Effect of time delay and syringe surface area on partial pressure of oxygen and oxygen saturation of arterial and venous blood gas samples. (an observational study)

A Thesis Study

Submitted for the fulfillment of master degree in Anaesthesia , Surgical ICU & Pain Management

By

Dina Samir Mahmoud Shaker
(M.B.B.CH. Faculty of Medicine-Cairo University)

Supervised by:

Dr Amina Aboul Ela Elew Mousa
Professor Assistant of Anaesthesia, Surgical ICU and Pain Management
Faculty of Medicine, Cairo University.

Dr Mamdouh Amin Abdul Hamid
Professor Assistant of Anaesthesia, Surgical ICU and Pain Management
Theodor Bilharz Research Institute.

Dr Ahmed Mohamed Lotfy Mostafa
Lecturer of Anaesthesia, Surgical ICU and Pain Management
Faculty of Medicine at Cairo University.

2019

Acknowledgment

*First of all, I should thank **Allah** the greatest and most merciful not only for helping me through this work, but also for helping me all through my life. Words will never be able to express my deepest gratitude to all those who helped me during preparation of this study.*

*I would like to express my deepest gratitude and thanks to **Prof. Dr Amina Abo Ella**, Professor Assistant of Anaesthesia, Surgical ICU and Pain Management , Faculty of Medicine, Cairo university for her kind continuous encourgement and great support through this work It was great honor to work under her supervision.*

*I am deeply grateful to **Prof. Dr Mamdouh Amin**, Professor Assistant of Anaesthesia, Surgical ICU and Pain Management Theodor Bilharz Research Institute, for his great support , valuable time and continuous advice.*

*My completion of this project could not have been accomplished without the support of **Dr Ahmed Lotfy**, Professor Assistant of Anaesthesia, Surgical ICU and Pain Management , Faculty of Medicine, Cairo university. I am also greatly indebted to him for his careful and great support and valuable effort that made this work come to light.*

*Finally, no words can express how thankful I am to my caring, loving, and **supportive family**. Without their encouragement and support, I could never have reached this current level of success.*

*It is an honour to dedicate this project to **Dr Ahmed Hasanain** , Professor Assistant of Anaesthesia, Surgical ICU and Pain Management , Faculty of Medicine, Cairo university for his extraordinary ideas , great effort and support in this study .*

Dina Samir

List of Contents

Abbreviations

Abbreviation	*Meaning*
ABG	Arterial blood gases
VBG	Venous blood gases
BGA	Blood gas analysis
PaO2(pO2)	oxygen tension
PaCO2(pCO2)	carbon dioxide tension
SaO2	oxyhaemoglobin saturation
sO_2	Oxygen saturation
TAT	Turnaround Time
MET	Medical Emergency Team
ICU	Intensive care unit
ED	Emergency Department
LOS	length of hospital stay
POC	point-of-care
POCT	point-of-care testing
CLT	Central laboratory testing
Ag-AgCl	silver-silver chloride
ISE	ion-selective electrodes
PCV	packed cell volume
BUN	Blood urea nitrogen
AARC	American Association for Respiratory Care
SD	standard deviation
R	the correlation coefficient of a sample
CI	Confidence Interval

Common Symbols and Acronyms Related to Blood Gas Testing are tabulated in Table 1 .

Haemoglobin Fractions abbreviation are tabulated in Table 2

List of Figures

List of tables

Abstract

Background: Blood gas analysis is an important test in many medical situations. Either arterial blood gases or venous blood gases are done in nearly all critically ill patients. Delays may occur in analysis of a drawn sample in the hospital due to the limited resources and as blood gas analyzers are not available in many areas in the hospital. Previous studies have found that the values of arterial blood gases were influenced by time delay and sample storage temperature in blood gas analysis

Methods: An arterial sample (from arterial line connected to 20G cannula) and a central venous sample (from a central venous catheter) were withdrawn. Each sample was 5ml and was divided into five parts: 1ml of blood in a heparinized syringe was analyzed immediately, 1ml of blood in a 3ml plastic syringe (2 syringes) , 1ml of blood in a capillary insulin syringe (2 syringes). The last 4 samples were analyzed after 30 mins and 1hour respectively. The samples while waiting analysis were left in ambient temperature. This was done to the arterial and venous samples simultaneously. This means that every patient had 10 samples analyzed and subjected to final analysis.

Results: There is a wide range between limits of agreement in oxygen saturation, PO2 and PCO2.The mean baseline PO2 in arterial samples was 159.9±113.6. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were 21(-99to142) and 38 (-112 to 188) respectively in the 3 ml syringe. The mean baseline PCO2 in arterial samples was 36±7.6. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were -0.4(-4.8 to4.1) and -3.8(-11.8 to 4.1) respectively in the 3 ml syringe.

Conclusion: We conclude that arterial and venous samples that are delayed at room temperature give different results from baseline results (analyzed immediately). This change occurs as early as 30 minutes. We recommend that blood gas analysis should be done immediately to obtain reliable measurements.

Background and Rationale

An Arterial Blood gas (ABG) is a test that measures the oxygen tension (PaO_2), carbon dioxide tension ($PaCO_2$), acidity (pH), oxyhaemoglobin saturation (SaO_2) and bicarbonate concentration (HCO_3) in arterial blood.

The methods for and importance of blood gas testing for pO_2 and pCO_2 were first described by Van Slyke in the early 1900's (1)

The term "Blood gas testing" traditionally refers to determining the partial pressures of the physiologically active gases in blood (pO_2, pCO_2), the blood pH, and the oxygen saturation of haemoglobin (SaO_2) (2)

However, current commercially-available instruments are capable of performing haemoglobin quantitation and co-oximetry, as well as measuring electrolytes (sodium, potassium, chloride, ionized calcium and magnesium), glucose, lactate, and creatinine, usually simultaneously (3,4)

The process of analysis and monitoring of arterial blood gas (ABG) is an essential part of diagnosing and managing the oxygenation status and acid–base balance of the high-risk patients intra-operatively, as well as in the care of critically ill patients in the Intensive Care Unit. Disorders of acid–base balance can lead to severe complications in many disease states, and occasionally the abnormality may be so severe as to become a life-threatening risk factor.

Thus, what is commonly referred to as a "blood gas" analysis may include a few or many tests, and may be performed in a central laboratory, at the point of care.

Measurements of blood gas analysis are most accurate when the sample is analyzed immediately. For a variety of reasons, immediate analysis is not always possible.

During in vitro storage, red blood cells metabolize glucose and produce lactic acid(5), while nucleated white blood cells, reticulocytes, and platelets consume oxygen and produce carbon dioxide (6)

Plastic syringes for blood collection and storage have the advantage of low cost and easy disposal; however, one disadvantage for blood gas analysis is that oxygen and carbon dioxide can diffuse through the plastic. The magnitude of these changes depends upon storage time, storage temperature, and the partial pressure gradients between the blood sample and that of the plastic.

The effect of plastic syringes on blood gas measurements obtained following sample storage is complicated by the tendency of oxygen and carbon dioxide to diffuse into and through plastic syringes (through the spaces between the interlaced macromolecules of the plastic). (7) In plastic syringes, the change in PO_2 over time is the balance between the metabolic consumption of oxygen and the net flux of oxygen between the blood and the plastic of the syringe.

The rate of metabolic oxygen consumption is determined by storage temperature and the concentration of white blood cells and platelets in the blood sample. The flux of oxygen between the blood sample and the plastic depends upon the PO_2 of the blood sample at the beginning of the storage, the temperature at which the blood is stored, the duration of storage, the volume of blood, and the syringe barrel exposure area.

The laboratory testing process has been traditionally divided into three phases. Testing begins in the pre-analytical phase, which includes all activities that

occur prior to the sample's insertion into the analytical instrument. The analytical phase follows, and includes the chemical reactions, fluidics, and other processes that occur in the analytic platform. Testing finishes with the post-analytical phase, which includes all events occurring after the test result is generated, such as data entry, transport of the result through various information systems, and interpretation of the result.

It has been recognized for years that the phase of testing most prone to errors is the pre-analytical phase (5,7–9)

The reasons why pre-analytical processes are more error-prone than processes in later testing phases are varied, as they include both patient factors, such as interferences in blood samples, and iatrogenic factors, such as the fact that specimen collection is an almost entirely manual process. To the contrary, analytical and post-analytical processes in the modern clinical laboratory are often automated, and thus reliable computer-based safeguards can be implemented.

Pre-analytical errors may be due to many factors such as : type of blood collected whether arterial , venous or capillary ,inappropriate sample container including material and surface area of syringe used, how long the time delay for analysis, improper sample storage temperature, sample contamination, presence of air bubbles in the sample and use of the incorrect type or amount of anticoagulant leading to sample hemolysis or coagulation.

In this study, we selected certain pre-analytic factors to observe their effects on both arterial and venous blood gas results. We selected the time delay as it frequently occurs as the working staff in the operative theatre, in the intensive care unit or in the laboratory may be busy in these hot areas. In addition, the laboratory may be far-away. We selected the surface area of the syringes as

insulin syringes and the 3-ml syringes are both available in our hospitals and to observe which is better to use.

Review of Literature

Sensors and Measurement Concepts

Introduction to General Measurement Concepts

Gas tension

Gas tension is the partial pressure of a gas in blood. Partial pressure refers to the pressure exerted by a single gas in a mixture of gases or in a liquid. The pressure of the gas is related to the concentration of the gas to the total pressure of the mixture. For example, the concentration of oxygen in the atmosphere is 0.21. Atmospheric pressure is 760 mm Hg (at sea level). The **partial pressure of oxygen** in the atmosphere can be calculated by multiplying the concentration of this gas in the atmosphere (0.21) by atmospheric pressure (760 mm Hg) (10)

Gas tension of oxygen in the atmosphere = 0.21 × 760 mm Hg = 160 mm Hg

pO_2 refers to the partial pressure or tension of oxygen; it may also be written as PO_2, PO_2, and pO_2.The reference range of pO_2 in arterial blood is 80–110 mmol/L. pCO_2 refers to the partial pressure or tension of carbon dioxide; it may also be written as PCO_2, PCO_2, or pCO_2. The reference range of pCO_2 for arterial blood is 35–45 mm Hg.

pH

The pH is a measure of the acidity or alkalinity of a solution and ranges from 1–14. Values less than 7.0 are acidic and greater than 7.0 are alkaline. An **acid** is a substance that produces or donates hydrogen ions [H^+] when dissolved in water, whereas a **base** or alkaline substance is one that produces or donates hydroxyl ions [OH^-] when dissolved in water. When there are equal numbers of [H^+] and [OH^-] ions, the solution is neutral and the pH is 7.0, as shown in the following equation (10):

$$H^+ + OH^- \longrightarrow H_2O$$

Relationship Between pH and H^+

The pH is the negative logarithm of the hydrogen ion concentration [H^+] in moles/liter. For example, a pH of 6, a slightly acidic solution, would have an [H^+] of 1.0×10^{-6}. Conversely, if a solution has an [H^+] concentration of 1×10^{-12}, the pH of this solution would be 12, which is alkaline.

The pH is measured in arterial blood to determine the degree of acidity or alkalinity. The acid–base balance of body fluids, including blood, is maintained through the hydrogen ion concentration. The reference range for the pH of arterial blood is 7.35–7.45 (10)

Table 1: Common Symbols and Acronyms Related to Blood Gas Testing

Symbol	Meaning
pO_2	partial pressure of oxygen; also written as pO_2, pO_2, or PO_2
pCO_2	partial pressure of carbon dioxide; also written as pCO_2, pCO_2, or PCO_2
$[H^+]$	hydrogen ion concentration
$[OH^-]$	hydroxyl ion concentration
Hb	Haemoglobin
COHb	Carboxyhaemoglobin
HHb	reduced or deoxygenated haemoglobin
O_2Hb	Oxygenated haemoglobin or oxyhaemoglobin
THb	Total Haemoglobin
sO_2	Oxygen saturation of haemoglobin
p50	pO_2 at which 50% of haemoglobin is saturated
ctO_2	Oxygen content
Qa	Quality assurance
QC	Quality control

pH Electrode and Reference Electrode

Glass electrodes are commonly used to measure pH. The pH measurement system uses the Sanz electrode, which consists of two half cells connected by a potassium chloride (KCl) bridge. The measurement half cell or electrode has a glass membrane with layers of hydrated and non-hydrated glass. It is permeable or sensitive to hydrogen [H^+] ions. This measurement electrode consists of silver-silver chloride (Ag-AgCl), which is then placed into a phosphate buffer of pH 6.840, and thus has a known [H^+] concentration. The reference half cell or electrode consists of mercury and mercurous chloride (Hg-HgCl) or calomel. This calomel electrode is placed into a solution of saturated KCl . (11)

The reference electrode provides steady voltage while the measuring electrode responds to the ions of interest in the sample. Thus, the reference electrode provides a baseline voltage against which the voltage measured by the measuring electrode is compared. A pH meter or voltmeter measures this potential difference, known as ΔE, between the two electrodes. This relationship is shown in the following equation:

$$\Delta E = \Delta E^0 + 0.05916/n \log a_1 \text{ at } 25°C$$

where:

- ΔE = potential difference
- ΔE0 = Standard potential of electrochemical cell
- n = charge of analyte ion a1 = activity of ion

There is a change of + 59.16 millivolts (mV) at 25°C for a 10-fold increase in [H^+] activity and a decrease in pH units. At 37°C, the change in one pH unit causes a 61.5 mV change in the electrical potential. (12)

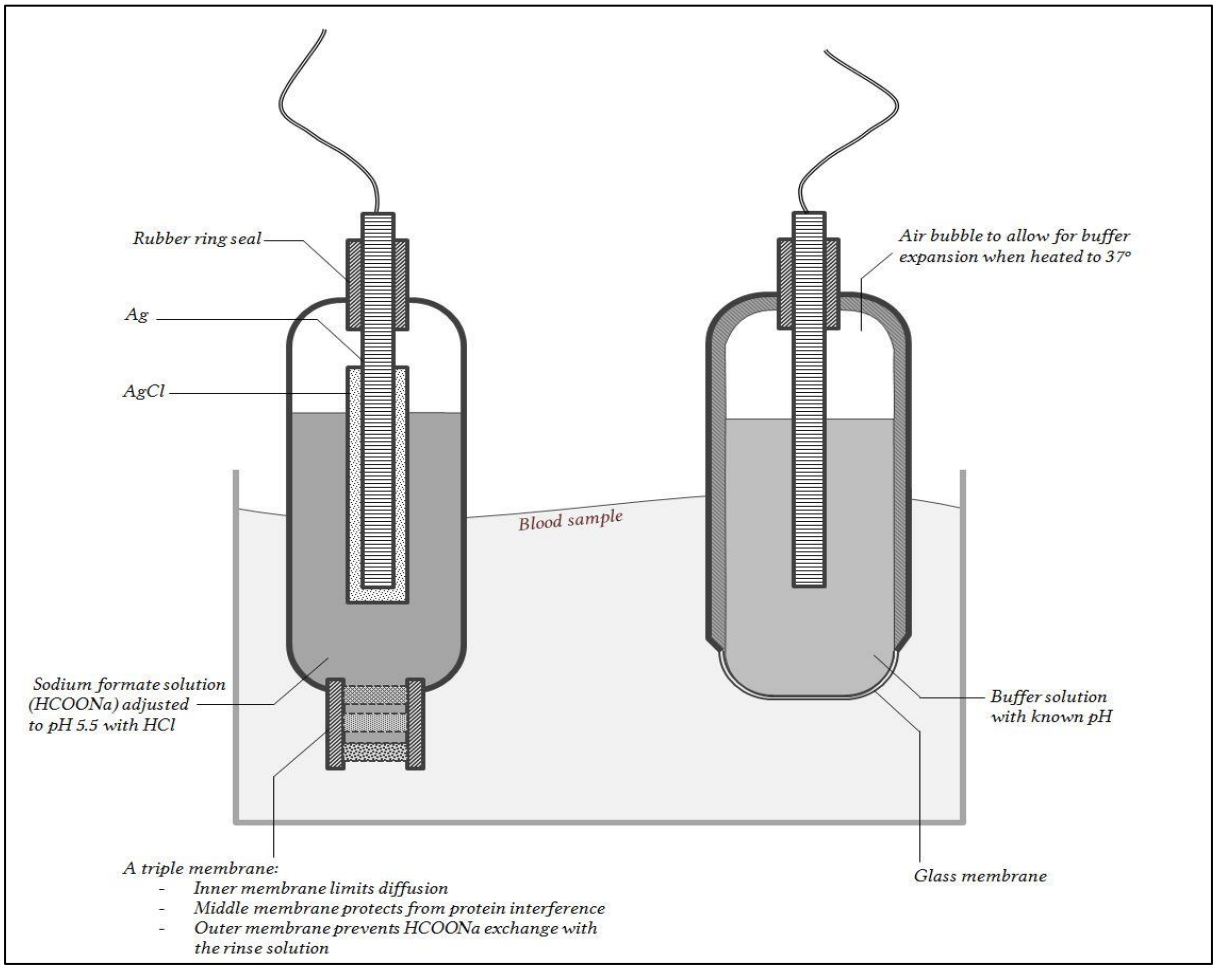

Figure 1: The Sanz electrode

Functional Requirement and Characteristics of the pH system

It is postulated that sodium ions in the hydrated glass drift out and are replaced by the smaller hydrogen ions that are present in the sample. This results in a net increase in the external membrane potential, which travels through the thin, dry membrane to the inner hydrated glass surface. Chloride ions in the buffer solution migrate to the internal glass layer, creating a potential difference at the pH electrode that, in turn, signals the external reference electrode. The difference in voltage is converted and displayed as the pH.

pCO_2 Electrode System

Functional Requirement and Characteristics of the pCO_2 Electrode

The pCO_2 electrode is a modified pH electrode that was first described by Stowe and later by Severinghaus (13); today it is known as a Stowe-Severinghaus electrode. The electrode has an outer semipermeable membrane consisting of Teflon or silicon elastic (Silastic). CO_2 diffuses into an electrolyte layer; a bicarbonate buffer covers the electrode glass. When CO_2 reacts with the buffer, carbonic acid forms, which then dissociates into a bicarbonate ion [HCO_3^-] and hydrogen ions [H^+]. (14)

$$CO_2 + H_2O \longrightarrow H_2CO_3 \longrightarrow H^+ + HCO^-$$

The hydrogen ions diffuse across the glass electrode and the change in $[H^+]$ activity is measured using the same principle as for the pH electrode. The pCO_2 is determined from the pH value using the Henderson-Hasselbalch equation:

$$pH = pk + \log [HCO^-]/pCO_2$$

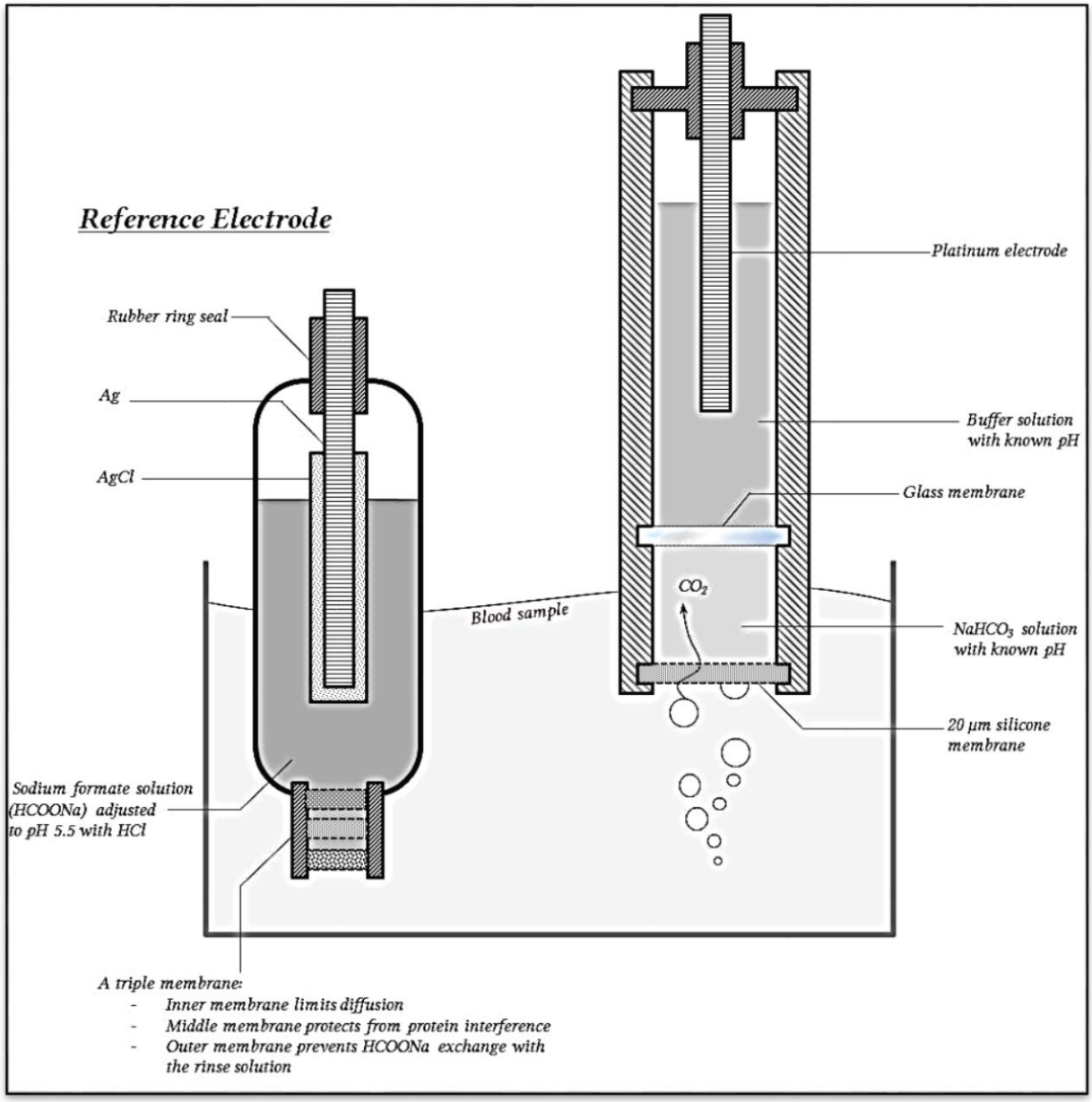

Figure 2: Stowe-Severinghaus electrode.

pO_2 Electrode System

Functional Requirement and Characteristics of the pO_2 Electrode

The partial pressure of oxygen (pO_2) is measured using the Clark electrode, which is a complete electrical cell. The Clark electrode consists of a small platinum **cathode** and a silver-silver chloride (Ag-$AgCl_2$) **anode** immersed in a phosphate buffer that contains additional potassium chloride. The platinum electrode is covered with a small layer of electrolyte and a thin gas-permeable membrane made of a material such as polypropylene. The membrane separates the test specimen from the electrode and is selectively permeable to oxygen, which diffuses into the electrolyte to contact the cathode. The cathode potential is adjusted to a constant voltage potential of –0.65 volts (V). When there is no oxygen present in the solution, the cathode is polarized and the current is approximately equal to 0 volts. When oxygen is present in the test specimen, a current is produced and oxygen diffuses from the sample solution and then through the membrane, where it is reduced (15). Electrons are drawn from the anode surface to the cathode to reduce the oxygen. The current is proportional to the pO_2 of the test solution.

The sensitivity of the pO_2 electrode is related to the thickness of the membrane and the size of the cathode area. A micro-ampmeter measures movement of electrons between the anode and cathode, which forms the electrical current. There are four electrons drawn for each mole of O_2 that is reduced. The reaction at the cathode is summarized as follows:

$$O_2 + 4e^- \longrightarrow 2\,O^-$$

$$2\,O^- + 2\,H_2O \longrightarrow 4\,OH^-$$

Next, elemental silver present at the anode is oxidized and then ionized, forming four electrons before combining with chloride to form silver chloride. The reaction at the anode is summarized as follows:

$$4\,Ag^+ + Cl^- \longrightarrow 4\,AgCl$$

$$4\,Ag \longrightarrow 4\,Ag^+ + 4\,e^-$$

Other gases may pass through the membrane, but the degree of the polarizing voltage does not permit them to be reduced at the cathode. The membrane prevents proteins and other oxidizing agents from reaching the cathode surface. Protein build-up on the membrane is an important source of measurement error; proteins may alter the diffusion of the gases and hinder the electrode response. The sensitivity of the electrode is related to the thickness of the membrane and the size of the cathode area.

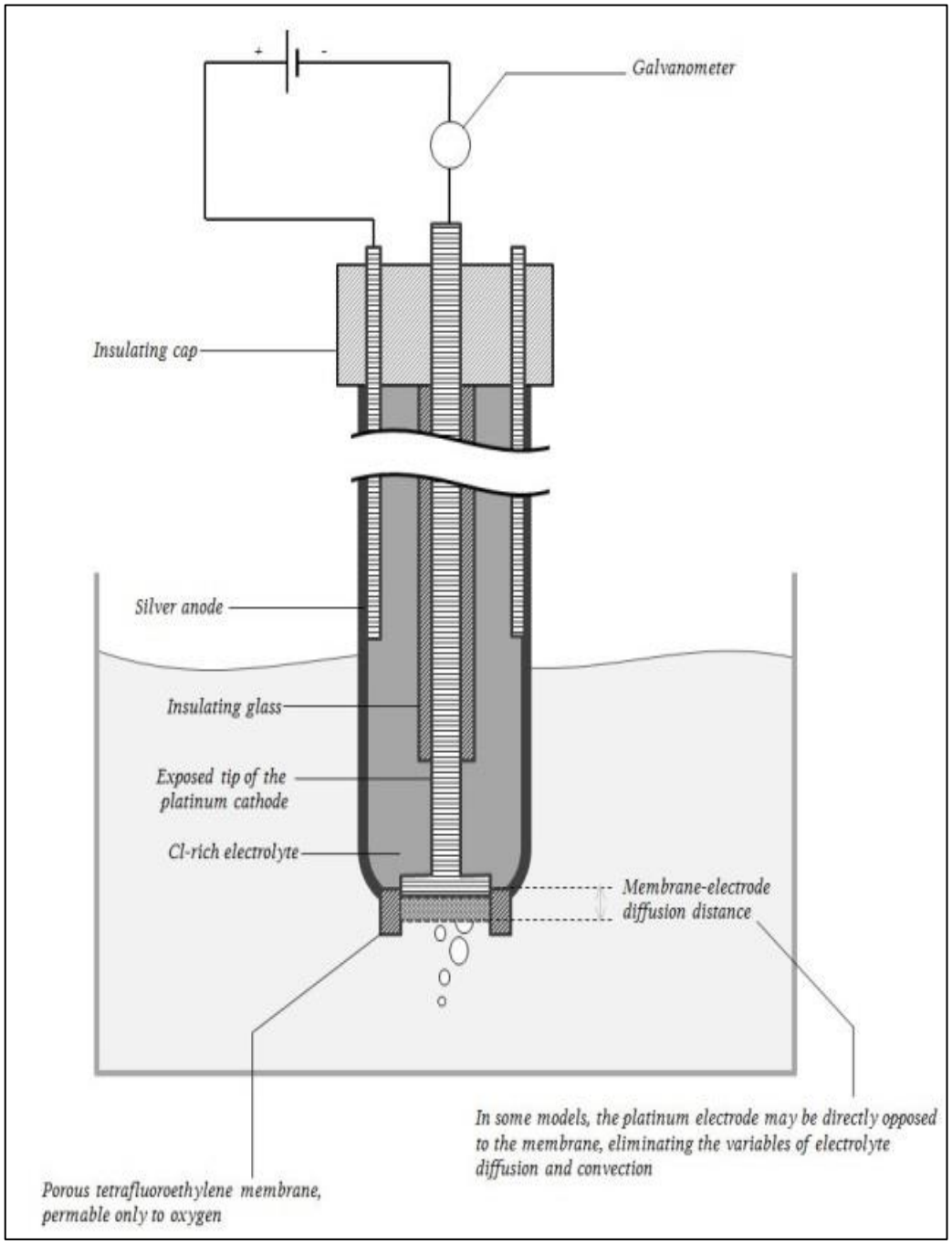

Figure 3: The Clark electrode.

Calculated Values

Haemoglobin/Oxygen Saturation

Oxygen saturation of haemoglobin is the percentage of oxygenated haemoglobin divided by total haemoglobin present capable of binding with oxygen. Oxygen saturation (sO_2) is calculated using the following equation:

$$sO_2\% = cO_2Hb/(cO_2Hb + cHHb) \times 100$$

where cO_2Hb is the concentration of oxyhaemoglobin and cHHb is the concentration of reduced or deoxyhaemoglobin; the sum of oxy- and deoxyhaemoglobin represents the total functional haemoglobin. Oxygen saturation is a derived value for most blood gas analyzers. It is only measured by analyzers that have hemoximetry capabilities. A hemoximeter directly measures the amount of haemoglobin present and capable of binding with oxygen (16). Therefore, the microprocessors of analyzers that calculate sO_2 assume normal oxygen affinity of the haemoglobin. It is important for the clinician to recognize this calculation may be approximate and should be interpreted with caution.

Oxygen saturation may also be calculated using the following formula:

$$sO_2\ \% = cO_2\ Hb/ctHb \times 100\%$$

where *ct*HB includes the carboxyhaemoglobin, methaemoglobin, and sulfhaemoglobin fractions. Using this equation, the sO_2% will never reach 100% if any of these nonfunctional haemoglobin fractions are present. This calculation should not be used because the

dishaemoglobins, mentioned above, are present in the blood and the findings may be misleading. For example, if a patient had 10% carboxyhaemoglobin, the sO_2 could not be any higher than 90%, even in fully saturated blood, which might indicate an increase in oxygen shunting to the lungs, which would not be accurate.

Total Haemoglobin Measurement: Oxyhaemoglobin and Dishaemoglobinemias

Total haemoglobin (*ct*Hb or tHb) must be measured because the value is needed to calculate several other blood gas values. Total haemoglobin is a measure of all of the haemoglobin fractions detected by the spectrophotometer of the analyzer. The haemoglobin molecule that is bound to oxygen is known as oxyhaemoglobin, whereas deoxyhaemoglobin or reduced refers to a haemoglobin molecule that does not contain oxygen. Carboxyhaemoglobin contains bound carbon monoxide instead of oxygen, and methaemoglobin is a haemoglobin fraction that contains iron in the ferric (Fe^{3+}) form. These fractions are summarized in ***Table 2***.

Blood gas analyzers measure total haemoglobin spectrophotometrically (17) . Once in the analyzer, the hemolyzer unit hemolyzes or ruptures the red blood cells in an aliquot of the specimen. A portion of the hemolyzed sample is transferred to a measuring chamber, also known as a cuvette. A tungsten halogen lamp or other light source provides polychromatic light, which is directed toward the sample in the cuvette. Depending on the concentration of haemoglobin in the sample, light is transmitted through the hemolyzed sample and toward the spectrophotometer. The specific wavelengths that transmit the color of each haemoglobin

fraction are selected through a monochromater. Transmitted light contacts photodetectors that produce a voltage that corresponds to the amount of light transmitted and photons of light produced. The microprocessor converts the voltage through calculations into the haemoglobin concentrations or fractions. Most blood gas analyzers detect oxyhaemoglobin, as well as deoxyhaemoglobin, carboxyhaemoglobin, and methaemoglobin fractions.

The haemoglobin unit must be calibrated using a known total haemoglobin standard. A calibration curve is electronically developed based on the voltage produced and is sent to the microprocessor. Sample results cannot be reported if the analyzer fails to calibrate successfully. Also, quality control using two different levels of a haemoglobin control must be performed with acceptable results before reporting patient values .(18)

Table 2: Haemoglobin Fractions

Haemoglobin Fraction	Abbreviation	Description	Comments
Total haemoglobin	cthb or thb	Concentration of total haemoglobin or all fractions measured by spectrophotometer	
Oxyhaemoglobin or fraction of oxyhaemoglobin	O_2hb or FO_2hb	Concentration of haemoglobin that is oxygenated	Normal adult haemoglobin includes 1.5–3.5% hba_2, less than 2% hbF, and ~95% hba, which is the major adult haemoglobin
Deoxyhaemoglobin or fraction of deoxyhaemoglobin	hhb or Fhhb	Concentration of haemoglobin that is deoxygenated and is not bound to oxygen	
Carboxyhaemoglobin or fraction of carboxyhaemoglobin	COhb or FCOhb	Concentration of haemoglobin that is combined with carbon monoxide	hb affinity for CO is 200 times higher than for O_2; increased in city dwellers and smokers; extreme elevation and anoxia in carbon monoxide poisoning
Methaemoglobin or fraction of methaemoglobin	Methb or FMethb	Concentration of haemoglobin that contains iron in its ferric (Fe^{3+}) state	Methb cannot bind oxygen; normally less than 1.5% of total hb; increased in cyanosis and hypoxia; causes include exposure to nitrates and/or benzocaine products
Fetal haemoglobin or fraction of fetal haemoglobin	hbF or FhbF	Concentration of haemoglobin F or fetal haemoglobin; hbF can bind oxygen very tightly	Makes up 50–80% of total haemoglobin at birth and less than 2% in adults; the concentration of hbF is increased in some haemoglobinopathies and in some cases of hypoplastic anemia, pernicious anemia, and leukemia
Sulfhaemoglobin or fraction of sulfhaemoglobin	Sulfhb or FSulfhb	Sulfur molecule attaches to haemoglobin and oxygen cannot be transported; may combine with CO to form carboxy-sulfhaemoglobin	Normally less than 2.0%; cyanosis when increased; occupational exposure to sulfur compounds and pollutants

Hematocrit Measurement

The hematocrit, previously known as the packed cell volume (PCV), is the percentage of red blood cells in the whole blood specimen. After centrifuging of a whole blood specimen components of blood is sediminted into three layers., the red blood cells sediment to the bottom, the white blood cells and platelets form a middle layer, and plasma forms the upper layer.

Bicarbonate Content

Bicarbonate constitutes a large fraction of the ions in plasma. Bicarbonate includes true bicarbonate, carbonate, and CO_2 bound in plasma carbamino compounds. True bicarbonates are the largest contributor to bicarbonate content.

Oxygen Content

Oxygen content can be measured directly or calculated by the oxygen content equation.

$$ctO_2 = (Hb \times 1.36 \times sO_2) (0.003 \times pO_2)$$

where:

- ctO_2 is the oxygen content

The Van Slyke equation(19). The base excess is useful in evaluating the patient's acid–base balance in metabolic disorders. A positive base excess occurs when there is a surplus of $HCO3^-$ and a negative base excess when there is a deficit of $HCO3^-$. The calculation of base excess requires the haemoglobin

value, pCO_2, and $HCO3^-$; the base excess at pH of 7.40, pCO_2 of 40 mm Hg, and Hb of 15 g/dL at a temperature of 37°C is zero.

$$\textit{Base excess} = (1.0 - 0.0143\ Hb)(HCO_3^-) - (9.5 + 1.63\ Hb)(7.4 - pH) - 24$$

Today, this value is automatically calculated by the microprocessor in the blood gas analyzer (20). The reference range for base excess in adults is from – 2 to + 3.

Biosensors and Methods Used in the Measurement of Analytes

Whole blood can be analyzed for many analytes, including the electrolytes potassium (K^+), sodium (Na^+), and calcium (Ca^{2+}) and metabolites such as glucose, lactate, blood urea nitrogen (BUN), and creatinine. The sensors used for these measurements are ion-specific or ion-selective electrodes(ISE). These sensors are membrane-based electrochemical transducers that respond to a specific ion. Biosensors are used in analyzers in the traditional clinical laboratory, but also in point-of-care (POC) testing devices. Biosensors use biologically sensitive material that contacts the appropriate transducer responsible for converting the biochemical signal into an electrical signal. **(*Figure 4*)**

Electrolytes are determined by **potentiometric** measurements, a form of electrochemical analysis (21). In potentiometry, the potential or voltage is measured between two electrodes in a solution. These potentials can also be produced when a metal and ions of that

- Hb is the haemoglobin in g/dL
- sO_2 is the oxygen saturation in %

- pO_2 is the partial pressure of oxygen in mmHg

However, the tO_2 can be determined with results obtained from an arterial blood sample using the **CO-oximetry** test panel, which includes fractional concentration of oxyhaemoglobin, reduced haemoglobin, carboxyhaemoglobin, and methaemoglobin. The sum total of these haemoglobin derivatives yields the total haemoglobin concentration. Many current blood gas analyzers either measure or calculate all variables needed to calculate the ctO_2.

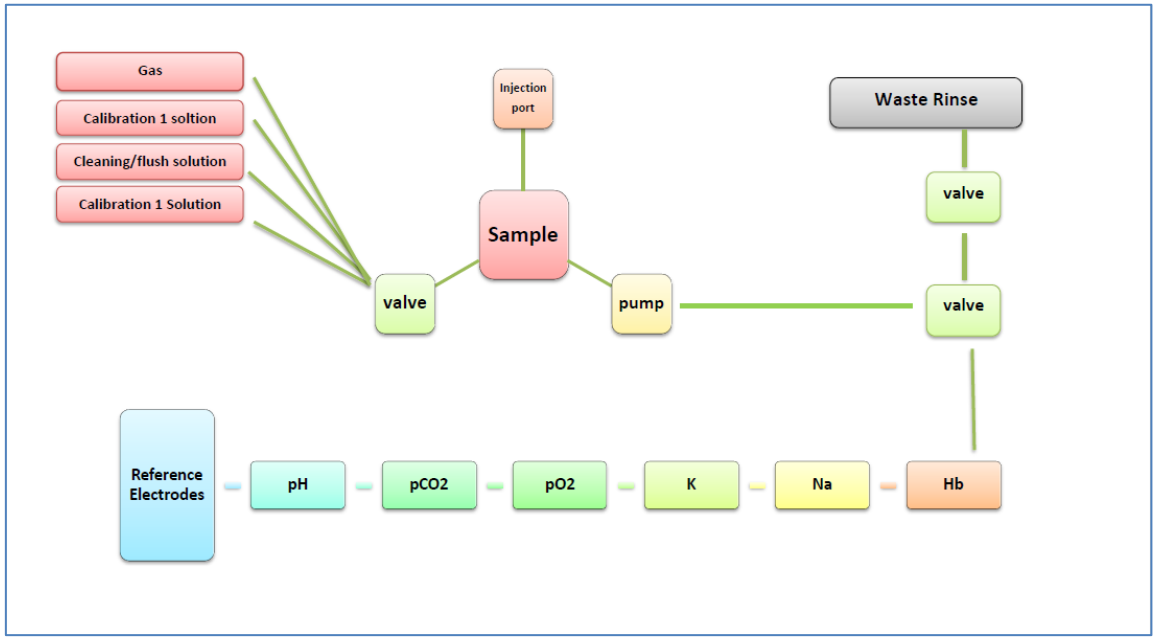

Figure 4: Schematic of a typical analyzer used to measure blood gas and electrolytes.

Base Excess

Base excess can be defined as the concentration of titratable base when a fluid is titrated to a pH of 7.40 at a pCO_2 of 40 mm Hg. However, in practical terms, the base excess is calculated using a nomogram or with metal are present in a solution. By using a membrane that is semipermeable to the ion, different concentrations of the ion can be separated. These systems use a reference and a measuring electrode. A constant voltage is applied to the reference electrode; the difference in voltage between the reference and measuring electrode is used to calculate the concentration of the ion in solution.

Ion-selective electrodes are based on a modification of the principal of potentiometry (21). The potential difference or electron flow is created by selectively transferring the ion to be measured from the sample solution to the membrane phase. The ISE measures the free ion concentration of the desired analyte on a selectively produced membrane. Membranes have a complex composition and contain organic solvents, inert polymers, plasticizers, and ionophors. Ionophors are molecules that increase the membrane's permeability to the specific ion.

Because ISEs produce a direct measurement, there is no need for reagents or the production of a standard curve. Results are precise, accurate, sensitive, and specific for the analyte that is being tested. Ion-selective electrodes are also cost effective, have a rapid analysis time, and are easily maintained and adapted toward automation.

The corresponding membrane component is unique for a specific ISE membrane. For example, the sodium ISE membrane contains silicate in glass; the potassium ISE membrane contains valinomycin; the chloride ISE membrane contains solvent polymeric membranes.

ISEs are also available for chloride (Cl^-), calcium (Ca^{2+}), magnesium (Mg^{2+}), and lithium (Li^+).

Amperometric methods measure the current flow produced from **oxidation-reduction** reactions. Types of **amperometry** include enzyme electrodes, such as the glucose oxidase method and the Clark pO_2 electrode, previously discussed. These types of designs are known as biosensors and are adaptable for testing in the clinical laboratory as well as for point-of-care (POC) testing.

Enzyme-based biosensor technology was first developed to measure blood glucose. A solution of glucose oxidase is placed between the gas-permeable membrane of the pO_2 electrode and an outer membrane that is semipermeable. (22) Glucose in the blood diffuses through the semipermeable membrane and reacts with the glucose oxidase. Glucose is converted by glucose oxidase to hydrogen peroxide and gluconic acid. A polarizing voltage is applied to the electrode, which oxidizes the hydrogen peroxide and contributes to the loss of electrons. Oxygen is consumed near the surface of the pO_2 electrode and its rate of consumption is measured. The loss of electrons and rate of decrease in pO_2 is directly proportional to the glucose concentration in the sample. Enzyme-based biosensors are also used to measure cholesterol, creatinine, and pyruvate.

There are also enzyme-based biosensors with potentiometric and conductimetric detection methods (23). Conductimetric methods utilize chemical reactions that produce or consume ionic substances and alter the electrical conductivity of a solution. In this technology, polymembrane ion-selective electrodes are used. Blood urea nitrogen (BUN), glucose, and creatinine may be measured using this technology. The BUN biosensor immobilizes the enzyme urease at the surface of an ammonium ISE; the urease catalyzes the breakdown

of urea to ammonia (NH_3) and CO_2. Subsequently the ammonia forms ammonium, which is detected by the ISE. The signal produced by the ISE is related to the concentration of blood urea nitrogen in the sample.

Biosensor systems can also use optical detection to measure glucose, bilirubin, and other analytes. The sensors include immobilized enzymes and indicator dyes and may be detected using spectrophotometer, fluorescence, reflectance, or luminescence. (24)

Effect of time delay on Laboratory Results

During the era of evidence based medicine, the medical team depends on the laboratory results for assisting in diagnosis and management of the patients to confirm a clinical impression. Accuracy of measuring the laboratory results depends on multiple factors and steps. The main goal of adequate understanding of each of the steps involved in the process is to achieve nearly optimal conditions and consequently to improve the accuracy and precision of each measurement.

The process involves several steps starting from preparation of the patient, collection of the samples, processing of the samples, estimation by auto analyzer and manual methods, reporting and interpretation of the values. Proper documentation of the samples with details of the patients is of critical importance and is implicit in the process.

Previously, turnaround time needed for labs to be done was not a major focus in clinical laboratories. However, nowadays there is increasing pressure to report results rapidly. To achieve rapid laboratory test (fast-tracking of samples) in emergencies, both technical and non-technical factors should be taken in consideration. World Class Service Industries are concerned to reduce the time delay to improve the technical factors (25–27). On the other hand, many hospitals developed Medical Emergency (MET) teams to improve the non-technical delay. This had a positive impact on expected patient outcomes (25,26). Although data on the actual effectiveness of this approach in reducing mortality is unclear, the deployment of these teams is regarded as “scientifically rational” (27). For example, rapid diagnosis and treatment of sepsis is associated with significantly better outcomes. (28)

For example, Procedures to hasten the turnaround time of laboratory tests have been deployed for Emergency Departments, specifically to improve throughput (29)(15) and, in the UK, to facilitate decision-making within the requisite 4-hour window. For example, point-of care testing has been shown to alleviate the effects of overcrowding on patient safety. (30) At the Royal Sussex County Hospital, samples received from the Emergency Department are fast-tracked to achieve a turnaround time of less than one hour.

Definition of Turnaround Time:

Inspection of the literature reveals a variety of different approaches to definition of TAT. TAT can be classified by test (e.g. potassium), priority (e.g. urgent or routine), population served (e.g. inpatient, outpatient, ED) and the activities included. This last area is the greatest source of variation in reporting of TAT. The steps in performing a laboratory test were outlined by Lundberg, who described the brain to brain TAT or "total testing cycle" as a series of nine steps: ordering, collection of the samples, identification of the samples, transportation, preparation, analysis by auto analyzers or manual methods, reporting the results, interpretation of the values and action. (11,12) The term "therapeutic TAT" is sometimes used to describe the interval between when a test is requested to the time a treatment decision is made. (13–15) Hence the entire process can be viewed as including factors which are – Pre analytical, Analytical and Post analytical. (25)

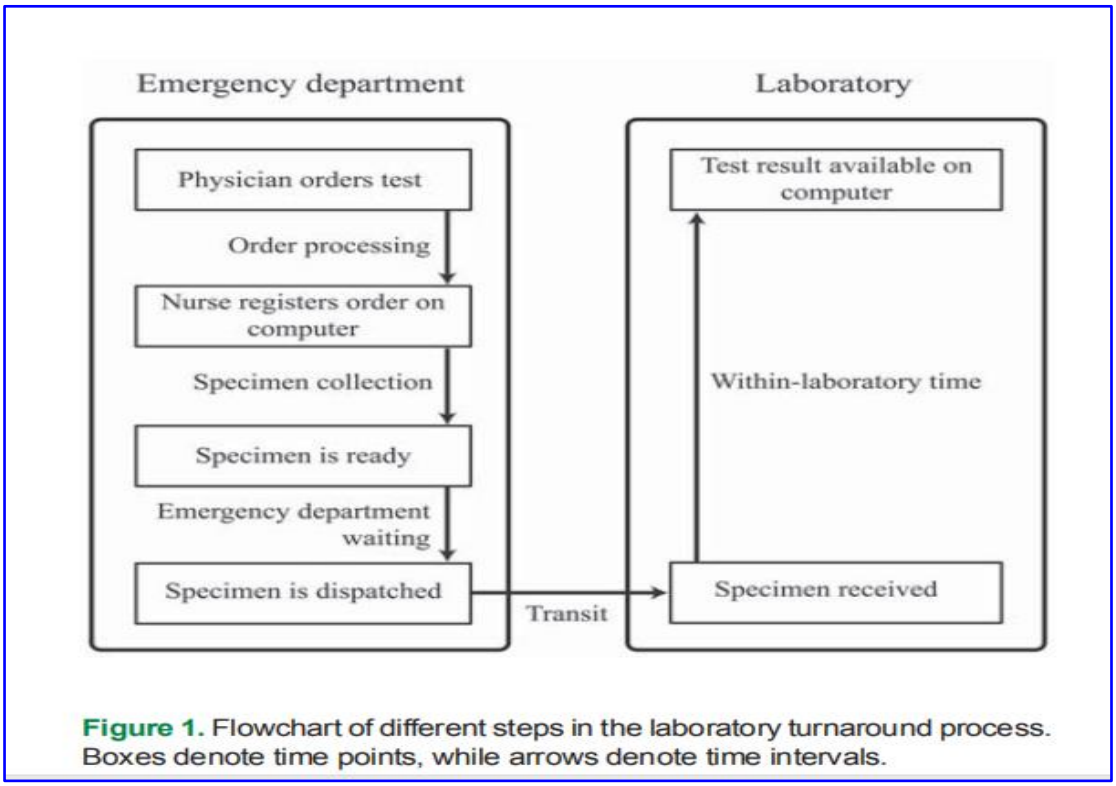

Figure 5: Flowchart of different steps in the laboratory turnaround process.

Importance of Turnaround Time:

Turnaround time (TAT) is one of the most noticeable signs of laboratory service and is often used as a key performance indicator of laboratory performance. This review summarizes the literature regarding laboratory TAT, focusing on the different definitions, measures, expectations, published data, associations with clinical outcomes and approaches to improve TAT. It aims to provide a consolidated source of benchmarking data useful to the laboratory in setting TAT goals and to encourage introduction of TAT monitoring for continuous quality improvement. A 90% completion time (sample registration to result

reporting) of for common laboratory tests is suggested as an initial goal for acceptable TAT.

The impact of turnaround time (TAT), has been reported to prolong length of hospital stay (LOS) in high-volume patient care settings such as the Emergency Department(ED). The TAT OP has a direct and significant relationship with patient wait times in the emergency department. Moreover, our findings indicate that improvement in core laboratory TAT OP will reduce ED patient LOS substantially. In addition, we found that the older methods for monitoring laboratory performance using test TAT means are not particularly useful in assessing the laboratory's impact on patient care. We conclude that the laboratory has a clear and significant role in the effective practice of real time, evidence-based medicine in the critical care setting.

Improving turnaround time (TAT) is a complex task involving education, equipment acquisition, and planning. All the steps from test ordering to results reporting should be monitored and steps taken to improve the processes.

Proper documentation of the samples with details of the patients is of critical importance and is implicit in the process

Clinical Outcome of Delay of Turnaround Time on samples:

Errors in pre-analytical, analytical and post-analytical practices account for 32 – 75% of laboratory errors.(26) It is a standard practice to process the serum immediately after the blood specimens reach the laboratory and proceed with the assay. However, the samples arrival in the laboratory is delayed due to transporting. The analysis may also be delayed due to the increased work load or the casual attitude of the technicians. The adverse effects of prolonged serum-clot contact time were known long back and immediate separation of the

serum from cells was advised. During a prolonged serum clot contact time, both the biological activity of the cells and trans-membrane diffusion can change the concentration of serum electrolytes. transport from the collection centre to the central lab.

Consequently, sample lysis takes place. Not only, there were significant alterations in the values of serum electrolytes on hemolysis (the most common analytes requested in the clinical biochemistry laboratory) but also, the significant alterations in the values of glucose and liver enzymes e.g. Aspartate transaminase when there was delay of 4 hours in estimation at room temperature.

Multiple projects arose from anecdotal concerns that delayed reporting of blood samples for critically ill patients was slowing the process of diagnosis and management, and therefore having a serious impact on patient care.

Timing Guidelines

Blood gas analysis is an important test in many medical situations. Either arterial blood gases or venous blood gases are done in nearly all critically ill patients. Usually heparinized samples are withdrawn in plastic syringes and analyzed in the blood gas analyzer. Before using a clinical parameter in therapeutic decisions, its accuracy is important.

Unfortunately, previous studies have found that the values of arterial blood gases were influenced by time delay and sample storage temperature in blood gas analysis

Stability of withdrawn samples for laboratory tests was tested in previous studies and reported to be stable in general, so out-of hospital withdrawn samples can be relied on. Delays may occur in analysis of a drawn sample in the hospital due to the limited resources and blood gas analyzers are not available in many areas in the hospital.

Stability of blood gases especially partial pressure of oxygen (PO_2) was a subject of debate and conflicting data. While some studies reported a decrease in PO_2 with delay of analysis, others claimed that PO_2 will increase with time delay, as atmospheric oxygen will penetrate plastic material of syringes.

Immediate

American Association for Respiratory Care. AARC recommended that the blood samples should be analyzed immediately after sampling (31,32)

Within 5 min

Ghanpur R et al., in 2016, recommend venous blood gas samples to be analyzed within 5 minutes. If a delay of >5 minutes cannot be avoided glass syringes should be used.(31)

Within 10 min

The American Association for Respiratory Care Clinical Practice Guideline stated that the specimen should be analyzed within 10-15 minutes of drawing if held at room temperature or within 1 hour for the iced sample' (31,32)

Chillar RK. Et al., in1980 , suggested that if a delay of more than 10 minutes is anticipated, the specimen must be immersed in an ice bath .Leukocytes and platelets continue to consume oxygen in the sample after it is drawn and can cause a significant fall in PaO_2 over time at room temperature, especially in the setting of leukocytosis or thrombocytosis . Cooling will prevent any clinically important effect for at least 1 hour by decreasing the metabolic activity of these cells. (33)

Within 15 min

Srisan P.,et al. , in 2011 ,suggested that for ABG and electrolytes analysis, the blood sample should be analyzed within 15 minutes and be stored at either room temperature or on ice. (34)

Within 30 min

The guideline of the American Association for Respiratory Care (AARC) states that a delay in analysis of a venous blood gas of 30 minutes is acceptable as reported by Davis MD et al. in 2013.(35) In the case of samples that must be

kept for longer than 30 min, they should be drawn and stored in a glass vessel and chilled to 0 – 4°C.

Clinical and Laboratory Standards Institute (formerly the National Committee for Clinical Laboratory Standards) recommends that samples taken in plastic syringes should not be iced, but instead kept at room temperature and analyzed within 30 minutes. If the analysis is delayed more than 30 minutes, glass syringes and coolant immersion are recommended. (36–39)

Within 60 min

A research article done by Elham Mohammadhoseini et al., in 2015 , to investigate the effects of samples storage temperature and time delay on blood gases, bicarbonate and PH results in human arterial blood samples and claimed that it is not necessary to store samples in iced water when analysis delayed up to one hour. (40)

It remains an acceptable practice for hospitals to routinely transport blood gas samples to central laboratories for analysis (41–43) and is reported by several studies.

Aim of the study

The objectives of this observational study are:

- To detect the effect of time delay and syringe surface area on PO2 in arterial and venous samples
- To detect the effect of time delay and syringe surface area on SO2 in arterial and venous samples
- To detect the effect of time delay and syringe surface area on other parameters of blood gases e.g. PH,CO2

Patients and Methods

Study design

Observational Study in which samples from ICU and Intraoperative patients were collected and analyzed.

Study setting and location

It was conducted at Theodor Bilharz Research Institute and Kasr Al-Ainy hospital, Faculty of medicine, Cairo University.

Study population

ICU and Major Intraoperative patients

Eligibility Criteria

Inclusion criteria

- Age above 18 years
- Critically ill-patients
- Intraoperative patients (Major Surgeries)

Exclusion criteria

- Haematological diseases e.g. sickle cell anaemia ,
- Burn patients with risk of carbon monoxide poisoning.
- ARDS
- Pulmonary oedema

Criteria for subject or study discontinuation:

- if any sample is clotted
- error in results of components of blood gases
- Presence of air bubbles in any sample

Study Procedures

Study Protocol

The methodology should include:

After approval from ethics and research committee, consents were obtained from every patient.

Any critically ill patient or intraoperative patient who fulfilled the inclusion criteria was enrolled in the study. An arterial sample (from arterial line connected to 20G cannula) and a central venous sample (from a central venous catheter) were withdrawn. Each sample was 5ml and was divided into five parts: 1ml of blood in a heparinized syringe was analyzed immediately, 1ml of blood in a 3ml plastic syringe (2 syringes) , 1ml of blood in a capillary insulin syringe (2 syringes). The last 4 samples were analyzed after 30 mins and 1hour respectively. The samples while waiting analysis were left in ambient temperature. This was done to the arterial and venous samples simultaneously. This means that every patient had 10 samples analyzed and subjected to final analysis.

Measurement tools

GEM Premier 3000 system with *Intelligent Quality Management (iQM)*

Measured Analytes: pH, pCO_2, pO_2, Na+, K+, Ca++, Glu, Lac, Hct. Derived (Calculated) Parameters:$HCO3^-$, $HCO3^-$ std, TCO_2, BE(B) *in vitro*, BE(ecf) *in vivo*, sO_2c, Ca++ (7.4), THbc, A-aDO_2, pAO_2, paO_2/pAO_2, RI, CaO_2, CvO_2, CcO_2, a-vDO_2, Qsp/Qt, P50

Study Outcomes

Primary outcome

The change in PO2 in arterial blood sample after 1 hour.

Secondary outcomes

- The change of PO2 in venous blood samples.
- The change in SO2 after 30 min and 1 hour in arterial and venous samples.
- The difference in PO2 and SO2 after 30 min and 1 hour between 3 ml syringe and capillary syringe.
- The change in the other parameters of blood gases e.g. PH, CO2

Statistical Analysis:

Sample size

Sample size was calculated using G-power software. The arterial partial pressure of oxygen was derived from a previous study and was 96.4±6 mmHg (9) we assumed that time delay will change this PO2 by at least 5 mmHg to be clinically significant. Considering a study power of 95% and a p-value of 0.05 to be significant, the sample size was calculated to be 45 blood samples.

Statistical analysis

Data will be presented as mean±SD (if numerical and normally distributed) and with median (range) (if not normally distributed). Categorical data will be presented as number and frequency. Paired t- test will be used to compare data if normally distributed, as it is one group of patient. Wilcoxon non-parametric test will be used if the data are not normally distributed. Categorical variables with the $\chi 2$ test. Pearson correlation was done and Bland-Altman was used to assess the agreement between 30 min and 60 min samples with baseline. The level of significance will be set at $P < 0.05$ for two-tailed tests. Statistical analysis will be performed using SPSS for Windows version 15.0 (SPSS Inc., Chicago, IL, USA).

Results

50 patients were screened in our study and 5 patients did not meet the inclusion criteria. Therefore, a total of 45 sets of samples were collected (10 for each patient). Each set of samples included 5 arterial and 5 venous samples. The arterial as well as the venous samples were as follows: one baseline sample, two samples delayed for 30 minutes of collection and two samples delayed for 60 minutes of collection. The two samples delayed for 30 minutes were collected one in plastic 3ml syringe and one in an insulin plastic syringe. This was done also for the samples of 60 minutes delay.

Table 3: Oxygenation variables. Data are presented as mean ± standard deviation.

	Baseline	30 minutes	60 minutes
Oxygen saturation (arterial sample) **- 3 mL syringe** **- Insulin syringe**	98 ± 2	- 97.9 ±1.8 - 95.6 ±13.3	- 96.8 ±3.1 - 96.6 ±2.9
Oxygen saturation (venous sample) **- 3 mL syringe** **- Insulin syringe**	73± 14.5	- 72 ±13.5 - 72 ±12.9	- 72 ±13 - 71 ±12.5
Oxygen tension (arterial sample) **- 3 mL syringe** **- Insulin syringe**	159.9± 113.6	- 138.6 ±66.8 - 137.4 ±75.1	- 122 ±56.6 - 116.6 ±52.6
Oxygen tension (venous sample) **- 3 mL syringe** **- Insulin syringe**	50 ± 46	- 45 ±14.4 - 44 ±11.8	- 45 ±11.3 - 44 ±10.2

Regarding oxygenation variables in arterial samples, ***the mean baseline PO2*** was 159.9±113.6. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were 21(-99to142) and 38 (-112 to 188) respectively in the 3 ml syringe. The same variables were 23(-66to 111) and 43(-103to 190) respectively in ±the insulin syringe.

The mean baseline SO2 was 98±2. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were 0.1(-2.3to2.4) and 1.3 (-3.4 to 5.9) respectively in the 3 ml syringe. The same variables were 0.4(-1.7 to2.5) and 1.4(-2 to 4.7) respectively in the insulin syringe.

Regarding venous sample, ***the mean baseline SO2*** was 73±14.5.The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were 0.3(-11.3 to 11.9) and 0.6(-13.3 to14.6) respectively in the 3 ml syringe. The same variables were 0.6(-11.5to 12.7) and 1.9(-11.2 to14.9) respectively in the insulin syringe.

Table 4: Correlation coefficient (r), mean bias, and limits of agreement between the baseline oxygenation and haemoglobin values with each of 30-minute values and 60-minute values.

	30 minutes		60 minutes	
	r (95% CI)	**Mean bias (limits of agreement)**	**r (95% CI)**	**Mean bias (limits of agreement)**
PaO2 (arterial samples)				
3ml	0.95 (0.91 – 0.97)	21 (-99 to 142))	0.88 (0.79 – 0.93)	38 (-112 to 188)
Insulin	0.95 (0.91 to 0.97)	23 (-66 to 111)	0.91 (0.84 to 0.95)	43 (-103 to 190)
SaO2 (all samples)				
3ml	0.95 (0.93 – 0.97)	0.2 (-8.1 to 8.5)	0.94 (0.91 – 0.96)	0.9 (-9.4 to 11.3)
Insulin	0.96 (0.95 – 0.98)	0.5 (-8.1 to 9.2)	0.95 (0.93 – 0.97)	1.6 (-7.8 to 11.1)
SaO2 (arterial samples)				
3ml	0.84 (0.72 to 0.91	0.1 (-2.3 to 2.4)	0.8 (0.66 – 0.89)	1.3 (-3.4 to 5.9)
Insulin	0.88 (0.78 – 0.93)	0.4 (-1.7.1 to 2.5)	0.81 (0.68 – 0.89)	1.4 (-2 to 4.7)
SaO2 (venous samples)				
3ml	0.83 (0.71 – 0.9)	0.3 (-11.3 to 11.9)	0.77 (0.62 – 0.87)	0.6 (-13.3 to 14.6)
Insulin	0.86 (0.75 – 0.92)	0.6 (-11.5 to 12.7)	0.83 (0.7 – 0.9)	1.9 (-11.2 to 14.9)

Table 5: Acid base variables. Data are presented as mean ± standard deviation.

	baseline	30 minutes	60 minutes
***p*H (all samples)** **- 3 mL syringe** **- Insulin syringe**	7.39 ±0.09	7.36 ±0.09 7.36 ±0.09	7.33 ±0.09 7.34 ±0.1
***p*H (arterial samples)** **- 3 mL syringe** **- Insulin syringe**	7.41 ±0.09	7.39 ±0.09 7.39 ±0.09	- 7.36 (0.1) - 7.36 (0.1)
***p*H (venous samples)** **- 3 mL syringe** **- Insulin syringe**	7.36 ±0.08	7.33±0.09 7.33 ±0.09	7.31±0.09 7.31 ±0.09
PCO2 (all samples) **- 3 mL syringe** **- Insulin syringe**	39.2±8.4	40.1±8.5 40.1 ±8.0	42.8±8.5 41.8 ±8.6
PCO2 (arterial samples) **- 3 mL syringe** **- Insulin syringe**	36 ±7.6	36.5±7.6 36.5 ±6.8	39.8±8.0 38.4±7.2
PCO2 (venous samples) **- 3 mL syringe** **- Insulin syringe**	42.4 ±8	43.8 ±7.7 43.8 ±7.6	45.9 ±7.9 45.2 ±8.6

Regarding acid base variables in arterial samples, ***the mean baseline PH***_was 7.41±0.09. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were 0.02(-0.02 to 0.06) and 0.06 (-0.03 to0.14) respectively in the 3 ml syringe. The same variables were 0.02(-0.02 to0.07) and 0.05(-0.03 to 0.12) respectively in the insulin syringe.

The mean baseline PCO2_was 36±7.6. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were -0.4(-4.8 to4.1) and -3.8(-11.8 to 4.1) respectively in the 3 ml syringe. The same variables were -0.6(-5.4 to 4.2) and -2.4(-7.6 to 2.8) respectively in the insulin syringe

In venous samples, ***the mean baseline PH***_was 7.36±(0.08). The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were 0.03(-0.04 to 0.1)and 0.05(-0.04 to 0.13) respectively in the 3 ml syringe. The same variables were 0.03(-0.04 to 0.07) and 0.05(-0.03 to 0.12) respectively in the insulin syringe.

The mean baseline PCO2_was 42.4±8. The mean Bias (limits of agreement) of the 30 minute value (from baseline) and the 60 minute value (from baseline) were -1.4(-8.5 to 5.8) and -3.5(-12.5 to 5.6) respectively in the 3 ml syringe. The same variables were -1.3(-9.7 to 7) and -2.8(-12.5 to7) respectively in the insulin syringe.

Table 6: Correlation coefficient (r), mean bias, and limits of agreement between the baseline acid-base values with each of 30-minute values and 60-minute values.

	30 minutes		60 minutes	
	r (95% CI)	Mean bias (limits of agreement)	r (95% CI)	Mean bias (limits of agreement)
***p*H (All samples)**				
3ml	0.95 (0.92 – 0.97)	0.02 (-0.03 to 0.08)	0.89 (0.84 – 0.93)	0.05 (-0.03 to 0.14)
Insulin	0.95 (0.93 – 0.97)	0.03 (-0.03 to 0.08)	0.9 (0.86 – 0.94)	0.05 (-0.03 to 0.13)
***p*H (arterial samples)**				
3ml	0.98 (0.96 – 0.99)	0.02 (-0.02 to 0.06)	0.9 (0.83 – 0.95)	0.06 (-0.03 to 0.14)
Insulin	0.95 (0.92 – 0.98)	0.02 (-0.02 to 0.07)	0.91 (0.84 – 0.95)	0.05 (-0.03 to 0.12)
***p*H (venous samples)**				
3ml	0.91 (0.84 – 0.95)	0.03 (-0.04 to 0.1)	0.88 (0.79 – 0.93)	0.05 (-0.04 to 0.13)
Insulin	0.92 (0.86 – 0.96)	0.03 (-0.04 to 0.1)	0.88 (0.8 – 0.94)	0.05 (-0.03 to 0.13)
PCO2 (all samples)				
3ml	0.92 (0.88 – 0.95)	-0.9 (-6.9 to 5.1)	0.83 (0.75 – 0.88)	-3.6 (-12.1 to 4.8)
Insulin	0.89 (0.84 – 0.93)	-1 (-7.8 to 5.8)	0.89 (0.8 – 0.94)	-2.6 (-10.4 to 5.2)
PCO2 (arterial samples)				
3ml	0.94 (0.89 – 0.97)	-0.4 (-4.8 to 4.1)	0.83 (0.71 – 0.91)	-3.8 (-11.8 to 4.1)
Insulin	0.93 (0.88 – 0.96)	-0.6 (-5.4 to 4.2)	0.92 (0.85 – 0.95)	-2.4 (-7.6 to 2.8)
PCO2 (venous samples)				
3ml	0.85 (0.74 – 0.92)	-1.4 (-8.5 to 5.8)	0.77 (0.62 – 0.87)	-3.5 (-12.5 to 5.6)
Insulin	0.8 (0.66 – 0.89)	-1.3 (-9.7 to 7)	0.81 (0.67 – 0.89)	-2.8 (-12.5 to 7)

Discussion

Our findings were that delay of blood gas samples for 30 minutes or 60 minutes at room temperature resulting in a significant difference from baseline samples in pO_2, pCO_2 and oxygen saturation.

Suggested physiological background behind this finding is that red blood cells and white blood cells consume oxygen and produce CO_2 during this time delay. The metabolism of blood cells, both aerobic and anaerobic, continues in stored blood. The CO_2 production from both aerobic and anaerobic metabolism leads to an increase in PCO_2 and a decrease in pH. In addition, decrease in pH may be due to lactate produced by anaerobic respiration of mature red blood cells. (44–46)

In agreement with our results, Srisan et al found that the pH, PaO2 significantly decreased while the PaCO2 significantly increased from baseline values. The decrease in pH was statistically significant from 30 minutes onwards at room temperature and from 45 minutes onwards on ice. (34)

Rajasekaran R et al also reported that pO_2 decreased by time delay at room temperature on samples stored at room temperature at 15, 30, 45 and 60 min when compared with baseline were 1.80 (-18 to 21.7), 3.84 (-20.5 to 28.2), 3.40 (-18.8 to 25.6) and 4.36 (-17.5 to 26.2) respectively. (47)

It is also stated in ***AARC Clinical Practice Guideline 2013,*** that since PaO_2 in samples drawn from subjects with very high leukocyte counts can decrease rapidly, immediate analysis is crucial in this type of patients. (35)

As pointed by ***Scott M et al***, delay in analysis of blood gases resulted in decrease of pO_2 by 2 mmHg/hr. at room temperature (or greater with elevated

white blood cells) (48,49) increase of pCO_2 by 1 mmHg/hr. at 22C (or greater with elevated white blood cells) (48,49) decrease of pH by 0.02-0.03 pH units/hr. at 22C (or greater with elevated white blood cells) (48,49)

Biswas et al reported that PO_2, decreased significantly by 20 minutes at 4°C (refrigerator) and 22°C (room temperature). In addition, there was no significant change in pH and PCO_2, for up to 30 minutes at 4°C and 22° C. (50)

Smeenk FW, et al found that .PaO_2 in plastic syringes deteriorated slightly but significantly (1.1 kPa; p=0.037 when compared to those in glass ones while there was significant increase in the $PaCO_2$ values in plastic syringes when compared with that in glass syringes. (51)

Nanji et al reported that decrease in pH, pO_2 and increase in PCO_2 kept at room temperature showed significant difference from 20 minutes onwards. (52)

Pretto et al have shown that PO_2, declined at an average rate of 0.33 mmHg/minute (19.8 mmHg/hour) on ice and 1.37 mmHg/minute (82.2 mmHg/hour) at 22°C. The changes in PCO_2 were less dramatic than those of $PO_2$with the average increment of 0.71 kPa (5.3 mmHg) over 2 hours. (53)

Beaulieu et al found decreases in PaO_2 when plastic syringes were stored at room temperature for 30 min, and increases when plastic syringes were stored in ice for 30 min. (54)

On the contrary, ***Wan XY et al*** reported that there is a trend of increasing values for $ScvO_2$, Pv-aCO_2, and Pv-aCO_2/Ca-vO_2 over time ($P < 0.001$). $ScvO_2$ and Pv-aCO_2/Ca-vO_2 values of the samples stored for 10, 20, 30, 40, 50 and 60 min were significantly higher relative to baseline ($P < 0.05$). (55)

Another study by ***Ghanpur R*** reported that SvO_2 increased with time delay and suggested that the plastic material of syringes is permeable to atmospheric oxygen. (31)

In addition, ***Knowles TP et al*** reported that compared to the samples that were analyzed immediately, the PO_2 of the samples stored in plastic syringes for 30 min at 22 degrees C and at 0-4 degrees C was significantly higher, with a clinically important magnitude of 11.9-13.7 mm Hg and no significant difference in pH or oxygen saturation among groups (37)

Picandet V et al found that blood collected in either of the plastic syringes resulted in a variation >10 mm Hg after 10 +/- 3 to 17 +/- 2 minutes, depending on the storage conditions. Plastic syringes kept at ambient temperature offered more stability for $PaCO_2$ analysis .There was no significant change in pH. (56)

Mahoney et al also found significant PO_2 increases when the samples were iced (8.4 mm Hg increase) and when they were stored at room temperature (2.6 mm Hg increase). (57)

Liss and Payne found significant PaO_2 increases in samples stored for 30 min at room temperature (2.4 mm Hg increase) and on ice (3.0 mm Hg increase). (36)

Schmidt and Muller-Plathe also studied the stability of blood gases and found that in plastic syringes the greatest increases for pO_2 occurred after storage at 4 degrees C, which can be explained by the increased solubility of oxygen and the higher O_2 affinity of haemoglobin at 4 degrees C. When stored at room temperature, the deviations in plastic syringes were smaller. The deviations of pCO_2 and pH were relatively small. (6)

Limitations

Limitations were:

We did not freeze samples because the delay that may occur in our unit mostly occur in room temp. The second limitation is that we checked changes at two time points only because if we do more, the sample number will be huge per patient.

Conclusion

We conclude that arterial and venous samples that are delayed at room temperature give different results from baseline results (analyzed immediately). This change occurs as early as 30 minutes.

The change was mainly decrease in PO_2 and increase in PCO_2.

We recommend that blood gas analysis should be done immediately to obtain reliable measurements.

References

1. Van Slyke DD. An Apparatus for Determination of the Gases in Blood and Other Solutions. Proc Natl Acad Sci U S A. 1921 Aug;7(8):229–31.

2. CLSI. Blood gas and pH analysis and related measurements; approved guidelines. CLSI document C46-A2. 2009;

3. Skurup Anne, Kristensen Tina, Wennecke Gitte. New creatinine sensor for point-of-care testing of creatinine meets the National Kidney Disease Education Program guidelines. cclm. 2007;46(1):3.

4. Bénéteau-Burnat Bénédicte, Pernet Pascal, Pilon Antoine, Latour Damien, Goujon Stéphanie, Feuillu Alain, et al. Evaluation of the GEM® Premier™ 4000: a compact blood gas CO-Oximeter and electrolyte analyzer for point-of-care and laboratory testing. cclm. 2007;46(2):271.

5. Greenbaum R, Nunn JF, Prys-Roberts C, Kelman GR. Metabolic changes in whole human blood (in vitro) at 37°C. Respiration Physiology. 1967 May 1;2(3):274–82.

6. Schmidt C, Müller-Plathe O. Stability of pO 2 , pCO 2 and pH in Heparinized Whole Blood Samples: Influence of Storage Temperature with Regard to Leukocyte Count and Syringe Material. Clinical Chemistry and Laboratory Medicine [Internet]. 1992 [cited 2019 Oct 18];30(11). Available from: https://www.degruyter.com/view/j/cclm.1992.30.issue-11/cclm.1992.30.11.767/cclm.1992.30.11.767.xml

7. Scott PV, Horton JN, Mapleson WW. Leakage of Oxygen from Blood and Water Samples Stored in Plastic and Glass Syringes. BMJ. 1971 Aug 28;3(5773):512.

8. Plebani M. The detection and prevention of errors in laboratory medicine. Ann Clin Biochem. 2009 Dec 1;47(2):101–10.

9. Hawkins R. Managing the Pre- and Post-analytical Phases of the Total Testing Process. Ann Lab Med. 2012 Jan;32(1):5–16.

10. Thomson JM. Blood collection and preparation: pre-analytical variation. In: Jespersen J, Bertina RM, Haverkate F, editors. ECAT Assay Procedures A Manual of Laboratory Techniques: European Concerted Action on Thrombosis and Disabilities of the Commission of the European

Communities [Internet]. Dordrecht: Springer Netherlands; 1992. p. 13–20. Available from: https://doi.org/10.1007/978-94-011-2992-3_2

11. Tietz N, Pruden L, Andersen S. Electrolytes In: Tietz, NW (ed.), Fundamentals of Clinical Chemistry. 1996;

12. ADAMS AP, MORGAN-HUGHES JO, SYKES MK. pH and blood—gas analysis. Anaesthesia. 1967 Oct 1;22(4):575–97.

13. Beetham R. A Review of Blood pH and Blood-Gas Analysis. Ann Clin Biochem. 1982 Jul 1;19(4):198–213.

14. Severinghaus JW, Astrup PB. History of blood gas analysis. I. The development of electrochemistry. Journal of Clinical Monitoring. 1985 Jul 1;1(3):180–92.

15. Severinghaus JW, Astrup PB. History of blood gas analysis. III. Carbon dioxide tension. Journal of Clinical Monitoring. 1986 Jan 1;2(1):60–73.

16. Reynafarje B, Costa LE, Lehninger AL. O2 solubility in aqueous media determined by a kinetic method. Analytical Biochemistry. 1985 Mar 1;145(2):406–18.

17. Gehring H, Duembgen L, Peterlein M, Hagelberg S, Dibbelt L. Hemoximetry as the "Gold Standard"? Error Assessment Based on Differences Among Identical Blood Gas Analyzer Devices of Five Manufacturers. Anesthesia & Analgesia [Internet]. 2007;105(6). Available from: https://journals.lww.com/anesthesia-analgesia/Fulltext/2007/12001/Hemoximetry_as_the__Gold_Standard___Error.5.aspx

18. TOOBIAK S, SHER EA, SHAKLAI M, SHAKLAI N. Precise quantification of haemoglobin in erythroid precursors and plasma. International Journal of Laboratory Hematology. 2011 Dec 1;33(6):645–50.

19. Kazmierczak Steven C. Laboratory Quality Control: Using Patient Data to Assess Analytical Performance. cclm. 2005;41(5):617.

20. Siggaard-Andersen O, Wimberley PD, Fogh-Andersen N, Gøthgen IH. Measured and derived quantities with modern pH and blood gas equipment: Calculation algorithms with 54 equations. Scandinavian Journal of Clinical and Laboratory Investigation. 1988 Jan 1;48(sup189):7–15.

21. Lang W, Zander R. The accuracy of calculated base excess in blood. Clinical chemistry and laboratory medicine. 2002;40(4):404–10.

22. Albert V, Subramanian A, Rangarajan K, Pandey RM. Agreement of two different laboratory methods used to measure electrolytes. Journal of laboratory physicians. 2011;3(2):104.

23. Ronkainen NJ, Halsall HB, Heineman WR. Electrochemical biosensors. Chemical Society Reviews. 2010;39(5):1747–63.

24. D'Orazio P. Biosensors in clinical chemistry. Clinica Chimica Acta. 2003;334(1–2):41–69.

25. Rauch CA, Nichols JH. Laboratory Accreditation and Inspection. Clinics in Laboratory Medicine. 2007 Dec 1;27(4):845–58.

26. Kiehlbauch JA, Hannett GE, Salfinger M, Archinal W, Monserrat C, Carlyn C. Use of the National Committee for Clinical Laboratory Standards guidelines for disk diffusion susceptibility testing in New York state laboratories. J Clin Microbiol. 2000 Sep;38(9):3341–8.

27. Browning J, Kaiser D, Durbin C. The effect of guidelines on the appropriate use of arterial blood gas analysis in the intensive care unit. Respir Care. 1989;34(4):269–76.

28. Jordan A. Blood gas: a brief anecdotal history by one who has been there. MLO: medical laboratory observer. 2012;44(9):30.

29. Li L, Georgiou A, Vecellio E, Eigenstetter A, Toouli G, Wilson R, et al. The effect of laboratory testing on emergency department length of stay: a multihospital longitudinal study applying a cross-classified random-effect modeling approach. Academic Emergency Medicine. 2015;22(1):38–46.

30. Shapiro BA, Mahutte CK, Cane R, Gilmour I. Clinical performance of a blood gas monitor: a prospective, multicenter trial. Critical care medicine. 1993;21(4):487–94.

31. Ghanpur R, Santamaria J, Dixon B. Plastic Blood Gas Syringes and Measurement Error in Central Venous Oxygen Saturations: SHOCK. 2016 Sep;46(3):287–9.

32. AARC Clinical Practice Guideline: Blood Gas Analysis and Hemoximetry: 2001 Revision & Update. 2001. 46(5):498–505.

33. Chillar RK, Belman MJ, Farbstein M. Explanation for apparent hypoxemia associated with extreme leukocytosis: leukocytic oxygen consumption. Blood. 1980;55(6):922–4.

34. Srisan P, Udomsri T, Jetanachai P, Lochindarat S, Kanjanapattanakul W. Effects of Temperature and Time Delay on Arterial Blood Gas and Electrolyte Measurements. 2011;94:7.

35. Davis MD, Walsh BK, Sittig SE, Restrepo RD. AARC Clinical Practice Guideline: Blood Gas Analysis and Hemoximetry: 2013. Respiratory Care. 2013 Oct 1;58(10):1694–703.

36. Hess CE, Nichols AB, Hunt WB, Suratt PM. Pseudohypoxemia secondary to leukemia and thrombocytosis. New England Journal of Medicine. 1979;301(7):361–3.

37. Knowles TP, Mullin RA, Hunter JA, Douce HF. Effects of Syringe Material, Sample Storage Time, and Temperature on Blood Gases and Oxygen Saturation in Arterialized Human Blood Samples. RESPIRATORY CARE. 2006;51(7):5.

38. Dukić L, Milevoj Kopčinović L, Dorotić A, Baršić I. Blood gas testing and related measurements: National recommendations on behalf of the Croatian Society of Medical Biochemistry and Laboratory Medicine. Biochem Med. 2016;318–36.

39. Blonshine S, National Committee for Clinical Laboratory Standards. Procedures for the collection of arterial blood specimens: approved standard. Wayne, Pa.: NCCLS; 2004.

40. Mohammadhoseini E, Safavi E, Seifi S, Seifirad S, Firoozbakhsh S, Peiman S. Effect of Sample Storage Temperature and Time Delay on Blood Gases, Bicarbonate and pH in Human Arterial Blood Samples. Iran Red Crescent Med J [Internet]. 2015 Mar 20 [cited 2019 Oct 18];17(3). Available from: http://ircmj.com/en/articles/16104.html

41. Kilgore M, Steindel S, Smith J. Estimating costs and turnaround times: presenting a user-friendly tool for analyzing costs and performance. Clinical laboratory management review: official publication of the Clinical Laboratory Management Association. 1999;13(4):179–87.

42. Jaschke K, Brown D, Clark A, Doull S, English A, Hoover N, et al. Speed of blood withdrawal and accurate measurement of oxygen content in mixed venous blood. American Journal of Critical Care. 2014;23(6):486–93.

43. Goswami B, Singh B, Chawla R, Gupta V, Mallika V. Turn around time (TAT) as a benchmark of laboratory performance. Indian Journal of Clinical Biochemistry. 2010;25(4):376–9.

44. Foster JM, Terry ML, Gunther H. Studies on the Energy Metabolism of Human Leukocytes. Blood. 1967 Aug 1;30(2):168–75.

45. Cline MJ. Metabolism of the Circulating Leukocyte. Physiological Reviews. 1965 Oct 1;45(4):674–720.

46. Lenfant C, Aucutt C. Oxygen uptake and change in carbon dioxide tension in human blood stored at 37 C. Journal of Applied Physiology. 1965 May 1;20(3):503–8.

47. Rajasekaran R, Arthur HS, Peter JV. Arterial blood gas tensions - Effect of storage time and. :1.

48. Baird G. Preanalytical considerations in blood gas analysis. Biochem Med. 2013;19–27.

49. SCOTT M. Electrolytes and blood gases. Tietz textbook of clinical chemistry [Internet]. 1999; Available from: https://ci.nii.ac.jp/naid/10018966491/en/

50. Biswas CK, Ramos M, Agroyannis B. Blood gas analysis: effect of air bubbles in syringe and delay in estimation. 1982;284:5.

51. Smeenk F, Janssen J, Arends B, Harff G, van den Bosch J, Schonberger J, et al. Effects of four different methods of sampling arterial blood and storage time on gas tensions and shunt calculation in the 100% oxygen test. Eur Respir J. 1997 Apr 1;10(4):910.

52. Nanji AA, Whitlow KJ. Is it necessary to transport arterial blood samples on ice for pH and gas analysis? Can Anaesth Soc J. 1984 Sep;31(5):568–71.

53. Pretto JJ, Rochford PD. Effects of sample storage time, temperature and syringe type on blood gas tensions in samples with high oxygen partial pressures. Thorax. 1994 Jun 1;49(6):610.

54. Beaulieu M, Lapointe Y, Vinet B. Stability of Po2, Pco2, and pH in fresh blood samples stored in a plastic syringe with low heparin in relation to various blood-gas and hematological parameters. Clinical Biochemistry. 1999 Mar 1;32(2):101–7.

55. Wan X, Wei L, Jiang Y, Li P, Yao B. Effects of time delay and body temperature on measurements of central venous oxygen saturation, venous-arterial blood carbon dioxide partial pressures difference, venous-arterial blood carbon dioxide partial pressures difference/arterial-venous oxygen difference ratio and lactate. BMC Anesthesiol. 2018 Dec;18(1):187.

56. Picandet V, Jeanneret S, Lavoie J-P. Effects of Syringe Type and Storage Temperature on Results of Blood Gas Analysis in Arterial Blood of Horses. Journal of Veterinary Internal Medicine. 2007 May;21(3):476–81.

57. Mahoney JJ, Harvey JA, Wong RJ, Van Kessel AL. Changes in oxygen measurements when whole blood is stored in iced plastic or glass syringes. Clin Chem. 1991 Jul 1;37(7):1244.

58. Liss HP, Piyne CP. Stability of Blood Gases in Ice and at Room Temperature. Chest. 1993 Apr 1;103(4):1120–2.

الملخص العربى

الخلفية:

تحليل غازات الدم اختبار هام جدا في العديد من الحالات الطبية حيث يتم عمل تحليل غازات الدم الشرياني أو غازات الدم الوريدي تقريبا في جميع المرضى المصابين بأمراض خطيرة اوفى وضع صحى حرج ويحتاجون الى رعاية طبية فائقة.وقد يحدث تأخير في تحليل عينة الدم في المستشفى بسبب محدودية الموارد ولأن أجهزة تحليل غازات الدم غير متوفرة في العديد من الاقسام داخل المستشفى. وقد اثبتت دراسات سابقة أن قيم غازات الدم الشرياني تأثرت بالتأخير الزمني ودرجة حرارة تخزين العينة في تحليل غازات الدم .

الطريقة:

تم سحب عينة شريانية وعينة وريدية مركزية . كانت كل عينة 5 مل ، وتم تقسيمها إلى خمسة أجزاء: تم تحليل 1 مل من الدم في محقنة هيبارين على الفور ، 1 مل من الدم في محقنة بلاستيكية 3 مل (2 محاقن) ، 1 مل من الدم في محقنة أنسولين شعري (2 محاقن). تم تحليل آخر 4 عينات بعد 30 دقيقة و 1 ساعة على التوالي. تم ترك العينات أثناء انتظار التحليل في درجة الحرارة المحيطة. وقد تم ذلك للعينات الشريانية والوريدية في وقت واحد. هذا يعني أن كل مريض لديه 10 عينات تم تحليلها وإخضاعها للتحليل النهائي.

النتائج:

هناك نطاق واسع بين حدود الاتفاق في تشبع الأكسجين ، PO2 و PCO2. كان متوسط خط الأساس PO2 في العينات الشريانية 159.9 ± 113.6. كان التحيز المتوسط (حدود الاتفاق) بقيمة 30 دقيقة (من خط الأساس) والقيمة 60 دقيقة (من خط الأساس) 21 (-99 إلى 142) و 38 (-112 إلى 188) على التوالي في حقنة 3 مل. كان متوسط خط الأساس PCO2 في العينات الشريانية 36 ± 7.6. متوسط التحيز (حدود الاتفاق) بقيمة 30 دقيقة (من الأساس) وقيمة 60 دقيقة (من الأساس) كانت -0.4 (-4.8 إلى 4.1.1) و -3.8 (-11.8 إلى 4.1) على التوالي في 3 مل حقنة.

الخلاصة:

نستنتج أن العينات الشريانية والوريدية التي تتأخر في درجة حرارة الغرفة تعطي نتائج مختلفة عن نتائج خط الأساس (يتم تحليلها على الفور). يحدث هذا التغيير في أقرب وقت 30 دقيقة. نوصي بإجراء تحليل غازات الدم على الفور للحصول على قياسات موثوقة

وعليه فإننا نوصى باجراء تحليل غازات الدم مباشرة بعد سحب العينات من المرضى للحصول على نتائج صحيحة وتقرير العلاج السليم.

تأثير تأخر الوقت و مساحة المحاقن على الضغط الجزئي للأكسجين و تشبع الأكسجين لعينات غازات الدم الشرياني و الوريدي

(دراسة قائمة على الملاحظة)

دراسة بحثية تمهيدا للحصول على درجة الماجستير فى التخدير و الرعاية الركزة و علاج الألم

مقدمة من

الطبيبة/ دينا سمير محمود شاكر

بكالوريوس الطب والجراحة كلية الطب جامعة القاهرة

تحت اشراف

الاستاذة الدكتورة/ أمينة أبو العلا عليو موسى

أستاذ مساعد التخدير و الرعاية الركزة و علاج الألم

كلية الطب قصر العيني –جامعة القاهرة

الاستاذ الدكتور / ممدوح أمين عبد الحميد

أستاذ مساعد التخدير و الرعاية الركزة وعلاج الالم

بمعهد تيودور بلهارس للأبحاث

الاستاذ الدكتور / أحمد محمد لطفي مصطفى

مدرس التخدير و الرعاية الركزة و علاج الألم

كلية الطب قصر العيني –جامعة القاهرة

2019