all the typical clinical features, including pigmentation of the mucous membrane of the mouth and lips, while the children, in order of age, demonstrate the same phenomena in decreasing severity and distinctness. The moles, placed so prominently in the list of important clinical features by Addison, Greenhowe, and others, are specially characteristic of this family group. If less significance is attached to them, then the correctness of our diagnosis is much more open to question.

The cases were shown at a meeting of the Medico-Chirurgical Society of Edinburgh on February 1st, 1900.

Why should it not be conceivable that a tuberculous lesion so often hereditary might not exist in the same family in every child? But why is it that in so many recorded cases of Addison's disease, amounting now to several hundreds, so seldom is any reference made to plurality of cases in one family?

The future will clear up the mystery as far as this family is concerned, and the record is published in the hope that by directing attention to investigation into family history, more carefully than has been done in the past, similar instances may be met with.

PRELIMINARY NOTE ON A NEW QUANTITATIVE METHOD FOR SERUM DIAGNOSIS.

BY

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For the successful application of the agglutination test ("Widal" method) in the diagnosis of disease, it is universally admitted that some dilution of the blood is desirable, a dilution which should be approximately known, and which varies in practice between 1 in 10 and 1 in 100, while for the investigation and comparison of the specific agglutinating power of the blood in different cases or diseases, and under varying conditions, an accurate quantitative method for carrying out the dilutions is a necessity.

Of the various methods employed for diluting, the following may be mentioned:

1. Dilution by means of a platinum loop (McWeeney, Delépine, and others). This at the best can be but approximate.
2. Dilution by means of a capillary pipette (Wright). This is more suitable for serum than for the whole blood.
3. The dry-blood method (Johnson). This is subject to many inaccuracies: for example, variability in the amount of solids in the blood and in the dryness of the film, and the difficulty of weighing out so small a quantity as 1 milligram.
4. The hæmoglobinometric method, the blood being diluted to a given standard colorimetrically by its contained hæmoglobin, a very variable factor.
5. The capillary-tube method (McFarland). Independently of McFarland, we carried out experiments with the capillary-tube method, and early abandoned it, for we found it impossible to obtain capillary tubes of even approximately the same internal diameter, while accurate calibration by capillarity is attended with great difficulties.

We do not mean to imply that the above methods are useless, all of them give satisfactory results for rapid clinical diagnosis or in those cases where an approximation suffices, but we believe that the capillary pipette method (No. 2 above) is the only one which has any claims to accuracy. We have, therefore, devised the following quantitative method, which is suitable not only for serum but also for the whole blood, and is capable of the greatest accuracy.

Ordinary vaccine tubes are taken, the large, the very small, and any irregular ones being discarded. We prefer those varying in diameter from about 0.9-1.2 mm., they are boiled in strong nitric acid, thoroughly rinsed in tap, and then in distilled water and dried; a large number can thus be treated at one operation. The selected tubes are charged with the blood to be examined, being preferably one-half to two-thirds filled, and are carefully sealed by melting their ends in a flame in the ordinary way. In the laboratory the length of the column of blood between the bases of the two menisci is first measured by means of a scale graduated to half millimetres assisted by a lens mounted to avoid parallax. The end of the tube is then cut off by means of a fine file or writing diamond, and its internal diameter is measured. This, the most important of all the manipulations, is carried out in the following manner:—

The tube is supported in the axis of the microscope by means of two iris diaphragms carried respectively by the stage and by the substage. These close concentrically and support the tube, at the same time cutting off all light reflected from the mirror. The opaque blood in the interior of the tube in the same way cuts off all light that might pass through the lumen of the tube. The only light that thus reaches the optical system is that transmitted by the glass walls of the tube. In the field of vision the lumen of the tube now appears as a black circle surrounded by a brilliantly-illuminated annulus. Owing to the extreme sharpness of the image the measurement of the diameter of the tube can be carried out with great accuracy by means of an eye-piece micrometer arranged to give readings to the one-thousandth of a millimetre. From the formula

$$V = D^2 \times L \times 0.7854,$$

Where V = volume of the cylinder,

D = diameter of the cylinder,

And L = the length of the cylinder,

the volume of the blood is calculated. If many measurements have to be made, a Fuller's slide-rule greatly expedites the calculations. It is of the utmost importance that the diameter of the tube be accurately measured, and therefore the adjustment of the eye-piece micrometer must be carried out with great care, and the scale by which it is set should be one the accuracy of which is undoubted. For, considering the formula ($V = D^2 \times L \times 0.7854$), it will be seen that any error in the measurement of D will be increased in a geometrical ratio in the final result, hence the comparative inefficiency of a capillary-tube method in which D is measured indirectly and with less accuracy.

We have controlled the accuracy of the method by filling tubes with distilled water, ascertaining the amount of water by direct weighing, and afterwards calculating the weight of the water by the formula given above; it is easy to obtain results which are within 0.5 per cent. of the true weight. Having ascertained the volume of blood, it remains to carry out the dilutions; this is accomplished as follows:

The capillary tube is placed in a vertical thick-walled glass tube of slightly larger bore, the lower end of which is submerged in the required amount of the diluting medium (measured out by a graduated pipette) contained in a hollowed-out block of glass, such as is used in histological and microscopical work. The capillary tube with its contained blood is then crushed *en masse* by means of a soft iron plunger, and the interior of the containing tube is washed out several times by drawing up the diluting medium into its lumen, either with the mouth or by means of an india-rubber teat. The powdered glass quickly falls to the bottom, and the mixture may be employed for conducting the serum reaction either by the microscopic or by the macroscopic method.

On a future occasion we propose giving further data and details, but we may say in conclusion that this method has been tested at the Jenner Institute of Preventive Medicine, and has proved to be easily carried out, and to give satisfactory results, while the time consumed in the whole of the manipulations (5 to 10 minutes) is comparatively short.

ON THE VALUE OF THE AGGLUTINATION TEST AS A MEANS OF DIAGNOSIS OF THE B. TYPHOSUS FROM COLIFORM ORGANISMS.

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THE difficulties which surround the correct interpretation of the agglutination test are well shown by the following bacteriological examination of a drinking water derived from a polluted upland surface. The water was sent for analysis as there had been several cases of enteric fever amongst the consumers, though no real epidemic, such as might be attributed to a polluted water supply, had occurred. On arrival at the laboratory the water was found to contain a large number of micro-organisms, so the usual plan of concentrating the sample by pumping one or two litres through a Berkefeld bougie had to be abandoned. Consequently 1 c.cm. of the water was added to each of twelve broth tubes containing 0.05 per cent. of carbolic acid; all the tubes were then incubated at 37° C. At the end of twenty-four hours some of the tubes were hazy in the upper half and some in the lower half, and three tubes were turbid throughout. These three tubes were then plated out in gelatine containing 0.05 per cent. carbolic acid. After seventy-two hours' incubation at 22° C. the plates made from two of the tubes showed almost pure cultures of coliform colonies, whilst the plates made from the third tube showed numerous colonies of *B. megaterium*. Nearly all the coliform colonies were fished and their cultural characteristics studied. It was then found that the coliform organisms could be arranged in three groups.