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THE MEASUREMENT OF BLOOD GAS TENSIONS

Measurement of Blood Oxygen Tension

By J. M. BISHOP, M.D., M.R.C.P.

Birmingham

THE measurement of the blood oxygen tension is not as simple as the measurement of oxygen content but improved techniques are now available and one such method will be described after a review of earlier methods.

The measurement of blood oxygen tension was first achieved by methods based upon the aerotonometer, in which a small bubble of gas is equilibrated with a large volume of blood, and the gas bubble subsequently analysed for oxygen. Krogh (1908) designed a micro-aerotonometer in which blood flowed from a cannulated artery around a bubble which was subsequently drawn into a capillary tube and analysed for oxygen. Barcroft and Nagahashi (1921) devised a method which was similar in principle but, being suitable for analysis of a blood sample contained in a syringe, it was the first method applicable to human experiments. Twenty years later Comroe and Dripps (1944) published a modification of this method and established its accuracy. Then Riley *et al.* (1945) described their use of the Roughton-Scholander syringe for the equilibration of blood with a bubble of gas and for its subsequent analysis. The essential advance was that the whole procedure was carried out in the same syringe. This method has proved of great value in physiological studies but has some disadvantages. It is not for occasional use and requires constant practice, considerable skill and perfect attention to detail. An experimenter may attain repeatability of results and yet show systematic errors, so that it is essential that each individual should assess his own personal accuracy by means of a series of tonometer experiments. The method is most accurate in the range of pO_2 between 40–100 mm Hg and is unsuitable for values above 100 mm Hg, which for many purposes is a serious limitation.

The methods using the principle of the aerotonometer, especially the earlier ones, were not easy to use and in consequence many workers at this time resorted to the highly inaccurate indirect method in which oxygen saturation was measured and the oxygen tension read from a standard oxygen dissociation curve —an inaccurate procedure in the range of values found in arterial blood, because of the shape of the dissociation curve.

When the oxygen tension exceeds 100 mm Hg, it cannot be estimated by any of these methods and it has been measured by determining the oxygen content of anaerobically separated plasma by the manometric method of Van Slyke, or one of several modifications designed to give greater accuracy (Berggren, 1942; Fasciolo and Chiodi, 1946). From a knowledge of the Bunsen solubility coefficient the oxygen tension may then be calculated. With the modifications described, oxygen content can be estimated to about 2% of the measured value, but the method is time-consuming and large errors are introduced by small degrees of hæmolysis.

None of these methods was entirely satisfactory and attention has been directed to a polarographic method, based on the principle evolved by Heyrovsky (1941) using a dropping mercury electrode. The method was first used for blood oxygen tension measurements by Baumberger (1938), and was further developed by Berggren (1942). In essence the method consists of measuring the current flowing in an electrolytic cell to which a potential has been applied, the current being proportional to the quantity of oxygen reduced at the cathode. As the applied potential becomes more negative, the current increases until a plateau is reached at which the current becomes limited by the rate of diffusion of oxygen to the cathode. The chief disadvantage of this type of electrode is that it is unsuitable

for whole blood, measurements having to be made on anaerobically separated plasma. Changes in drop size and variations in other constituents of the plasma result in unpredictable variations in readings, although Bartels (1951) by the use of the potentiometric method overcame some of these objections.

Similar observations were subsequently made using stationary electrodes, usually of platinum. The principles underlying this method are similar and were studied by Kolthoff and his colleagues (Kolthoff and Lingane, 1946). The method was first applied in physiological measurements by Davies and Brink (1942). When blood is in contact with a bare platinum electrode, the rate of reduction of oxygen, and hence the current, changes in time in a manner not fully understood, although it appears to be due largely to the presence of red cells in contact with the electrode. The rate of reduction of oxygen at the platinum surface is then determined by the rate at which oxygen is liberated from combination with hæmoglobin molecules within the red cells in contact with the platinum and the electrode is also sensitive to changes in pH and carbon dioxide tension. Attempts to minimize these effects have followed three principles. First, the electrode may be covered in order to separate it from the blood. Secondly, the potential is applied discontinuously to the cathode and for a short time so that significant oxygen tension gradients within the blood do not develop. Finally the blood may be stirred or otherwise kept in motion relative to the electrode. This also reduces the oxygen tension gradients which develop within the blood as in the case of the dropping mercury electrode. Several workers have combined two or more of these principles in their attempts to measure whole blood oxygen tension. Davies and Brink (1942) used a recessed platinum electrode, the recess being filled with agar, and observed the current ten seconds after applying the potential. They also reported briefly on a cellophane-covered electrode. Olson *et al.* (1949), using cell suspensions but not blood, tried to overcome these difficulties by applying the potential as short pulses and recording the current transients. Morgan and Nahas (1950) used a rotating platinum electrode covered with a film of silicone but failed to eliminate sensitivity to changes in carbon dioxide tension and other unexplained variations. Drenckhahn (1956) used an electrode covered with collodion and reported that it was not influenced by changes of carbon dioxide tension within the physiological range. Clark *et al.* (1953) covered the platinum electrode with a cellophane membrane and reported encouraging preliminary results. Mochizuki and Bartels (1955) used the switched potential method and an uncovered electrode consisting of many platinum points and the blood was not stirred. Oxygen tensions up to 400 mm Hg could be measured but the instrument was still sensitive to changes in hæmoglobin concentration and to some extent to changes in carbon dioxide tension.

The proposal by Clark (1956) that the electrode and reference anode should be incorporated in a cell and isolated electrically from the blood by a membrane permeable to oxygen but not permeable to water or ions was a major advance. It made possible the construction of an instrument which is stable over long periods of time and uninfluenced by other constituents of the blood. Such an instrument is also capable of measuring the oxygen tension of a gas mixture as well as in a liquid phase. Methods for measuring blood oxygen tension using such an electrode have been described by Sproule *et al.* (1957), Kreuzer *et al.* (1958), Severinghaus and Bradley (1958), and by Polgar and Forster (1959). These methods have differed in the manner in which the instrument has been calibrated and the way in which blood has been brought into contact with the electrode.

During the past two years, a convenient and reliable apparatus has been developed and we have reported briefly upon an earlier form of the instrument (Bishop and Pincock, 1958). There are several variables which must be taken into account and these will be briefly discussed before describing the apparatus.

(*a*) *Temperature sensitivity.*—The current output of the electrode rises by approximately 5% per degree centigrade rise in temperature. This effect is due in about equal proportions to the effect of temperature on the platinum cathode and to the effect on the diffusivity of the membrane. Accurate temperature regulation of the electrode system is therefore essential.

(*b*) *Pressure sensitivity.*—The output of the electrode is very sensitive to negative pressure transients, presumably due to displacement of the membrane. Sustained pressure alterations produce less effect but are important under some conditions. These effects can be minimized by correct design of the electrode tip and by firm application of the membrane.

(*c*) *Flow sensitivity.*—When blood or any other liquid at a given oxygen tension is in contact with the membrane of the electrode, the output of the electrode is less than if a gas mixture with the same oxygen tension were substituted. This is due to the fact that the electrode consumes a small quantity of oxygen and oxygen has there-

fore to diffuse through the liquid to reach the membrane, the oxygen tension at the membrane surface being below that in the body of the liquid. The magnitude of this difference can be reduced in two ways, the first of which is the use of a membrane of relatively low diffusibility. The second way is to stir the liquid, when it is found that with increasing rates of movement the difference between gas and liquid readings diminishes, but does not disappear altogether because a boundary layer of stationary fluid always remains. Furthermore, when water and blood of the same oxygen tension are compared, the greater viscosity of blood results in a thicker boundary layer and hence the electrode output is always lower than for water, although the difference is proportionately constant. This is important when calibrating because a correction must be applied if water is used for this purpose.

(*d*) *Response time.*—This is very largely dependent on the diffusibility of the membrane which may be varied by changing the thickness or the material used. Although very small cathodes have been reported to give a fast response time, this has not been related to platinum area within the range studied.

Thus several features in the design require a compromise. The apparatus illustrated in Figs. 1 and 2 has been evolved through a series of modifications. Details of the construction show the electrode held in a stainless steel block, a recess in which, covered by a glass plate, forms the chamber, 1 ml in volume, into which blood is introduced. Water circulates through the Perspex water jacket from a temperature-regulated water bath, which is maintained at 38°±0·1° C. The electrode tip has a taper which permits the membrane to be applied firmly and held in place by a rubber O-ring. The membrane is of polyethylene 0·125 mm thick. The blood is stirred magnetically at a speed of 2,000 rev/min by a glass-coated soft iron slug, the dimensions of which are such that the clearance between it

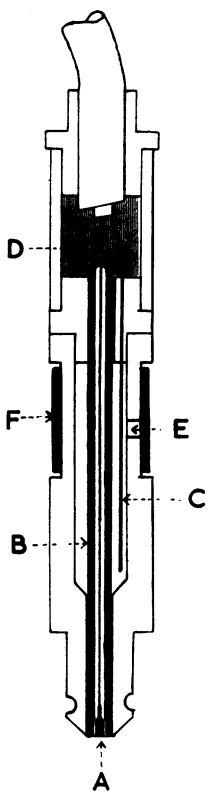

FIG. 1.—Details of electrode construction. The body of the instrument is made of Perspex. A, platinum cathode fused in glass (B). C, silver-silver chloride anode. D, epoxy resin cast around electrode connections to cable. E, hole for introduction of electrolyte and covered by rubber diaphragm (F).

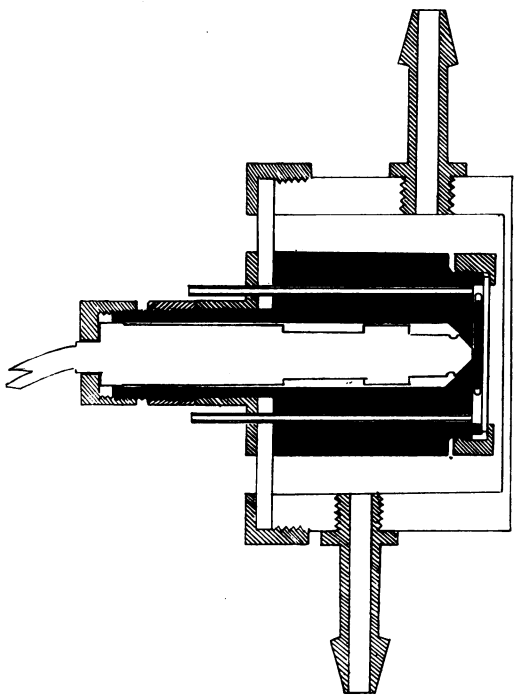

FIG. 2.—Section through the assembled instrument. The electrode is in place in the stainless steel block (solid black) which is surrounded by a water jacket. The recessed measuring chamber is shown with its glass cover and contained stirring slug.

and the membrane is 0·05 mm. The voltage-current relationship for such an electrode shows a plateau between −0·4 and −0·7 volt. In use, a potential of −0·6 volt is applied to the electrode and the current resulting flows through a variable series resistance. The potential drop across this resistance is measured by means of an electrometer (E.I.L. 33B Vibron electrometer) and recorded by an Elliott recorder. The time response of the electrode is not exponential, there being a prolonged "tail". In practice the response is complete in four minutes, this also allowing for the blood to assume the temperature of the water jacket. Calibration is by means of water equilibrated with gas mixtures at two levels of oxygen tension by bubbling in suitable vessels held in the water bath. Water is transferred to the electrode chamber by siphonage through nylon tubes.

The current output is linearly related to oxygen pressure from zero to one atmosphere. To assess the overall accuracy of the method specimens of blood and of water have been prepared in a double tonometer rotating in a water bath from which the water perfusing the water jacket of the apparatus was drawn. The blood was equilibrated by continuously passing a gas mixture of known composition through the tonometer for 25 minutes. Both blood and water were in turn siphoned into the chamber and the oxygen tension estimated in duplicate in the usual manner. Thirty-seven pairs of estimations were made and in the case of water the points were distributed about the line of identity. In the case of blood this was not so, the estimated value always falling below the tonometer value. The relationship between the tonometer and estimated values was constant and a linear regression line was calculated. The precision of the method was satisfactory and in the range 0–150 mm Hg the standard deviation about the regression line was 2·1 mm Hg. Some of this error is related to the preparation of the blood in the tonometer and its transfer to the measuring instrument, so that the error of the measurement has probably been overestimated.

By the use of this instrument we have a versatile method which is suitable for the measurement of the oxygen tension in blood, and incidentally in other fluids, and also in gas mixtures. It is accurate and will measure oxygen tension from zero to one atmosphere of oxygen. It is simple to use and gives results quickly; ten samples can be analysed in one hour. The instrument has been in use in physiological studies during the past year and continues to work satisfactorily.

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Measurement of Blood Carbon Dioxide Tension

By J. F. NUNN, Ph.D., F.F.A. R.C.S.

London

IN the gas phase, tension is synonymous with partial pressure, which is defined by Dalton's law as the pressure a gas would exert if it occupied the volume alone. The tension of a gas in a liquid is a concept more familiar to the biologist than the physicist. Krogh (1908) has defined it as the partial pressure of the gas in an atmosphere which is in diffusion equilibrium with the liquid. This is, in fact, the basis of many of the methods of measurement of carbon dioxide tension (pCO_2) of blood.

There are at least three reasons for measuring carbon dioxide tension in preference to content. Firstly, the tissues of the body are sensitive to