the average survival time for the group was 60 days. Gross inspection at autopsy showed the pancreas to be firm and atrophic; the gall bladder and bile ducts were dilated and the liver was firm and rough. Microscopic examination of the pancreas usually showed only mild degrees of interstitial pancreatitis and fibrosis.

The remaining 7 dogs with intact anastomoses had a significant degree of clinical jaundice at the time of death even though a patent channel was demonstrated from the gall bladder through the pancreatic duct system into the duodenum at autopsy. This circumstance suggests that although some bile was probably passing through the pancreatic duct system, the flow was not free enough to maintain the serum bilirubin at normal levels.

Summary and conclusions. 1. The entire flow of bile was successfully diverted through the pancreatic duct system into the intestine in a total of 11 dogs. Eight of these dogs did not develop any significant degree of pancreatitis even though the average observation period for this group was more than 60 days, and only one dog in the series developed a full-blown acute pancreatic necrosis. 2. The results of these experiments indicate that the presence of bile in the pancreatic duct system under physiologic pressures is tolerated for long periods of time without any serious damage to the pancreas in most cases.

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Received August 18, 1955. P.S.E.B.M., 1955, v90.

Lactic Dehydrogenase Activity in Blood.* (21985)

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The observation that during experimental and clinical myocardial infarction glutamic oxaloacetic transaminase is released from cardiac muscle resulting in increased enzyme activity in the serum(1-5), suggested that other cardiac tissue enzymes behave similarly during myocardial infarction. Although present in other tissues in greater activity, lactic dehydrogenase, the enzyme concerned primarily with the reduction of pyruvic acid to lactic acid, is present in appreciable activity in cardiac musculature. In order to ascertain whether lactic dehydrogenase (hereafter referred to as LD) activity is increased in the serum during myocardial infarction, it was necessary to first demonstrate its presence in human and animal blood, and to delineate variations in LD activity in the blood of normal and diseased man.

Methods and materials. Lactic dehydrogenase is concerned with the reduction, in the presence of reduced diphosphonucleotide (DPNH), of alpha-keto and of alpha, gamma-diketo acids; maximum reduction by LD has been shown to occur with pyruvic acid. Inasmuch as LD catalyzes the reduction of various alpha-keto and alpha, gamma-diketo acids, the reduction of a substrate in the presence of LD cannot be used to specifically identify an alpha-keto or an alpha, gamma-diketo acid. However, the catalytic reduction of a known amount of a specific keto acid may be used to quantitatively estimate LD activity(6). The *LD activity* of human serum was measured

* Supported by grants from the New York Heart Assn. and the U.S.P.H.S. (H-1978).

spectrophotometrically. To 2.4 ml of pH 7.4 phosphate buffer, 0.1 m of serum and 0.1 m of reduced diphosphonucleotide (DPNH) (2.5 mg per ml) were added. After 20 minutes, 0.1 ml of sodium pyruvate (2.5 mg per ml) was added. The optical density at wavelength 340 mμ was followed for 3 to 5 minutes at intervals of 10, 15, or 30 seconds depending on the rate of reaction. The rate of decrease of optical density representing the rate of oxidation of DPNH was taken as the measure of LD activity of serum. The reaction was followed in a Beckman model DU spectrophotometer using a tungsten light source and at room temperature, 24 to 27°C. The activity is expressed as units per ml of serum per minute. One unit equals a decrease in optical density of 0.001 per minute per ml under the conditions described.

When DPNH and pH 7.4 phosphate buffer were added to serum without the addition of pyruvate, the optical density of the mixture decreased slowly for up to 20 minutes indicating the oxidation of a finite quantity of DPNH. This initial oxidation of DPNH by serum without added pyruvate appears to depend on the presence of alpha-keto and alpha, gamma-diketo acid in the serum (in the presence of serum LD). The reaction stopped when the keto and diketo acids were enzymatically reduced to lactate by DPNH. The addition of more DPNH will not restart the reaction, but the addition of pyruvate resulted in a steady decrease of optical density which was taken as a measure of LD activity of the serum. The optimal concentration of DPNH was found to be 0.1 ml (2.5 mg DPNH per ml in water made basic to pH 9.0 with sodium hydroxide) in a total reaction mixture of 3.0 ml (3.8×10^{-7} M). At pH 7.2 to 7.6, serum LD was optimally active and this is in substantial agreement with that reported for LD from other tissue sources(6-8). The LD activity of serum was maximal when 0.1 ml of pyruvate (2.5 mg per ml of distilled water) was used in a total reaction mixture of 3.0 ml (0.23×10^{-5} M). The rate of LD activity was directly proportional to the quantity of serum used. No activators of LD were used although it has been reported(9) that amino-DPNH potentiates LD activity. Competitive inhibitions of LD by alpha, gamma-diketo valerie acid has been reported(6) but the enzyme is insensitive to sulphydryl reagents(10). Serum samples stored from 30 minutes to 96 hours at room temperature or for periods of from 1 hour to 1 week in the refrigerator (0° to 5°C) showed no significant alteration in serum LD activity; the heat stability noted is in essential agreements with reports of LD derived from other sources(7, 11).

TABLE I. Distribution of Lactic Dehydrogenase Activity in Venous Serum of Normal Adults.

LD activity (units)	No. of individuals	%
250-290	7	4.3
300-340	18	11.2
350-390	25	15.5
400-440	32	20.0
450-490	22	13.7
500-540	16	10.0
550-590	17	10.6
600-640	7	4.3
650-690	7	4.3
700-740	2	1.2
750-790	4	2.5
800-840	3	1.9
850-890	1	0.7
Total	161	100.2

Mean LD activity, 470 units ± 130 units.

Results. 1. *Lactic dehydrogenase activity* in *blood* of human adults. The venous serum LD activity in 161 normal human adults varied from 260 to 850 units with a mean activity of 470 per m + 130 units (Table I). The LD activity found in whole venous blood hemolysates from normal human adults ranged from 16,000 to 67,000 units with a mean value of 34,000 ± 12,000 units per ml. The whole blood hemolysates were noted to be on the average approximately 100 times

TABLE II. Lactic Dehydrogenase Activity of Heparinized Plasma, Serum, and Whole Blood Hemolysate of the Same Venous Blood Samples.

Hematocrit (mm)	Whole blood hemolysate (units/ml)	Plasma (units/ml)	Serum (units/ml)
	×1000		
42	22	370	380
43	26	370	380
45	40	800	750
50	33	300	400
50	24	500	420
50	31	330	290
51	16	370	390
52	30	480	430
53	27	830	340
54	30	500	360

TABLE III. Estimated Lactic Dehydrogenase Activity of Tissues of the Dog.

Tissue	Units/g wet tissue (×1000)
Kidney	640
Skeletal muscle	600
Liver	390
Heart	240
Pancreas	150
Spleen	140
Brain	130
Lung	25

more active than the serum of the corresponding venous blood samples, but no consistent relationship was noted between serum and corresponding whole blood hemolysate LD activities, and none between whole blood hemolysate activity and hematocrit (Table II). The LD activity of plasma obtained from heparinized venous blood samples was not significantly different from the activity of serum from a corresponding venous blood sample, oxalated plasma was considerably less active suggesting oxalate inhibition and/or interference. In no instance was LD activity absent in the sera of normal human adults tested or in any of the sera of hospitalized patients with various disease states. The fasting did not appear to significantly influence the LD activity, and no consistent alteration in LD activity was observed during glucose tolerance studies in normal or in diabetic individuals. Day to day serum LD activities in the same individual varied 30% (within the normal range). Duplicate determinations on the same serum samples varied by 10% or less. LD activity was not connected with sex.

A sampling was made of patients hospitalized at Memorial Center with various diseases and serum LD activity determined in each instance. Activities within the normal range were obtained on sera from a limited selection of patients with infectious, degenerative, neoplastic and other disease states. High activities were observed in patients with myocardial infarction, diabetic acidosis, acute stem cell leukemia, chronic myelogenous leukemia and hepatitis.

2. *Lactic dehydrogenase activity* in the *tissues* and *blood* of experimental animal. LD activity was demonstrable in all the serum samples obtained from several experimental animals. LD activity in the venous sera of the experimental animals studied varied from 380 units per ml in the rabbit to 3100 units per ml in the rat with progressively diminishing values in between these 2 extremes in the dog, hamster, mouse and guinea pig. Using aliquot samples of macerated homogenized dog tissues obtained immediately after the death of the animal, the LD activity of various tissues was estimated and are summarized in Table III.

Following the intravenous administration of 0.13 ml of lactic dehydrogenase† diluted with saline to 5.0 ml (equivalent to approximately 240,000 units of LD activity or about one gram of dog heart muscle homogenate) to a 12.5 kilo dog, the serum was sampled at periodic intervals and examined for LD activity as indicated in Fig. 1.

Fig. 2 describes the serum LD alterations following experimental myocardial infarction, produced by the closed chest technic(5). (The detailed observations of serum and cardiac tissue LD activity elevations seen in experimental graded myocardial infarction and in clinical myocardial infarction will be the subject of a subsequent report.) As seen in the case of glutamic oxaloacetic transaminase, serum LD rises during the course of myocardial infarction. Similar alterations in serum LD activity have been observed in clinical myocardial infarction (Fig. 3).

Discussion. Lactic dehydrogenase activity

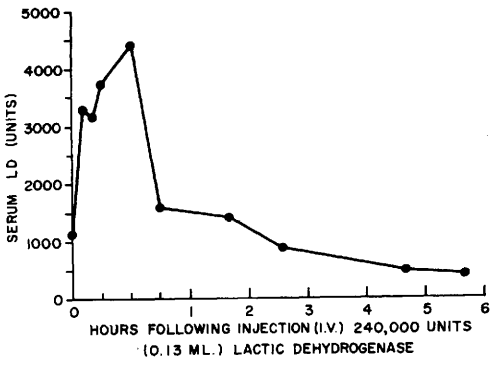

FIG. 1. Serum lactic dehydrogenase activity in a 12.5 kilo dog following the intravenous injection of approximately 240,000 units of lactic dehydrogenase.

† Obtained from Nutritional Biochemical Corp, Cleveland, O.

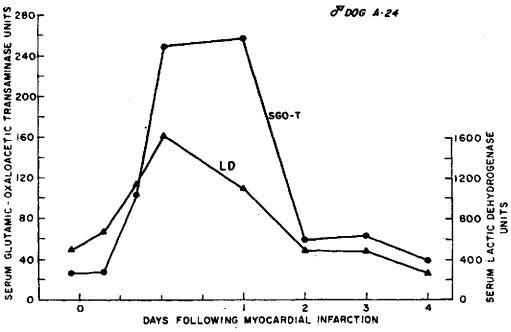

FIG. 2. Serum lactic dehydrogenase alterations during the course of closed chest experimental myocardial infarction in the dog. Comparison with serial changes in serum glutamic oxaloacetic transaminase is shown.

is present in all human sera and in all whole blood hemolysates examined. The previously reported serum glutamic oxaloacetic transaminase activity alterations in the course of myocardial infarction appear to be mimicked by similar changes in LD activity. That the latter changes are parallel but not related to serum glutamic oxaloacetic transaminase activity alterations is indicated by a comparison of serum glutamic oxaloacetic transaminase and LD activity observed during the course of clinical hepatitis. The LD and serum glutamic-oxaloacetic transaminase activity alterations following myocardial infarction are comparable in direction and degree but probably independent of one another. The fact that elevation of both LD and serum glutamic oxaloacetic transaminase follow heart muscle injury suggests that these enzymes are liberated from the damaged muscle cells into the blood stream. If such is the case for these

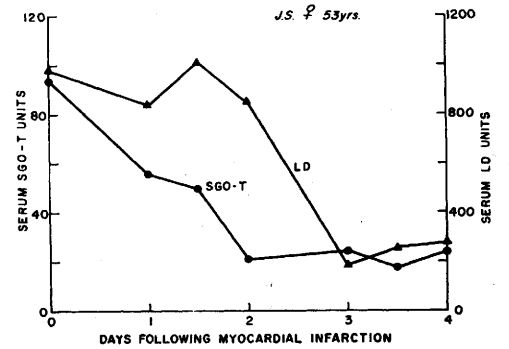

FIG. 3. Serum lactic dehydrogenase alterations during the course of clinical anterior myocardial infarction. Comparison with serial changes in serum glutamic oxaloacetic transaminase is shown.

two enzymes, there is every reason to expect that other cardiac tissue enzymes, when present in sufficient concentrations in the heart muscle, would also be released following damage to the heart muscle cells with a consequent change in the serum enzyme activity. This generalization is presently under investigation.

Summary and conclusions. 1. Lactic dehydrogenase activity is present in the venous serum of normal human adults. Normal activity ranges from 260 to 850 units per ml with a mean value of 470 ± 130 units per ml. 2. Venous whole blood hemolysates of normal adults have a lactic dehydrogenase activity varying between 16,000 to 67,000 units per ml with a mean value of 34,000 ± 12,000 units per ml. 3. Alterations in serum lactic dehydrogenase have been studied in a selected group of disease states. 4. Experimental and clinical myocardial infarction are associated with a rise in serum lactic dehydrogenase activity. 5. Lactic dehydrogenase like serum glutamic oxaloacetic transaminase rises in a characteristic fashion following myocardial infarction.

The authors wish to acknowledge the technical assistance of Martin Podgainy. Dr. P. Ruegsegger participated in the study of experimental myocardial infarction in dogs.

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