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ACTIVITY IN MYOCARDIAL ISCHÆMIA**

**BY**

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## Serum $\gamma$ -Glutamyl Transpeptidase Activity in Myocardial Ischæmia

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The enzyme,  $\gamma$ -glutamyl transpeptidase, catalyses the transfer of  $\gamma$ -glutamyl groups from  $\gamma$ -glutamyl peptides to other peptides, L amino acids, and water. A high level of activity has been found in normal renal, pancreatic, pulmonary, and hepatic tissues (Goldberg *et al.*, 1960). High levels of this enzyme have been found in sera of patients with hepatic and pancreatic disorders (Rutenburg, Goldberg, and Pineda, 1963) and in diabetes mellitus (Goldberg *et al.*, 1963), but in the absence of disease affecting these organs Agostoni, Ideo, and Stabilini (1965) found high serum levels following myocardial infarction, with a peak on about the tenth day. This led us to study the behaviour of this enzyme in the sera of a more comprehensive group of patients with ischæmic heart disease. It was thought that changes in levels of this enzyme might help in distinguishing patients with myocardial infarction from those presenting similarly but showing no evidence of myocardial necrosis, as judged by the appearance of serial electrocardiograms and the observation of the serum activity of other enzymes whose place in the diagnosis of myocardial infarction is well established.

### SUBJECTS AND METHODS

Thirty-seven consecutive patients admitted to hospital with suspected myocardial infarction were studied.

Blood was taken for laboratory investigations as soon as possible after admission, every morning for the first seven days, and thereafter on alternate days as long as the patient was in hospital.

The serum bilirubin, thymol turbidity, alkaline phosphatase, serum albumin, and total globulins were estimated on admission. The erythrocyte sedimentation rate (Westergren) and white cell count were measured

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daily until they returned to normal levels, and thereafter twice weekly.

The serum aspartate aminotransferase (SGOT), alanine aminotransferase (SGPT)†, lactate dehydrogenase (LDH), hydroxybutyrate dehydrogenase, and  $\gamma$ -glutamyl transpeptidase were estimated daily during the hospital stay of the majority of the patients, and where possible serum  $\gamma$ -glutamyl transpeptidase levels were measured when the patients later attended the out-patient clinic.

An electrocardiogram was taken at the time of admission, two days later, and thereafter at weekly intervals while the patient was in hospital.

All cases of myocardial infarction and angina pectoris, except one, received anticoagulant therapy with phenindione for a minimum of one month.

*Clinical Assessment.* After admission each patient was placed in one of three groups: (i) acute myocardial infarction, (ii) severe angina with no evidence of recent myocardial infarction, or (iii) a group with chest pain due to non-cardiac causes.

Clinical assessment included consideration of the erythrocyte sedimentation rate, white cell count, and serum aspartate aminotransferase, but the variations in the other serum enzyme levels were not correlated with the patient's condition until after a final diagnosis had been made at the time of discharge from hospital.

A patient was placed in the acute myocardial infarction group when serial electrocardiograms showed pathological Q waves and the typical evolutionary ST segment and T wave changes of acute myocardial infarction. Additional supportive evidence that myocardial infarction had occurred was found on clinical grounds, i.e. when chest pain of cardiac character lasted for more than one hour, when the patient was admitted in a shocked condition or developed a pyrexia of over 38°C. (100°F.), and on laboratory grounds, when the serum aspartate aminotransferase was temporarily raised.

† Formerly known as, glutamate-oxaloacetate transaminase and glutamatepyruvate transaminase, respectively (International Union of Biochemistry, 1961).

A patient whose serial electrocardiograms showed depression of the ST segment or non-progressive T wave inversion, without increases in serum aspartate aminotransferase levels, was placed in the severe angina group.

*Laboratory Methods and Normal Ranges.* Serum aspartate aminotransferase and alanine aminotransferase were estimated by the method of Reitman and Frankel (1957), serum lactate dehydrogenase by the method of Wróblewski and LaDue (1955), and serum hydroxybutyrate dehydrogenase by the method of Elliott and Wilkinson (1961).

The normal ranges found in 50 hospital staff for serum aspartate aminotransferase and alanine aminotransferase were 7–30 units per ml. and 5–26 units per ml., respectively. In 24 hospital staff the normal range for lactate dehydrogenase was 145–380 units per ml. and for hydroxybutyrate dehydrogenase it was 120–300 units per ml.

The method used for the estimation of serum  $\gamma$ -glutamyl transpeptidase was similar to that of Agostoni *et al.*, which was based on the method of Goldberg *et al.* (1963) but using DL  $\gamma$ -glutamyl  $\beta$ -naphthylamide as substrate. The units are defined as the number of micromoles of  $\beta$ -naphthylamine liberated per 100 ml. of serum in 2 hours, and the normal range in 50 hospital staff, aged between 18 and 60 years, was 4–29 units per ml., which is in agreement with the findings of Agostoni *et al.* The distribution was positively skewed. Direct comparison with the work of Goldberg *et al.* is not possible because of the different substrate, N (DL  $\gamma$ -glutamyl) aniline that they used. However, we found no sex variation, though Goldberg *et al.* reported significantly higher values in men than in women.

## RESULTS

Twenty patients were considered to have had an acute myocardial infarction, 11 had severe angina without infarction, and 6 had chest pain due to other conditions.

### *Patients with Acute Myocardial Infarction*

The 20 patients with myocardial infarction consisted of 17 men and 3 women between 30 and 70 years: 10 had anterior infarction, 9 posterior infarction, and one was considered to have sustained an acute myocardial infarction on clinical grounds, despite a normal electrocardiogram. Three patients died during the study: 2 suffered further myocardial infarction 2 weeks after admission to hospital, and the third developed a ventricular septal defect following extensive antero-septal infarction and died from intractable heart failure 18 days later.

### *Enzyme Levels.*

*Serum aspartate aminotransferase (SGOT).* A raised level of this enzyme was seen in 19 of the 20 patients with myocardial infarction 27 to 40

hours after infarction (Fig. 1a): 14 of them still showed high levels three days after infarction, and it was not until the seventh day that all had normal levels.

*Serum alanine aminotransferase (SGPT).* A small rise, usually coinciding with the serum aspartate aminotransferase peak, was seen in 10 out of 20 patients. In 2 of them a further rise occurred about a week after infarction, and this was considered to be due to venous congestion of the liver, secondary to the onset of congestive cardiac failure (Rowell and Smith, 1959; Richman, Delman, and Grob, 1961).

*Serum lactate dehydrogenase (LDH) and hydroxybutyrate dehydrogenase.* Raised levels of serum lactate dehydrogenase were noted in 18 of the 20 patients on the second day after myocardial infarction (Fig. 1b); 15 of them showed serum levels in excess of 400 units per ml. seven days after infarction and 9 on the twelfth day. Only one did not show a level of serum lactate dehydrogenase in excess of 400 units per ml. on at least one day after infarction.

The changes in serum hydroxybutyrate dehydrogenase levels paralleled those of lactate dehydrogenase.

*Serum  $\gamma$ -glutamyl transpeptidase.* A significant rise in serum levels of this enzyme was seen in 16 of the 20 patients during the period of study (Fig. 2). Unlike Agostoni *et al.*, who reported that serum  $\gamma$ -glutamyl transpeptidase levels were normal on the first four days after infarction, we found that out of 7 patients from whom samples were taken within 12 hours of the clinical episode of acute infarction, 5 had abnormal serum levels of this enzyme. Moreover, these patients gave no history of cardiac pain before the episode of acute infarction.

A sudden rise in serum  $\gamma$ -glutamyl transpeptidase level frequently occurred between the third and eighth day after infarction (average 5 days) and in all 16 patients the highest level was recorded between the fourth and fourteenth day (average 8 days) (Fig. 1c). These levels gradually fell during the following month. In 11, the peak serum  $\gamma$ -glutamyl transpeptidase coincided with the highest erythrocyte sedimentation rate.

No correlation could be obtained between the increase in serum  $\gamma$ -glutamyl transpeptidase level and the patient's age, duration of chest pain, duration of pyrexia, presence of shock on admission, or the extent of myocardial necrosis, as assessed by the electrocardiogram.

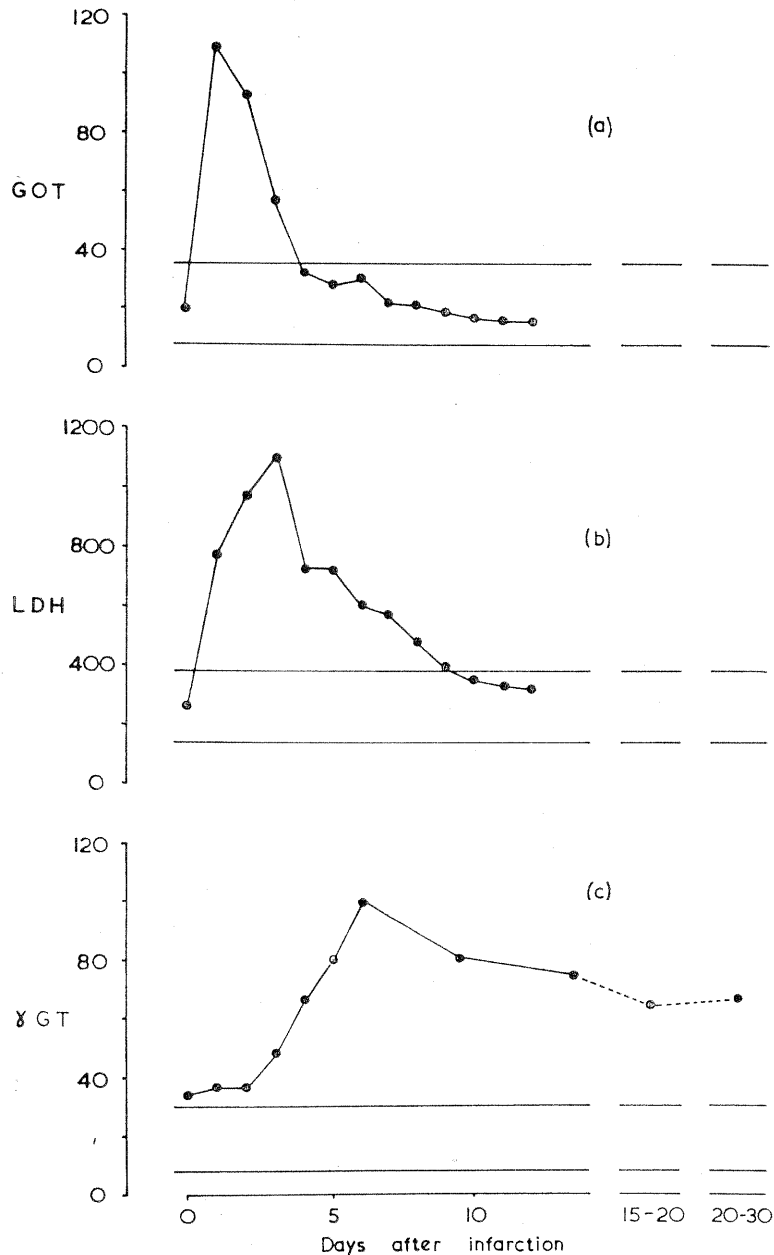


FIG. 1.—Mean values of (a) serum aspartate aminotransferase (GOT), (b) lactate dehydrogenase (LDH), and (c)  $\gamma$ -glutamyl transpeptidase ( $\gamma$  GT) in 20 patients after myocardial infarction. The continuous lines show the upper limit of the normal ranges.

Of 10 patients seen 30–120 days after discharge from hospital, raised serum levels of  $\gamma$ -glutamyl transpeptidase were still found in 2, though there was no clinical evidence of recurrence of infarction, or that congestive cardiac failure had developed.

#### *Patients with Severe Angina*

In all, 11 patients, 10 men and 1 woman, aged between 42 and 69 years, were admitted with cardiac pain lasting on no occasion for longer than half an hour but occurring with such increasing frequency

that a period of bed-rest was necessary. Patients could be grouped according to the length of history of chest pain: 4 gave a history of angina for less than 8 weeks, and 7 a history beginning between 1 and 10 years before admission.

#### *Enzyme Levels.*

*Serum aspartate aminotransferase, alanine aminotransferase, lactate dehydrogenase, and hydroxybutyrate dehydrogenase.* In this group 10 of the 11 patients showed no changes on serial estimation of

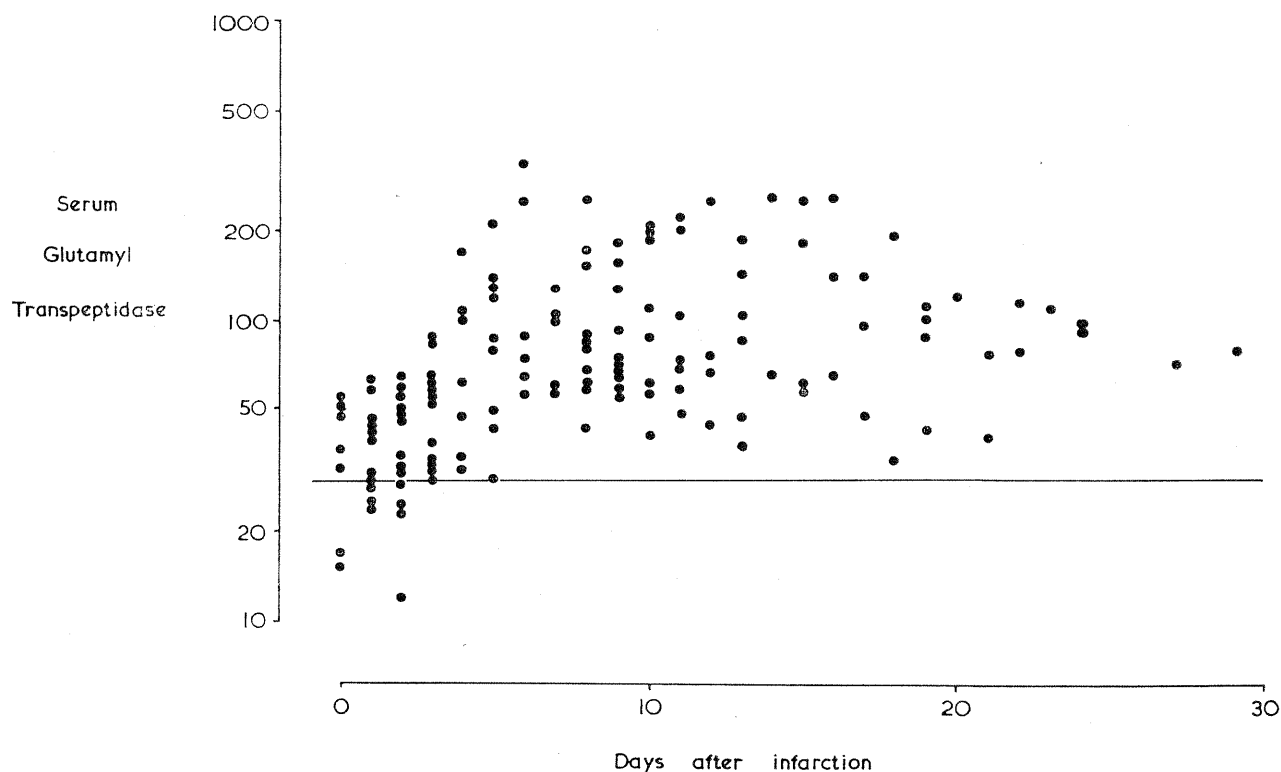


FIG. 2.—Serum  $\gamma$ -glutamyl transpeptidase activities in 16 patients, showing a rise in serum  $\gamma$ -GT after myocardial infarction. The continuous line indicates the upper limit of the normal range.

any of these serum enzyme levels. One patient, however, showed raised levels of all four enzymes. This was a man admitted with a long history of angina, but in addition mitral regurgitation and congestive cardiac failure were found on clinical examination. Enzyme levels were very high on admission but fell to normal in three weeks, after successful treatment of the cardiac failure. These enzyme levels were considered to be due to hepatic congestion.

*Serum  $\gamma$ -glutamyl transpeptidase.* Excluding the patient described above with hepatic congestion, whose serum  $\gamma$ -glutamyl transpeptidase was also very high, 7 out of 10 patients with severe angina had raised levels of this enzyme. During the period of study these levels fell slightly (Fig. 3).

All 4 patients giving a short history of angina, and 3 with a long history, showed raised serum levels of  $\gamma$ -glutamyl transpeptidase. One patient in the latter group is of particular interest in that a comparison of his electrocardiogram taken on admission with one recorded eight weeks previously showed that antero-septal infarction had occurred during this period.

#### *Patients with Chest Pain Not Due to Myocardial Ischaemia*

Six patients admitted with a presumptive diagnosis of myocardial infarction did not have ischaemic heart disease. One was a man with acute non-specific pericarditis. Although serum levels of aspartate amino-transferase, alanine aminotransferase, lactate dehydrogenase, and hydroxybutyrate dehydrogenase remained normal throughout the illness, the serum  $\gamma$ -glutamyl transpeptidase rose from a level of 82 units per ml. on admission to 184 units per ml. 10 days later. The serum activity of this enzyme fell to normal after two months, and this coincided with the return to normal of the electrocardiogram.

Two patients were admitted with pulmonary embolism. Neither patient showed a rise of serum  $\gamma$ -glutamyl transpeptidase, though one had a rise of serum aspartate aminotransferase on the second day, and levels of serum lactate dehydrogenase and hydroxybutyrate dehydrogenase in excess of normal were recorded for more than a week. One patient was a man who died from congestive cardiac failure two days after admission, and high levels of all the enzymes studied, including  $\gamma$ -glutamyl transpepti-

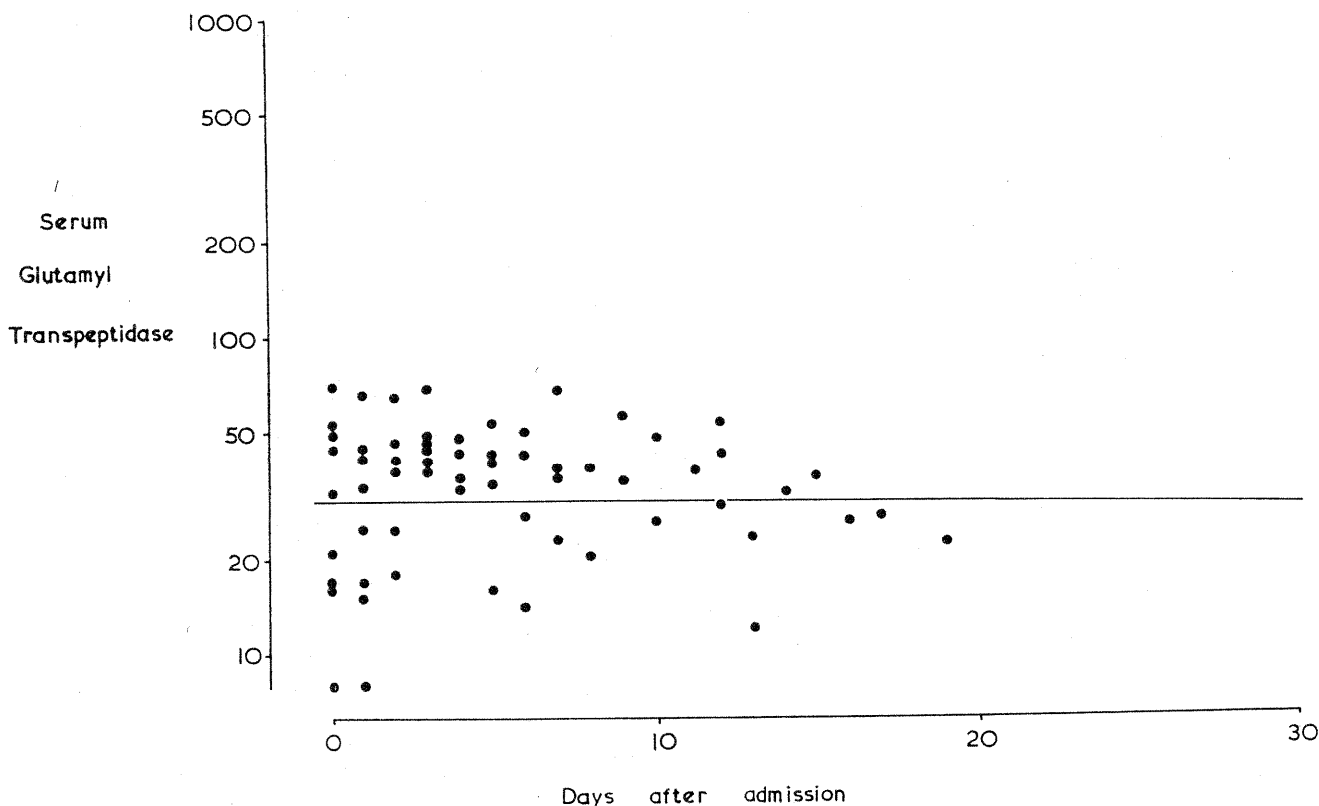


FIG. 3.—Serum  $\gamma$ -glutamyl transpeptidase activities in the patients with severe angina pectoris. The continuous line indicates the upper limit of the normal range.

dase, were found. Another man was admitted with left ventricular failure but there was no evidence of venous congestion of the liver. However, signs of right heart failure had been noted 10 days before admission when appropriate treatment had been initiated. In this case, though all the other enzymes studied showed no change, the serum  $\gamma$ -glutamyl transpeptidase was 68 units per ml. on admission and the serum activity of this enzyme fell slowly during the period of observation.

One patient was admitted with acute non-icteric cholecystitis and normal levels of each enzyme studied were recorded.

#### DISCUSSION

The typical case of myocardial infarction showed a sudden rise of the serum  $\gamma$ -glutamyl transpeptidase level between the third and eighth day after infarction. However, the diagnostic value of estimating the serum activity of this enzyme in acute myocardial infarction is limited by the finding that 4 out of 20 patients showed no rise above the normal level and that 5 were admitted with levels already in excess of normal. In view of these findings we feel that the estimation of serum  $\gamma$ -glutamyl transpeptidase is probably only of use in the diagnosis of acute myocardial infarction after the first week,

when the other serum enzymes may have returned to normal levels, and when the electrocardiogram may still not show acute changes. However, in view of the high levels of serum  $\gamma$ -glutamyl transpeptidase found in some cases up to two months after myocardial infarction, the finding of a raised level of this enzyme in a case of suspected recurrent infarction is of doubtful value if it is taken during this time.

In the group of patients we investigated, the serum lactate dehydrogenase and hydroxybutyrate dehydrogenase were more consistently raised after acute myocardial infarction, and 15 out of 20 were still raised seven days after the incident. It was technically more satisfactory to estimate the serum hydroxybutyrate dehydrogenase rather than the serum lactate dehydrogenase, as enzyme inhibitors did not develop so readily in the reagents.

A high proportion of patients with severe angina had raised serum  $\gamma$ -glutamyl transpeptidase levels. All our patients with angina could be classified in the group described by Master *et al.* (1947) as suffering from "Acute Coronary Insufficiency". It was thought that patients with this condition did not show raised levels of serum aspartate aminotransferase (LaDue and Wróblewski, 1955; Chinsky and Sherry, 1957) or lactate dehydrogenase (Wróblew-

ski, 1957) unless myocardial necrosis had occurred. Wood (1961) suggested that though coronary arterial occlusion had occurred in these cases, myocardial infarction was not the inevitable sequel, especially if treatment was initiated at an early stage of the illness. However, Goble and O'Brien (1958) found fluctuating levels of serum aspartate aminotransferase in patients with acute coronary insufficiency when daily measurements of this enzyme were made over a period of 10 days, and they concluded that their patients were undergoing diffuse myocardial necrosis during that time. Although the serum aspartate aminotransferase and lactate dehydrogenase levels remained unchanged and within normal limits in the patients whom we studied, it is probable that those showing raised  $\gamma$ -glutamyl transpeptidase levels had had one or more episodes of myocardial necrosis sometime before admission to hospital. This view is supported by the findings in the patient who was known to have had a myocardial infarction within the two months before admission to hospital. All 4 patients giving a short history of angina had a raised serum  $\gamma$ -glutamyl transpeptidase level, and this suggests that there is a much higher incidence of myocardial necrosis in patients with this clinical presentation than has been previously recognized.

Goble and O'Brien stressed that serial determinations rather than a single measurement of serum aspartate aminotransferase should be made in the assessment of cases of acute coronary insufficiency. However, in view of the slow fall in the level of serum  $\gamma$ -glutamyl transpeptidase after an episode of myocardial necrosis, there may be a place for the measurement of this enzyme as a single determination in patients with this condition.

The raised level of  $\gamma$ -glutamyl transpeptidase present in the patient with marked congestive cardiac failure is explained by the presence of venous congestion of the liver and hepatic anoxia (Rutenburg *et al.*, 1963). However, the raised level of this enzyme found in the man with left ventricular failure is more difficult to understand, but in view of the characteristically slow fall of this enzyme to normal levels after myocardial infarction, it may represent the only remaining evidence that liver damage had occurred at the time when right-sided heart failure was noted 10 days before.

The considerably raised serum  $\gamma$ -glutamyl transpeptidase levels found in a case of acute non-specific pericarditis without a rise of the other serum enzyme levels deserves further investigation. In this patient there was no evidence of liver disease, and it may be that this enzyme was liberated from the epicardial region of the heart.

Thus it appears that this enzyme is raised in a

variety of cardiac conditions in which myocardial necrosis is a feature, and in particular this study suggests that it is possible to demonstrate a high incidence of myocardial necrosis in acute coronary insufficiency.

#### SUMMARY

The serum levels and serial changes of the enzyme  $\gamma$ -glutamyl transpeptidase have been studied in a group of patients with ischaemic heart disease. These changes have been compared with changes in serum aspartate aminotransferase (SGOT), alanine aminotransferase (SGPT), lactate dehydrogenase, and hydroxybutyrate dehydrogenase.

Twenty patients had an acute myocardial infarction and 16 showed a rise in the level of serum  $\gamma$ -glutamyl transpeptidase. This rise occurred between the third and eighth day after infarction, with a peak between the fourth and fourteenth day. These levels gradually fell during the following month, though 2 out of 10 patients still had raised levels two months after infarction.

Eleven patients had severe angina without evidence of recent myocardial infarction and 8 showed increases in serum  $\gamma$ -glutamyl transpeptidase above normal levels.

The diagnostic and pathological implications of these findings are discussed.

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#### REFERENCES

- Agostoni, A., Ideo, G., and Stabilini, R. (1965). Serum  $\gamma$ -glutamyl transpeptidase activity in myocardial infarction. *Brit. Heart J.*, **27**, 688.
- Chinsky, M., and Sherry, S. (1957). Serum transaminase as a diagnostic aid. *Arch. intern. Med.*, **99**, 556.
- Elliott, B. A., and Wilkinson, J. H. (1961). Serum " $\alpha$ -hydroxybutyric dehydrogenase" in myocardial infarction and in liver disease. *Lancet*, **1**, 698.
- Goble, A. J., and O'Brien, E. N. (1958). Acute myocardial ischaemia: significance of plasma-transaminase activity. *Lancet*, **2**, 873.
- Goldberg, J. A., Friedman, O. M., Pineda, E. P., Smith, E. E., Chatterji, R., Stein, E. H., and Rutenberg, A. M. (1960). The colorimetric determination of  $\gamma$ -glutamyl transpeptidase with a synthetic substrate. *Arch. Biochem.*, **91**, 61.
- , Pineda, E. P., Smith, E. E., Friedman, O. M., and Rutenberg, A. M. (1963). A method for the colorimetric determination of  $\gamma$ -glutamyl transpeptidase in human serum; enzymatic activity in health and disease. *Gastroenterology*, **44**, 127.
- International Union of Biochemistry (1961). *Report of the Commission on Enzymes*. Pergamon Press, London.

- LaDue, J. S., and Wróblewski, F. (1955). The significance of the serum glutamic oxalacetic transaminase activity following acute myocardial infarction. *Circulation*, **11**, 871.
- Master, A. M., Dack, S., Grishman, A., Field, L. E., and Horn, H. (1947). Acute coronary insufficiency: An entity. *J. Mt Sinai Hosp.*, **14**, 8.
- Reitman, S., and Frankel, S. (1957). A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Amer. J. clin. Path.*, **28**, 56.
- Richman, S. M., Delman, A. J., and Grob, D. (1961). Alterations of indices of liver function in congestive heart failure with particular reference to serum enzymes. *Amer. J. Med.*, **30**, 211.
- Rowell, N. R., and Smith, A. J. (1959). Multiple serial enzyme studies in acute myocardial infarction. *Brit. med. J.*, **2**, 459.
- Rutenburg, A. M., Goldberg, J. A., and Pineda, E. P. (1963). Serum  $\gamma$ -glutamyl transpeptidase activity in hepatobiliary pancreatic disease. *Gastroenterology*, **45**, 43.
- Wood, P. (1961). Acute and subacute coronary insufficiency. *Brit. med. J.*, **1**, 1779.
- Wróblewski, F. (1957). Clinical significance of serum enzyme alterations associated with myocardial infarction. *Amer. Heart J.*, **54**, 219.
- , and LaDue, J. S. (1955). Lactic dehydrogenase activity in blood. *Proc. Soc. exp. Biol. (N.Y.)*, **90**, 210.