C′ units, and with other immune systems, and these are being assembled for publication.

Summary. A quantitative method for the determination of complement fixation is briefly described and data are given for its application to one immune system.

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Histochemical Demonstration of Liver Glycogen in Human Diabetic Acidosis by Liver Biopsy.

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In spite of the extensive experimental and clinical literature which has been devoted to diabetes mellitus, few observations have been concerned with liver glycogen in human diabetes. These were based on autopsy material or on single specimens removed at operation. Two recently developed technics make the direct study of the liver glycogen content in the human feasible under clinical conditions. The 3-inch Silverman biopsy needle[1] makes it possible to obtain enough tissue for histologic examination; and Gomori's histochemical method for the demonstration of glycogen[2] appears to permit a quite accurate estimate of the glycogen content.

Method. A core of liver tissue is secured by the Silverman needle, at varying times during the treatment of patients in diabetic acidosis. Although needle biopsy is a reasonably safe procedure, it should be performed cautiously, and only after defects of the clotting mechanism have been ruled out. Moreover, the risk may be greater in unconscious patients.

The tissue, fixed for 24 hours in absolute alcohol, is stained by Gomori's method. Blood sugar levels are determined by the method of Folin and Wu,[3] and plasma CO_2 combining powers by the manometric Van Slyke technic.[4]

Results. Liver biopsies have been performed in diabetic patients with acidosis, and the histologic results correlated with the clinical and biochemical status of the patient. An example of the data so obtained is shown in Fig. 1. This patient was a 35-year-old Negro woman, not previously known to be diabetic, who had had polyuria and polydipsia for 4 months. During the 10 hours prior to admission she had been comatose. No evidence of infection was found. Fig. 1-A shows the liver glycogen prior to treatment. At this time, the blood sugar level was 358 mg/100 cc, the CO_2 combining power 27.8 vol % (11.6 mM/L) and there was 4+ glycosuria and 2+ acetonuria. Note that although the glycogen content is greatly diminished, a small amount is still present. Many large vacuoles, indicating fatty metamorphosis, are seen in the liver cells of the midzonal portions of the lobules.

Fig. 1-B shows the appearance of the liver 7½ hours later. By this time, the patient had received 300 units of regular insulin, 4000 ml saline and 300 g of glucose by vein. Her urine had been acetone-free for 2½ hours, but still showed 4+ test for sugar. The CO_2 combining power was 43.2 vol % (19.3 mM/L). Note the striking restoration of glycogen. Fat vacuoles are still numerous. A third biopsy (Fig. 1-C) was taken after 6 days, when the diabetes had been well controlled for 3 days. Note that there

1 Silverman, I., *Am. J. Surg.*, 1938, **40**, 671.

2 Gomori, G., *Am. J. Clin. Path.* (Tech. Sect.), 1946, **10**, 177.

3 Folin, O., and Wu, H., *J. Biol. Chem.*, 1920, **41**, 367.

4 Van Slyke, D. D., and Neill, J. M., *J. Biol. Chem.*, 1924, **61**, 523.

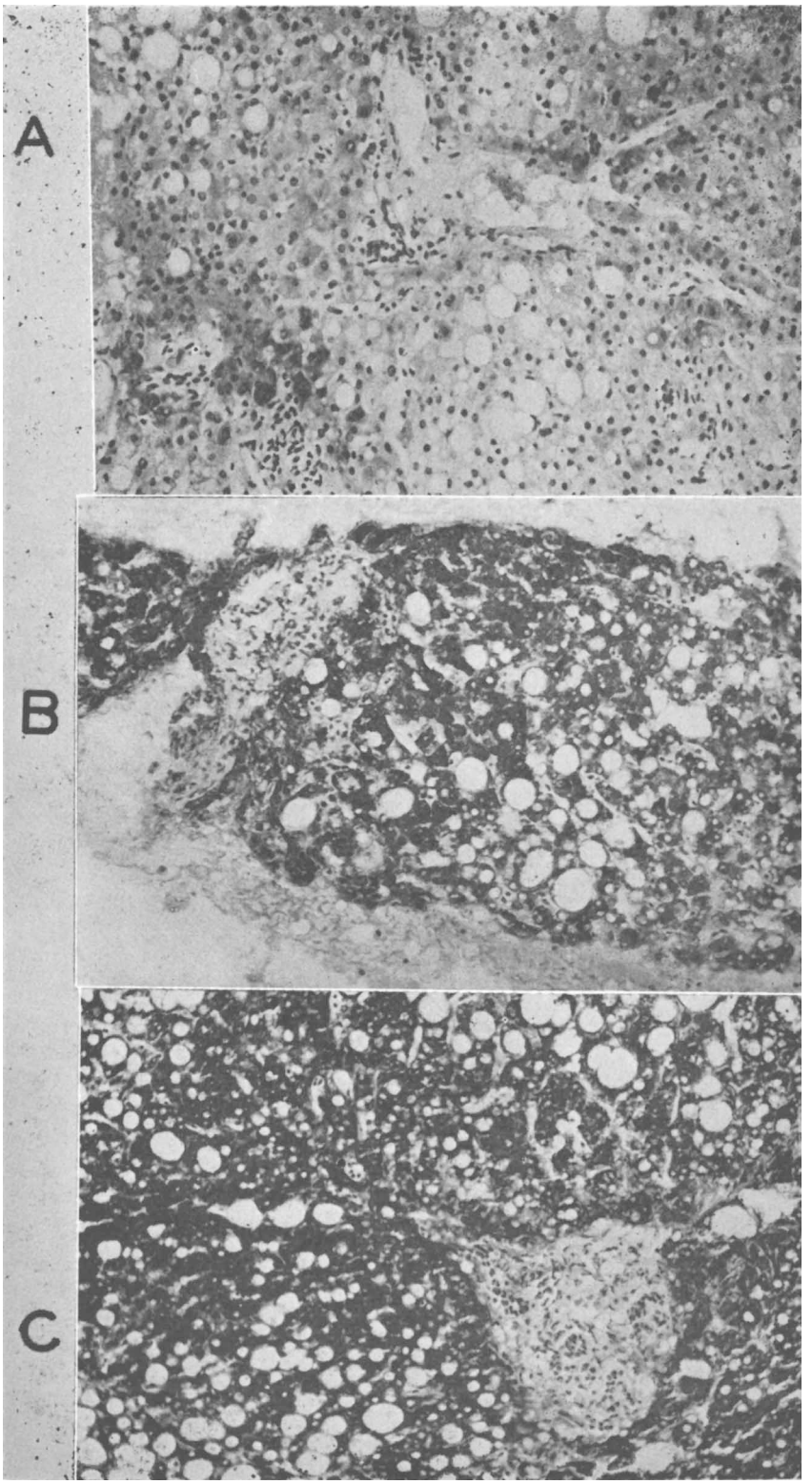

FIG. 1.

A. Glycogen depletion and fatty metamorphosis in liver during acidosis. B. Restoration of glycogen with persistent fatty metamorphosis after 7½ hours of treatment. C. Slight further increase in glycogen 6 days later. The black granules in the cytoplasm represent glycogen. Gomori's glycogen test, × 160.

is only a slight increase in glycogen content and no change in the degree of fatty metamorphosis.

Discussion. In order to evaluate these findings, knowledge of normal values and their fluctuations is essential. The only observations of the glycogen content of the liver in human beings have been obtained at operation, when the metabolism had been deranged by operative trauma, anesthesia, and by previous illness.[5] Autopsy material may not give an accurate picture because of the rapidity of postmortem glycogenolysis, and because even brief agonal states are known to cause drastic changes in the liver glycogen of animals. We are, therefore, accumulating a series of observations in normal individuals in the postabsorptive state, after meals, and after the administration of various drugs and stresses.

A quantitative correlation between histochemical and chemical methods for the determination of liver glycogen would be desirable. The study by Deane, Nesbett and Hastings[6] indicates that a good correlation can be obtained. To this end, we are performing simultaneous quantitative and histochemical determinations of glycogen in livers obtained from rats under various experimental conditions, as well as from human tissue obtained at autopsy. To date, it appears that a roughly quantitative correlation can be observed, and it is hoped that ultimately this can be done with a fair degree of accuracy.

Summary. Liver biopsy specimens obtained during diabetic acidosis and its treatment were examined by the Gomori technic for glycogen. Severe glycogen depletion was found before treatment. Restoration of glycogen content occurred after a few hours of therapy.

[5] MacIntyre, D. S., Pedersen, S., and Maddock, W. G., *Surgery*, 1941, **10**, 716.

[6] Deane, H. W., Nesbett, F. B., and Hastings, A. B., PROC. SOC. EXP. BIOL. AND MED., 1946, **63**, 401.

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Scleroma: Complement Fixation Test.

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From the time that rhinoscleroma was discovered in 1870 by von Hebra,[1] the disease has been the subject of a good deal of controversy. The name of the disease itself has been considered inadequate, and in 1932, at the Second International Congress Otorhinolaryngology[2] held in Madrid, it was changed to the more inclusive term, scleroma. The lack of definite information on the etiology of the disease has probably been the main source of confusion. Despite the inclusion of the supposed causative organism, *Klebsiella rhinoscleromatis*, in Bergey's Manual,[3] this bacillus is far from acceptable as the etiologic agent. Wilson and Miles[4] say, ". . . there is very little evidence that this organism is primarily responsible for it. There is no means by which it can be distinguished with certainty from other members of the capsulated group; and, since we know that members of this group may be present in the nose of healthy persons, it is difficult to prove

[1] von Hebra, R., *Wien. med. Wchnschr.*, 1870, **20**, 1.

[2] Rapp, Cong, *Madrid Internat. d'Oto-rhenolaryng.*, 1932.

[3] Bergey, D. H., Breed, R. S., Murray, E. G. D., and Hitchens, A. P., *Bergey's Manual of Determinative Bacteriology*, Baltimore, 1939.

[4] Wilson, G. S., and Miles, A. A., *Topley and Wilson's Principles of Bacteriology and Immunity*, 3rd Ed., Baltimore, 1946.