COMPLEX MODEL OF BLOOD ACID-BASE BALLANCE

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Annotation

Originally, the classic Siggaard-Andersen nomogram, widely used in clinical practice for the assessment of acid-base balance, experimentally obtained at 38°C with the precondition of normal plasma protein concentrations. However, a nomogram is used in clinical practice to calculate from the data measured in blood samples tempered at 37°C. We made a simulation recalculation of the baseline experimental data to 37°C and set a new nomogram for 37°C. Compared with the original nomogram, there are no significant deviations, if BE does not deviate by more than 10 mmol/l; the results are, however, different with the deviations exceeding 15 mmol/l. We suggested an algorithm and a program, which enables calculation of BE from pH and pCO_2 according to the original as well as adjusted normograms. However, the data, having been a base of the normogram, count with normal plasma protein concentrations. Furthermore, we combined Figge and Fencl plasma acid-base balance model with the data based on Siggaard-Andersen nomogram, adjusted to 37°C. Thus, BE was not only defined in the dependence on haemoglobin concentrations, but also on plasma protein and phosphate concentrations. At these conditions, BE corresponds to SID changes according to the "modern conception" of acid-base balance by Stewart. Moreover, the model obviously suggests that the independence of SID and pCO_2 is not applicable for the whole blood. The model is a core of a wider model of acid-base balance in the organism, enabling realisation of the pathogenesis of acid-base balance disturbances, which is in accordance with our earlier publication of the balance approach to the interpretation of acid-base balance.

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Key words

Acid-base balance, formalised description, simulation model, blood gases, educational simulators

1. Introduction

Acid-base balance in the organism is controlled by two balances – carbon dioxide flow balance (respiration control) and strong acid production/excretion balance (regulation of acidification processes in the kidney). Both flows are connected via buffer systems. The balance disturbances result in pH changes in blood liquids. Drifts in the chemical balances of buffer systems, transport of substances between the buffer systems, H^+/Na^+ H^+/K^+ exchange between the cell and the interstitial liquid (and, in a long-term scale, washing out $NaHCO_3$, $KHCO_3$ and, later, $CaCO_3$ and $CaHPO_4$ from the bone mineral mass in chronic acidemia) are only suppressive mechanisms in acid-base disturbances. The basic regulation organs, able to control acid-base balance (by their effect on CO_2 and H^+/HCO_3^- flows) include the respiratory system and kidney.

From the clinical point of view, the arterial blood buffer system is an important indicator of the status of acid-base balance. CO_2 retention or depletion during the change of carbon dioxide balance as well as H^+/HCO_3^- retention or depletion during the changes of strong acid production/excretion balance develop into the drift of the chemical balance in bicarbonate and non-bicarbonate buffer systems.

Labelling the total concentration of non-bicarbonate bases $[Buf^-]$ –which, in fact, are the buffer bases of plasma proteins and phosphates (and haemoglobin concentrations in the whole blood) – then the total concentration of non-bicarbonate buffer bases forms the Buffer Base (BB) value:

$$BB=[HCO_3^-]+[Buf^-]$$

2. Classical approach of the "Danish School" for the assessment of acid-base disturbances

The variations in pCO_2 result in pH changes; If the titration curve of pCO_2 and pH changes is plotted in the semi-logarithmic scale, these titration curves verge on lines in the range of life-compatible pH values. This precondition was a base of blood acid-base balance tests introduced in the first half of 1950s by Paul Astrup. At that time, there were no electrodes which enable direct measurement of plasma pCO_2. There were, however, relatively accurate methods of pH measurement. Astrup's method of pCO_2 analysis (1956) was based on the following procedure: first, blood pH was measured, then, the sample was automatically equilibrated by O_2/CO_2 mixture with accurately set pCO_2. The blood sample was equilibrated with a high pCO_2 gas mixture and the equilibration was followed by measuring pH. Then, the blood was equilibrated with a mixture with low carbon dioxide partial pressure and the equilibration was followed by another pH measurement. The points obtained were plotted into a semi-logarithmic graph to create a line, used to read out pCO_2 according to baseline pH (see Fig. 1).

The Buffer Base concept made by Singer and Hastings (1948) was further improved by Siggaard-Andersen in (1960,1962), who introduced the difference of Buffer Base and its normal value - Normal Buffer Base (NBB) - as a clinically relevant factor:

$$BE=BB-NBB$$

At normal circumstances, BE values (for blood samples with any haemoglobin concentrations) are zero. They are changed during a buffer reaction with strong acid or base added.

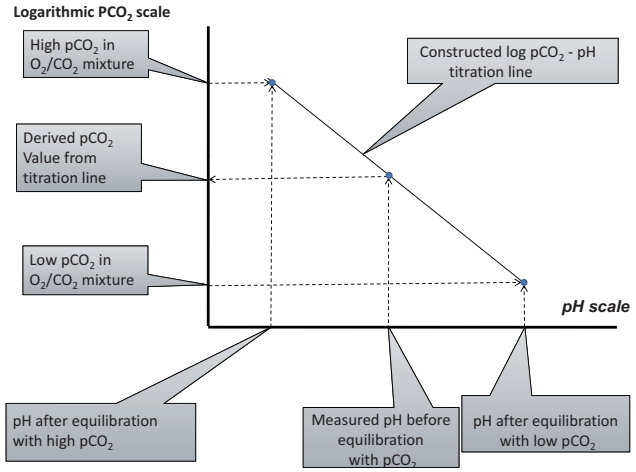

Fig. 1 *The titration curve of pH/PCO$_2$ variations after blood equilibration with carbon dioxide is practically a line. This therefore enabled pCO$_2$ determination in the tested blood sample according to the titration curve plotted after blood equilibration with low and high partial CO$_2$ pressure.*

Siggaard Andersen used the equilibrium titration curves to determine BB and BE. He added defined amounts of strong acids or bases to blood samples with various haematocrit concentrations, changing their BE. Then, the samples were titrated and the results were plotted in log PCO$_2$/pH coordinates. The titration curves (being lines in the semi-logarithmic coordinates) of the blood samples with various haematocrit and the same BE always crossed in the same points (see Fig. 2). Similarly, the titration curves of the samples with various haematocrit concentrations (and various BE), but

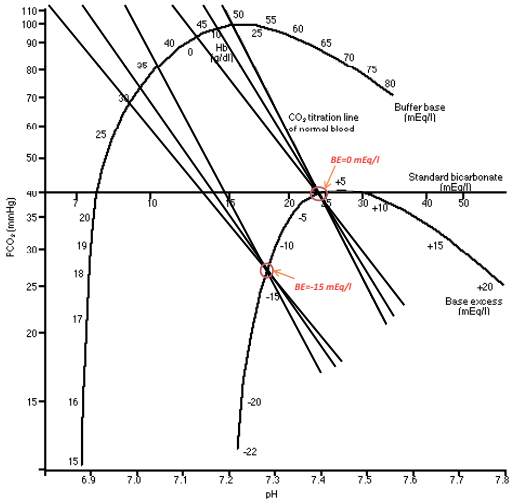

Fig. 2 *Siggaard-Andersen nomogram. The titration curves (lines in the semi-logarithmic scale) have different slopes after blood titration with carbon dioxide, depending on haemoglobin concentrations. The curves with the same BE cut each other in one point. The intersections of these points were a base for experimental determination of BE curve (Base Excess). Similarly, BB curve (Buffer Base) was experimentally determined as the intersection of the points where the titration curves of the blood samples with the same BB cut each other. The nomogram was experimentally created for 38°C. The tested blood sample is tempered to the standard temperature of 37°C in modern automats for the tests of acid-base balance. At present, the determination of BE and BB, however, uses (digitalised) data based on the original Siggaard-Andersen nomogram.*

with the same BB crossed in the same points, too.

Thus, a nomogram with BE and BB curves with semi-logarithmic coordinates was obtained; the curves enabled the determination of BE and BB in the samples having been tested.

Siggaard-Andersen used this procedure to find experimentally the dependence of hydrogen ion [H$^+$] concentrations or pH on pCO$_2$ and haemoglobin (Hb) concentrations; the results obtained were used to create clinically applicable nomograms expressing the following dependence:

[H$^+$]= function (pCO$_2$,BE,Hb)

In the assessment of acid-base disturbances by BE and pCO$_2$, it should be taken into consideration that the increase or the fall in CO$_2$ affects neither the total concentration of the buffer bases (BB) nor BE . The increase results in the increase in carbonic acid concentration, dissociating into bicarbonate and hydrogen ions, which are, however, completely bound to non-carbonate buffer bases [Buf$^-$]; the increase in bicarbonate concentrations therefore corresponds with the same fall in non-bicarbonate buffers with the total [HCO3$^-$]+[Buf$^-$] concentrations and, thus, BB as well as BE remaining practically unchanged. BB and BE are therefore considered pCO$_2$ independent. This applies for plasma exactly but not exactly for the whole blood – pCO$_2$ affects haemoglobin oxygenation. However, as deoxygenated haemoglobin has higher affinity to protons than oxygenated haemoglobin (the oxygenated blood therefore contains virtually higher non-bicarbonate buffer concentrations), the total concentration of buffer bases BB also depends on haemoglobin oxygen saturation (susceptible by pCO$_2$).

Hence, to make acid-base balance models, it is beneficial to define ***standardised Buffer Base oxy-value (BBox)*** as BB, potentially found in the blood sample with full oxygen saturation of oxyhaemoglobin (i.e. full 100% oxygen saturation of haemoglobin). Similarly, the ***standardised Base Excess oxy-value (BEox)*** is defined as BE measured in the blood sample with full oxygen saturation of oxyhaemoglobin (Kofránek, 1980). Thus, ***BEox*** is really pCO$_2$ – independent.

It is necessary to say that the independence of pCO$_2$ and BEox does not apply for “in vivo” whole blood completely, as the increase in pCO$_2$ is connected with higher increase of bicarbonates in plasma compared with that in the interstitium; thus, part of the bicarbonates is transported into the interstitiary liquid during the increase in pCO$_2$ (with a mild fall in BEox in acute pCO$_2$ increase).

BB and BE (or BBox and BEox) change after addition of a strong acid (or strong base) or bicarbonates to the blood sample. Addition of one millimol of a strong acid to one litre of blood results in BE fall by one millimol; addition of one millimol of bicarbonates (or withdrawal of one millimol of hydrogen ions by a reaction with a strong base) results in BB and BE (BBox and BEox) increase by one millimol.

The variations in dissolved CO$_2$ plasma concentrations (expressed as pCO$_2$) and BE therefore characterise carbon dioxide flow balance and the variation in strong acid production/excretion balance, respectively. Thus, pCO$_2$ and BE characterise the respiratory and metabolic parts of acid-base balance, respectively.

To use pH, pCO_2 and BE in clinical practice for the diagnosis of acid-base balance, so called compensation diagrams were created, expressing the effect of adaptation responses of the respiratory and renal systems to acid-base disturbances (Dell and Winters, 1970, Goldberg et al., 1973, Siggaard-Andersen, 1974, Grogono et al., 1976).

Siggaard-Andersen nomogram (expressed in the form of approximate equations) became a base for algorithm assessment in a number of laboratory automats for the measurement of acid-base balance. A certain problem was that the experimental measurements for the construction of Siggard-Andersen nomogram were carried out at 38°C. Modern devices for the measurement of acid-base balance (allowing direct measurement of pCO_2, pH and pO_2) usually give data for samples adjusted to 37°C.

However, a more serious problem was that the titration done to create an experimental nomogram was carried out with blood with normal plasma protein concentrations (72 g/l). If the plasma protein concentrations are lower (which is not rare in critically ill patients), the points on the nomogram are shifted and all the clinical counts derived from this nomogram are incorrect.

Later, Siggaard-Andersen published certain corrections, considering various plasma protein concentrations (Siggaard-Andersen, 1977, Siggaard-Andersen et al. 1985, Siggaard-Andersen, Fogh-Andersen, 1995); however, they were not included into clinical practice properly.

3. Steward´s "modern" approach

The abovementioned inaccuracies of the classical approach to the assessment of acid-base balance resulted in the attempt to find new methods of the description and assessment of blood acid-base balance in 1980s. The most used method was Stewart´s one (1983), improved later for clinical practice by Fencl et al. (1989, 1993, 2000).

Unlike Siggaard-Andersen´s method, Stewart´s description is limited to plasma only; however, it enables accurate description of hypo- and hyperalbuminaemia, dilute acidosis as well as concentration alkalosis. Stewart´s calculations are based on the combination of physical-chemical equations. The original Stewart´s calculations are based on simple preconditions:

1. The equation for water must apply:

$$[H^+]\,[OH^-] = K'w$$

2. The constancy of the sum of weak acid concentrations (Buf^-), and their dissociated buffer bases (Buf^-):

$$[Buf^-]+[HBuf] = [Buf_{TOT}]$$

3. Dissociation balance of non-bicarbonate buffer system:

$$[Buf^-]\,[H^+] = K_{BUF} \times [HBuf]$$

4. Dissociation balance of bicarbonate buffer:

$$[H^+]\,[HCO_3^-] = M \times pCO_2$$

5. Dissociation balance between bicarbonate and carbonate:

$$[H^+]\,[CO_3^{2-}] = N \times [HCO_3^-]$$

6. Electroneutrality:

$$SID + [H^+] - [HCO_3^-] - [Buf^-] - [CO_3^{2-}] - [OH^-] = 0$$

with SID meaning the value of "strong ion difference" (residual anion) – defined as the difference between the concentrations of fully dissociated anions and cation (expressed in mEq/l). Practically, the value can be found out by the following equation:

$$SID = [Na^+] + [K^+] + [Mg^{2+}] + [Ca^{2+}] - [Cl^-]$$

Combining these two equations, the result is the fourth degree algebraic equation, enabling calculation of hydrogen ion concentrations in dependence on SID, the total concentration of weak acids and their buffer bases $[Buf_{TOT}]$ and pCO_2 (the dependent variable is underlined in the equation, independent variations and constants are in bold and italic, respectively):

$$\underline{[H^{\pm}]}^4 + (\mathbf{SID} + K_{BUF}) \times \underline{[H^{\pm}]}^3 + (K_{BUF} \times (\mathbf{SID} - \mathbf{[Buf_{TOT}]}) - K'w - M \times \mathbf{pCO_2}) \times \underline{[H^{\pm}]}^2 - (K_{BUF} \times (K'w^2 + M \times \mathbf{pCO_2}) - N \times M \times \mathbf{pCO_2}) \times \underline{[H^{\pm}]} - K'w \times N \times M \times \mathbf{pCO_2} = 0$$

Solving of the equation gives hydrogen ion concentration, depending on the respiratory part of acid-base balance – i.e. pCO_2, and, moreover, on the respiratory part of SID independent metabolic parameters as well as on the total concentration of non-bicarbonate bases and acids $[Buf_{TOT}]$:

$$pH = \text{function}\,(\,pCO_2,\ SID,\ [Buf_{TOT}]\,)$$

The total concentration of non-bicarbonate bases $[Buf_{TOT}]$ is related to the total plasma protein (albumin) concentration. More detailed studies consider the total phosphate concentrations, too. The results of these studies are relationships enabling (by means of a computer programme) calculation of pH (and other variables such as bicarbonate concentrations etc.) from pCO_2, SID, and total phosphate [Pi] and plasma albumin $[Alb_{TOT}]$ concentrations (see, for example, Watson, 1999):

$$pH = \text{function}\,(\,pCO_2,\ SID,\ [Alb_{TOT}],\ [Pi]\,)$$

One of the most detailed quantitative analyses of plasma acid-base balance (Figge, 2009) improving Figge-Fencl's model (Figge et al. 1992) even corrects the effect of externally added citrate [Cit] in the plasma sample used for the laboratory test.

$$pH = \text{function}\,(\,pCO_2,\ SID,\ [Alb_{TOT}],\ [Pi],\ [Cit]\,)$$

4. Benefits and drawbacks of Stewart´s approach

Mathematical relationships between the variables derived from the quantitative physical-chemical analysis enable calculation of dependent variables – pH, being a base for other dependent variables, i.e. bicarbonate concentrations – from independent variables (i.e. pCO_2, SID, albumin and phosphate concentrations or, as the case may be, concentrations of the citrate added to the plasma sample).

Stewart´s approach enables more detailed description of some of the pathophysiological conditions (the effect of hypo- and hyperalbuminaemia on acid-base balance, dilution acidosis or concentration alkalosis) and, at first site, gives the clinicians the feeling of better insight into the ethiology of acid base disturbances. To determine "independent" variables, used for the calculation of other acid-base parameters, it is

necessary to do explicit measurements of phosphate, Na^+, Cl^-, HCO_3^- and other ion concentrations, which clinicians work in their diagnostic forethought with.

On the contrary, the drawbacks of Stewart´s theory include the fact that he works with plasma only. Moreover, some Stewart´s followers, fascinated by the possibility to calculate acid-base parameters - pH (and proper concentrations of bicarbonates, carbonates and non-bicarbonate acids) – from independent variables (pCO_2, SID, [Alb_{TOT}], [Pi]), often make objectively incorrect conclusions in their interpretation. In the calculation, the independence of baseline variables, particularly SID, is meant not in a causal but in a strictly mathematical meaning. This is, however, often forgotten in clinical-physiological practice, which often results in incorrect interpretation of the causality relationship between the causes of acid-base disturbances.

5. "Mathematical wizardry" of Stewart´s followers

A number of Stewart´s followers considered his mathematical relationships as "oracle" – incorrect causal relationships are deducted from substantially correct mathematical relationships. The causality of mathematical calculations (where independent variables are calculated from dependent ones) is confused with the causality of pathophysiological relationships.

For example, some authors deduct that one of the elementary causal relationships of acid-base disturbances are changes in SID concentrations. Sirker et al. (2001) even states that "the transport of hydrogen ions through membranes (via hydrogen channels) does not affect their actual concentration. Direct removal of H^+ from one compartment can alter neither the value of any independent variable nor [H^+] concentration… the equilibrium dissociation of water balances any fluctuations in [H^+] concentrations and serves as an inexhaustible source or sink for H^+ ions".

There is no rational explanation for the opinion that SID (as a mathematical construct, not a physical-chemical characteristic) affects [H^+] concentrations in a certain mechanistic way to keep electroneutrality – any buffer reaction is a shifted chemical balance only; thus, there is no way how they could affect the electroneutrality themselves (without membrane transport).

6. Are both approaches significantly different?

Excited debates lead by supporters of both theories in international journals (e.g. Dubin et al. 2007, Dubin 2007, Kaplan 2007, Kurz et al., 2008, Kelum 2009) might suggest that both theories are completely different and their applicability will be proved during the time. In fact, both theories are complementary. If similar conditions of their applicability are observed (i.e. they are used for plasma with normal albumin and phosphate concentrations only), the results are, in fact, identical. It is obvious that if one of the theories is used out of the area which it was proposed for, it fails and the other theory seems to be more accurate. For example, reduced protein concentrations do not correspond to the conditions determined experimentally for Siggaard-Andersen nomogram; if this nomogram is used for BE assessment in patients with hyperalbuminaemia, incorrect values are obtained. In this case, the use of Stewart´s method prevents incorrect diagnosis. On the other hand, Stewart does not calculate with the effect of such an important blood buffer - haemoglobin in erythrocytes. Stewart´s approach is applicable neither for the calculation of the amount of infusion solutions for the correction of the acid-base disturbance nor for the assessment of the grade of respiratory and renal compensation of the acid-base disturbance. During the bedside diagnostics it is advisable to consider both theories and to realise their benefits and limits (Kelum, 2005).

The accordance and differences of both approaches are as follows.

Both Stewart and Siggaard-Andersen use pCO_2 as a parameter describing the respiratory part of acid-base balance. According to the "Danish School", the metabolic part is represented by BB or its deviation from the norm – BE. According to Stewart, the metabolic part is represented by SID as the difference of fully dissociated positively and negatively charged anions and cations – in the respect of keeping the principle of electroneutrality, it might seem at first sight that, numerically, SID is identical with plasma BB (Fig. 3).

$SID = [HCO3^-] + [Buf^-] = BB$

But is it true really? Siggaard-Andersen (2006) states so. However, focused on the importance of non-bicarbonate bases, certain differences can be seen.

Plasma non-bicarbonate bases include phosphates and plasma proteins – particularly albumin (the effect of globulins on acid-base balance is insignificant). The albumin hydrogen ion can be bound to the following negatively charged amino acids (Figge, 2009): cysteine, glutamic and aspartic acid, tyrosine and carboxyl end of protein polymer. Labelling these binding sites as Alb^- , the binding of hydrogen ions can neutralise the electric charge (as presumed in the classical Stewart´s theory):

$Alb^- + H^+ = HAlb$

Hydrogen ions van, however, be bound to imidazol cores of histidine as well as to arginine, lysine and NH_2–end of an albumin molecule. Labelling these binding sites as Alb, then the binding of hydrogen ions results in the creation of positive charge:

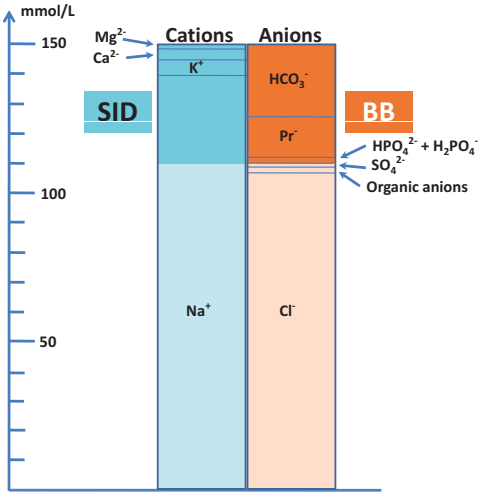

Fig. 3 *SID and BB are nearly identical. The variations in SID and BB are completely identical: dSID=dBB.*

$Alb + H^+ = HAlb^+$

Labelling the total concentrations of non-bicarbonate bases by Stewart and Siggaard-Andersen as $[Buf_{st}^-]$ and $[Buf_{sa}^-]$, respectively, a small difference can be observed (the concentrations are considered in miliequivalents):

$[Buf_{st}^-] = [PO_4^{3-}] + [HPO_4^{2-}] + [H2PO_4^-] + [Alb^-] – [HAlb^+]$

$[Buf_{sa}^-] = [PO_4^{3-}] + [HPO_4^{2-}] + [H2PO_4^-] + [Alb^-] + [Alb]$

The concentration of non-bicarbonate bases is a bit higher by Siggaard-Andersen, as the relationship $[Alb]>[HAlb^+]$ applies at physiological conditions. This obviously suggests the difference between normal SID (around 38 mmol/l) and normal plasma BB (stated as 41.7 mmol/l).

However, as it applies that the variation in [Alb] concentrations is related to the variation in $[HAlb^+]$ concentrations:

$d[Alb]=-d[HAlb^+]$

the variation in the concentrations of non-bicarbonate bases by Siggaard-Andersen will be identical with that of non-bicarbonate bases by Stewart:

$d[Buf_{st}^-] = d[Buf_{sa}^-]$

The variation in BB or BE is therefore the same as that of SID:

dBB=dSID

Thus, it would meaningful for clinical purposes to calculate normal SID for various plasma protein and phosphate concentrations: NSID=function ($[Alb_{TOT}]$, [Pi]), similarly as Siggard-Andersen calculates NBB as a variable dependent on haemoglobin concentrations. It would not be complicated in any respect.

However, the problem is that what circulates in the blood vessels is not plasma only, but plasma and erythrocytes. A more accurate quantitative analysis requires considering the whole blood and it is also necessary to re-evaluate and connect both the approaches.

The outcome of the connection will be the sufficiently quantified Figge-Fencl's model of plasma (Figge, 2009) and experimental data for the whole blood, included in Siggaard-Andersen nomogram.

7. Formalisation of Siggaard-Andersen nomogram

The first step necessary for the realisation of this connection is to formalise Siggaard-Andersen nomogram.

The literature describes a number of equations which formalise Sigaard-Andersen nomogram with higher or lower accuracy (e.g. Siggaard-Andersen et al. 1988). Lang and Zander (2002) compared the accuracy of BE calculation in 7 approximations of various authors. The most accurate approximation was that of Van Slyk equation by Zander (1995). Surprisingly, it was, however, shown that the formalisation of Siggaard-Andersen nomogram from 1980, used in a lot of our models in the past, approximated Siggaard-Andersen nomogram with higher accuracy than the relationships having been published later (Fig. 4)

It is possible to try further specification of our approximation.

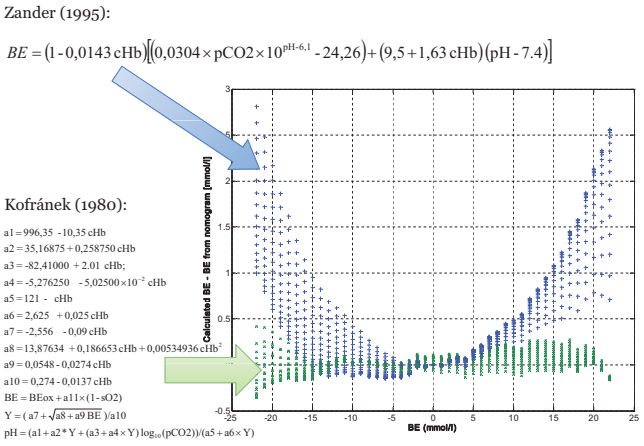

Fig. 4 *The comparison of the accuracy of BE curve approximation by Siggard-Andersen nomogram for various haemoglobin concentrations and BE. Approximation by Kofránek (1980): '×' and by Zander (1995): '+'.*

However, the situation in 1980s was a bit different. At that time, the struggle was focused on the finding of such approximations which would not require a big memory (regarding the opportunity of their use in laboratory devices and the capacity of microprocessors at that time). At present, the approximation of experimental curves is carried out by means of the approximation of the original curve Siggaard-Andersen nomogram by splines.

The aim is to create approximation of the function

pH=BEINV(cHb,BEox,sO2,pCO2)

where cHb is haemoglobin concentration (in g/100 ml blood), BEox is BE (mmol/l) with 100% haemoglobin saturation (being therefore independent on haemoglobin oxygen saturation), sO_2 is haemoglobin oxygen saturation and pCO_2 is carbon dioxide partial pressure (torr).

Hence, the spline approximation of the coordinates of BE and BB curves on the curve Siggaard-Andersen nomogram (Fig 5. and 6) is created first, being a base for the calculation of pH according to BEox, haemoglobin concentration cHB, haemoglobin oxygen saturation sO_2 and pCO_2 (Fig. 5). The calculation of pH takes advantage of the fact that the titration curves are practically lines in the semi-logarithmic scale (log pCO_2, pH).

Function BEINV (Fig. 7) enables simulation of blood titration with carbon dioxide at various haemoglobin concentrations and haemoglobin oxygen saturation (at standard temperature 38°C and normal plasma protein concentrations).

The calculation of BE and BEox from pH and pCO_2, haemoglobin concentration and haemoglobin oxygen saturation is based on the iteration calculation using the abovementioned equations. This calculation is a base of ABEOX function.

8. Correction of Siggaard-Andersen nomogram to 37°C

Siggaard-Andersen nomogram was created at the standard temperature of 38°C. However, the standard temperature for measuring acid-base balance parameters is 37°C in modern diagnostic devices. Nevertheless, Siggard-Andersen

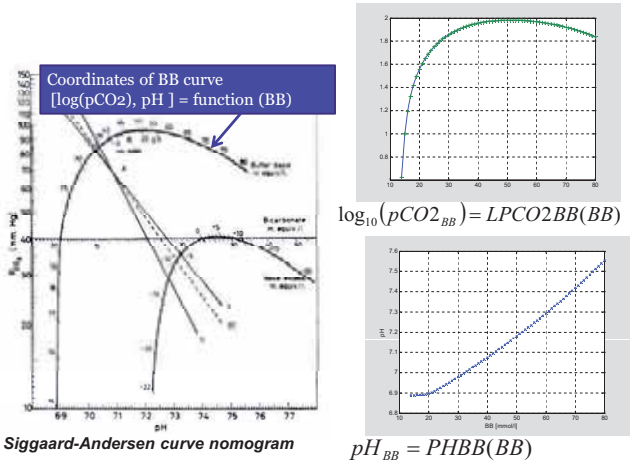

Fig. 5. Approximation of BB curve from Siggaard-Andersen nomogram by means of splines.

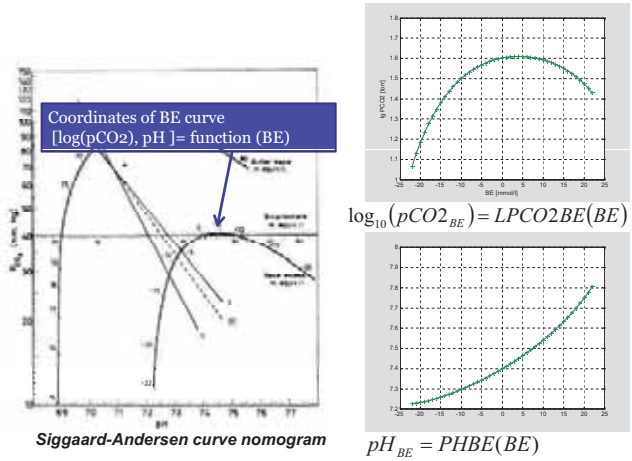

***Fig. 6** Approximation of BE curve from Siggaard-Andersen nomogram by splines.*

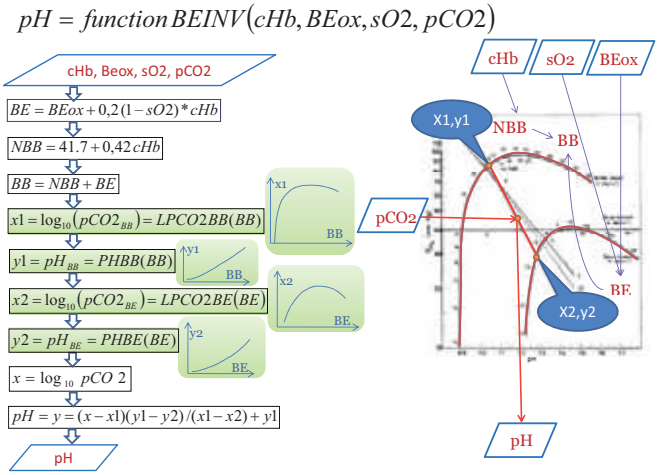

***Fig. 7** Algorithm of the calculation of titration curves by Siggaard-Andersen nomogram formalised by means of splines.*

nomogram is used for the assessment of measured nomograms without any correction. Moreover, this nomogram is used for identification of the models created for 37°C in a number of works (e.g. Reeves and Andreassen 2005).

On the contrary, models of plasma acid-base balance, e.g. Watson´s (Watson, 1999) or Figge-Fencl´s (Figge, 2009) models have been identified for 37°C. Thus, it was necessary to correct Siggaard-Andersen nomogram from 38°C to the standard temperature of 37°C.

In clinical practice, the temperature corrections of pH and pCO_2 from t° to the standard temperature of 37°C are based on simple relationships, e.g. (Ashwood et al. 1983):

$pH_{37°C} = pH_{t°} - 0.0147\ (37-t°)$

$\log_{10}(pCO_{2\,37°C}) = 0.019\ \log_{10}(pCO2_{\,37°C})(37-t°)$

For proper temperature corrections of Siggaard-Andersen nomogram it is advisable to use the more accurate relationship by Ashwood et al. (1983):

$pH_{37°C} = (pH_{t°} - 0.0276(37-t°) - 0.0065\ (7.4)\ (37 - t°) + 0.000205\ (37^2 - t°^2))/(1 - 0.0065(37-t°))$

$\log_{10}(pCO_{2\,37}) = \log_{10}(pCO_{2\,t°}) + (0.02273 - 0.00126\ (7.4 - pH_{37°C}))\ (37 - t°) - 0.0000396(37^2 - t^2)$

However, to correct Siggaard-Andersen nomogram from 38°C to 37°C, it is insufficient to transfer simply $\log_{10}pCO_2$ and pH, representing the coordinates of BE and BB curves in Siggaard-Andersen nomogram, from one temperature to another.

The trouble is that, according to the definition, BE is calculated as a titrable base in blood titration to the standard values (pCO_2=40 torr and pH=7.4). BE is zero at these standard values. Thus, the zero point of the BE curve, where all titration curves of blood with various haematocrit cut each other, lies in the coordinates of pH=7.4, and pCO_2=40 torr. Using a simple re-calculation of the values from 38°C to 37°C, the zero point of the BE curve is transferred to pCO_2=38.2195 torr and pH=7.421 then (Fig. 8). Our aim is, however, to achieve that pCO_2 and pH corresponding to zero BE are 40 torr and 7.4 on the curve for 37°C.

Thus, standard pH and pCO_2 are re-calculated from 37°C to 38°C as follows:

$pH_{37°C} = 7.4_{\ 37°C}$ -> $pH_{38°C} = 7.3878_{\ 38°C}$

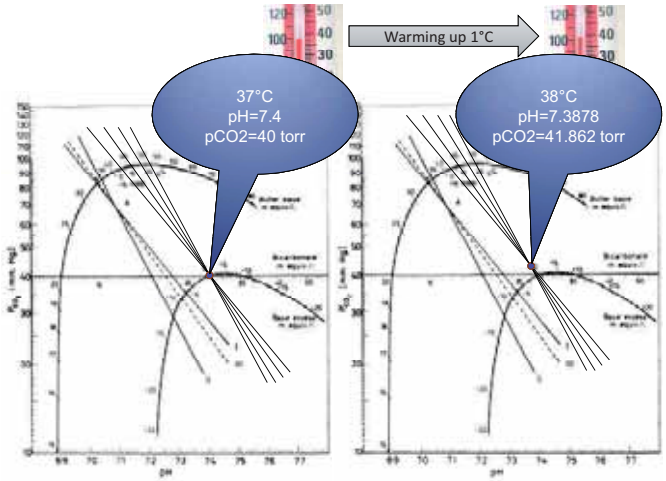

***Fig. 8** In the points $pH_{37°C}$ = 7.4 and $pCO_{2\,37°C}$ = 40 torr, there is an intersection of plasma and erythrocyte titration curves with various haematocrit and BE=0 mmol/l. After the temperature increase by one degree centigrade, all lines are shifted with the intersection in the same point ($pH_{38°C}$ = 7.3878 and $pCO_{2\,38°C}$ = 41.862 torr); BEs are, however, non-zero and differ for each blood sample.*

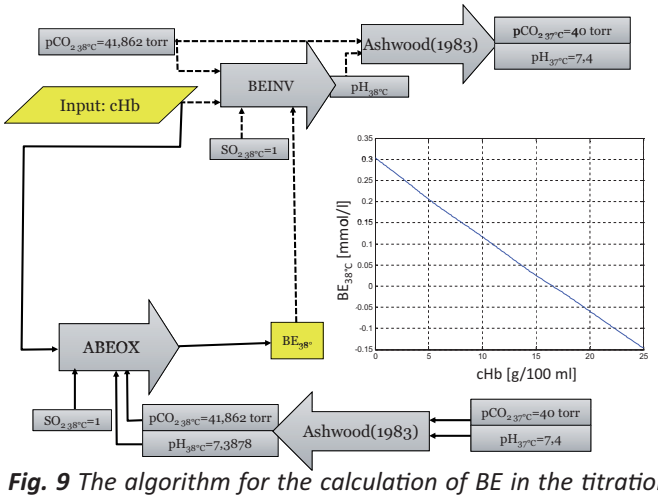

Fig. 9 *The algorithm for the calculation of BE in the titration curves with various haemoglobin concentrations (cHb) for 38°C, with the corresponding zero BE at 37°C (the curves cut each other in the point of pH=7.4 and pCO_2=40 torr at 37°C).*

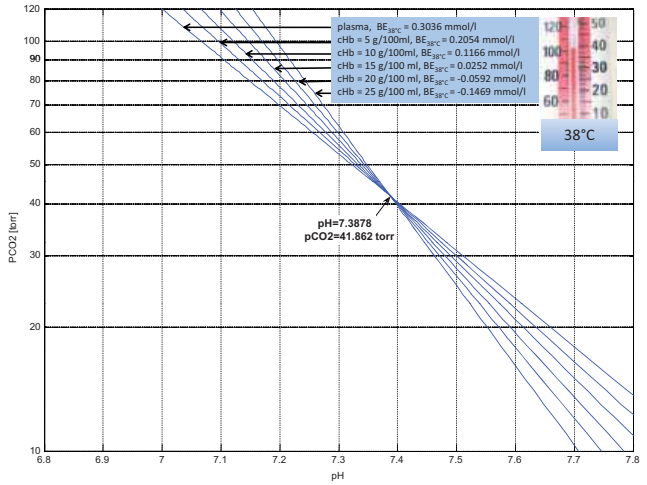

Fig. 10 *The titration curves at 38°C with various haemoglobin concentrations and BE, calculated by the algorithm described in the previous picture, cut each other in the point whose pH and pCO_2 coordinates correspond to pH=7.4 and pCO_2 = 40 torr after cooling the blood by one degree.*

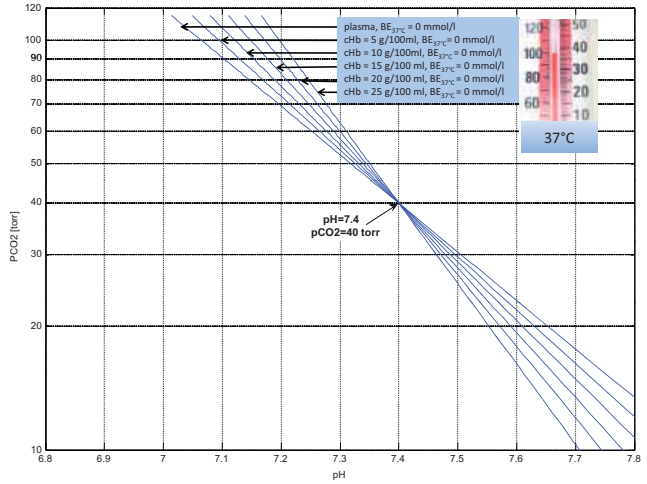

Fig. 11 *The titration curves at 37°C – pH and pCO_2 from particular curves in the previous figure were re-calculated from 38°C to 37°C. The curves cut each other in the zero point of BE curve for 37°C, which lies on the coordinates pCO_2=40 torr and pH=7.4.*

$pCO_{2\ 37°C}$ = 40 $torr_{38°C}$ -> $pCO2_{\ 38\ °C}$ = 41.862 $torr_{\ 38°C}$

All titration curves of fully oxygenated blood with various haematocrit will cut each other in these points (in fact, these curves will be lines in the semi-logarithmic scale). Their BE will be set to zero at 37°C. Their BE will be non-zero at 37°C, depending on haemoglobin concentration (Fig 8). For the algorithm of the calculation, see Fig. 9.

If the titration curves of the values calculated by this procedure are modelled, it is obvious that they cut each other in one point at 38°C (Fig. 10).

The re-calculation of the data of the titration curves from 38°C to 37°C by the abovementioned relationships derived by Ashwood et al. (1983) enables to obtain a set of curves (or lines in the semi-logarithmic scale), cutting each other at the standard values of pH=7.4 and pCO_2=40 torr (see Fig. 11). According to the definition, BE (at 37°C) will be therefore zero in all cases. At 38°C, their BE will be different, depending on haemoglobin concentration (see Fig. 12).

To obtain a set of the values characterising the BE curve for Siggaard-Andersen nomogram corrected to 37°C, it is advisable to carry out simulation experiments with carbon dioxide blood titration in blood samples with various haemoglobin concentrations for each $BE_{37°C}$, in the condition of full oxygen saturation (see the calculation algorithm scheme in Fig. 13). Correction factor $dBE_{38°C}$ (depending on haemoglobin concentration and corresponding to BE zero value at 37°C) was always added to each $BE_{37°C}$. This correction shift was a base for $BE_{38°C}$.

$$BE_{38°C} = BE_{37°C} + dBE_{38°C}$$

A set of $pH_{38°C}$ was calculated from a set of $BE_{38°C}$ and $pCO_{2\ 38°C}$ by Siggaard-Andersen nomogram (by means of BEINV algorithm – see Fig. 7). $pCO_{2\ 38°C}$ and $pH_{38°C}$ were then re-calculated to the values corresponding to 37°C.

This procedure enabled obtaining the titration curves for 37°C. The intersections of the curves with the same $BE_{37°C}$ and various haematocrit characterise the BE curve of Siggaard-Andersen nomogram corrected to 37°C (see Fig. 14).

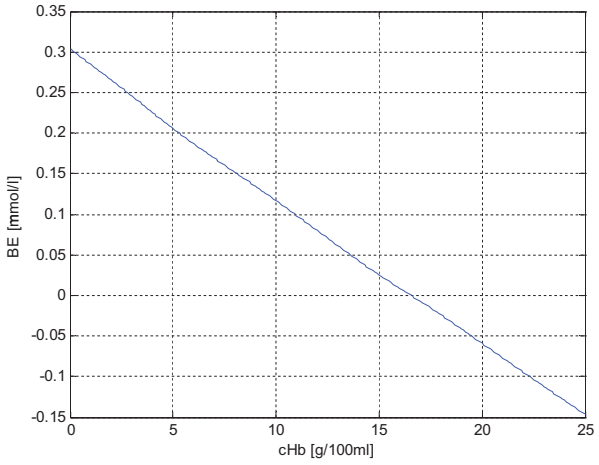

Fig. 12. *The dependence of BE on haemoglobin concentrations at pH=7.3878 and pCO_2 =41.862 torr according to the data from Sigaard-Andersen nomogram at 38°C (in fully oxygen-saturated blood). At 37°C, these values correspond to the standard values of pH=7.4 and pCO_2=40 torr, at which BE will be zero (at 37°C).*

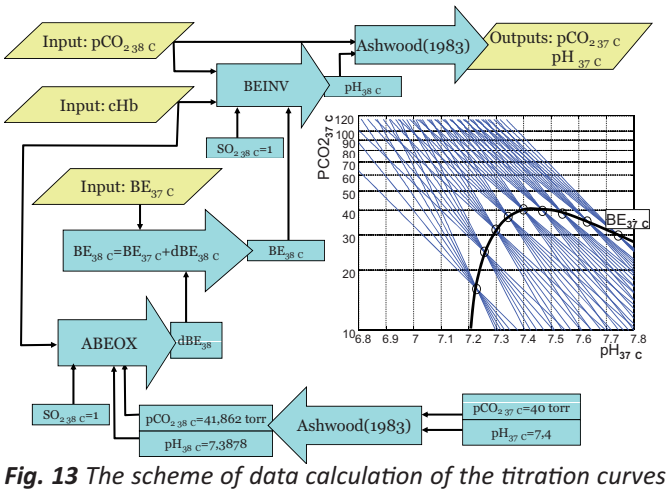

Fig. 13 *The scheme of data calculation of the titration curves for various haemoglobin concentrations and BE at 37°C. First, the re-calculation of the normal values pH=7.4 and pCO_2=40 torr from 37°C to 38°C is done. These values and the given haemoglobin concentration (supposed to be fully saturated by oxygen) is a base for the calculation of the correction shift of BE ($dBE_{38°C}$) corresponding to zero BE value at 37°C. The given $BE_{37°C}$ at 37°C is re-calculated to $BE_{38°C}$ at 38°C; this value and the set of $pCO_{2\ 38°C}$ values for the given haemoglobin concentrations (supposing fully saturated blood with oxygen) are a base for the calculation of $pH_{38°C}$. These values are then re-calculated to $pH_{37°C}$ and $PCO_{2\ 37°C}$, characterising the titration curve for the given haemoglobin concentration and selected BE at 37°C.*

For new coordinates of BE curves, see Fig. 15 and 16.

The calculation of new coordinates of BB curves (i.e. the coordinates where the curves – or lines in the semi-logarithmic scale – of blood samples with the same BB cut each other is simpler. In anaerobic heating (or cooling) must apply that:

$d[HCO_3^-] = -d[Buf^-]+d[H^+]$,

as $d[H^+]<<d[HCO_3^-]$,

thus, it applies that $d[HCO_3^-]=-d[Buf^-]$, i.e. BB do not vary; thus:

$BB_{37°C}=BB_{38°C}$

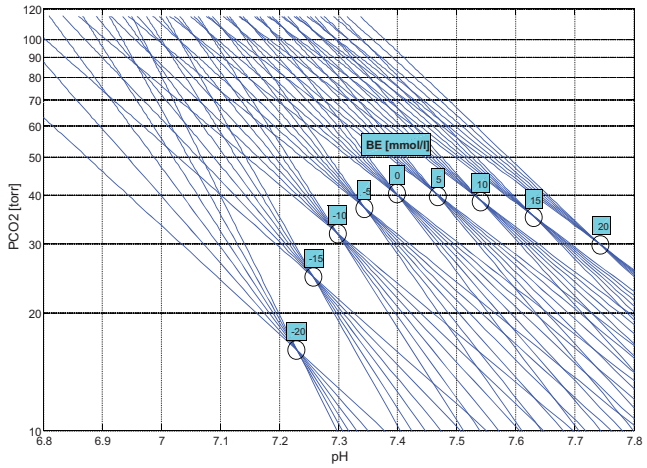

Fig. 14 *The titration curves for haemoglobin concentrations (0, 5, 10, 15, 20, 25 g/100 ml) and various BE at 37°C cut each other in the points characterising BE curve on Siggaard-Andersen nomogram corrected to 37°C.*

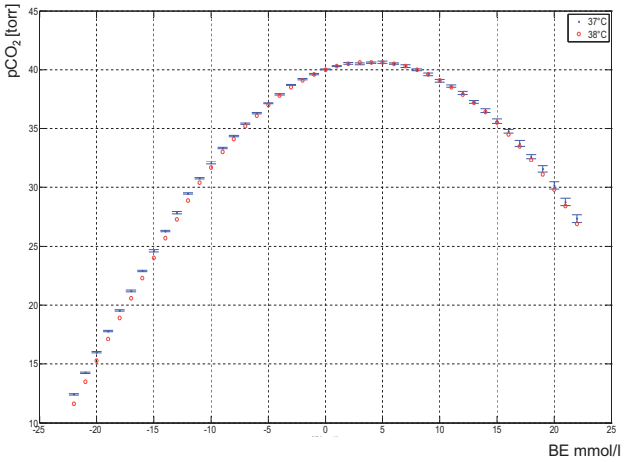

Fig. 15 *Results of BE curve correction from 38°C to 37°C – new coordinates in pCO_2 axis.*

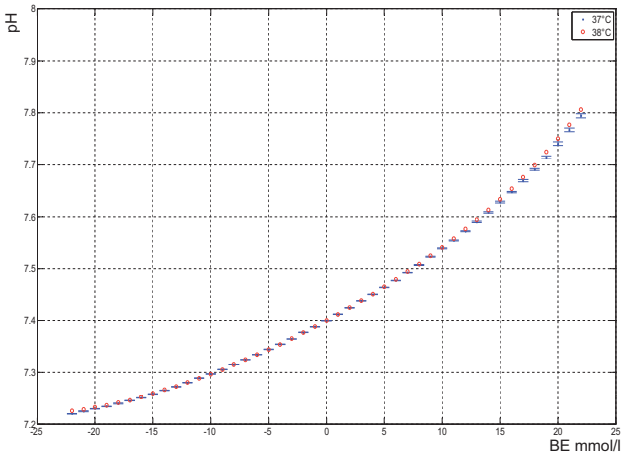

Fig. 16 *Results of BE curve correction from 38°C to 37°C – new coordinates in pH.*

The $pH_{38°C}$ and $pCO_{2\ 38°C}$ on the titration curve with a given BB are re-calculated from 38°C to 37°C to new $pH_{37°C}$ a $pCO_{2\ 37°C}$ by Ashwood et al. (1983) – however, the titration curve will correspond to the same BB (but to a different BE value).

It therefore suggests that the coordinates of the points of the BB curve of Siggaard-Andersen nomogram for 37°C can be obtained by the transformation of the coordinates of the points on the BB curve of the original Siggard-Andersen nomogram (representing the coordinates of the intersections of the titration curves with the same BB vale at 38°C) into new values by Ashwood et al. (1983).

BBs depend on BE normal BB (NBB). Although $BB_{38°C}$ and $BB_{37°C}$ are the same, it is possible to show that their normal values ($NBB_{37°}C$ and $NBB_{37°}C$) are different for 37°C and 38°C:

$NBB_{37°C}=BB_{37°C}-BE_{37°C} = BB_{38°C} - BE_{37°C}$

As (see above):

$BE_{37°C}=BE_{38°C} - dBE_{38°C}$

then:

$NBB_{37°C}=BB_{38°C} - BE_{38°C} + dBE_{38°C} = NBB_{38°C} + dBE_{38°C}$

The value of $dBE_{38°C}$ shift is calculated by the algorithm stated in Fig. 13 and depends on haemoglobin concentration. The

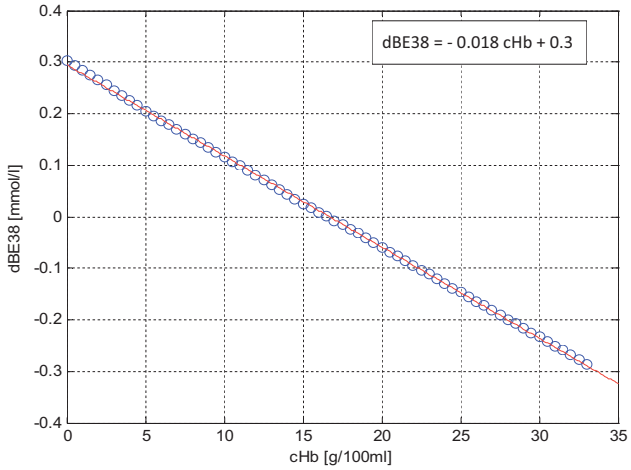

Fig. 17 *Linearization of the dependence of BE shift on haemoglobin concentration (cHb) expressed in g/100 ml during temperature change from 37°C to 38°C.*

consequent dependence can be linearised by the following relationship (Fig. 17)

dBE=0.3 – 0.018 cHb

where cHb is haemoglobin concentration in g/100ml.

$NBB_{38°C}$ is calculated by the known, in clinical practice used, relationship (Siggaard-Andersen, 1960):

$NBB_{38°C}$ = 41.7 + 0.42 cHb

The substitution of $NBB_{37°C}$ results in a slightly different relationship:

$NBB_{37°C}$ = 42.0 + 0.402 cHb

$BB_{37°C}$ value will be calculated from $BE_{37°C}$ and haemoglobin concentration:

$BB_{37°C}$ = 42.0 + 0.402 cHB + $BE_{37°C}$

For the comparison of the curve Siggaar-Andersen nomograms for 37°C and 38°C, see Fig. 18 and Table 1.

In clinical laboratory practice, data (pH and pCO_2) are measured at the standard temperature of 37°C; however,

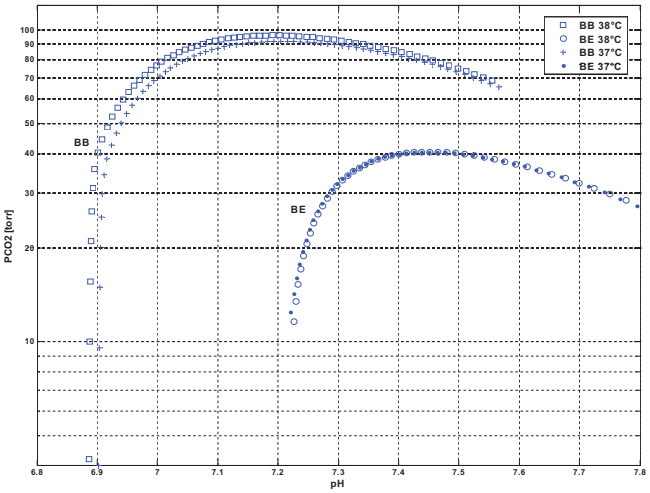

Fig. 18 *Correction of the values on BE and BB curves of Siggaard-Andersen nomogram (created originally for 38°C) to the standard temperature 37°C.*

BE [mmol/l]	37°C		38°C	
	pH	pCO_2 [torr]	pH	pCO_2 [torr]
-22	7,226	11,6	7,221	12,4
-21	7,229	13,5	7,225	14,2
-20	7,233	15,3	7,230	16,0
-19	7,237	17,1	7,235	17,8
-18	7,242	18,9	7,241	19,5
-17	7,247	20,6	7,246	21,2
-16	7,253	22,3	7,252	22,9
-15	7,259	24	7,258	24,6
-14	7,266	25,7	7,265	26,3
-13	7,273	27,3	7,272	27,9
-12	7,281	28,9	7,280	29,4
-11	7,289	30,4	7,289	30,8
-10	7,297	31,7	7,297	32,1
-9	7,306	33	7,306	33,3
-8	7,315	34,1	7,315	34,4
-7	7,324	35,2	7,324	35,4
-6	7,334	36,1	7,334	36,3
-5	7,344	37	7,344	37,2
-4	7,354	37,8	7,354	37,9
-3	7,365	38,5	7,365	38,7
-2	7,377	39,1	7,377	39,2
-1	7,388	39,6	7,388	39,6
0	7,4	40	7,400	40,0
1	7,412	40,3	7,412	40,3
2	7,425	40,5	7,424	40,5
3	7,438	40,6	7,438	40,5
4	7,451	40,6	7,450	40,6
5	7,465	40,6	7,463	40,7
6	7,479	40,5	7,477	40,5
7	7,494	40,3	7,492	40,3
8	7,509	40	7,507	40,0
9	7,525	39,6	7,523	39,6
10	7,541	39,1	7,539	39,1
11	7,558	38,5	7,555	38,6
12	7,576	37,9	7,572	38,0
13	7,594	37,2	7,590	37,3
14	7,613	36,4	7,608	36,5
15	7,633	35,5	7,628	35,6
16	7,654	34,5	7,648	34,7
17	7,676	33,5	7,669	33,7
18	7,699	32,3	7,691	32,6
19	7,724	31,1	7,714	31,6
20	7,75	29,8	7,740	30,2
21	7,777	28,4	7,767	28,8
22	7,806	26,9	7,795	27,3

BB [mmol/l]	37°C		38°C	
	pH	pCO_2 [torr]	pH	pCO_2 [torr]
14	6,903	4,0	6,887	4,2
15	6,904	9,6	6,888	10
16	6,904	14,9	6,889	15,6
17	6,905	20,1	6,89	21
18	6,906	25,1	6,891	26,2
19	6,908	29,8	6,893	31,1
20	6,911	34,3	6,896	35,8
21	6,916	38,6	6,901	40,3
22	6,923	42,7	6,908	44,6
23	6,932	46,6	6,917	48,7
24	6,940	50,3	6,925	52,6
25	6,949	53,9	6,934	56,3
26	6,958	57,2	6,943	59,8
27	6,967	60,4	6,952	63,1
28	6,976	63,3	6,961	66,2
29	6,985	66,1	6,97	69,1
30	6,994	68,7	6,979	71,8
31	7,004	71,1	6,989	74,3
32	7,013	73,4	6,998	76,7
33	7,022	75,6	7,007	79
34	7,032	77,6	7,017	81,1
35	7,042	79,4	7,027	83
36	7,051	81,0	7,036	84,7
37	7,060	82,5	7,046	86,3
38	7,070	83,9	7,056	87,7
39	7,080	85,1	7,066	89
40	7,090	86,3	7,076	90,2
41	7,100	87,3	7,086	91,3
42	7,110	88,3	7,096	92,3
43	7,120	89,0	7,106	93,1
44	7,131	89,7	7,117	93,8
45	7,141	90,3	7,127	94,4
46	7,151	90,7	7,137	94,9
47	7,162	91,1	7,148	95,3
48	7,173	91,4	7,159	95,6
49	7,183	91,6	7,169	95,8
50	7,194	91,7	7,18	95,9
51	7,205	91,8	7,191	96
52	7,215	91,8	7,202	96
53	7,226	91,7	7,213	95,9
54	7,237	91,5	7,224	95,7
55	7,248	91,2	7,235	95,4
56	7,260	90,8	7,247	95
57	7,271	90,4	7,258	94,6
58	7,282	89,9	7,269	94,1

BB [mmol/l]	37°C		38°C	
	pH	pCO_2 [torr]	pH	pCO_2 [torr]
59	7,294	89,5	7,281	93,6
60	7,306	88,9	7,293	93
61	7,318	88,2	7,305	92,3
62	7,330	87,4	7,317	91,5
63	7,342	86,7	7,329	90,7
64	7,354	85,9	7,341	89,9
65	7,365	85,0	7,353	89
66	7,378	84,1	7,366	88
67	7,391	83,0	7,379	86,9
68	7,404	82,0	7,392	85,8
69	7,417	80,8	7,405	84,6
70	7,430	79,7	7,418	83,4
71	7,443	78,5	7,431	82,2
72	7,456	77,3	7,444	80,9
73	7,469	76,0	7,457	79,6
74	7,482	74,7	7,47	78,2
75	7,496	73,3	7,484	76,7
76	7,510	71,8	7,498	75,2
77	7,523	70,3	7,512	73,6
78	7,537	68,8	7,526	72
79	7,551	67,2	7,54	70,4
80	7,566	65,6	7,555	68,7

Table 1 *Coordinates of BE and BB curves for original (37°C) and corrected (37°C) Siggaard-Andersen nomogram .*

they are assessed (BE calculation) by means of Siggaard-Andersen nomogram, created originally for 38°C. Thus, the comparison of the course of the titration curves according to the original and corrected Siggaard-Andersen nomogram (Fig. 19) is interesting in the view of clinical outcomes. It is obvious that noticeable deviations occur as late as with BE under 10 mmo/l and more significant ones at BE exceeding 15 mmol/l.

9. Erythrocytes and plasma

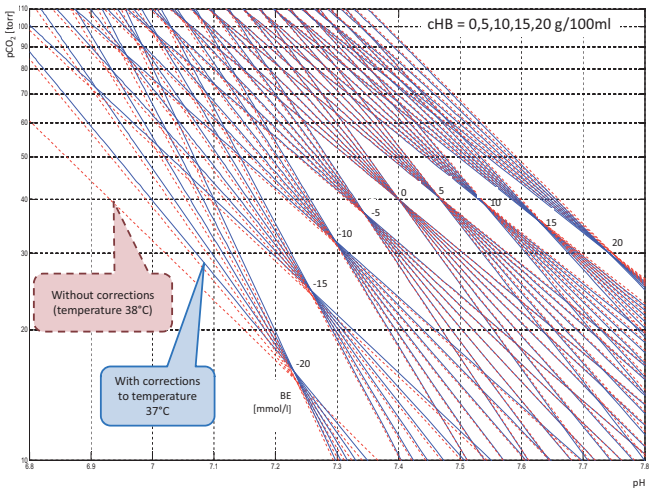

Fig. 19 *Comparison of the titration curves calculated according to original and corrected Siggaard-Andersen nomogram.*

Now, Siggaard-Andersen nomogram is formalised for the same temperature, which detailed models of plasma acid-base balance, created by Stewart´s model, are identified for. These models (e.g. Figge 2009), anyhow considering the details of the effect of the dissociation constants of particular amino acids in an albumin molecule, entirely neglect the effect of such a substantial buffer as haemoglobin in erythrocytes. On the other hand, the drawback of the models based on experimental data derived from Siggaard-Andersen nomogram, is a precondition of normal plasma protein concentration.

The aim of this work is to connect both approaches into one model, potentially usable as a subsystem of the complex model of homeostasis in the organism with the possibility to simulate complex osmotic, ion, volume and acid-base disturbances.

First, using the experimental data from Siggaard-Andersen nomogram, the titration curves of plasma and erythrocytes should be separated – the result should be a model of the buffer behaviour of erythrocytes, connected with the detailed model of plasma acid-base balance, created by Stewart´s approach, regarding various plasma protein and phosphate concentrations.

Siggaard-Andersen verified experimentally that the curves of plasma and blood samples with various haematocrit and the same BE cut each other in one point on the BE curve (see Fig. 2). Similarly, the curves of blood samples with the same BB cut each other in one point on the BB curve. It raises a question, ***why the BB and BE titration curves cut each other in the same points on Siggaard-Andersen nomogram?***

To reply this question, it is necessary to realise that blood titration with carbon dioxide results in the increase in bicarbonate concentrations in plasma and erythrocytes during the increase in pCO_2.

Regarding the plasma itself by Stewart – then, during ***plasma titration with carbon dioxide***, the sum of bicarbonates and all non-bicarbonate buffer bases, forming ***BB_p*** and ***SID***, are ***unchanged*** (Fig. 20) – ***SID*** and ***pCO_2*** are therefore ***mutually independent variables***, which, together with another independent variable, plasma protein concentration, determines the value of the dependent variable – pH.

This basic Stewart´s canon does not apply ***in blood*** (see Fig. 21) – ***in the titration with carbon dioxide, plasma SID, corresponding*** (with the abovementioned objections) ***with BB_p, varies***. The increase in pCO_2 causes the increase in BB_p (and SID), whereas the decrease in pCO_2 causes the decrease in BB_p. As the erythrocyte has more non-bicarbonate bases (particularly due to haemoglobin) than plasma, and the dissociation reaction of carbonic acid is more shifted to the right, there is a higher increase in bicarbonate concentrations in erythrocytes than in plasma. Bicarbonates are transported into plasma by the concentration gradient (by exchange for chloride ions). Thus, the increase in CO_2 concentrations is associated with the decrease or increase in BB concentrations in erythrocytes or plasma, respectively.

Blood titration with carbon dioxide helps achieve pCO_2 at which BB concentrations in erythrocytes and plasma equilibrate ($BB_e = BB_p$). This value determines the place where the titration curves with the same total BB and various haematocrit (Hk) will cut each other on Siggaard-Andersen nomogram.

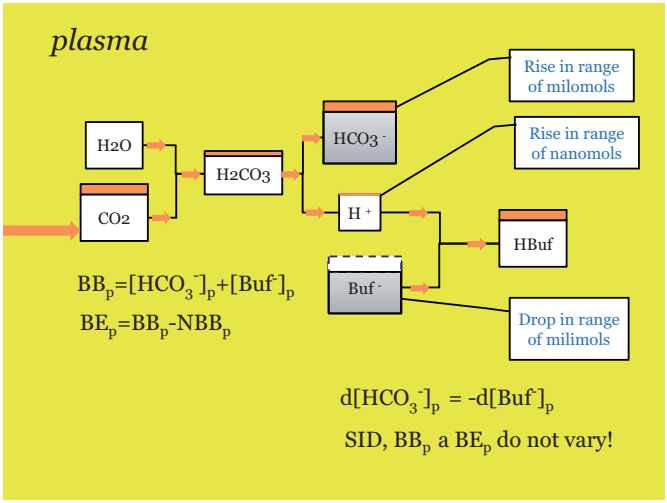

Fig. 20. *Plasma titration with carbon dioxide – BE_p, BB_p and SID do not vary. Thus, pCO_2 and SID are mutually independent.*

As:

$$BB = BB_p (1 - Hk) + BB_e Hk = BB_p + Hk (BB_e - BB_p)$$

The second member of the sum is zero with $BB_e = BB_p$ and the whole blood BB does not depend on haematocrit. With this pCO_2 (and proper plasma pH) when $BB_p=BB_e$, the blood exert any value of haematocrit; all titration curves of blood samples with various haematocrit therefore cut each other in this point. Thus, ***the BB curve on Siggaard-Andersen nomogram is a geometric site of the points where plasma and erythrocytes have the same buffer base concentrations***, as at $BB_e=BB_p$ the whole blood *BB* does not depend on haematocrit (Hk):

A similar consideration applies for the BE curve, too. As:

$$BE=BE_p (1 - Hk) + BE_e Hk = BE_p + Hk (BE_e - BE_p)$$

the second member of the sum equals zero at $BE_e=BE_p$ then and the whole blood BE does not depend on haematocrit (*Hk*) or haemoglobin concentration. Thus, ***the BE curve on Siggaard-Andersen nomogram is a geometric site of the points with the same BE in the whole blood and plasma,*** as the whole blood BE does not depend on haematocrit at proper pCO_2 and pH, when $BE_e=BE_p$.

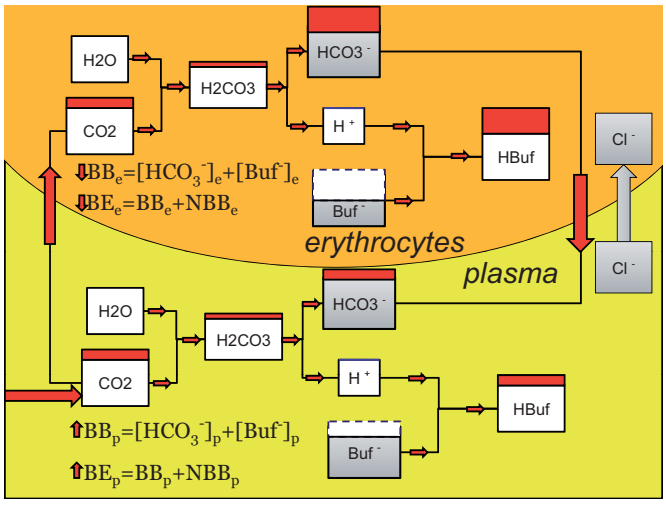

Fig. 21 *Blood titration with carbon dioxide – SID varies during pCO_2 changes (thanks to the exchange of HCO_3^- for Cl^-). SID and pCO2 in the whole blood are not mutually independent.*

The BE curve can also be interpreted in other way. Regarding the fact that BE is the difference between BB and normal proper NBB for the given haemoglobin concentration, then the precondition of the equality of BE in plasma and erythrocytes means:

$BB_e - NBB_e = BB_p - NBB_p$

This can be specified:

$BB_e - BB_p = NBB_e - NBB_p = constant$

This means that ***the BE curve can be interpreted as the geometric site of the points (i.e. pCO_2 and pH values) with a constant difference between BB in erythrocytes and plasma, which equals the difference between the proper values in erythrocytes and plasma (pCO2=40 torr and plasma pH=7.4).***

If the equation $NBB_{38°C} = 41.7 + 0.42$ cHb applies (Siggaard-Andersen, 1962), then haemoglobin concentration in erythrocytes cHb = 33.34 g/100ml is $NBB_e - NBB_p = 0.42 \times 33.34 = 14$ mmol/l (according to our correction of Siggaard-Andersen nomogram, this value was $0.402 \times 33.34 = 13.4$ mmol/l for 37°C).

Siggaard-Andersen used the mixture of O_2 - CO_2 for blood titration with fully oxygen-saturated blood – in fact, the BE curves are those for fully oxygenated blood – i.e. the abovementioned standardised oxyvalues of Base Excess – BEox (Kofránek, 1980). BE or BB exert a linear increase in haemoglobin oxygen desaturation:

BE = BEox + 0.2 cHB (1-sO2)

where *cHb* is haemoglobin concentration [g/100ml] and sO_2 is haemoglobin oxygen saturation (Siggaard-Andersen 1988).

10. Separation of plasma and erythrocyte titration curves on Siggaard-Andersen nomogram

It is recommended to test if it is possible to make a model of blood acid-base balance from the experimental data on Siggaard-Andersen nomogram as a combination of the models of plasma and erythrocyte titration curves (Fig 22). The titration curves (plotted as lines in the semi-logarithmic scale) can be read out direct from the nomogram. The titration curves of erythrocytes can be obtained from the nomogram as follows: chose the blood concentration of haemoglobin 33.34 g/100 ml, which is the value with haematocrit having the value of one. The titration curve of this "virtual blood" with carbon dioxide follows pH variations (measured on the outer side of the erythrocyte) during pCO_2 changes. The titration curve of the blood with a given haemoglobin and, thus, haematocrit concentrations cHb (in g/100ml blood).

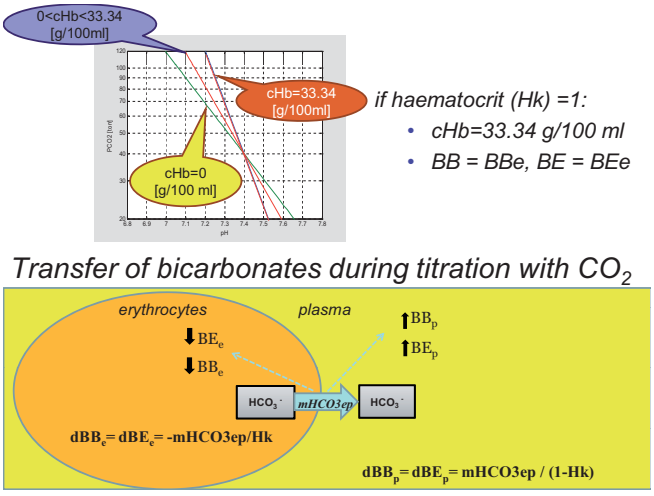

Fig. 22 *The transfer of bicarbonates and variations in plasma and erythrocyte BB and BE during the titration with carbon dioxide. The titration curve of blood (a line in the semi-logarithmic scale) is calculated from the combination of plasma and erythrocyte titration curves and from the transfer of bicarbonates between erythrocytes and plasma, which change proper BE and BB in plasma and erythrocytes (depending on haematocrit).*

Hk=cHb/33.34

(supposing the normal haemoglobin concentration in erythrocytes 33.34 g/100ml) will lie between the titration curves of plasma and erythrocytes in the semi-logarithmic coordinates $\log_{10}(pCO_2)$ – pH. It will cut the curves for plasma and erythrocytes in a point of the BE curve. As non-bicarbonate buffers (haemoglobin and phosphates) have a higher buffer capacity in erythrocytes than those in plasma (plasma proteins and phosphates), and non-bicarbonate bases in erythrocytes bind more hydrogen ions than those in plasma during blood titration with increasing concentrations of carbon dioxide, the concentration of bicarbonates increases more significantly in the erythrocyte than in plasma. The consequence is the transfer of bicarbonates between the erythrocyte and plasma (accompanied with a counter chloride transport). Labelling the amount of bicarbonates in 1 litre, transferred from erythrocytes into plasma during blood titration with carbon dioxide: mHCO3ep [mmol/l], then the variations in plasma BE and BB is:

$dBB_p = dBE_p = mHCO3ep/(1-Hk)$

The corresponding variation of BE in erythrocytes is:

$dBB_e = dBE_e = -mHCO3ep/Hk$

Choosing, for example, haemoglobin concentration 15 g/100 ml (and haematocrit concentration 15/33.34=0.4449) for the transfer of 1mmol of bicarbonate, there will be an increase and decrease in plasma and erythrocyte BE as well as BB concentrations by 1/(1-0.4449)=1.8015 mmol/l and by 1/0.4449=2.2477 mmol/l, respectively. There will be left and right shifts on plasma and erythrocyte titration curves (see Fig. 23), respectively – their intersection corresponds with the point on the titration curve with haemoglobin concentration 15 g/100 ml, in which 1 ml of bicarbonates were transferred from erythrocytes into plasma during the increase of pCO_2 from the baseline value of 40 tor. As seen in Figure 23, this intersection lies on the titration curve with haemoglobin concentration 15 g/100 ml, modelled according to the data in Siggaard-Andersen nomogram (by means of the abovementioned function BEINV). Similarly, this curve includes the intersections of the left and right s of plasma and erythrocyte curves after the transfer of 2 and 1 mmol of bicarbonates from erythrocytes into plasma (during pCO_2 increase) and from plasma into erythrocytes (during pCO_2 increase), respectively.

Figures 24 and 25 show the results of the modelling of the titration curves for blood titration with carbon dioxide at BE -10 mmol/l and 10 mmol/l. Fig. 26 shows the results of the modelling of blood titration with carbon dioxide in the range of BE -20 to 20 mmol/l.

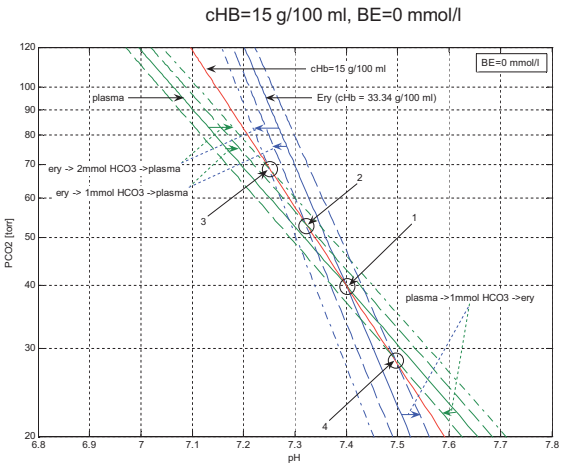

Fig. 23 *Model of the titration curves of plasma, erythrocytes and blood with haemoglobin concentration 15 g/100 ml with BE=0 mmol/l. The plasma and erythrocyte curves cut each other in point (1) and on Base Excess in point BE=0, respectively. The transfer of bicarbonates from erythrocytes into plasma during blood titration with carbon dioxide shifts the plasma and erythrocyte curves to the right and to the left (with the increase and decrease in plasma and erythrocyte BE and BB values), respectively. The curves cut each other in points (2) and (3) on the titration curve with haemoglobin concentration 15g/100 ml. The decrease in pCO_2 causes the transfer of bicarbonates from plasma to erythrocytes with following decrease in plasma BE and BB, which results in the right shift of the titration curve and increase in erythrocyte BB with the left shift of erythrocyte curve. The curves cut each other on the blood titration curve (in point 4) with haemoglobin concentration 15 g/100 ml, modelled by the data in Siggaard-Andersen nomogram. This suggests that the titration curves can be modelled by the intersections of the shifts on plasma and erythrocyte titration curves.*

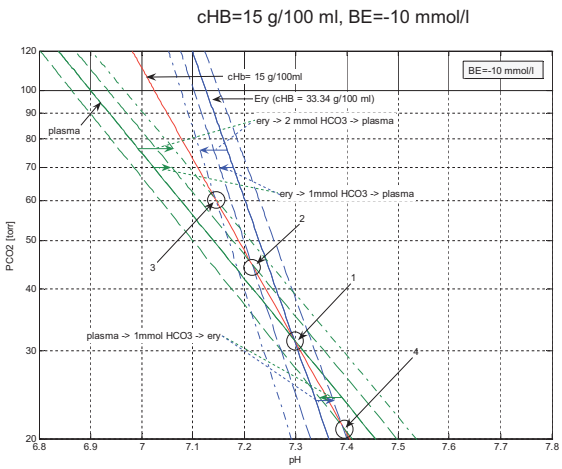

Fig. 24 *Model of the titration curves of plasma, erythrocytes and blood with haemoglobin concentration 15 g/100 ml with BE=-10 mmol/l. If the blood titration curve of the titration with carbon dioxide is modelled by means of the intersections of the shift of blood and erythrocyte titration curves caused by the transfer of bicarbonates between the erythrocyte and plasma, points of the titration curve are obtained (similarly as in the previous figure), which cover the titration curve of blood with haemoglobin concentration 15 g/100 ml, modelled by Siggaard-Andersen nomogram.*

It has been shown that the titration curves modelled by means of the intersections of the shifts of plasma and erythrocyte titration curves (due to the transfer of bicarbonates between the erythrocyte and plasma) copy the titration curves modelled direct by Siggaard-Andersen nomogram with a sufficient accuracy.

It therefore means that the modelling of blood titration with carbon dioxide can be based on the combination of plasma and erythrocyte titration curves. The modelling of blood titration with **varied plasma protein concentration** can be based on the combination of **plasma titration curve** with various plasma protein concentrations (for which, however, Siggaard-Andersen nomogram does not apply) – for example by Figge-Fencl ´s model (Figge, 2009), and **erythrocyte titration curve** (obtained from the experimental data of Siggaard-Andersen nomogram, corrected to 37°C).

11. Connection of the erythrocyte model by Siggaard-Andersen nomogram, corrected to 37°C and Figge-Fencl's model of plasma

Fig. 27 shows erythrocyte titration curves with various BE by Siggaard-Andersen nomogram – the erythrocytes are modelled as blood with haemoglobin concentration 33.34 g/100ml (corresponding to the proper haematocrit value of 1). In the semi-logarithmic scale, these curves are lines with variable slopes (k) and offset (h), depending on BE concentrations in erythrocytes (BE_{er}).

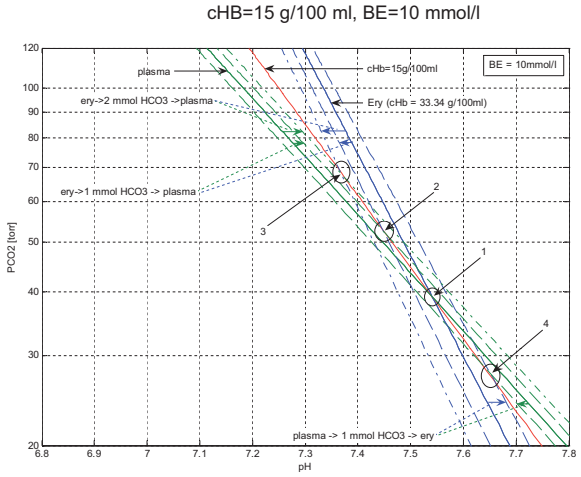

Fig. 25 *Model of the titration curves of plasma, erythrocytes and blood with haemoglobin concentration 15 g/100 ml with BE=10 mmol/l. Similarly as in the previous figures, the intersections of the shifts of the plasma and erythrocyte titration curves caused by the transfer of bicarbonates between the erythrocyte and plasma cover the titration curve of blood with haemoglobin concentration 15 g/100 ml, modelled by Siggaard-Andersen nomogram.*

The erythrocyte titration curves will be approximated according to the following relationships:

$\log10(pCO2) = k\ pH + h$

$k=f(BE_{er})$

$h=g\ (BE_{er})$

Functions "f" and "g" are approximated by polynomic regression according to the data from Siggaard-Andersen nomogram, corrected to 37°C (see Fig. 28 and 29).

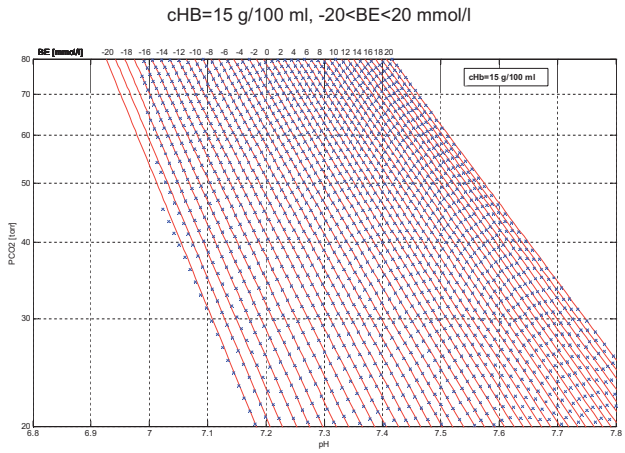

Fig. 26 *Model of the titration curves of plasma, erythrocytes and blood with haemoglobin concentration with various BE concentrations ranged from -20 to 20 mmol/l by Siggaard-Andersen nomogram (continuous lines). The crosses represent the titration curves modelled as the intersections of the shifts of plasma and erythrocyte titration curves caused by the transfer of bicarbonates between the erythrocyte and plasma. This means that the whole blood titration curves on Siggaard-Andersen nomogram can be calculated from the plasma and erythrocyte titration curves with sufficient accuracy (modelled as blood with limit haematocrit 1 and haemoglobin concentration 33.34 g/100 ml).*

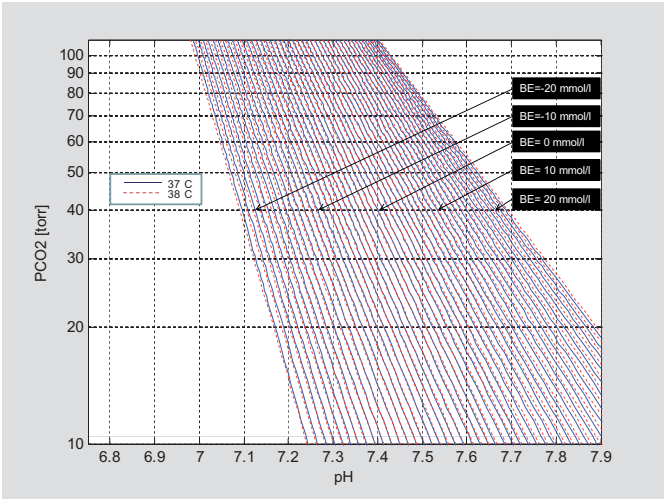

Fig. 27 *Erythrocyte titration curves (lines in the semi-logarithmic scale) by Siggaard-Andersen nomogram at 38°C and after correction to 37°C at various BE concentrations.*

pH (pH of the outer side of erythrocytes), depending on pCO_2 and BE in erythrocytes (BE_{er}), is calculated by means of eryBEINV function; for its algorithm, see Fig. 30.

pH=eryBEINV(pCO_2,BE_{er})

The erythrocyte model is connected with the plasma model . Figge-Fencl´s model (Figge, 2009), combined, in addition, with the effect of globulin concentrations (calculated by means of their "buffer value" by Siggaard-Andersen, 1995), was selected as a plasma model. BEINV function calculates blood pH in dependence on pCO_2, total phosphate (Pitot), albumin (Alb), globulin (Glob) and haemoglobin concentrations as well as on standardised oxyvalues BEox, (i.e. BE found in fully oxygenated blood), pCO_2 and haemoglobin oxygen saturation:

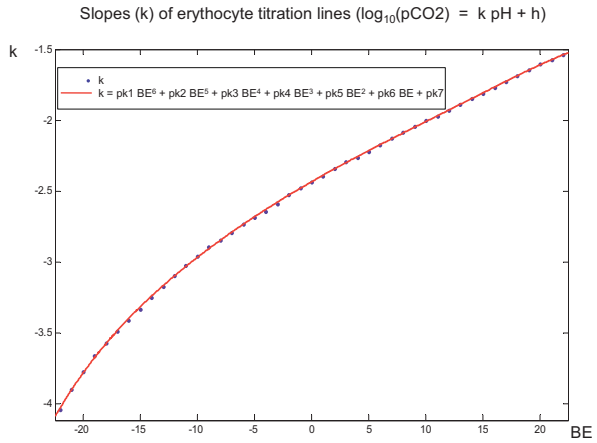

Fig. 28 *Polynomic regression of the variable slopes of erythrocyte titration lines.*

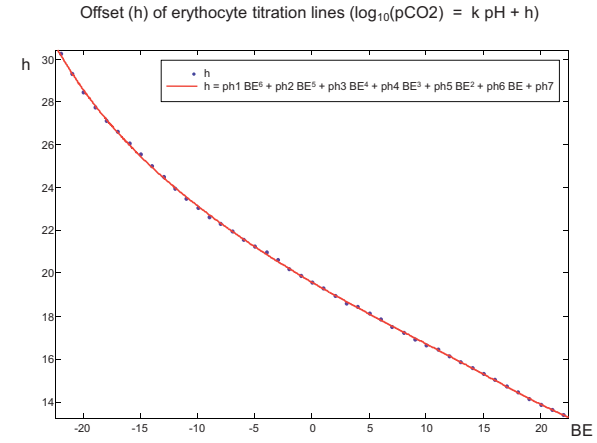

Fig. 29 *Polynomic regression of the variable offset of erythrocyte titration lines.*

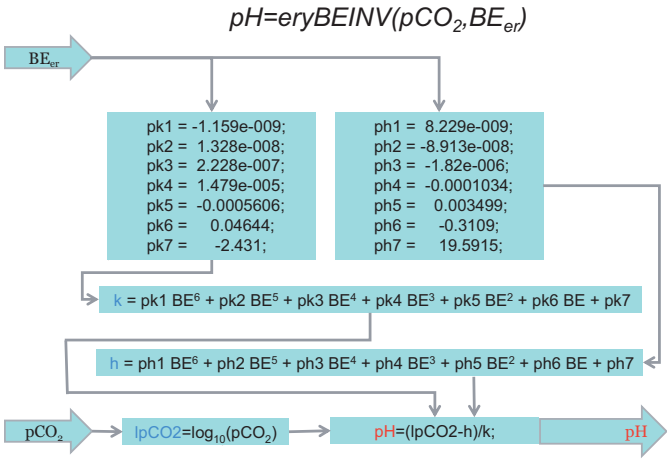

Fig. 30 *Algorithm of the calculation of erythrocyte titration curves*

pH=bloodBEINV(Pitot,Alb,Glob,cHb,BEox,pCO_2,sO_2)

For the principle of the calculation and for the algorithm itself, see Fig. 31 and 32, respectively.

First, BE is calculated according to the grade of desaturation (from sO_2) and BEox. This value is considered initial for plasma and erythrocytes (BE). pH is calculated from pCO_2.

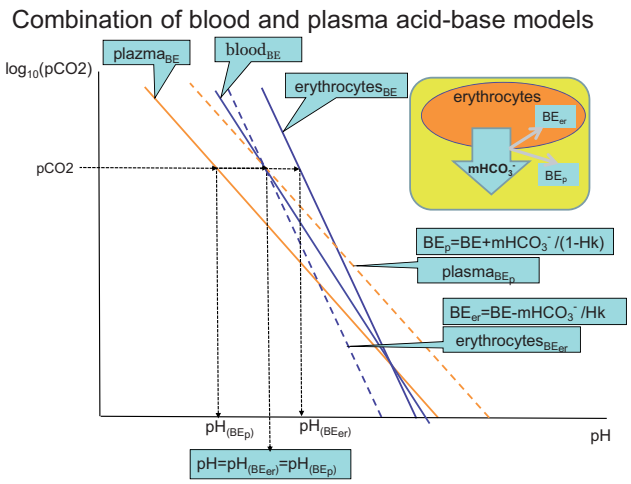

Fig. 31 *Principle of the calculation of the whole blood titration curves. At given BE plasma and erythrocytes titration curves (plasma$_{BE}$ and erythrocytes$_{BE}$) have a different slopes, hence at given pCO$_2$ a different pH can be calculated. Searched blood titration curve (blood$_{BE}$) lies between plasma$_{BE}$ and erythrocytes$_{BE}$ curves. In the blood at given haematocrit (Hk) plasma and erythrocyte BE (BE$_p$ and BE$_{er}$) is shifted because of HCO$_3^-$/Cl$^-$ erythrocyte-plasma exchanges. New titration curves of plasma and erythrocytes (plasma$_{BEp}$, erythrocytes$_{BEer}$) can be calculated. Algorithm seeks the intersection of blood$_{BE}$, plasma$_{BEp}$, and erythrocytes$_{BEer}$ curves at given pCO$_2$.*

However, the plasma titration curve has a smaller slope than that for erythrocytes (see Fig. 31) and plasma pH ($H_{(BEp)}$) is calculated according to plasma BE (BE_p); pH on the outer side of erythrocytes ($pH_{(BEer)}$), calculated according to erythrocyte BE (BE_{er}), is different. Then, the transfer of bicarbonates between plasma and erythrocytes is calculated by iteration – the transfer causes variations in plasma (BE_p) and erythrocyte (BE_{er}) BEs – the ratio of BE varaitions in erythrocytes and plasma depend on haematocrit. The iteration converges to the final value in plasma calculated according to both erythrocyte and plasma BEs ($pH = pH_{(BEp)} = pH_{(BEer)}$).

The algorithm also calculates the normal SID (NSID) – i.e. the SID, in which pH=7.4 with the given haemoglobin, albumin and phosphate concentrations and pCO_2=40 torr.

There is a wider definition of BE in this model compared with classical Siggaard-Andersen´s nomogram interpretation – its normal value depends not only on haemoglobin concentrations but also on albumin, globulin and phosphate concentrations - like Siggaard-Andersen´s van Slyke equation (Siggaard-Andersen, 1977, 2006). Unlike in classical plasma models by Stewart and his followers, this model enables to demonstrate that the relationship between SID a pCO_2 does not apply in the whole blood. The model (and the related formalised relationships) can be used in a number of clinical-physiological calculations.

For the model, including its source text and the description of all used mathematical relationships and algorithms, see ***www.physiome.cz/acidbase***.

11. Conclusion

Siggaard-Andersen nomogram was recalculated from original 38°C to standard 37°C. The experimental data of Fige and Fencl´s model of plasma acid-base balance was combined with the data based on Siggaard-Andersen nomogram,

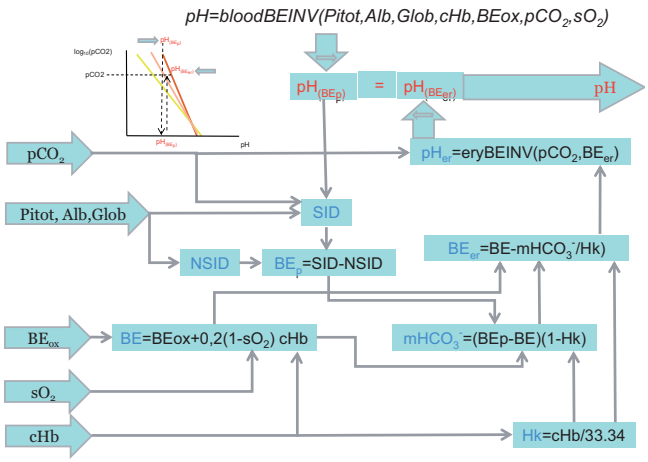

Fig. 32 *Algorithm of the calculation of the whole blood titration curves.*

corrected to 37°C. It was obtained a model of blood acid-base balance combining the plasma model with variable albumin, globulin and phosphate concentrations and connected with the erythrocyte model. The model is a core of an extent model of acid-base balance which enables the realisation of pathogenesis of acid-balance disturbances in compliance with the bilance approach to the interpretation of ABB disturbances, published earlier (Kofránek et al., 2007).

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