TECHNICAL METHODS

Comment on "The Estimation of Magnesium in Serum Using Titan Yellow"*

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The modifications suggested by Neill and Neely (1956) of a method for the estimation of magnesium in serum seem unnecessarily to complicate a simple analytical procedure which should be more widely employed. In this laboratory it has been found that the method of Orange and Rhein (1951), slightly modifed, is suitable for general use. Orange and Rhein (1951) recommend that polyvinyl alcohol be used instead of gum ghatti solution as a dispersing agent. Both polyvinyl alcohol and gum ghatti are suitable for stabilizing the magnesium-dye lake, but, as they point out, polyvinyl alcohol is a pure chemical which is easily dissolved and which accentuates the final colour, increasing the sensitivity by about 15%. If the micro-method of Orange and Rhein is used with double quantities of all reagents, beginning with 0.2 ml. of serum, a final volume of 10 ml. is obtained. This can be read at 560 mµ in the "unicam" SP 600 spectrophotometer in cells with a 40 mm. light path if they are raised slightly in the carrier by small blocks.

In their method Neill and Neely (1956) read the blank and the unknown separately distilled water and against utilize the difference of these results. We presume that the authors used this procedure because the colour obtained from the unknown when read against the blank (which is always high) was not sufficient to give accurate readings by their method. Using our modification of the Orange and Rhein method, the better procedure of reading the unknown directly against the blank gave results that were accurate and reproducible.

Neill and Neely find, in their experiments, that calcium ions intensify the colour of the magnesium-dye lake, and therefore suggest that if calcium is omitted from the standards an over-estimation of the serum magnesium concentration results. They therefore add 50 μ g. of calcium to each standard (which contain respectively 5, 10, 15, 20, and 25 μ g. of magnesium). Since the calcium concentration of serum is approximately five times greater than the magnesium concentration, by this procedure only in the tube containing 10 μ g. of magnesium is the calcium/magnesium ratio the same as in serum.

The Orange and Rhein method has been tested for calcium interference in two ways: (1) $50 \mu g$. of calcium was added to each magnesium standard (as in the Neill and Neely procedure); (2) calcium was added to the stock magnesium standard to make the calcium/magnesium ratio the same as in serum: accordingly in all the dilute standards the calcium/magnesium ratio stayed at 5:1. Fig. 1 shows the results obtained. It may be concluded that if serum magnesium is estimated by the Orange and Rhein method, calcium is without effect on the colour of the final solution. This is in agreement with the original findings of Orange and Rhein (1951).

References

Neill, D. W., and Neely, R. A. (1956). Journal of Clinical Pathology, 9, 162. Orange, M., and Rhein, H. C. (1951). J. biol. Chem., 189, 379.

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FIG. 1.—Effect of calcium ions on the determination of magnesium. Magnesium determined (a) in the absence of calcium (●); (b) with a constant quantity of calcium (Neill and Neely procedure) (■); (c) with a constant calcium/magnesium ratio (○).