SOME PROBLEMS ASSOCIATED WITH MEASUREMENT OF PHYSIOLOGICAL BLOOD GASES

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INTRODUCTION

BLOOD GAS ANALYSES have been possible for almost a century, but until approximately 15 years ago, methods for the measurement of pH, PCO_2 and PO_2 in blood samples were cumbersome and difficult to perform. With the development of precision electrodes for blood-gas analysis, it has become possible to obtain accurate values for these parameters. Also, within the past decade, physicians have become more aware of the importance of blood-gas analysis in the assessment of the respiratory and acid-base status of their patients.

MEASUREMENT OF pH AND BLOOD GASES

(I) pH

The glass electrode is used for pH measurement (Figure 1).¹ This electrode consists of two half-cells, each of which develops a potential when connected together. The reference half-cell (calomel electrode) maintains a constant potential, while the glass half-cell (or electrode) develops a potential which is proportional to the hydrogen ion concentration present in the test solution.²

The instrument is calibrated by setting the span of the electrode on two phosphate buffers of known pH. To ensure accuracy it is important that the temperature of the apparatus be kept at 37 °C. This is accomplished by means of a circulating water jacket around the electrode.

(II) PCO_2

There are two methods for determination of PCO_2 . (i) CO_2 electrode, (ii) Astrup Interpolation Technique.

(i) CO_2 Electrode (Figure 2)¹

This consists of a standard pH glass electrode over which is placed a thin film of aqueous NaHCO_3 . A teflon membrane separates the blood sample from the NaHCO_3 solution and the electrode. CO_2 diffuses across the Teflon membrane and so changes the pH of the bicarbonate solution. This change in pH is then measured by the glass electrode.^{3,4}

(ii) Astrup Interpolation Technique

This technique is based on the observation that there is a linear relationship between pH and $\log \text{PCO}_2$ of any particular blood sample with pH falling as PCO_2 increases.⁵ Two aliquots of the blood sample are equilibrated in a tonometer with two different gases of known CO_2 concentra-

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FIGURE 1. The measurement of pH. Arrangement of electrodes and pH meter.

FIGURE 2. The carbon dioxide electrode.

tion, usually 4 per cent and 8 per cent CO_2 ⁶ (Figure 3).¹ The pH of each equilibrated aliquot is read with a glass electrode system, and a buffer line is drawn by plotting the pH readings against the relevant PCO_2 values on a logarithmic scale. This represents the buffer line of that particular blood sample. The pH of a third aliquot of the blood sample is then interpolated in the buffer line, and the PCO_2 is read off the ordinate.

FIGURE 4. The oxygen electrode.

calibrated with gases, P_{O_2} values for blood should be corrected upwards by a small factor to ensure accuracy. This correction factor is called the "blood-gas factor", and is necessary due to decreased diffusing capacity of O_2 in blood as compared with gas.⁹

PURPOSE OF STUDY

Recently it was brought to our attention that there appeared to be discrepancies in blood-gas values obtained from various laboratories at the Vancouver General Hospital, on samples obtained from the same patient at the same time. A study was thus undertaken to determine the nature of these discrepancies and to attempt to eliminate them.

There are five major areas in the hospital where blood-gas analyses are performed, involving seven machines. The areas involved were designated by letter and the methods for blood-gas analysis in each laboratory were as follows:

Area A (1 and 2): Two Radiometer machines with pH, PCO_2 and PO_2 electrodes;

Area B: One Instrumentation Laboratory machine with pH, PCO_2 and PO_2 electrodes;

Area C: One Beckman analyzer with pH, PCO_2 and PO_2 electrodes;

Area D (1 and 2): Two Radiometer machines with pH and PO_2 electrodes, PCO_2 determined by Astrup Interpolation Technique;

Area E: One Radiometer machine with pH and PCO_2 electrodes, PCO_2 determined by Astrup Interpolation Technique.

FIGURE 5. Phase I - pH values.

PHASE I OF STUDY

METHODS

Phase I of the study was carried out to determine under standard conditions whether or not significant discrepancies in values did in fact exist from area to area. Thirty blood samples were taken. Blood samples were drawn from anaesthetized patients or patients before induction of anaesthesia, or in the recovery room.

The samples were drawn from a single radial artery puncture with a #20 needle, into a 5 cc glass syringe heparinized from the same heparin ampoule. Care was taken to ensure anaerobic collection, and the samples were immediately placed in ice and delivered to the various laboratories. Analysis was within 30 to 45 minutes of collection, and all laboratories analyzed the sample at the same time.

Area A₁ was arbitrarily taken as the standard and results for pH, Pco₂ and Po₂ from each other area were statistically compared with this standard, using the Student "t" test. A 95 per cent confidence limit was chosen and calculated values above this level were considered statistically significant. The means of all pH, Pco₂ and Po₂ values from each area were then plotted in bar graph form.

RESULTS

The mean pH values ranged from 7.36 to 7.41. The differences between Area A₁ and C, D₁ and D₂ were statistically significant (Figure 5). However, it is doubtful whether patient therapy would be adversely affected by such differences in pH between areas. The mean of Pco₂ values showed a wider range (from 34 to 43 mm Hg) (Figure 6). Again, using Area A₁ as the standard, there was a significant difference in means between this area and Areas B, C, D₁ and D₂. The range of mean Po₂ values was very wide from area to area (81 to 99 mm

FIGURE 6. Phase I – PCO₂ values.

Hg), with areas D1, D2 and E having statistically significant difference in means, as compared with the mean Po₂ for Area A1 (Figure 7).

Having determined that significant discrepancies did exist between areas, especially for Po₂, a search for the possible causes of this discrepancy was undertaken. Investigation revealed that areas A, B and C calibrated their O₂ electrodes with gaseous standards (either room air or analyzed tank gases) and areas D and E calibrated with liquids – specifically, water at 37° C equilibrated with room air. The areas employing gas calibrations of their Po₂ electrodes had not been correcting their Po₂ readings for blood upwards by any blood-gas factor. Radiometer Company has stated in one of their briefs that the blood-gas factor to be employed for their O₂ electrodes should be 1.03 ± 2.0 .¹⁰ Thus, Area A, using Radiometer equipment, began to correct their blood Po₂ readings upward by this factor. As well, this area changed the source of their analyzed gases. They had previously used gases analyzed by the Scholander method, but changed to tanks from another company which analyzed the gases by chromatography, which is considered to be a very accurate method of gas analysis.

Area D maintained the temperature of the oxygen electrode at slightly higher than 37° C, which would tend to raise the Po₂ readings. This, in turn, was corrected. We concentrated mainly on areas A and D, since these are the two laboratories which are not autonomous; that is, they may be involved in analyzing samples from the same patient. We therefore wished to ensure that readings from these two areas were reliably consistent with one another.

FIGURE 7. Phase I - PO₂ values.

PHASE II OF STUDY

METHODS

The second phase of the study was carried out after the corrections in Areas A and D were completed. A tonometer was employed. This is an apparatus used to equilibrate a blood sample with a known concentration of gases, in this case oxygen and carbon dioxide. The gases flow into a chamber containing the blood sample, which is rotated to expose a large surface of blood to the gases. At the end of the equilibration, the Po₂ and Pco₂ of the blood sample will be known.

Blood samples were equilibrated with a known gas (12 per cent O₂ and 5 per cent CO₂) previously analyzed by gas chromatography. These tonometered samples were transported in ice to the various areas for analysis.

The results were again analyzed employing the Student "t" test, using the known Po₂ and Pco₂ of the tonometered sample as the control and comparing the results obtained from each area with these values.

FIGURE 8. Phase II - PCO₂ values.

RESULTS

There were no statistically significant differences for PCO₂ (Figure 8). The mean PCO₂ values now ranged from 32 to 36 mm Hg. The mean PO₂ values continued to show a range from area to area of 11 mm Hg (Figure 9). Area B results for PO₂ were significantly lower than control. This could be alleviated somewhat by employing a blood-gas correction factor to readings in this area, which uses gases to calibrate its PO₂ electrode. Area D readings were significantly higher than control values, and reasons for this were not apparent.

DISCUSSION

From this study, we have shown that in spite of careful attention to detail, the results obtained in blood gas analyses may be inaccurate. The major sources of error occur in two areas:

- (1) Obtaining and handling of the blood sample.
- (2) Analysis of the sample.

Certain details must be carefully observed in order to prevent errors in obtaining and handling the sample (Table I):

(a) Arterial blood should be used whenever possible, as values from capillary or arterialized venous blood may be inaccurate during low flow states, in newborns during the first 24 hours of life, and often in newborns with respiratory distress syndrome.^{11,12}

FIGURE 9. Phase II - PO₂ values.

TABLE I
PREVENTION OF ERRORS IN OBTAINING
SAMPLE

1. Use arterial blood
2. Glass or plastic syringes may be used
3. Heparin must fill only syringe deadspace
4. Avoid traction on plunger
5. Avoid air bubbles in sample
6. Store in ice until analyzed

(b) Glass or plastic (polypropylene) syringes may be used to collect the sample. However, if any delay in analysis of the sample is anticipated, glass is to be preferred, as the diffusion rate of gases through glass is less than that through plastic. The type of plastic used in manufacture of the syringe may also be important, as the diffusion rate of gases through polypropylene is less than that through polyethylene. If a plastic syringe is used, one must disengage the plunger from the barrel before use, so that arterial blood will flow freely into the syringe during collection.¹³

(c) Heparin should fill only the dead space of the syringe.

(d) One must not use excessive traction on the plunger of the syringe, as this may cause gases to be drawn out of solution and lower gas values.²

(e) One must avoid air bubbles in the sample, which will change blood gas values.

TABLE II
PREVENTION OF ERRORS IN ANALYSIS
OF SAMPLE

1.	Accurate calibrating methods
2.	Apparatus temperature 37° C
3.	Change membranes frequently
4.	Mix sample thoroughly
5.	Meter must stabilize before reading
6.	Blood gas factor

(f) The sample must be stored in ice until the time of analysis in order to slow the metabolic rate of cellular elements in the sample.

In order to prevent errors in the analysis of the blood sample these points should be observed (Table II):

(a) Accurate calibrating methods must be used for the electrodes. If gases are to be used it must be ensured that these gases have been accurately analyzed prior to use. If a liquid is to be used for calibration of the Po_2 electrode, it is usually water from the water bath surrounding the electrode system. It must be ensured that the water jacket has been on for a sufficient period of time to warm to 37° C, and for equilibration with room air to have occurred.

(b) The water jacket surrounding the electrodes must be kept at exactly 37° C.¹⁴

(c) Faulty membranes or faulty glass electrodes must be replaced.

(d) The blood sample must be thoroughly mixed before aliquots are introduced into the electrodes for analysis.

(e) The electrode meter must be allowed to stabilize before a reading is taken. This is especially important for the CO_2 electrode, since, depending upon the thickness of the Teflon membrane, up to three minutes may be required for meter stabilization.

(f) Po_2 readings should be corrected upwards by a blood gas factor, on readings from machines which employ gases to calibrate the electrode.⁹

CONCLUSION

The results of this study show that significant discrepancies may exist in blood gas values from various areas within a large general hospital. Potential sources of error in blood gas values involve both collection of the sample and its handling before analysis, and factors in the laboratory itself involving both equipment and personnel.

It is important for physicians to realize that, in spite of strict attention to detail, blood gas values may not be accurate, and clinical judgment must always be judiciously employed to interpret the results obtained.

RÉSUMÉ

Cette étude démontre que, malgré une attention minutieuse, l'analyse des gaz du sang peut être erronée. Les principales sources d'erreur sont de deux ordres.

- (1) Le prélèvement et la manutention de l'échantillon de sang.
- (2) L'analyse de l'échantillon.

Il s'impose d'observer soigneusement certains détails pour prévenir des erreurs au cours du prélèvement et de la manutention de l'échantillon (Table I) :

- (a) Autant que possible, il faut utiliser du sang artériel, car les données du sang capillaire ou du sang veineux artérialisé peuvent être inexactes dans les états où la circulation est lente, chez les nouveaux nés au cours des 24 premières heures de vie et souvent chez les nouveaux nés présentant un syndrome de détresse respiratoire.^{11,12}
- (b) Pour prélever l'échantillon on peut employer des seringues de verre ou de plastique (polypropylène); toutefois, si l'on prévoit un délai avant l'analyse, une seringue de verre est préférable car le taux de diffusion des gaz à travers le verre est inférieur à celui à travers le plastique. La sorte de plastique utilisé pour faire la seringue est également importante car le taux de diffusion des gaz est inférieur à travers le polypropylène comparé à celui à travers le polyéthylène. Si l'on emploie une seringue de plastique, il est à conseiller d'enlever le piston avant l'usage de façon à ce que le sang artériel coule librement dans la seringue durant le prélèvement.¹³
- (c) L'héparine doit combler seulement l'espace mort de la seringue.
- (d) Il ne faut pas exercer une traction trop forte sur le piston ce qui pourrait extraire les gaz du sang et donner des résultats inférieurs.²
- (e) Il faut éviter d'avoir des bulles d'air dans l'échantillon ce qui changerait les valeurs des gaz sanguins.
- (f) L'échantillon doit être conservé sur la glace jusqu'au moment de l'analyse pour ralentir le métabolisme des cellules dans l'échantillon.

Pour éviter des erreurs dans l'analyse de l'échantillon de sang, il faut observer trois points (Table II) :

- (a) Il faut utiliser des méthodes précises de calibration des électrodes. Si l'on doit employer des gaz, il faut s'assurer que ces gaz ont été analysés de façon précise avant l'usage. Si l'on emploie un liquide pour calibrer l'électrode de la Po_2 , il faut employer l'eau du bac qui entoure le système d'électrode. Il faut s'assurer que le chauffe-eau fonctionne depuis un certain temps pour que la température de l'eau soit à 37° et qu'elle ait eu le temps de devenir en équilibre avec l'air de la pièce.
- (b) Le chauffe-eau autour des électrodes doit être maintenue à 37° exactement.¹⁴
- (c) Il faut remplacer les membranes et les électrodes défectueuses.
- (d) L'échantillon de sang doit être agité et bien mélangé avant l'introduction des électrodes pour analyse.
- (e) Le compteur de l'électrode doit avoir le temps de s'arrêter avant qu'une lecture soit faite. Cela devient plus spécialement important pour l'électrode du CO_2 puisque, selon l'épaisseur de la membrane de Teflon, il faut un délai qui peut aller jusqu'à trois minutes avant que le compteur ne se stabilise.
- (f) Les lectures de la Po_2 doivent être augmentées par un facteur de gaz sanguin à partir des lectures sur les machines qui emploient des gaz pour calibrer les électrodes.⁹

CONCLUSION

Les résultats de cette étude démontrent que, dans un grand hôpital, il peut exister des différences appréciables entre les valeurs des gaz sanguins d'un endroit à l'autre. Les sources éventuelles d'erreurs dans les valeurs des gaz sanguins impliquent aussi bien le prélèvement que la manutention de l'échantillon avant l'analyse et, dans le laboratoire, elles impliquent aussi bien le matériel que le personnel. Il est *bien important* pour les médecins de se rappeler que, malgré toute l'attention apportée à la technique, les valeurs des gaz sanguins peuvent être erronées et que c'est à la lumière du jugement clinique qu'il faut interpréter les résultats obtenus.

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