess lipase activity.

If the composition of the intraluminar lipids obtained in the present study (Table III) accurately reflects the extent of lipolysis, intestinal hydrolysis of triglycerides by

TABLE IV. Composition* of Intestinal Mucosal Lipids of Normal Dogs and Dogs Deprived of Pancreatic Juice.

Lipid component	Duodenum-jejunum		Ileum	
	Normal	Depancreatized	Normal	Depancreatized
		A. Fat-free		
	(8)†	(8)	(8)	(8)
Total lipid	2269 ± 116	2075 ± 94	2151 ± 73	2190 ± 116
PL	1734 ± 54	1571 ± 84	1600 ± 64	1590 ± 84
TCH	257 ± 10	242 ± 15	273 ± 12	262 ± 14
FCH	243 ± 11	223 ± 11	246 ± 10	234 ± 10
ECH	14 ± 4	19 ± 7	27 ± 5	15 ± 6
FFA + G	278 ± 17	262 ± 16	278 ± 16	351 ± 30
FFA	57 ± 6	64 ± 5	62 ± 6	88 ± 7
MG	20 ± 2	17 ± 3	20 ± 2	25 ± 2
DG + TG‡	201 ± 13	181 ± 13	196 ± 13	238 ± 22
		B. Cottonseed oil		
	(7)	(8)	(7)	(8)
Total lipid	2955 ± 113	2394 ± 169	2683 ± 159	2560 ± 188
PL	1752 ± 69	1600 ± 57	1658 ± 79	1520 ± 60
TCH	263 ± 11	224 ± 12	273 ± 12	236 ± 15
FCH	241 ± 9	204 ± 10	245 ± 10	206 ± 13
ECH	22 ± 4	20 ± 5	28 ± 4	30 ± 7
FFA + G	940 ± 106§	580 ± 122‖	752 ± 138§	804 ± 154§
FFA	144 ± 24	108 ± 15	96 ± 12	119 ± 16
MG	53 ± 7	31 ± 6	34 ± 6	44 ± 6
DG + TG	743 ± 99	431 ± 112	622 ± 136	641 ± 135

* Values are mean ± S.E. of mean and represent milligrams of lipid/100 g of wet musosa. Abbreviations as in Table II, with addition of G, glycerides, which is the sum of MG and DG + TG.

† Number of samples given in parentheses.

‡ Combined di- and triglycerides were determined by difference between other fractions and total lipid.

§ $P < .01$.

‖ $P < .05$.

TABLE V. Weights and Composition* of Mucosal Free Fatty Acids Plus Glycerides Corrected for Endogenous Lipid.

Lipid component	Duodenum-jejunum		Ileum	
	Normal	Depancreatized	Normal	Depancreatized
Lipid wt, mg	662	308	474	453
FFA, %	13.2	14.3	7.2	6.8
MG, %	5.0	4.6	3.0	4.2
DG + TG, %	81.8	81.1	89.8	89.0

* Lipid weights are mg/100 g wet mucosa and composition values are percentage by weight. Abbreviations as in Table II.

depancreatized dogs is considerably less than normal. This difference cannot be explained on the basis of a faster than normal gastric emptying time by the operated animals, because gastric evacuation of lipid by both groups of dogs was quite similar (Table I).

A number of sources of gastrointestinal lipase activity other than that of the pancreas have been suggested. These include gastric lipase(13), intestinal lipases(14,15), and intestinal bacteria(12). That gastric lipase was probably not an important source of lipolytic activity in this study is pointed out by the fact that only slight hydrolysis of fat occurred in the stomach of both groups of dogs. With regard to intestinal lipase, Dinella *et al* (14), who found hog intestinal homogenates to have lipase activity as measured by the hydrolysis of olive oil, suggested that, although the activity of intestinal lipase was only 5% that of pancreatic tissue, total intestinal lipase activity may approach that of pancreatic lipase because of the relatively large intestinal mucosal mass. The present study does not support this idea because, even if all hydrolysis observed in the operated animals is the result of intestinal lipase activity, the extent of lipolysis is only a small fraction of that when pancreatic lipase is available.

One observation regarding certain physical characteristics of intestinal contents recovered from the normal and operated animals is worth reporting. The chyme collected from all regions of the intestine of the depancreatized dogs was in the form of a thick paste, whereas that from normal dogs was quite fluid in nature. A paste is not a medium from which optimal absorption might be expected to occur and, as has been previously suggested(2), the external secretion of the pancreas may have a function other than digestive. This idea was based on the fact that duodenal installation of $NaHCO_3$ in depancreatized dogs fed oleic acid reduced fecal fat excretion to a normal level. Thus, the fluid component of the pancreas, as well as the enzymes contained therein, may be important for optimal absorption of fat.

The fact that most of the lipid absorbed by the intestinal segments of normal dogs was in the form of higher glycerides (Table V) is in accord with the idea that absorbed lipids are for the most part converted to triglycerides in the cells of the intestinal epithelium. Recovery and analysis of the mucosal lipids of depancreatized dogs was prompted by results obtained in an earlier study(4), which suggested that intracellular esterification of the absorbed products of fat digestion is retarded in the absence of bile. However, the composition of the absorbed mucosal lipids of the depancreatized dogs was the same as normal, indicating that there is no defect in the intracellular absorption mechanism when pancreatic juice is absent from the intestine.

Summary. The role of pancreatic juice in fat digestion and absorption was investigated by studying the intestinal intraluminar and mucosal lipid changes which occur after feeding cottonseed oil triglycerides to normal dogs and dogs deprived of pancreatic juice by partial pancreectomy. As based on the low concentrations of intraluminar free fatty acids and monoglycerides, intestinal digestion of cottonseed oil by the depancreatized dogs was considerably less than normal. It is concluded that, relative to pancreatic lipase, other sources of gastrointestinal lipase activity contribute only to a minor extent to the hydrolysis of fat. The composition of mucosal lipids of dietary origin, which consisted primarily of higher glycerides, was the same for normal and depancreatized dogs. This result suggests that intercellular esterification of the absorbed products of fat digestion is normal in the absence of pancreatic juice.

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1. Pessoa, V. C., Kim, K. S., Ivy, A. C., Am. J. Physiol., 1953, v174, 209.

2. Annegers, J. H., A.M.A. Arch. Int. Med., 1954, v93, 9.

3. Cohen, B. J., Proc. Soc. Exp. Biol. and Med., 1961, v107, 40.

4. Knoebel, L. K., Ryan, J. M., Am. J. Physiol., 1963, v204, 509.

5. Markowitz, J., Archibald, J., Downie, H. G.,

Experimental Surgery, Williams and Wilkins, Baltimore, 1954, 357.

6. Borgstrom, B., Acta Physiol. Scand., 1952, v25, 101.

7. Fiske, C. H., Subarrow, Y., J. Biol. Chem., 1925, v66, 375.

8. Sperry, W. M., Webb, M., ibid., 1950, v187, 97.

9. Pohle, W. D., Mehlenbacher, V. C., J. Am. Oil Chem. Soc., 1950, v27, 54.

10. Umber, F., Brugsch, T., Arch. f. Exp. Path. u. Pharm., 1906, v55, 196.

11. Nothmann, M., Wendt, H., ibid., 1931, v162, 472.

12. Handelsman, M. B., Ann. Int. Med., 1937-38, v11, 1479.

13. Douglas, G. J., Reinauer, A. J., Brooks, W. C., Pratt, J. H., Gastroenterology, 1953, v23, 452.

14. Dinella, R. R., Meng, H. C., Park, C. R., J. Biol. Chem., 1960, v235, 3076.

15. Tidwell, H. C., Johnston, J. M., Arch. Biochem. & Biophys., 1960, v89, 79.

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Organic Phosphate Compounds of Erythrocytes from Individuals with Cirrhosis of the Liver.* (29787)

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Comparatively little information is available concerning the metabolism of the red cells of cirrhotic individuals. Pitcher and Williams(1) noted that the red cells of patients with various types of liver diseases, including cirrhosis, had low levels of reduced glutathione despite the presence of normal activities of glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase. Bock *et al*(2) reported that the aldolase, triose isomerase, phosphoglyceraldehyde dehydrogenase, lactic acid dehydrogenase, and methemoglobin reductase activities were significantly increased in the washed red cells of cirrhotic patients. Italian investigators(3) noted that the concentration of ATP† was markedly decreased, ADP was significantly decreased, while AMP was nearly normal. The decrease in ATP appeared to be proportional to the severity of the disease syndrome.

The present investigation was undertaken to determine the concentrations of a number of phosphorylated compounds in the washed red cells of patients with cirrhosis of the liver.

Materials and methods. The following diagnoses were made on the 21 patients studied: Laennec's cirrhosis (12), post-necrotic cirrhosis(4), cirrhosis associated with hemochromatosis (2), cardiac cirrhosis (2), and acute yellow atrophy associated with serum hepatitis (L.B). Healthy young adults served as controls. In all cases, blood was drawn during the fasting state into acid-citrate-dextrose solution. The bloods of 5 patients were collected and stored in glass-stoppered tubes to determine the effect of storage at 4°C. The preparation of the erythrocytes, extraction with trichloroacetic acid, and the anion exchange analysis of the phosphorylated compounds(4) have been described(5). The phosphate concentrations are expressed as μmoles/g Hb since phosphorylated compounds have been shown to form complexes with hemoglobin(6).

Results and discussion. The analytical data for P_{total}, P_{org}, P_i, DPG, ATP, ADP, AMP, HDP, and IMP of freshly drawn erythrocytes from control and cirrhotic subjects are plotted in Fig. 1 and the statistical evaluation is presented in Table I. The statistically significant increase in P_{org} concentration

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† Abbreviations used in this paper are: P_{org}, organic phosphate; P_i, inorganic phosphate; P_{total}, total phosphate; DPG, 2,3-diphosphoglycerate; ATP, ADP, AMP, adenosine tri-, di-, monophosphate; HDP, hexose diphosphate; IMP, inosine monophosphate.