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## Calibration and Quality Control of Equipment Used for Measuring O<sub>2</sub> Concentration

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### *Introduction*

Materials employed in the calibration and quality control of equipment used for measuring O<sub>2</sub> concentration must have a defined O<sub>2</sub> concentration and should be as easy to handle as possible. In addition, the main steps in the analysis should also be carried out with the test material. The procedures most widely used today are:

#### *Van Slyke Procedure*

Extraction of O<sub>2</sub> from the blood with a coarse vacuum, chemical binding of O<sub>2</sub>, manometric or volumetric determination.

#### *Galvanic Cells*

Elution of O<sub>2</sub> from the blood using a carrier gas, physical measurement with a galvanic cell.

#### *O<sub>2</sub> Cuvette*

Chemical reaction of O<sub>2</sub> with an indicator, photometry.

The main materials available for calibration are air, equilibrated distilled water or blood, and a potassium iodate solution (KIO<sub>3</sub>).

*Materials for Calibration*

The materials described here are shown in table 1.

In many cases, air is especially suitable for the calibration of equipment, since it is always available and has a defined O<sub>2</sub> concentration (20.95 ml/dl), which is also roughly that of blood. However, the disadvantages are two-fold: One is the fact that an important step in the analysis, gas extraction or elution, does not take place. The second is that measuring the volume of a gas sample is always problematic because of its dependence upon temperature. In addition, in order to calculate the amount of O<sub>2</sub> entering the analysis equipment (STPD conditions, i.e. 0°C, 760 mm Hg, dry), the exact temperature, barometric pressure and relative humidity must be known.

Distilled water, equilibrated at a defined temperature with pure oxygen, is very suitable for calibration, since the O<sub>2</sub> concentration and thus the amount of O<sub>2</sub> are precisely indicated, if the O<sub>2</sub> solubility and barometric pressure are known. Due to the low O<sub>2</sub> solubility, however, sample volumes must be used which are approximately 10 times larger than a corresponding blood sample.

*Table 1.* Calibration and quality control: O<sub>2</sub> concentration

Sample material	Equilibration	Calculation of O <sub>2</sub> (STPD)
Air (10–100 µl, 20.95 ml/dl)	–	barometric pressure, temperature and relative humidity. No extraction!
Distilled water (100–500 µl, 2.41 ml/dl/atm at 37°C)	100% O <sub>2</sub> at 37°C	O <sub>2</sub> solubility (STPD) and barometric pressure (pH <sub>2</sub> O = 47 mm Hg)
Human blood (10–100 µl)	defined pO <sub>2</sub> , pCO <sub>2</sub> at 37°C 'normal blood'	Hb concentration, O <sub>2</sub> binding curve, Hüfner number, physical- ly dissolved O <sub>2</sub>
KIO <sub>3</sub> solution (5.767 mmol/l = 20 ml/dl)	–	10 µl = 2.00 µl O <sub>2</sub> STPD

The ideal material for calibration is human blood equilibrated at 37°C, although the procedure involved is somewhat more elaborate. In addition to equilibrating the blood in a tonometer at 37°C at an O<sub>2</sub> partial pressure of around 150 mm Hg and physiological pCO<sub>2</sub>, the O<sub>2</sub> concentration can be calculated very accurately if the exact Hb concentration and the pO<sub>2</sub> value are known (the exact calculation is described in this book, p. 203). The blood used, however, must be 'normal blood', i.e., the concentrations of COHb and MetHb must be within the physiological range. Although this material requires elaborate preparation for use in calibration, all the steps in the analysis are reproduced optimally.

A KIO<sub>3</sub> solution of defined O<sub>2</sub> concentration can only be used when the O<sub>2</sub> concentration is measured photometrically (O<sub>2</sub> cuvette). In this case, defined oxidation equivalents are available which are quantitatively consumed by the O<sub>2</sub> indicator (very strong reducing agent). Since the standard, KIO<sub>3</sub>, is available at the highest purity and can be weighed out, it represents a gravimetric standard with optimal properties. At a concentration of 5.767 mmol/l the O<sub>2</sub> concentration is exactly 20 ml/dl, whereby the very small proportion of physically dissolved oxygen (due to contact of the solution with air) is barely significant. In addition, within a wide range of temperatures and barometric pressures the concentration of the physically dissolved O<sub>2</sub> has no influence on the O<sub>2</sub> concentration of the solution, so that the total O<sub>2</sub> concentration remains constant. These relationships are shown in table 2. Thus the O<sub>2</sub> concentration of a KIO<sub>3</sub> solution with 5.767 mmol/l changes by less than 1% when the temperature is varied between 16 and 26°C and the barometric pressure between 730 and 770 mm Hg.

*Table 2.* Quality control with a KIO<sub>3</sub> solution (10 µl). Chemically bound and physically dissolved oxygen (µl STPD) from a 10 µl sample of a KIO<sub>3</sub> solution with a concentration (gravimetric) of 5.767 mmol/l. At a constant amount of chemically bound O<sub>2</sub> (1.939 µl) the total oxygen only varies to a small extent with variations in the physically dissolved O<sub>2</sub> (influenced by barometric pressure and temperature; variation clearly less than 1%).

Temperature (°C)	Barometric pressure (mm Hg)		
	770	750	730
16	2.009 µl		2.005 µl
21		2.000 µl	
26	1.997 µl		1.993 µl

*Materials for Quality Control*

Any material to be used for the quality control of equipment should optimally be a stable, easily stored solution with a defined and constant O<sub>2</sub> concentration which is roughly in the range of that of normal blood. This last prerequisite is necessary so that all steps in the analysis can be followed during quality control, namely, measurement of the sample volume (i.e., 10 or 100 μl), airtight introduction of the sample volume, and the carrying out of all the steps of the analysis. This is the only way that errors in taking and transferring the sample as well as errors in the analysis can be recognized and eliminated. In all the procedures for measuring O<sub>2</sub> concentration, these conditions can only be fulfilled by equilibrated 'normal blood', even if this procedure involves considerable preparation.

In the case of photometric measurement of the O<sub>2</sub> concentration (O<sub>2</sub> cuvette), however, the KIO<sub>3</sub> solution discussed above is an optimal standard for quality control purposes.

*Summary*

Various materials are available for the calibration and quality control of equipment used for measuring O<sub>2</sub> concentration: air, equilibrated distilled water or human blood, as well as a KIO<sub>3</sub> solution. For regular quality control, human blood is recommended, and for photometric measurement, a defined KIO<sub>3</sub> solution.