TECHNICAL METHODS

A SIMPLE VISUAL TURBIDIMETRIC ESTIMATION OF SERUM GAMMA GLOBULIN

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It was shown in our paper (page 260) comparing electrophoresis and precipitation methods that the γ -globulin fraction of serum could be determined by precipitation with a saline ammonium sulphate reagent giving results which were in good agreement with those obtained by electrophoresis. It was noticed in these precipitations that the turbidity produced by the addition of the reagent appeared to be directly proportional to the amount of γ -globulin, as determined by subsequent micro-Kjeldahl analysis. Since this might provide a simple method of estimating this fraction, it was decided to investigate further the correlation between serum γ -globulin and the turbidity produced.

Method

Serum, 0.1 ml., is added to 4.9 ml. of saline ammonium sulphate solution (prepared by dissolving 193 g. A.R. ammonium sulphate and 40 g. sodium chloride in distilled water and making up to 1 litre) in a test tube, which is inverted several times to mix, and allowed to stand for 15 minutes to develop maximum turbidity. The turbidity is then read visually against the turbidity standards of Kingsbury, as in the thymol turbidity test, the units of turbidity being calculated in the same way. Where the turbidity is high, the mixture is diluted by adding a further 5, 10, or 15 ml. of reagent and the reading obtained multiplied by the corresponding dilution factor. These proportions are slightly different from those used in the chemical estimation since, if the latter are used, the turbidity produced is outside the range of the standards.

Alternatively a fresh volume of 0.1 ml. of serum may be added to 9.9 ml., 14.9 ml., or 19.9 ml. of reagent, and the reading obtained multiplied by the appropriate dilution factor.

γ -Globulin was estimated chemically by the saline ammonium sulphate method.

Results.—The γ -globulin and the γ -globulin turbidity were determined in 70 sera, mostly from children under the age of 14 years, including normal children and others suffering from a variety of conditions. In Fig. 1 are shown the chemical and turbidity determinations plotted against each other. There is a linear relation, the results falling on or near a

line which cuts the axis at a point equivalent to 0.21 g. per 100 ml. From the results, the following formula was calculated:

γ -Globulin (g. per 100 ml.) = 0.08 U. + 0.21 where U = units of γ -globulin turbidity.

FIG. 1.—On this graph one very high result (80 units—6.72 g. per 100 ml.) has not been shown although it fell close to the line.

Discussion

Kunkel (1947) has described a similar method for the turbidimetric estimation of γ -globulin, although the precipitating reagent is of a different type. It is not claimed that the method described above is as accurate as the micro-Kjeldahl estimation of the globulin precipitate. It has the great merit of ease and simplicity, requiring only common reagents and apparatus. No special correction was needed for lipaemic sera, since lipaemia did not appear to alter the γ -globulin turbidity. The standard deviation of the two methods over the whole range of results is ± 0.13 g. per 100 ml., and the average percentage difference is 11.6. For results under 1.2 g. per 100 ml., the standard deviation is ± 0.11 g. per 100 ml. and the average percentage difference is 12.3, whereas for results above 1.2 g. per 100 ml., although the standard deviation is ± 0.19 g. per 100 ml., the average percentage difference is 9.0.

The method should be useful not only as a liver function test as Kunkel, Ahrens, and Eisenmenger (1948) have shown, but also in following up changes in γ -globulin in such conditions as nephritis and rheumatic fever. It is useful also as a method for the rapid survey of γ -globulin levels in large groups, and we are at the moment engaged in determining normal levels in infants and children.

Whilst this work was in progress, a similar method has been described by Popper and de la Huerga (1950), who, however, use a spectrophotometer or photoelectric colorimeter for measuring turbidities. We do not think this is necessary, since the accuracy of the method is not great enough to warrant it. If greater accuracy is required, it is necessary to carry out a micro-Kjeldahl analysis.

REFERENCES

- Kunkel, H. G. (1947). *Proc. Soc. exp. Biol., N.Y.*, **66**, 217.
— Ahrens, E. H., and Eisenmenger, W. J. (1948). *Gastroenterologia, Basel*, **11**, 499.
Popper, H., and de la Huerga, J. (1950). *J. Lab. clin. Med.*, **35**, 459.

Histopathologic Observations in a Fatal Case of Q Fever. PERRIN, T. L. (1949). *Arch. Path.*, **67**, 361.

In this paper are described the histological findings in a previously recorded case. A 43-year-old cattle-worker died after a 15-day illness characterized by fever, cough, nasal congestion, headaches, and mental confusion. Necropsy revealed a diffuse consolidation of lungs, microscopy showing an essentially mononuclear exudate, with congestion and haemorrhages, but neutrophil granulocytes were not infrequent and there was a fibrin network. No rickettsiae or other microorganisms could be found. Focal hypoplasia was seen in the sternal marrow. In the myocardium there was fairly extensive ischaemic fibrosis due to occlusion of a major coronary vessel. No significant change was found in other organs. It is concluded that the lung changes were essentially due to the Q-fever virus and possibly also the marrow hypoplasia; however, the cardiac condition certainly seems to have had an unfavourable influence in a disease usually benign in the healthy adult.

Diffuse Cystic Lungs of Granulomatous Origin. A Histological Study of Six Cases. CUNNINGHAM, G. J., and PARKINSON, T. (1950). *Thorax*, **5**, 43.

Detailed histological studies on 6 cases of honeycomb lungs are reported, and brief clinical records of the patients are given. All cases showed a granulomatous process in the walls of the cysts and in the intercystic spaces. In the acute phase this process was characterized by a highly cellular histiocytic response, and in the chronic phase by widespread fibrosis. In the intermediate stages, foamy macrophages were present in large numbers, sometimes being the predominant cell. It is suggested that the cystic spaces are produced by weakening of the walls of the smaller bronchioles as a result of granulomatous infiltration. In the present material the distinction between cysts of alveolar

and bronchiolar origin is impossible. The nature of the granulomatous process is discussed. [Authors' summary.]

Diagnostic Value of Histologic Lesions of Striated Muscle in Rheumatoid Arthritis.

SOKOLOFF, L., WILENS, S. L., BUNIM, J. J., and MCEWEN, C. (1950). *Amer. J. med. Sci.*, **219**, 174.

Biopsy or necropsy specimens or muscle from the deltoid and gastrocnemius were examined by serial section in 57 cases of rheumatoid arthritis, 10 of ankylosing spondylitis, 21 of acute rheumatism, 101 of various other conditions, some involving the joints, and in 13 healthy volunteers. Focal cellular lesions similar to those described by other workers were found in 56% of the cases of rheumatoid arthritis, but also in 3 of the normal controls and in 25% of the non-rheumatoid group as a whole. Various other laboratory investigations were performed, such as determination of the erythrocyte sedimentation rate and estimation of the serum content of streptococcal agglutinins and agglutin of sensitized sheep erythrocytes. None of these threw any light on the mechanism of production of the histological lesions, and the authors conclude that muscle biopsy is of little value as a diagnostic procedure in rheumatoid arthritis.

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Correction

Dr. LEVIN writes: I must apologize for an error, which was in our typescript and was no fault of the printers.

In the paragraph headed "Method" of "A Simple Visual Turbidimetric Estimation of Serum Gamma Globulin" (*J. clin. Path.*, **3**, 284) the first line should read: "Serum, 0.1 ml., is added to 2.4 ml. of saline ammonium sulphate solution" . . . instead of "Serum, 0.1 ml., is added to 4.9 ml. of saline ammonium sulphate solution."