Chapter 1

General introduction

1.1 The role of clinical chemistry in medicine

Medicine is an art and a science in the service of fellow human beings [1]. On the basis of collected empirical data and information, clinicians select specific diagnoses, rule out other differential diagnoses and eventually make decisions about which and how specific therapeutic interventions are made for the benefit and health of their patients. For a proper interpretation, collected data and information must be compared with other, already existing, data and information to assess the exact value of the clinician's findings. Moreover, a clinician compares observed medical data of a patient with knowledge obtained during his or her training as a clinician and with the experience obtained by working with other patients.

A prerequisite in this paradigm, however, is that collected empirical data on which the diagnoses of a clinician are based must be as objective as possible. Clinical chemistry takes a pivotal role in this in the sense that the chemical characterisation of a patient's body fluid is one of the ways in medicine that can provide such objective data. Since the beginning of this century, clinical chemistry has evolved into a separate and independent discipline in the field of medicine [2-4]. Nowadays, most often a single central clinical chemistry laboratory takes care of the 'analytical needs' of one or more hospitals.

Tasks of the clinical chemist typically include the improvement of existing methods of chemical analysis, the development of new analytical methods and providing the clinician with as much information as possible on the basis of chemical analyses. Especially this last task forms the basis of what has become known as *chemometrics*, a branch of clinical chemistry that uses mathematical and statistical methods to extract a maximum of information from chemical analyses [5, 6].

This thesis presents a multivariate chemometric approach to the problems that are currently associated with the interpretation and evaluation of those laboratory measurements that are used to assess the arterial acid-base status of a patient in an intensive care unit (ICU).

1.2 Arterial acid-base measurements in the ICU

The ICU of today is a highly specialised ward in which expert medical, nursing and technical staff provides medical services to severely ill patients. It is characterised as a high-tech environment in which the real-time monitoring of vital functions plays a central role. The origin of the ICU can be traced back to the second half of the 19^{th} century when special rooms, adjacent to the operating room, were used primarily for the purpose of postoperative care [7]. In the course of time, these recovery rooms evolved into specialised respiratory care units and shock and trauma units, eventually leading to the present day ICU. The modern ICU provides integrated cardiopulmonary support for both medical and surgical patients suffering from severe respiratory and / or cardiac problems as a result of disease or trauma.

The most frequently ordered chemical test in the ICU is the arterial blood gas measurement [8]. Arterial blood gas measurements comprise those measurements of the patient's arterial blood that are used for the evaluation and interpretation of the patient's oxygen and acid-base status. Basic arterial blood gas measurements include: the partial pressure of oxygen (PaO_2), the oxygen saturation of haemoglobin, the pH of arterial blood, the partial pressure of carbon dioxide ($PaCO_2$) and the bicarbonate-ion concentration ($a[HCO_3^-]$). The first two measurements (PaO_2 and oxygen saturation) are used to evaluate the oxygen status, while the other three are used for the interpretation of the arterial acid-base status.

In a strict sense, the term blood gas measurements is incorrect, since only PaO_2 and $PaCO_2$ are true gas measurements and in modern chemical analysers $a[HCO_3^-]$ is not measured but calculated from measured pH and $PaCO_2$. Moreover, two other derived acid-base parameters are generally considered part of the set of arterial blood gas measurements. These parameters are the standard bicarbonate-ion concentration (SB) and the base excess (BE). Their derivation and rationale are described in section 1.4.3 in more detail.

Since the second half of this century, the analysis of arterial blood for the purpose of acid-base characterisation has become a vital part of intensive care medicine. The importance of the acid-base characterisation of arterial blood is illustrated by the severe polio epidemic that struck Copenhagen (Denmark) in 1952 [9]. During this epidemic, hospitals in Copenhagen had to cope with a large number of patients needing intensive artificial respiration as a result of paralysis of the respiratory muscles. For a proper setting of the artificial respiration, the complete acid-base status of the patient had to be known. At that time, arterial blood of patients was seldom sampled for the purpose of performing blood gas measurements [10]. Arterial blood gas measurements were mainly performed in physiological laboratories and were not part of daily clinical practice. Techniques of measurement were cumbersome and needed large equipment.

The clinical necessity of quickly knowing the patient's arterial acid-base status for the purpose of a proper adjustment of the artificial respiration inspired Poul Astrup to develop his equilibration method [9]. This method allowed a relatively quick determination of the three basic acid-base parameters by only measuring the pH of an arterial blood sample and the pH of the sample equilibrated at two known $PaCO_2$ gas tensions. The original $PaCO_2$ is calculated by interpolation [11, 12]. Since then, techniques of analysis developed and arterial acid-base measurements have become routine and indispensable in the daily clinical care of intensive care patients.

1.3 Basic acid-base physiology

In chemical terms, acids are substances that are capable of donating hydrogen (H^+) ions while bases are substances capable of accepting H^+ ions. The amount of H^+ ions in the arterial blood determines its actual acidity. Acidity is measured as pH, which is, according to the definition of Sörensen, the negative logarithm of the H^+ concentration ($[H^+]$) [9].

The regulation of the amount of H^+ ions in the arterial blood and consequently its pH is one of the most powerful controlling mechanisms in the human body. Under normal physiologic conditions, the pH of arterial blood is kept within well-defined limits. This tight regulation of the H^+ concentration in arterial blood is essential since H^+ ions are highly reactive with negatively charged parts of molecules. Changes in H^+ concentration (intracellular as well as extra-cellular) therefore have a profound influence on the molecular configuration and consequently on protein function [13]. Hence, maintaining a constant pH ensures an optimal working condition for enzymes and other proteins. Moreover, large deviations in pH may have effects on the nervous system. If the body becomes too acidic, the nervous system can become so depressed that death can occur. On the other hand, if the body becomes too alkaline, the nervous system can become overexcited, resulting in death from tetanus of the respiratory muscle [14].

Two mechanisms exist to regulate pH of arterial blood: long term physiological buffering and short term chemical buffering. Physiological buffering is the redistribution, production, excretion and/or retention of (non-)volatile acids and bases by means of physiological processes. Chemical buffering is the result of the presence of weak acids and their conjugated bases in the arterial blood. Examples of chemical buffers in arterial blood are: inorganic phosphate, organic phosphate and haemoglobin.

One of the most important chemical buffer systems in the blood, however, is the bicarbonate ion (HCO_3^-)/carbon dioxide (CO_2) buffer system. It is mainly the presence of this buffer system that makes it possible for the human body to cope with the constant load of exogenous acids and bases and the vast amount of both volatile and non-volatile acids that are continuously generated as a result of normal metabolism.

The equation describing the HCO_3^-/CO_2 buffer system in blood is:

$$CO_2 + H_2O \rightleftharpoons H_2CO_3 \rightleftharpoons H^+ + HCO_3^-$$

(1–1)

The left-hand side of this chemical reaction represents the formation of carbonic acid (H_2CO_3) from CO_2 and H_2O. Therefore, although CO_2 itself is not an acid, an elevation of the CO_2 in the blood increases the acidity of the blood through the formation of H_2CO_3 which immediately dissociates into protons (H^+) and bicarbonate ions (HCO_3^-).

Since the concentration of H_2CO_3 is so low in relation to the concentration of dissolved CO_2 and the concentration of HCO_3^-, the law of mass action for the HCO_3^-/CO_2 buffer system is:

$$K = \frac{[H^+] \times [HCO_3^-]}{[CO_2] \times [H_2O]}$$

(1–2)

where K is a constant.

Because H_2O is relatively constant in body fluids, it can be omitted from the equation and incorporated into the constant K, further indicated as K′ [13]. Rewriting the resulting equation to solve $[H^+]$ yields the equation that Lawrence Joseph Henderson (1878-1942) first described in 1909 [10]:

$$[H^+] = K' \times \frac{[CO_2]}{[HCO_3{}^-]}$$

(1–3)

The concentration of dissolved CO_2 in blood ($[CO_2]$) is proportional to the partial pressure of CO_2 (PCO_2) in the gas with which the blood is in equilibrium. Therefore, $[CO_2]$ can be replaced by the partial pressure of CO_2 in the blood. Partial pressures are either measured in millimetres mercury (mmHg) or kilo-Pascal (kPa) where 1 mmHg = 0.133 kPa. The constant relating $[CO_2]$ in mmol/l to the PCO_2 is called the solubility constant. The solubility constant for $[CO_2]$ in plasma is 0.03 mmol per litre per mmHg or 0.225 mmol per litre per kPa.

Moreover, applying the pH concept of Sörensen, in 1917 Karl Albert Hasselbalch (1874-1962) introduced the Henderson-Hasselbalch equation:

$$pH = pK' + \log \frac{[HCO_3{}^-]}{PCO_2}$$

(1–4)

where $pK' = 6.10$ and α is the solubility constant for $[CO_2]$ in plasma.

From equation 1–4 it is apparent that pH is the resultant of the ratio $[HCO_3{}^-]/PCO_2$. Both PCO_2 and $[HCO_3{}^-]$ can effectively be regulated by lungs and kidneys, respectively [15]. This feature in particular makes the $HCO_3{}^-/CO_2$ buffer system so effective in maintaining a constant arterial blood pH. Knowing pH, PCO_2 and $[HCO_3{}^-]$ in the arterial blood of a patient is vital when interpreting the acid-base status of arterial blood. It gives information on both the respiratory and metabolic component of an acid-base disturbance and their joint effect on the acidity of the arterial blood.

Although Sörensen introduced the electrochemical measurement of H^+ ions as early as in 1909, it was not until 1932 that pH glass electrodes were produced commercially and used on a regular basis. Before that time, pH of blood was indirectly obtained from measuring total CO_2 and PCO_2 in the blood with the manometric Van Slyke apparatus that Donald Dexter van Slyke (1883-1971) introduced in 1924 [9]. Around 1960 the CO_2 electrode was introduced into clinical chemistry. Today, chemical analysers measure pH and PCO_2 and calculate $[HCO_3^-]$ with the use of the Henderson-Hasselbalch equation (see Equation 1 –4).

1.4 The clinical interpretation of acid-base parameters

An impairment in either the respiratory or metabolic function (or both) of the body may result in so-called acid-base disturbances [13]. For a proper treatment of these disturbances it is essential for an ICU clinician to be aware of the exact acid-base status of the arterial blood of an ICU patient. With the analysis of measured and calculated arterial acid-base parameters, the ICU clinician aims to find the underlying cause(s) of one or more acid-base disturbances in order to remove it with specific therapeutic interventions. Moreover, for patients receiving artificial respiration, the acid-base analysis of arterial blood is essential for setting the kind and degree of artificial respiration.

1.4.1 General nomenclature and terminology

Acid-base disorders can be divided into *primary*, *secondary* and *combined* acid-base disturbances. Primary acid-base disturbances are the result of impairment of either the respiratory function or the metabolic function of the body. Impairments of the respiratory function result in primary *respiratory* acid-base disturbances, whereas impairments in metabolic function result in *non-respiratory* or *metabolic* disturbances. Both respiratory and metabolic disturbances can be further divided into disturbances that tend to lower the pH, resulting in *acidemia*, and disturbances that tend to raise the pH, resulting in *alkalemia*. These acid-base disturbances are called *acidoses* and *alkaloses*, respectively. Hence, the terms acidosis and alkalosis refer to underlying pH-deranging physiologic processes, whereas the terms acidemia and alkalemia merely indicate the actual acidity of arterial blood. Multiple single primary acid-base disturbances can be present at the same time, resulting in *combined* acid-base disturbances.

Moreover, as a response to primary acid-base disorders, the human body is capable of initiating compensating mechanisms. Primary respiratory disturbances trigger mechanisms in the kidneys that actively regulate the reabsorbtion of excreted HCO_3^- ions, thereby inducing metabolic compensating effects. Also, primary metabolic dysfunction eventually triggers the breathing centre, resulting in an adjustment of the respiration and consequently the $PaCO_2$. These compensating processes result in *secondary* acid-base disturbances. The capability of the body to compensate for primary acid-base disturbances prevents large changes in the pH of arterial blood even though pathological processes may be present.

Respiratory compensations are very rapid and effective within minutes, while metabolic compensations can take up to three days to be fully effective. A metabolic compensation can, however, when in full working order, completely compensate a primary respiratory disturbance, while a respiratory compensation can only partially compensate primary metabolic acid-base disturbances.

It is apparent that for a proper treatment of an acid-base disturbance, the complete acid-base status of a patient should be known to a clinician. Although the body can compensate primary acid-base disturbances to a certain extent, therapeutic measurements must be taken as soon as possible to eliminate any primary acid-base disturbance. Moreover, severely ill patients on the ICU most often receive some form of artificial respiration. Being on mechanical ventilation means that the body cannot fully employ respiratory compensating mechanisms, making the ICU clinician even more responsible for keeping the pH of the arterial blood within acceptable boundaries.

For most ICU patients, an arterial blood gas analysis is performed on a routine basis, for instance every 3 or 6 hours. However, the interpretation of acid-base data is still regarded as difficult since several pieces of information must be evaluated at the same time in their clinical context. Multiple primary disturbances can be present at the same time, concealed by various degrees of compensation, making the diagnosis and monitoring of acid-base data a complex task.

This complexity is illustrated by the coexistence of two distinct methods for interpreting arterial acid-base parameters. One method uses *in vivo* information to interpret pH, $PaCO_2$ and $[HCO_3^-]$, while the other method makes use of pH, $PaCO_2$ and a calculated *in vitro* parameter called base excess (BE). This latter method was developed around 1960 by Poul Astrup and Ole Siggaard-Andersen from Denmark and is therefore also known as the *Scandinavian view* [16].

Schwartz and Relman of the Tufts University School of Medicine in Boston (USA) criticised the *in vitro* approach and made a case for pH, $PaCO_2$ and $[HCO_3^-]$ [17]. This method is therefore also known as the *North American view*. The controversy between the two schools, which Bunker called ‘The Great Trans-Atlantic Acid-Base Debate’, still exists today, although many attempts were made to bridge the gap [16, 18-22].

1.4.2 The North American view; $[HCO_3^-]$ and in vivo CO_2 buffer lines

In the North American view, a high value of $PaCO_2$ indicates a primary respiratory acidosis or a respiratory compensation for a metabolic alkalosis, while a low value of $PaCO_2$ indicates a primary respiratory alkalosis or a respiratory compensation for a primary metabolic acidosis. The metabolic component of an acid-base status is assessed with $[HCO_3^-]$. A high value of $[HCO_3^-]$ indicates a primary metabolic alkalosis or a metabolic compensation for a primary respiratory acidosis while a low $[HCO_3^-]$ indicates a primary metabolic acidosis or a metabolic compensation for a primary respiratory acidosis. However, $[HCO_3^-]$ cannot be used as a true metabolic parameter, since changes in $PaCO_2$ also effect $[HCO_3^-]$.

The concept of the North American view is that *in vivo* data is used to calculate the expected rise or fall in $[HCO_3^-]$ and/or $PaCO_2$ that occur in specific acid-base disorders. The empirically derived *in vivo* information has been compiled from a large number of clinical studies in which the normal compensatory reactions to each of the primary acid-base disorders has been investigated and quantified [23-30]. An observed value of $[HCO_3^-]$ or $PaCO_2$ below or above the expected value of $[HCO_3^-]$ or $PaCO_2$ is an indication for the presence and nature of a metabolic component or respiratory component of an acid-base disorder. Table 1 –1 presents the empirically found expected compensatory rise and fall in $[HCO_3^-]$ and $PaCO_2$ for the primary acid-base disturbances.

Table 1–1. Compensations to primary acid-base disturbances in the North American view [13].

disorder	primary change	compensatory response
metabolic acidosis	↓ $[HCO_3^-]$	1.2 mmHg decrease in $PaCO_2$ for every 1 mmol/l fall in $[HCO_3^-]$
metabolic alkalosis	↑ $[HCO_3^-]$	0.7 mmHg elevation in $PaCO_2$ for every 1 mmol/l rise in $[HCO_3^-]$
respiratory acidosis	↑ $PaCO_2$	
acute		1 mmol/l elevation in $[HCO_3^-]$ for every 10 mmHg rise in $PaCO_2$
chronic		3.5 mmol/l elevation in $[HCO_3^-]$ for every 10 mmHg rise in $PaCO_2$
respiratory alkalosis	↓ $PaCO_2$	
acute		2 mmol/l decrease in $[HCO_3^-]$ for every 10 mmHg fall in $PaCO_2$
chronic		5 mmol/l decrease in $[HCO_3^-]$ for every 10 mmHg fall in $PaCO_2$

1.4.3 The Scandinavian view; standard bicarbonate and base excess

The North American view requires calculations to be performed at the bedside of a patient. Moreover, to predict the amount of rise or fall in primary acid-base values, the acid-base disturbance of a patient should be known *a priori*. To overcome the 'problems' of bedside calculations and the paradox of classifying an already known acid-base disturbance, Astrup and Siggaard-Andersen developed the concept of the standard bicarbonate and the base excess as true metabolic acid-base parameters [11].

In 1960, Astrup described his equilibration method for the rapid measurement and calculation of the primary acid-base parameters pH, $PaCO_2$ and

$[HCO_3^-]$ [12 , 31]. In a microtonometer a blood sample is equilibrated with two known CO_2 gas mixtures, one with a high $PaCO_2$ and one with a low $PaCO_2$. Plotting $PaCO_2$ and measured pH at both $PaCO_2$ values in a log $PaCO_2$–pH diagram, and connecting the two points with a line yields the *in vitro* CO_2 equilibration curve. By measuring pH of the original blood sample and putting it in the plot, the actual $PaCO_2$ of the blood sample can be read from the CO_2 equilibration curve. With the Henderson-Hasselbalch equation $[HCO_3^-]$ can be calculated.

With the log $PaCO_2$–pH chart and the *in vitro* CO_2 equilibration curve of a patient, $[HCO_3^-]$ can be calculated at any desired $PaCO_2$ value. Astrup proposed to use the $[HCO_3^-]$ of a blood sample at a $PaCO_2$ of 40 mmHg as a true metabolic parameter, since this would be the concentration that would have been found in the blood sample if the influence of the respiration was eliminated. He called it the standard bicarbonate concentration or SB.

At the same time, Siggaard-Andersen completed his titration experiments in which he determined the CO_2 equilibration curves of normal blood and blood with known amounts of non-volatile acids and bases at a fixed $PaCO_2$ of 40 mmHg. Based on these experiments he added to the log $PaCO_2$–pH diagram of Astrup a curved line representing the amount of non-volatile acid or base needed to titrate the blood sample at a $PaCO_2$ of 40 mmHg to a pH of 7.40 at a temperature of 37 °C. Astrup and Siggaard-Andersen called this the base excess or BE. Positive base excess values indicate a relative deficit of non-volatile acids while negative base excess values indicate a relative surplus of non-volatile acids. A base excess of 0 means that there is no metabolic component in the acid-base disorder. In modern analysers, BE is calculated from pH, $PaCO_2$, $[HCO_3^-]$ and the haemoglobin concentration of the arterial blood sample at hand.

The most important argument against the use of standard bicarbonate and base excess is that they are determined *in vitro*. The *in vitro* CO_2 equilibration curve is the equilibration curve of whole blood in a tube or syringe. It has been shown that *in vivo* buffering of protons is different from the in *vitro* buffering of protons [17]. This is mainly because *in vivo* buffering takes place in the extracellular fluid in which the haemoglobin concentration (a powerful chemical buffer) is lower than in whole blood. Both Siggaard-Andersen himself and Severinghaus proposed to calculate BE not with the measured haemoglobin concentration of the sample, but with a haemoglobin concentration of 5 g/dl, which is the Hb concentration relative to the total volume of extracellular fluid of the body [32, 33]. This BE is also known as BEecf (Base Excess of extracellular fluid), SBE (Standard Base Excess) and BE5 (Base Excess at a haemoglobin concentration of 5 g/dl.

With BE as a true metabolic parameter, classifying acid-base disturbances is now straightforward. Figure 1 –1 and Table 1 –2 show all possible acid-base classifications based on pH, $PaCO_2$ and BE.

Table 1–2. Classification of acid-base disorders in the Scandinavian view. The signs '-', '+' and '=' indicate an observed value being respectively below, above or within its 95% normal reference interval. See also Figure 1–1.

	pH	$PaCO_2$	BE	classification
1	-	+	=	respiratory acidosis
2	-	+	+	partly compensated respiratory acidosis
3	=	+	+	compensated respiratory acidosis OR compensated metabolic alkalosis OR combined respiratory acidosis and metabolic alkalosis
4	+	+	+	partly compensated metabolic alkalosis
5	+	=	+	metabolic alkalosis
6	+	-	+	combined respiratory and metabolic alkalosis
7	+	-	=	respiratory alkalosis
8	+	-	-	partly compensated respiratory alkalosis

	pH	$PaCO_2$	BE	classification
9	=	-	-	compensated respiratory alkalosis OR compensated metabolic acidosis OR combined respiratory alkalosis and metabolic acidosis
10	-	-	-	partly compensated metabolic acidosis
11	-	=	-	metabolic acidosis
12	-	+	-	combined respiratory and metabolic acidosis
	=	=	=	normal
x	unclassifiable			

To determine whether an observed value for an acid-base parameter is too low, normal or too high, standard univariate 95% reference intervals are used. Table 1 –3 presents the associated upper and lower cut-off values for the univariate 95% reference intervals of arterial pH, $PaCO_2$, BE and $[HCO_3^-]$.

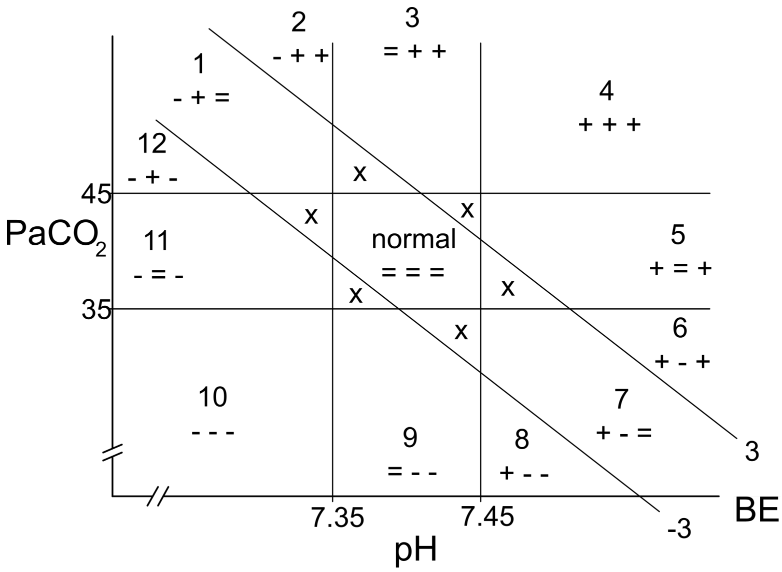

Figure 1–1. Areas of acid-base classification according to the method presented in Table 1–2. Combinations of low, high or normal observed values yield 12 specific acid-base disorder regions. In the normal region, all values are within their standard 95% normal reference intervals. The 'x' regions are formally not classifiable [34]. In these regions, one of the three observed acid-base values is outside its 95% univariate reference interval.

Table 1–3. Upper and lower limits of the standard 95% normal reference intervals for the acid-base variables in arterial blood.

Acid-base variable	lower limit	upper limit
pH	7.35	7.45
$PaCO_2$	35 mmHg	45 mmHg
BE	-3 mmol/l	3 mmol/l
$[HCO_3^-]$	21 mmol/l	27 mmol/l

1.5 Objective and scope of this thesis

Two main problems are currently associated with the interpretation and evaluation of arterial acid-base measurements in an intensive care setting.

The first problem occurs when classifying the acid-base variables pH, $PaCO_2$ and BE according to the method described in section 1.4.3. A strict adherence to the classification rules as described in Table 1 –3 reveals that some combinations of observed values for the three acid-base variables can formally not be classified. This was found when an attempt was made to computerise the classification scheme of Astrup and Siggaard-Andersen in a rule-based expert system [34]. Typically, the *unclassifiable* situation occurs when only one of the three observed acid-base values is outside its 95% univariate reference interval, while the other two are within their 95% univariate intervals. In Figure 1 –1, this situation is represented by the triangular regions denoted by ‘x’.

The second problem originates from the use of the 95% univariate reference interval as the standard statistical model for evaluating the ‘normalcy’ of observed arterial acid-base values from intensive care patients.

A first critical note on the use of 95% reference intervals is that the determination of the respective reference intervals and the characteristics of the reference population are completely unknown. In general, reference intervals are derived from a representative sample of a (often) ‘healthy’ reference population [35]. The process of defining the reference criteria, the selection of reference individuals, analytical considerations and the use of statistical techniques for defining valid 95% univariate reference intervals are described in detail [36-40]. Nothing is known, however, about the determination of the 95% univariate reference intervals that are presented in Table 1 –2. If we assume that the intervals are defined on a ‘healthy’ reference population, what is the value of these intervals in an intensive care setting where it is to be expected that most of the observed acid-base values will be outside these ‘health’-based intervals?

A second critical note concerns the number of reference intervals used. Traditionally, the interpretation of the acid-base status involves the use of three separate 95% reference intervals for evaluating the acid-base variables: pH, $PaCO_2$ and $[HCO_3^-]$ in the North American view, or pH, $PaCO_2$ and BE in the Scandinavian view. From the Henderson-Hasselbalch equation (Equation 1 –4), however, it is apparent that the relationship between pH, log PCO_2 and log $[HCO_3^-]$ is a linear one. This can best be appreciated when Equation 1 –4 is rewritten as:

$$\mathrm{pH} - \log\,[\mathrm{HCO_3}^-] + \log \mathrm{PCO_2} = \mathrm{pK}' - \log \alpha$$

(1–5)

with pK′ and log α both being constant.

Moreover, in Chapter 2 it will be demonstrated that the relationship between pH, $PaCO_2$ and BE is also (almost) linear. Consequently, as Madias [41] already pointed out, it is illogical and fundamentally wrong that three separate 95% univariate reference intervals are used, while only two of the three variables can change independently.

A third critical note on the use of univariate 95% reference intervals is that the 95% univariate interval is not the proper statistical model for evaluating arterial acid-base values. Theoretically, the use of more than one 95% univariate reference interval in case of a simultaneous evaluation of multiple variables – which is the case when interpreting arterial acid-base values – is prone to error and leads *a priori* to more false positive and false negative observations [42-44]. This will be illustrated in detail in Chapter 4.

This thesis describes a new multivariate statistical reference model for evaluating and classifying arterial acid-base variables in an intensive care environment that addresses all of the above mentioned problems. The essence of the model is that a single 95% multivariate statistical reference region is defined on a large reference population consisting of acid-base data coming from intensive care patients themselves. Furthermore, the multivariate reference model is not defined on the original acid-base measurements but rather on the values obtained after applying a mathematical data reduction transformation procedure. Finally, based on the outcome of this transformation, a new way of classifying pH, $PaCO_2$ and BE values will be proposed that will have no *unclassifiable* categories, unlike the method described in 1.4.3.

The outline of this thesis is as follows. In Chapter 2, the mathematical data reduction technique will be introduced, together with the results of various transformed large acid-base data sets coming from several ICUs. In Chapter 3, a two-dimensional graphical representation of the three acid-base variables will be presented, based on the mathematical transformation as described in

Chapter 2. Also, the new classification model for pH, $PaCO_2$ and BE combinations will be described. Then, in Chapter 4, the technique for defining a 95% multivariate patient-based reference region for the acid-base variables will be described. Chapter 5 presents the computational methods involved in the data reduction transformation procedure and the construction of the multivariate reference model. It also presents the prototype computer programs that were built for defining multivariate acid-base reference regions and describes their use in daily clinical practice. Chapter 6 exemplifies the use and practicability of the proposed graphical representation of acid-base data using measurements from three intensive care patients. In Chapters 7 and 8, the results of the clinical evaluation of the multivariate acid-base reference regions and classification model can be found. The thesis is concluded with a general discussion.

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