The Association of Clinical Pathologists: 83rd General Meeting

The 83rd general meeting of the Association of Clinical Pathologists was held at Imperial College, London, on 25, 26, and 27 September 1969. As well as several simultaneous sessions, when free communications were presented, there were also two half-day symposia and a slide seminar. The first symposium was on 'Drug addiction and the clinical pathologist' (Chairman: Professor W. D. M. Paton), and the second on 'Iron' at which Dr J. Fielding took the chair. Professor C. V. Harrison was the Chairman at the slide seminar on 'Unusual granulomata'. The last day of the meeting was devoted to a joint ACB/ACP meeting which took the form of a symposium on the 'Biochemical consequences of the pill' to which speakers were invited. This symposium will be published in full in 1970. The Presidential address, 'Research and development', was delivered by Professor N. H. Martin and is published virtually in full on pages 628-33 of this issue. Abstracts of many of the free communications follow.

OBSERVATIONS ON THE VITAMIN B₁₂ BINDING POWER OF SERUM

A. L. NEEDHAM AND H. G. H. RICHARDS Using a radioactive vitamin B₁₂ (Co cyanocobalamin) the authors have shown that the unsaturated B₁₂-binding capacity (UBBC) of serum tends to increase with the amount of B₁₂ offered to the serum protein. To obtain a realistic value for UBBC the authors have devised the following technique:—

A Three B₁₂ 'donor' solutions were prepared in 1 ml quantities to contain (1) 40 pg B₁₂, (2) 65 pg B₁₂, and (3) 115 pg B₁₂. Of these amounts 15 pg was radioactive B₁₂ in each case.

B Binding of 'donor' B₁₂ to serum protein was effected by adding 20 µl of serum to 1 ml aliquots of each of the three donor solutions above in duplicate and allowing to stand for 30 minutes at 4°C.

C Free B₁₂ was removed by adding 0.1 ml of albumin-coated charcoal to each tube, mixing and rotating for 30 minutes at 4°C, then centrifuging. The supernatant contained the bound, and the charcoal the free B₁₂. The 'free' and 'bound' portions were counted separately in a gamma scintillation counter, recording the 'free' and 'bound' counts which, summated, gave the total counts.

The results were plotted on graphs in which the abscissa recorded the ratio total counts : bound counts, while the ordinate recorded the corresponding amount of B₁₂ added to the serum (in picograms). The three recorded points for each test form a slope which is the hypotenuse of a right-angled triangle. The values for y (the vertical side) and x (the horizontal side) are determined, and the UBBC calculated from the formula

$$\text{UBBC} = \frac{y}{x}$$

In 21 normal subjects the UBBC ranged between 1,080 and 1,650 pg/ml. In three groups of conditions (neoplastic states, infective conditions, and leukaemias), the UBBC

generally increased with the leucocyte count; values up to 9,000 pg/ml were obtained.

UNDERESTIMATION OF VITAMIN B₁₂ IN SERUM BY A MICROBIOLOGICAL METHOD (*Lactobacillus leichmannii*): COMPARISON WITH A RADIOISOTOPIC METHOD

J. L. RAVEN, M. B. ROBSON, AND P. BARKHAN (*Department of Haematology, Guy's Hospital, London*) A radioisotopic method, using intrinsic factor, ⁵⁷CoB₁₂ and coated charcoal, may give higher vitamin B₁₂ values for some sera, especially post-gastrectomy sera, than a standard microbiological method (*Lactobacillus leichmannii*) (Raven, Robson, Walker, and Barkhan, 1969). Further studies have shown that one of the explanations for this discrepancy is the use of different extraction procedures to separate vitamin B₁₂ from its binding proteins in serum, the radioisotopic method employing autoclaving at pH 2 and the microbiological method deproteinization at pH 5-5.6. Appreciable amounts of vitamin B₁₂ (as measured by the radioisotopic method) have been found in the protein precipitate which is normally discarded during the extraction procedure of the microbiological method. Furthermore, when the radioisotopic method is modified so as to use the same extraction procedure as the microbiological method, the serum vitamin B₁₂ values are then lower and there is a much better correlation between the results obtained by the two methods. It has also been possible to modify the extraction procedure of the microbiological method so as to obtain higher serum vitamin B₁₂ values by this method. This modification involves the incubation of diluted serum with papain, before the stage of autoclaving and protein precipitation.

REFERENCE

Raven, J. L., Robson, M. B., Walker, P. L., and Barkhan, P. (1969). *J. clin. Path.*, **22**, 205.