TECHNICAL METHODS

A Technique of Blood pH Estimation

E. G. HARDY

From the Department of Surgery, University of Durham

(RECEIVED FOR PUBLICATION JUNE 23, 1959)

The blood pH can be measured electrometrically by means of a glass electrode and calomel reference electrode provided (a) the blood is kept from contact with air and (b) the measurement is made at 38° C. (Wynn and Ludbrook, 1957). Alternatively to (b), but less satisfactorily, the pH can be estimated at an arbitrary temperature (say 20° C.) and a correcting factor employed.

To satisfy the above requirements a closed circuit of small total volume may be used. The apparatus shown here has the advantage that it can be assembled from standard equipment in any department which has access to adequate glass-blowing facilities. The method can be used for clinical determination of blood pH; it is well suited also for performing

repeated estimations in small animals, such as the rabbit, without depleting the circulation, since each sample can be re-injected after use.

An exploded sketch is shown in Fig. 1 and a photograph of the assembled apparatus in Fig. 2. A 1 mm. bore glass capillary tube is bent to a flattened U so that it can be suspended by means of a suitable holder in a water-bath at the desired constant temperature. Two small chambers with lateral ports are made, one for the glass electrode and the other for the electrolyte junction to the calomel half-cell. To avoid expensive ground-glass joints these components are fitted into their respective ports by means of sleeves cut from P.V.C. tubing. A commercially available miniature glass electrode of standard pattern is used.

One difficulty was to devise an electrolyte junction which could be made easily and cheaply and which would stand the pressures caused by re-injection of blood against arterial resistance. These requirements are met by packing the lumen of a glass tube very tightly with chemically clean glass-blower's (asbestos) wool impregnated with agar gel embodying 3.5 M KCl.

FIG. 1.—Sketch of the apparatus. A=glass electrode. B=KCl bridge. C=asbestos fibre/agar KCl junction. D=plastic sleeves. E=1 mm. bore glass capillary tube. F=1 mm. bore polyethylene tubing. G=manifold made of two three-way adaptors. H=heparinized saline.

FIG. 2.—A = blood pH assembly. B = "perspex" holder.

Tight packing is achieved by ramming the asbestos very firmly against a constriction made at a suitable level in the tube. The agar gel minimizes the packing being soaked with blood. Several of these junctions are kept in a jar of 3.5 M KCl, and the one in use is changed for each series of estimations; after use it is washed with distilled water and returned to the jar of KCl. Electrolytic continuity with the calomel electrode assembly is by a P.V.C. tube containing 3.5 M KCl.

Method of Use

Polyethylene tubes of 1 mm. bore are attached to each end of the apparatus and the system is filled with heparinized saline. For use in the experimental animal the femoral artery is cannulated with similar tubing, which can be joined to one tube of the

apparatus by an appropriately sized hollow needle with the butt removed. The other tube of the apparatus is connected by way of a needle to a manifold constructed of two three-way adaptors (see Fig. 1); two 10 ml. syringes, one containing heparinized saline, are attached. By appropriate manipulations the saline filling the system can be drawn into the first syringe, followed by arterial blood into the second syringe. Suitable tilting of the apparatus helps these manoeuvres and allows the chambers to be completely and uniformly filled with fresh arterial blood. By reversing the actions the blood can be re-injected and the apparatus left full of heparinized saline. With repeated estimations, of course, there is some loss of blood into the first syringe, and some saline enters the circulation. Since the total volume of the system (including the polyethylene tubing) is only 1.5 ml., however, this interchange is not large. The apparatus can be disconnected from the animal and washed out at any stage of the experiment.

Precautions and Results

With certain precautions, consistent reproducible results have been obtained within the limits of performance of the pH meter used.

Buffers used to standardize the meter must be washed out of the apparatus with great thoroughness. Air bubbles must be trapped or evacuated. During the actual reading the arterial cannula should be clamped and disconnected temporarily from the apparatus. If this is not done interference may occur, even when the animal is earthed. Before each reading, the sample should be allowed to stand in the apparatus for about five minutes, to permit equilibration of the temperature of the blood with that of the water-bath. In practice this can be checked by ensuring that the reading has reached a sustained stable level.

For clinical estimation of blood pH, the sample is taken in a lubricated heparinized syringe which is then sealed. The estimation is made very simply by filling the empty apparatus slowly from the syringe, tilting it during filling so as to avoid trapping air in the chambers.

REFERENCE

Wynn, V., and Ludbrook, J. (1957). *Lancet*, 1, 1068.