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DOMESTIC SWINE IN PHYSIOLOGICAL RESEARCH
V. Construction of Acid-Base Alignment Nomograms to Estimate Buffer Base and Base Excess Concentrations in Arterial Blood from Immature Pigs

JOHN P. HANNON, PhD

DIVISION OF COMBAT CASUALTY CARE

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20. Abstract:

Inaccurate results are obtained when nomograms or other procedures based on the acid-base characteristics of human blood are used to estimate the buffer base or base excess concentration of porcine blood. The inaccuracies are due primarily to the higher plasma pH and bicarbonate concentration that characterize normal porcine as compared to normal human blood. Smaller errors are introduced by lower porcine plasma protein levels and a slightly higher body temperature. To address these problems, the acid-base characteristics of a population of immature domestic pigs were used to construct a blood acid-base alignment nomogram with scales to estimate porcine buffer base concentration. It was based on average plasma bicarbonate concentration of 31.6 mEq/l and plasma albumin and globulin levels of 25.4 and 32.2 g/l, respectively. A measurement temperature of 38 C was assumed. This nomogram was used subsequently to construct a blood acid-base alignment nomogram with scales to estimate porcine base-excess concentration. It was based on the assignment of zero base excess to blood with a pH of 7.50 and a P_{CO_2} of 40 torr. Construction details, including tabular data reflecting the acid-base characteristics of porcine plasma and erythrocytes, are provided.

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ABSTRACT

Inaccurate results are obtained when nomograms or other procedures based on the acid-base characteristics of human blood are used to estimate the buffer base or base excess concentration of porcine blood. The inaccuracies are due primarily to the higher plasma pH and bicarbonate concentration that characterize normal porcine as compared to normal human blood. Smaller errors are introduced by lower porcine plasma protein levels and a slightly higher body temperature. To address these problems, the acid-base characteristics of a population of immature domestic pigs were used to construct a blood acid-base alignment nomogram with scales to estimate porcine buffer base concentration. It was based on average plasma bicarbonate concentration of 31.6 mEq/l and plasma albumin and globulin levels of 25.4 and 32.2 g/l, respectively. A measurement temperature of 38 C was assumed. This nomogram was used subsequently to construct a blood acid-base alignment nomogram with scales to estimate porcine base-excess concentration. It was based on the assignment of zero base excess to blood with a pH of 7.50 and a P CO₂ of 40 torr. Construction details, including tabular data reflecting the acid-base characteristics of porcine plasma and erythrocytes, are provided.

Key Words: acid-base alignment nomograms, porcine blood, buffer base and base excess estimation.



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PREFACE

Previous reports in this series on Domestic Swine in Physiological Research have included the following titles:

- I. A Biomedical Model
- II. Electrolyte Values for Arterial Serum from Young Anesthetized Pigs Maintained Under Steady-State Ventilatory Condition
- III. Blood Gas and Acid-Base Values of Arterial Blood from Young Anesthetized Pigs Maintained under Steady State Conditions
- IV. A Blood Acid-Base Curve Nomogram for Immature Pigs

The next report will be concerned with the blood volume of conscious immature animals and the role of the spleen as an erythrocyte storage organ.

The author sincerely appreciates the many editorial and format suggestions provided by Lottie B. Applewhite and the numerous hours spent by Sue Zuckerbrot in typing, proofreading, and assembling this report.

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DOMESTIC SWINE IN PHYSIOLOGICAL RESEARCH
V. CONSTRUCTION OF ACID-BASE ALIGNMENT NOMOGRAMS TO ESTIMATE BUFFER
BASE AND BASE EXCESS CONCENTRATIONS IN ARTERIAL BLOOD FROM IMMATURE
PIGS

Arterial acid-base characteristics were determined recently for a population of immature domestic swine (1). The measurements, made on blood samples obtained under near-basal conditions, revealed two major deviations from the characteristics of human blood obtained and measured under similar circumstances: porcine pH averaged 7.496, bicarbonate concentration 31.6 mEq/l. In both instances, the values exceeded commonly accepted human norms, namely a pH of 7.40 and a bicarbonate concentration of 24.5 mEq/l (2-4).

Because of these species differences, conventional procedures did not provide an accurate estimate of porcine base excess concentration. Such procedures were designed originally for human (3,4) and subsequently for canine (5) blood and are based on the assignment of a zero base excess value to blood with a pH of 7.40 and a P_{CO_2} of 40 torr. The latter value would be appropriate for swine (average P_{CO_2} of 40.6 torr) but the former would not. Consequently, when attempts were made to use these procedures with porcine blood, positive base excess values were always obtained. An automated blood gas analyzer programmed for human samples, for example, gave an average porcine base excess concentration of 7.7 mEq/l (1).

In an effort to define and estimate base excess concentration more accurately, constant P_{CO_2} titration curves were prepared and a curve nomogram appropriate to porcine blood was constructed (1) according to the procedures described by Siggaard-Andersen (3,4). In this nomogram, base excess was assigned a zero value when blood pH was 7.50 and P_{CO_2} was 40 torr. The higher pH value associated with zero base excess caused a rightward displacement of the porcine nomogram compared to a human nomogram constructed in the same study. Base excess loci in the porcine nomogram, in addition to a rightward displacement, also reflected higher acid buffering capacity for porcine than human blood.

The present report describes construction of alignment nomograms for estimating the buffer base and base excess concentrations in blood from young pigs. This effort stemmed from the limited utility of curve nomograms in many blood gas laboratories. Their use requires equilibration of blood sample pairs to two known CO_2 tensions followed by pH measurements. This procedure is not only time-consuming, but oftentimes the requisite tonometry equipment is not available. Alignment nomograms, in contrast, can be used to estimate buffer base

or base excess concentration from pH and $P\text{CO}_2$ measurements made on a single blood sample. The pH and $P\text{CO}_2$ measurements are easily obtained with modern blood gas equipment.

METHODS

The Buffer Base Alignment Nomogram

The buffer base concept and a procedure for its estimation with an alignment nomogram was originally described in 1958 by Singer and Hastings (6). They defined blood buffer base concentration $[\text{BB}^-]_b$ as the sum of all buffer anion concentrations. Included were contributions from plasma bicarbonate, albumin and globulin plus intraerythrocytic bicarbonate (included carbamino CO_2) and hemoglobin. Construction of the nomogram for porcine blood proceeded through three stages. It followed essentially the same procedures, with certain modifications to be indicated later, as those described by Singer and Hastings (6). First, the Henderson-Hasselbalch equation was graphically represented in the form of an alignment nomogram. Next, acid-base data associated with a range of plasma pH values and erythrocyte concentrations were calculated and tabulated. Finally, these data were used in a sequence of calculations which culminated in the positioning of an array of buffer base loci and scales within the Henderson-Hasselbalch alignment nomogram.

Graphic Representation of the Henderson-Hasselbalch Equation

Alignment nomograms which facilitated the rapid and reasonably accurate estimation of various blood gas and acid-base values in human blood were introduced in 1924 by Henderson et al (7), and similar procedures were used shortly thereafter by Van Slyke and Sendroy (8) to prepare a simple graphic representation, or nomogram, of the Henderson-Hasselbalch equation. The nomogram was based on the same general principles as those used in slide rule construction, i.e., it allowed the estimation of any one variable contained in the equation when the other two were known. Construction of the porcine acid-base nomogram was patterned after the Van Slyke and Sendroy procedure (8), modified somewhat to reflect the pH-dependent alterations in the pK' of carbonic acid dissociation at 38 C reported by Severinghaus et al (9).

The first construction step involves the positioning of two parallel logarithmic scales on either side of linear graph paper. (A 25 x 38 cm graph sheet graduated 10 mm per cm [K and E type 47-1513] is well-suited for this purpose.) Values on the right-hand scale, representing $P\text{CO}_2$, increase in magnitude from top to bottom of the graph sheet while those on the left-hand scale, representing bicarbonate concentration, increase from bottom to top. The logarithmic form and the direction of these two scales expresses the relationship of pH to $P\text{CO}_2$ and $[\text{HCO}_3^-]$ in the Henderson-Hasselbalch equation; i.e.,

$$\text{pH} = \text{pK}' + \log \frac{[\text{HCO}_3^-]}{S_0(\text{P}_{\text{CO}_2})} \quad (1)$$

where S_0 represents the CO_2 solubility factor which, for plasma at 38 C, has a value of 0.0301 mM/l/unit (torr) change in P_{CO_2} (10). A calculator program to facilitate preparation of the logarithmic scales is described in the Appendix.

After the P_{CO_2} and $[\text{HCO}_3^-]$ scales are positioned, the Henderson-Hasselbalch equation is used to delineate the pH scale. The procedure is illustrated in Figure 1. Accordingly, two or more pairs of P_{CO_2} and $[\text{HCO}_3^-]$ values compatible with a pH of 6.8 and a pK' of 6.113 (6) are calculated and connected by straight lines. The intersection of these lines defines the location of pH 6.8 on the scale. The same procedure, except for a change in pK' to 6.060 (9), is used to locate the pH 8.0 position. Subsequently, either of the two techniques can be used to locate the scale positions for intermediate pH values. The more laborious technique involves repetition of the procedure just described, using pK' values (9) which are appropriate to each of the intermediate pH values. Alternatively, the distance in millimeters between pH 6.8 and 8.0 is divided by 120 to determine the millimeter increment corresponding to a pH increase of 0.01 unit and successively adding these increments to the scale starting at pH 6.8. The second technique, although slightly less accurate since it assumes equally spaced pH increments, is well within acid-base measurement errors and is readily accomplished with a simple calculator program (Appendix).

Blood Acid-Base Characteristics as Influenced by Plasma pH and Erythrocyte Concentration

Alterations in the pH of plasma (pH_p) and the erythrocyte fraction (V_p) of blood have pronounced effects on blood acid-base status. The second stage of buffer base nomogram construction entails a systematic tabulation of these effects.

The interrelationships of pH_p , plasma pK' (pK'_p), the antilogarithm of $\text{pH} - \text{pK}'_p$ (H-K_p), and intraerythrocytic pH (pH_i), measured at a blood temperature of 38 C, are shown in the first four columns of Table 1. The values for pK'_p were taken from the report of Severinghaus et al (9), and those for pH_i from the report of Dill et al (11)--corrected to 38 C by the factor -0.016 pH units per degree increase in temperature (12). Values in the fifth column, plasma protein anion concentration, were calculated by the equation of

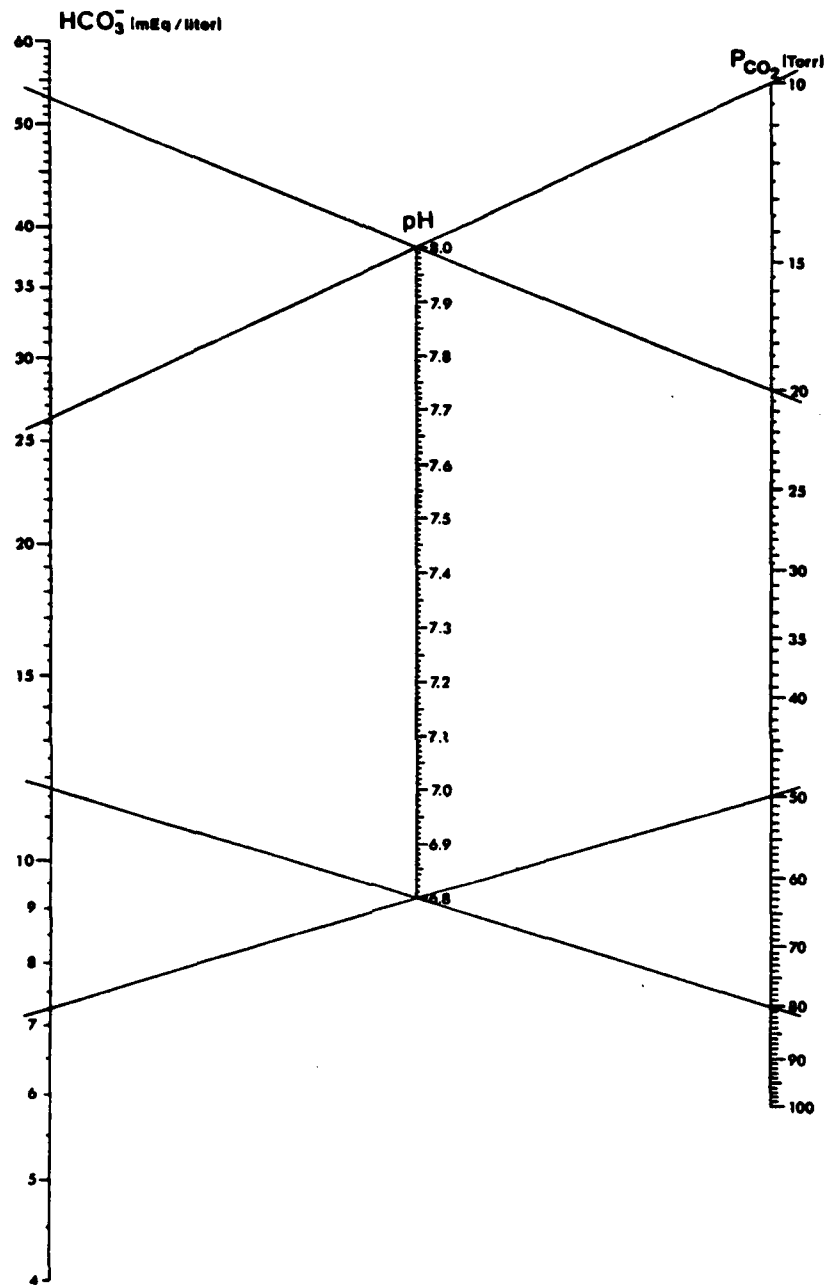


Figure 1. Procedure for delineating the pH scale in an acid-base alignment nomogram. Sets of P_{CO_2} , pK^1 values for a desired pH locus are used to calculate corresponding $[\text{HCO}_3^-]_p$ values. The intersection of lines connecting P_{CO_2} and $[\text{HCO}_3^-]_p$ pairs establish the desired pH locus.

Van Slyke et al (13) modified for use with protein concentrations expressed in g/l:

$$[A^- + G^-]_p = 0.125(g A/l)(pH_p - 5.16) + 0.077(g G/l)(pH_p - 4.89) \quad (2)$$

where A refers to albumin and G to globulin. Values for A (25.4 g/l) and G (32.7 g/l) were obtained from an earlier report on immature domestic swine (1). Values in the last column (Table 1), erythrocytic hemoglobin anion concentration $[Hb^-]_e$, were calculated by the equation of Dill et al (11), modified to include a factor of 20 (14) for converting concentration from mEq/mM Hb to mEq/l of erythrocytes:

$$[Hb^-]_e = 20[-0.5(pH_e)^2 + 10.625(pH_e) - 48.16] \quad (3)$$

Values in the last two columns of Table 1 were used subsequently to calculate total protein anion concentrations of blood $(Pr^-)_b$ as it was affected by pH_p and the V_e of blood. The following equation was used:

$$[Pr^-]_b = V_e [Hb^-]_e + (1 - V_e) [A^- + G^-]_p \quad (4)$$

The results of these calculations, which were expedited by a simple calculator program (Appendix), are shown in Table 2. Values for CO_2 solubility (S) at each V_e level, as reported by Van Slyke et al (10), are included at the bottom of Table 2.

The final tabular data needed for computations leading to the positioning of buffer base loci in the Henderson-Hasselbalch nomogram were the ratios (F) of plasma CO_2 content to blood CO_2 content as influenced by pH_p and V_e . These ratios (Table 3) were calculated by the equation of Van Slyke and Sendroy (8):

$$F = \frac{1}{V_e(d-1) + 1} \quad (5)$$

where d refers to the distribution ratio of erythrocyte CO_2 content to plasma CO_2 content, as determined with the Van Slyke apparatus (15). The d values used in equation 5 were obtained from Figure 2, which in turn was based on a similar figure contained in the report of Van Slyke and Sendroy (8), modified (dashed portions of curve) to include pH values ranging from 6.8 to 8.0.

Table 1. Interrelationships of Plasma pH (pH_p), pK' , (pK'_p), Antilog of $\text{pH}-\text{pK}'$ ($(\text{H}-\text{K})_p$), Erythrocyte pH (pH_e), Plasma Protein Anion Concentration ($[\text{A}^- + \text{G}^-]_p$), and Erythrocytic Hemoglobin Anion Concentration ($[\text{Hb}^-]_e$)

pH_p	pK'_p	$(\text{H}-\text{K})_p$	pH_e	$[\text{A}^- + \text{G}^-]_p$	$[\text{Hb}^-]_e$
6.8	6.113	4.864	6.737	9.96	8.54
6.9	6.109	6.180	6.814	10.52	14.47
7.0	6.105	7.852	6.891	11.09	20.28
7.1	6.101	9.977	6.964	11.66	25.68
7.2	6.097	12.680	7.037	12.22	30.97
7.3	6.093	16.106	7.106	12.79	35.87
7.4	6.088	20.512	7.173	13.35	40.54
7.5	6.083	26.122	7.242	13.92	45.26
7.6	6.079	33.189	7.308	14.49	49.68
7.7	6.074	42.267	7.372	15.05	53.89
7.8	6.070	53.703	7.437	15.62	58.07
7.9	6.065	68.391	7.495	16.18	61.74
8.0	6.060	87.096	7.553	16.75	65.33

Table 2. Protein Anion Concentration of Blood $[\text{Pr}^-]_b$ in mEq/l as a Function of Plasma pH (pH_p) and the Erythrocyte Function (V_e) of Blood and The Effect of V_e on CO_2 Solubility (S) in Blood as $\text{mM}/\text{unit (torr)}$ change in P CO_2

pH_p	V_e						
	0	0.1	0.2	0.3	0.4	0.5	0.6
6.8	9.96	9.80	9.66	9.52	9.38	9.24	9.10
6.9	10.52	10.92	11.30	11.70	12.09	12.49	12.89
7.0	11.09	11.99	12.91	13.83	14.75	15.67	16.60
7.1	11.66	13.04	14.45	15.85	17.26	18.66	20.06
7.2	12.22	14.08	15.95	17.83	19.71	21.58	23.46
7.3	12.79	15.08	17.39	19.70	22.01	24.32	26.63
7.4	13.35	16.05	18.77	21.49	24.21	26.93	29.66
7.5	13.92	17.04	20.17	23.31	26.44	29.58	32.72
7.6	14.49	17.99	21.51	25.03	28.55	32.07	35.60
7.7	15.05	18.92	22.80	26.69	30.57	34.46	38.35
7.8	15.62	19.85	24.09	28.34	32.59	36.83	41.08
7.9	16.18	20.72	25.28	29.83	34.39	38.95	43.51
8.0	16.75	21.59	26.45	31.31	36.17	41.03	45.89
$S_{(0-0.6)}$	0.0301	0.0296	0.0292	0.0287	0.0282	0.0277	0.0273

Table 3. Ratio (F) of Plasma to blood CO₂ Concentrations as a Function of Plasma pH and the Erythrocyte Fraction of Blood

pH _p	V _e						
	0	0.1	0.2	0.3	0.4	0.5	0.6
6.8	1.000	1.027	1.055	1.085	1.117	1.151	1.187
6.9	1.000	1.029	1.060	1.093	1.128	1.165	1.205
7.0	1.000	1.032	1.065	1.101	1.139	1.181	1.224
7.1	1.000	1.034	1.070	1.109	1.151	1.196	1.245
7.2	1.000	1.037	1.076	1.119	1.166	1.216	1.271
7.3	1.000	1.040	1.083	1.129	1.180	1.236	1.297
7.4	1.000	1.043	1.089	1.140	1.196	1.258	1.326
7.5	1.000	1.046	1.097	1.152	1.214	1.288	1.360
7.6	1.000	1.050	1.104	1.165	1.233	1.310	1.396
7.7	1.000	1.053	1.112	1.179	1.253	1.338	1.435
7.8	1.000	1.057	1.121	1.193	1.276	1.370	1.479
7.9	1.000	1.061	1.130	1.290	1.300	1.405	1.530
8.0	1.000	1.065	1.140	1.226	1.326	1.443	1.583

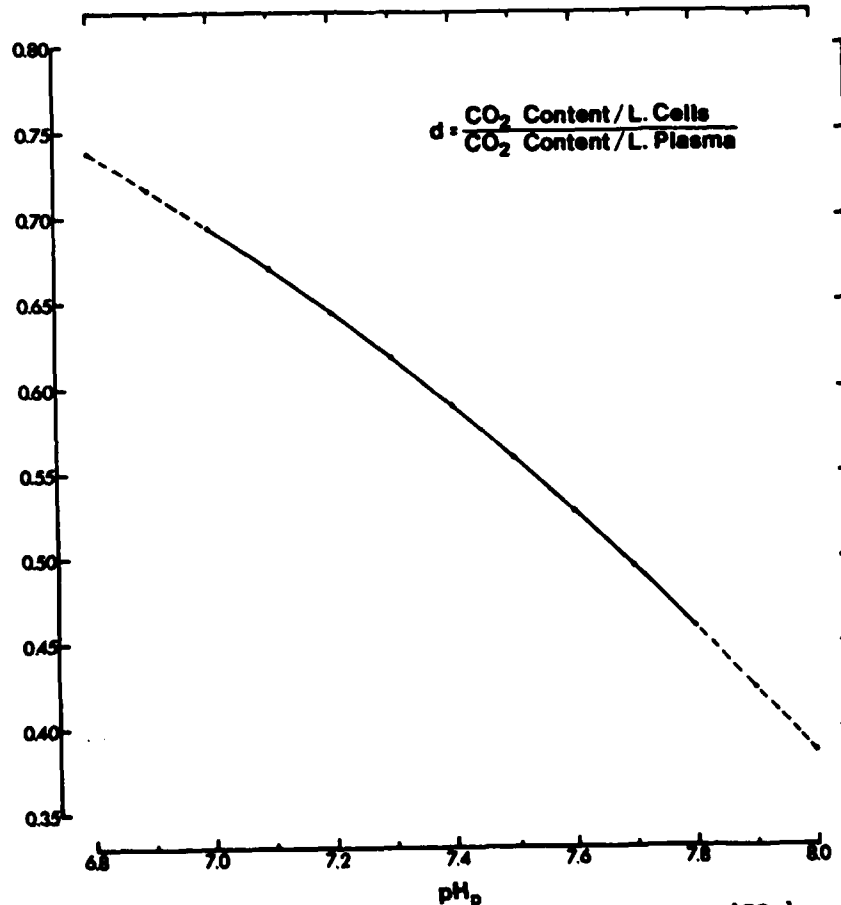


Figure 2. Ratio of erythrocyte CO₂ content, [CO₂]_c, to plasma CO₂ content, [CO₂]_p, as a function of pH_p. The ratio, termed d value, reproduced from Van Slyke et al (8), with values extended (dashed lines) from pH_p = 6.8 to pH_p = 8.0.

Positioning of Buffer Base Loci in the Henderson-Hasselbalch Nomogram

In one respect, the procedure for positioning buffer base loci in the Henderson-Hasselbalch nomogram is similar to that used in delineating the pH scale; in both instances, the location of specific scale values is delineated by the intersection of lines connecting two or more pairs of $P\text{ CO}_2$ and $[\text{HCO}_3^-]_p$ values. In the case of pH, the procedure is relatively uncomplicated since the scale values describe characteristics of a one-component system, namely plasma. In the case of buffer base concentration, however, scale values must characterize a two-component system--blood, which may contain variable amounts of erythrocytes and plasma. To describe accurately the buffer base characteristics of blood, therefore, a graded series of scales is needed, each scale describing the characteristics of blood with a specified erythrocyte fraction, V_e , and by inference plasma fraction, $1-V_e$.

Each pair of $P\text{ CO}_2$ and $[\text{HCO}_3^-]_p$ values used in describing a specific buffer base locus is obtained by means of a series of calculations. The first step is to calculate a blood bicarbonate value compatible with the locus. This is accomplished by inserting appropriate values from Table 2 into the equation defining buffer base concentration. For example, if a $[\text{BB}]_b$ value of 60 mEq/l is to be positioned on a scale for blood with a V_e of 0.2 and the pH_p for the particular $P\text{ CO}_2$ and $[\text{HCO}_3^-]_p$ pair is to be 7.8, then

$$\begin{aligned} [\text{HCO}_3^-]_b &= [\text{BB}]_b - [\text{Pr}^-]_b \\ &= 60 - 24.09 \\ &= 35.91 \end{aligned} \quad (6)$$

This value reflects bicarbonate contributions from both plasma and erythrocytes, but the magnitude of each contribution cannot be immediately determined because $P\text{ CO}_2$ is unknown at this point. An approximate $[\text{HCO}_3^-]_p$ value, however, can be obtained by means of an appropriate F ratio, $[\text{CO}_2]_p : [\text{CO}_2]_b$ (Table 3). Thus,

$$\begin{aligned} \text{approx. } [\text{HCO}_3^-]_p &= F[\text{HCO}_3^-]_b \\ &= 1.121(35.91) \\ &= 40.26 \text{ mEq/l} \end{aligned} \quad (7)$$

Next, an approximate $P\text{ CO}_2$ can be obtained by rearrangement of the Henderson-Hasselbalch equation and the insertion of appropriate values for plasma CO_2 solubility, S_o (Table 2), and the antilog of $\text{pH}_p - \text{pK}'_p$, $(\text{H-K})_p$ (Table 1). Accordingly,

$$\begin{aligned}
 \text{approx. } P_{\text{CO}_2} &= \frac{\text{approx. } [\text{HCO}_3^-]_p}{S_0(H-K)_p} \\
 &= \frac{40.26}{(0.0301)(53.703)} \\
 &= 24.90
 \end{aligned}
 \tag{8}$$

The P_{CO_2} approx. and S for blood at a V_e of 0.2 (Table 2), can now be used to estimate $[\text{CO}_2]_b$:

$$\begin{aligned}
 [\text{CO}_2]_b &= [\text{HCO}_3^-]_b + S_{0.2}(P_{\text{CO}_2}) \\
 &= 35.91 + (0.0292)(24.90) \\
 &= 36.64 \text{ mEq/l blood}
 \end{aligned}
 \tag{9}$$

This value for $[\text{CO}_2]_b$, along with the appropriate F ratio, can now be used to calculate $[\text{CO}_2]_p$.

$$\begin{aligned}
 [\text{CO}_2]_p &= F[\text{CO}_2]_b \\
 &= (1.121)(36.64) \\
 &= 41.07 \text{ mEq/l plasma}
 \end{aligned}
 \tag{10}$$

Since the Henderson-Hasselbalch equation can be expressed in the following form,

$$\text{pH} = \text{pK}' + \log \frac{[\text{CO}_2]_p - S_0(P_{\text{CO}_2})}{S_0(P_{\text{CO}_2})}
 \tag{11}$$

a more accurate value for P_{CO_2} can be obtained through equation rearrangement:

$$\begin{aligned}
 P_{\text{CO}_2} &= \frac{[\text{CO}_2]_p}{S_0(1 + (H-K)_p)} \\
 &= \frac{41.07}{(0.0301)(54.703)} \\
 &= 24.94 \text{ torr}
 \end{aligned}
 \tag{12}$$

This P_{CO_2} value can now be used to calculate $[\text{HCO}_3^-]_p$:

$$\begin{aligned}
 [\text{HCO}_3^-]_p &= [\text{CO}_2]_p - S_0(P_{\text{CO}_2}) \\
 &= 41.07 - 0.0301(24.94) \\
 &= 40.32 \text{ mEq/l plasma}
 \end{aligned}
 \tag{13}$$

Equations 12 and 13 provide one pair of $P\text{CO}_2$ and $[\text{HCO}_3^-]_p$ values which can be used to plot a line on the Henderson-Hasselbalch nomogram. This line passes through the locus of $[\text{BB}]_b = 60 \text{ mEq/l}$ at a V_e of 0.2. One or more additional pairs of $P\text{CO}_2$ and $[\text{HCO}_3^-]_p$ values, and connecting lines, are needed to delineate the position of this locus within the nomogram. Equations 6 through 13 are used to obtain these additional values, in each instance starting with a selected pH value and appropriate $[\text{Pr}^-]_p$, $(\text{H}-\text{K})$, and F values from Tables 1, 2, and 3. The entire procedure is repeated for each $[\text{BB}]_b$ locus to be plotted for $V_e = 0.2$, as illustrated in Figure 3 for $[\text{BB}]_b$ values of 35 and 60 mEq/l, and for all other V_e scales to be included in the nomogram. Loci positioning, obviously, would represent a formidable task if each $P\text{CO}_2$ and $[\text{HCO}_3^-]_p$ pair were calculated by hand. The task, however, can be greatly simplified and expedited by means of a calculator program (Appendix).

The completed acid-base nomogram for porcine arterial blood is shown in Figure 4. In constructing the buffer base scales, loci representing buffer base concentrations of 15, 20, 25, 30, 35, 40, 45, 50, 60, 70, and 80 mEq/l were plotted at V_e values of 0, 0.1, 0.2, 0.3, 0.4, 0.5, and 0.6. Lines depicting intermediate $[\text{BB}]_b$ values were obtained by plotting, with $P\text{CO}_2$ and $[\text{HCO}_3^-]_p$ pairs, appropriate points on the $V_e = 0$ and $V_e = 0.6$ scales.

Positioning of Base Excess Loci on the Henderson-Hasselbalch Nomogram

Once the buffer base nomogram is constructed, the positioning of base excess loci and the delineation of base excess scales for the Henderson-Hasselbalch nomogram becomes a relatively simple task. By definition (2,3), blood base excess concentration, $[\text{BE}]_b$, is equal to the difference between buffer base concentration observed $[\text{OBB}]_b$ in a blood sample and the normal buffer base concentration, $[\text{NBB}]_b$, that would be expected in the sample. That is,

$$[\text{BE}]_b = [\text{OBB}]_b - [\text{NBB}]_b \quad (14)$$

Since $[\text{NBB}]_b$ values can vary from species to species, the first task in constructing $[\text{BE}]_b$ scales is to establish the $[\text{NBB}]_b$ characteristics of the blood, in this instance porcine blood. The task is accomplished by taking average population values for arterial $P\text{CO}_2$, pH, and $[\text{HCO}_3^-]_p$ and connecting these value with a straight line on the Henderson-Hasselbalch nomogram. The point on each buffer base scale intersected by this line defines the normal buffer base value for that scale. Accordingly, from measurements made in an earlier population study (1), appropriate normal values for swine arterial blood would be $P\text{CO}_2 = 40$ torr, $\text{pH} = 7.50$, and $[\text{HCO}_3^-]_p = 31.6 \text{ mEq/l}$. A line connecting these values, as illustrated in Figure 5, intersects the V_e (plasma) buffer base scale at about 45.5 mEq/l and the $V_e = 0.6$ scale at about 55.8 mEq/l. A positive $[\text{BE}]_b$ of 5 mEq/l, according to equation 14,

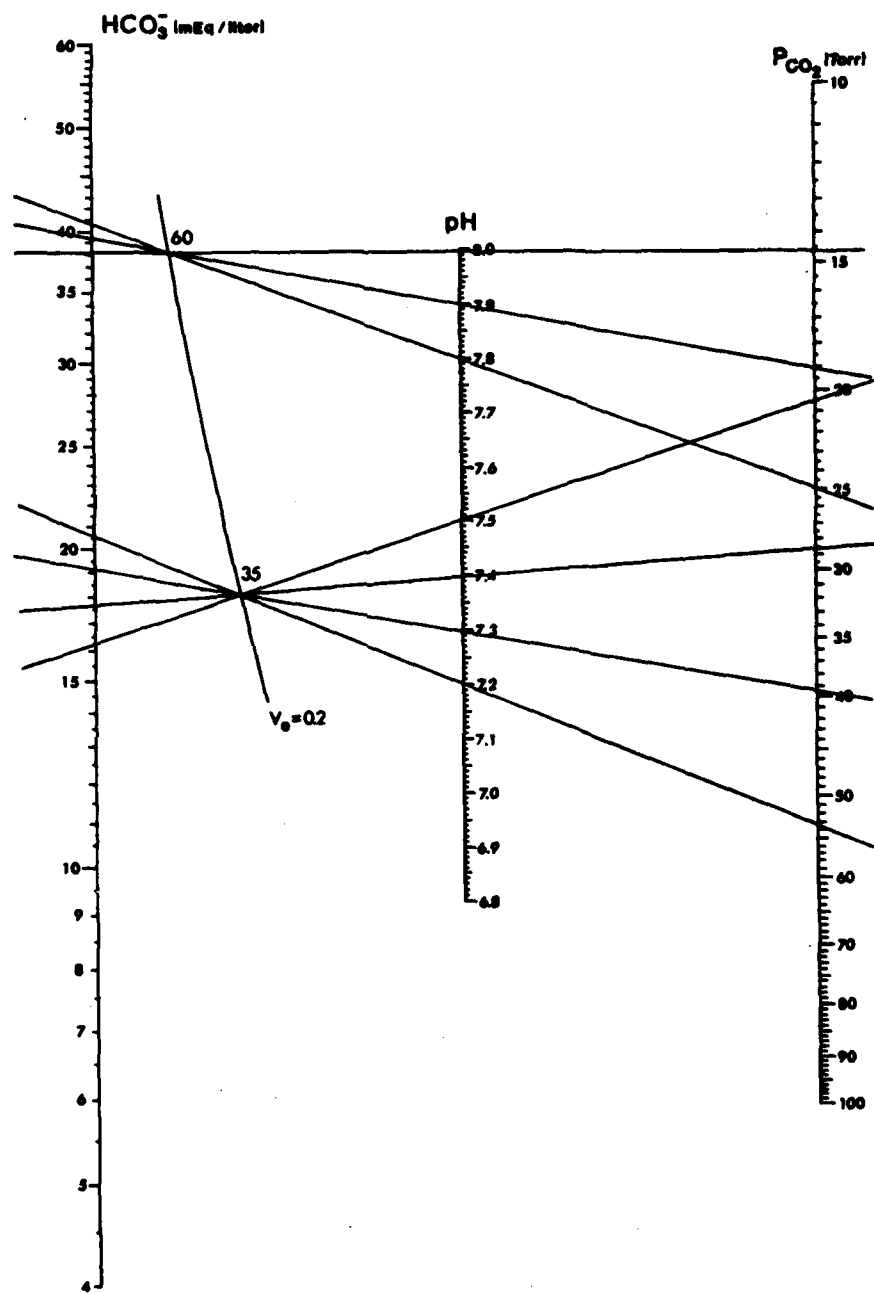


Figure 3. Procedure for plotting buffer base loci in the Henderson-Hasselbalch acid-base alignment nomogram. A series of equations (see text) is used to calculate two or more pairs of P_{CO_2} and $[\text{HCO}_3^-]_p$ values. The intersection of lines connecting these pairs defines the locus of a selected $[\text{BB}]_p$ on a selected V_c scale.

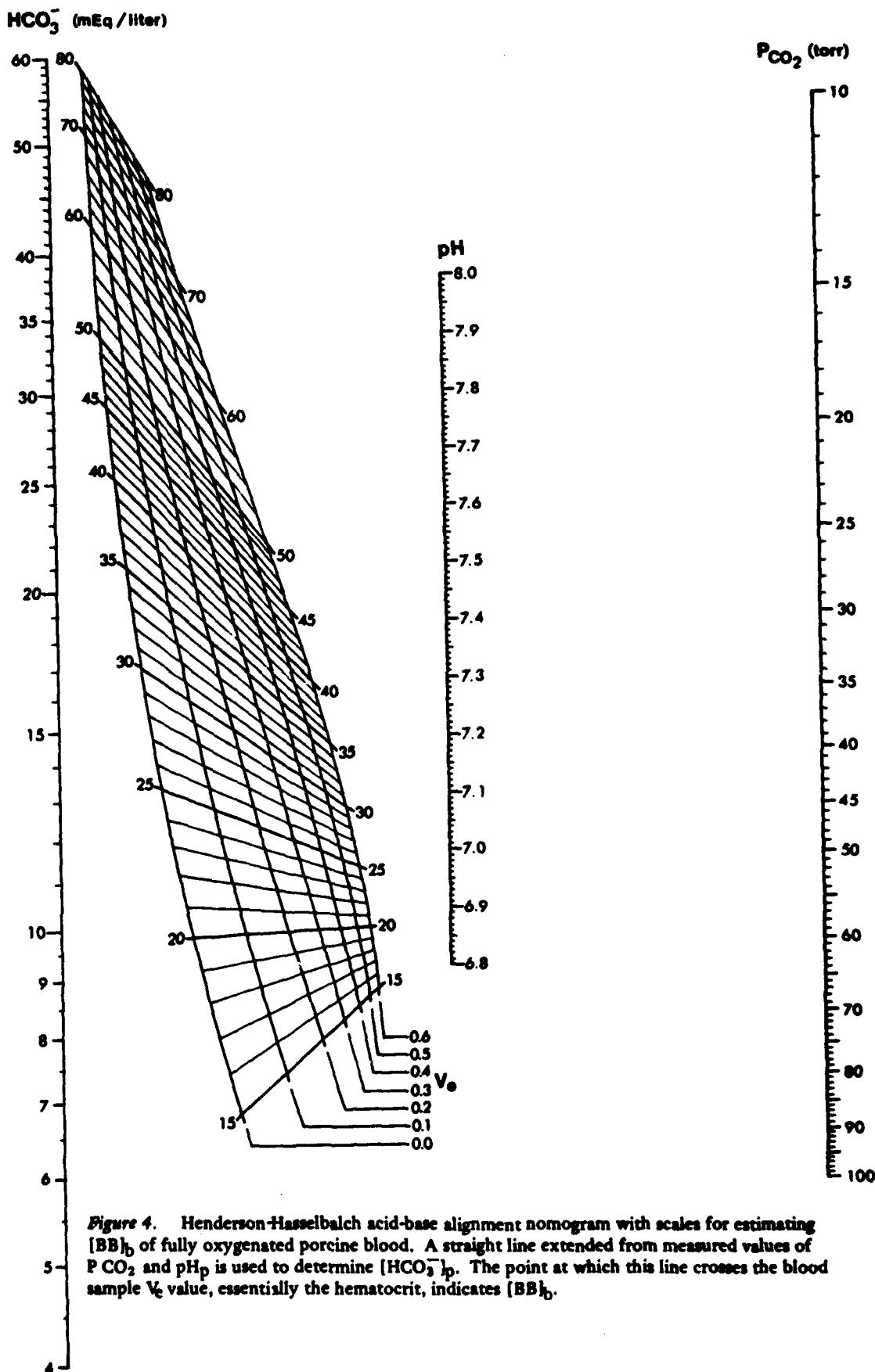


Figure 4. Henderson-Hasselbalch acid-base alignment nomogram with scales for estimating $[\text{BB}]_b$ of fully oxygenated porcine blood. A straight line extended from measured values of P CO_2 and pH_p is used to determine $[\text{HCO}_3^-]_p$. The point at which this line crosses the blood sample V_c value, essentially the hematocrit, indicates $[\text{BB}]_b$.

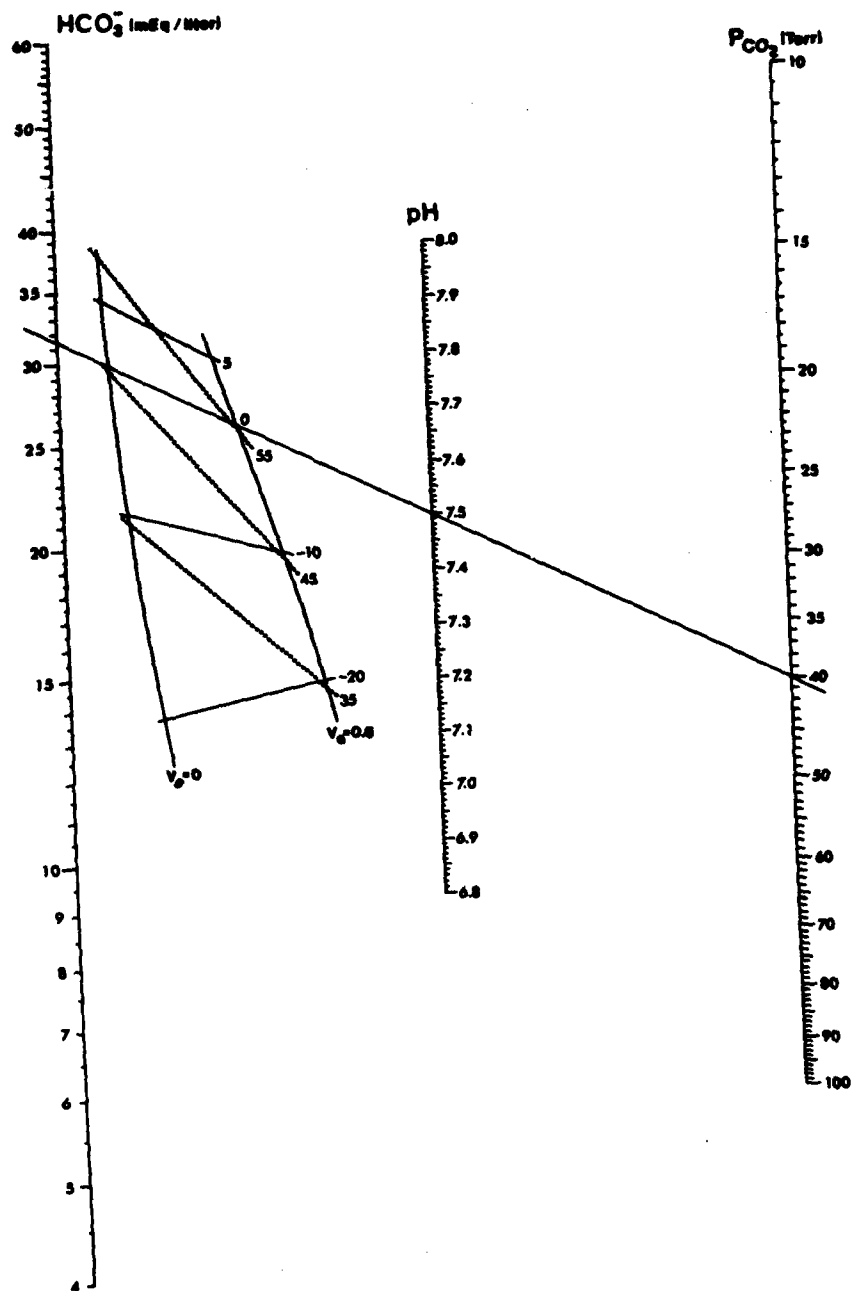


Figure 5. Procedures for plotting base excess loci. A line connecting arterial P CO_2 , pH_p , and $[\text{HCO}_3^-]_p$ values corresponding to those found in normal porcine population defines normal buffer base concentration $[\text{NBB}]_b$ at $V_c = 0$ and $V_c = 0.6$, 45.5 mEq/l and 55.7 mEq/l, respectively. Loci for $[\text{BE}]_b$ values of 5, -10, and -20 would occur on $V_c = 0$ scale at $[\text{BB}]_b$ values of 50.5, 35.5, and 25.5 mEq/l, respectively, and on the $V_c = 0.6$ scale at $[\text{BB}]_b$ values of 60.7, 45.7, and 35.7 mEq/l, respectively. Continuous lines depict $[\text{BE}]_b$ values; dashed lines depict $[\text{BB}]_b$ values.

would be located on the $V = 0$ buffer base scale at 50.5 mEq/l and on the $V = 0.6$ buffer base scale at 60.8 mEq/l. Negative base excess (base deficit) values of -10 and -20 mEq/l would be located, respectively, on the $V = 0$ buffer base scale at 35.5 and 25.5 mEq/l and on the $V = 0.6$ scale at 45.8 and 35.8 mEq/l. Lines connecting equivalent base excess values at $V = 0$ and $V = 0.6$ are used to establish points on the intermediate V scales. This process is continued until scales covering $[BE]_b$ values ranging from 20 to -30 mEq/l are constructed.

The completed Henderson-Hasselbalch alignment nomogram with base excess scales appropriate to porcine blood is illustrated in Figure 6.

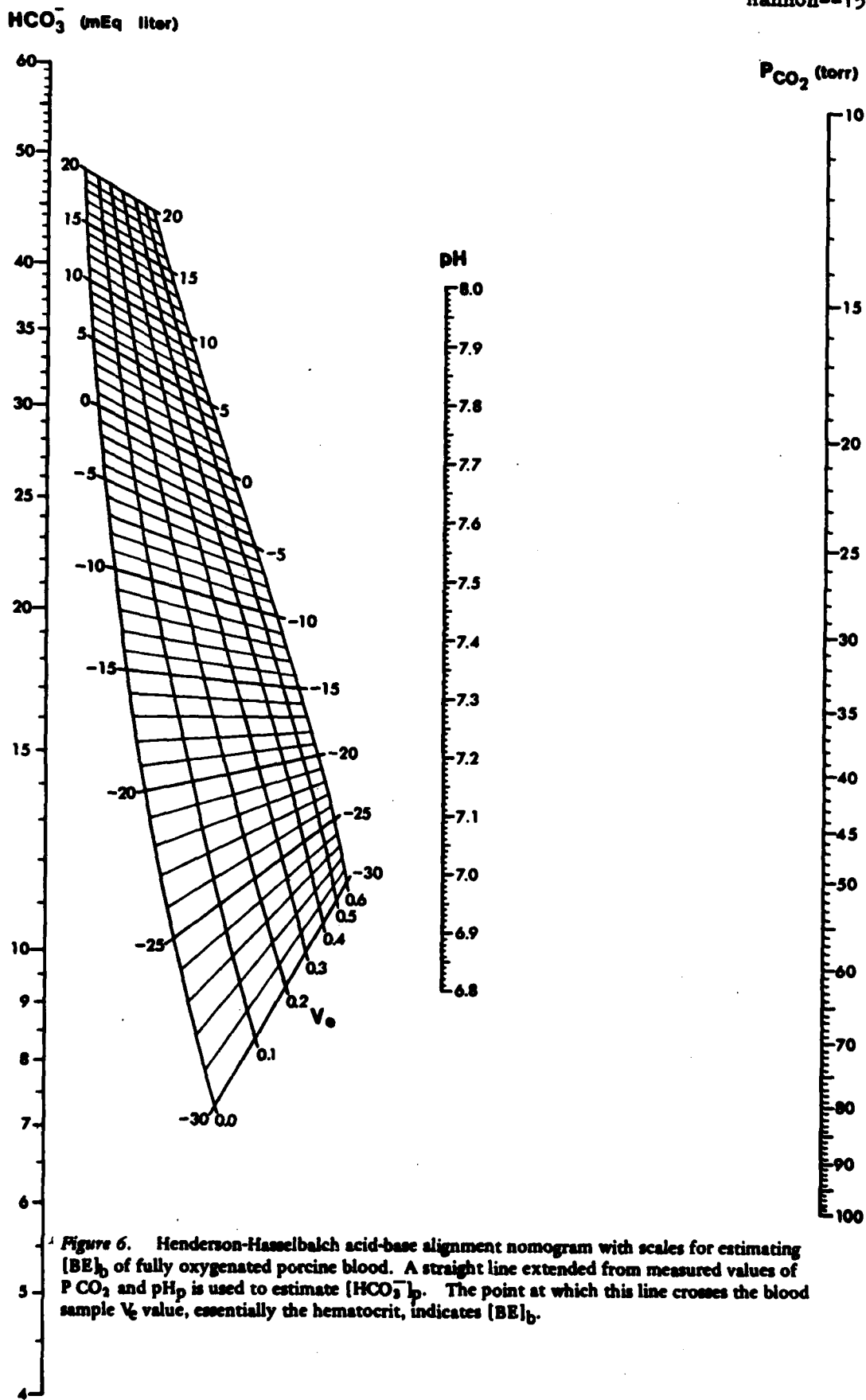


Figure 6. Henderson-Hasselbalch acid-base alignment nomogram with scales for estimating $[\text{BE}]_b$ of fully oxygenated porcine blood. A straight line extended from measured values of P_{CO_2} and pH_p is used to estimate $[\text{HCO}_3^-]_b$. The point at which this line crosses the blood sample V_c value, essentially the hematocrit, indicates $[\text{BE}]_b$.

COMMENT

As might be anticipated, the alignment nomograms describing buffer base and base excess characteristics of porcine arterial blood differ in several respects from similar nomograms for human blood (6,16). These differences are due primarily to the substantially higher pH and $[\text{HCO}_3^-]_p$ values that characterize normal porcine blood, relative to normal human blood. Porcine blood, as a consequence, exhibits greater acid buffering capacity than human blood (1).

Two additional factors also contribute, but to a lesser extent, to the differences in the porcine and human nomograms. One of these is the temperature at which blood acid base measurements are made. A value of 38 C was selected here for porcine blood, whereas Singer and Hastings (6) used a value of 37 C in constructing their nomogram to estimate buffer base concentration of human blood. Siggaard-Andersen (16) used the buffer characteristics of hemoglobin reported by Singer and Hastings (6) to construct an alignment nomogram for estimating base excess concentration at 38 C; no attempt apparently was made to correct hemoglobin buffer characteristics for a 1 C increase in blood temperature. The other factor concerned population characteristics for plasma protein anion concentration. Both Singer and Hastings (6) and Siggaard-Andersen (16) used a value of 72 g/l and an assumed albumin:globulin ratio of 1.40 to calculate $[\text{Pr}^-]_p$ concentration according to a general equation reported by Van Slyke et al (13). Here, the anion concentrations of albumin and globulin were calculated separately and subsequently added together to determine $[\text{Pr}^-]_p$ values. However, the total plasma protein concentration of immature pigs tends to be lower than that of humans; a population average of 57.6 g/l (1) was used to establish acid-base characteristics in the present study.

In constructing the buffer base nomogram for porcine blood, a high degree of precision was obtained when the plasma loci were plotted, but the precision tended to decrease as V_e increased. Such inaccuracies were particularly evident when extreme high or low $P\text{CO}_2$ and $[\text{HCO}_3^-]_p$ values were used for loci placement. This loss of precision presumably was due to inaccuracies in the various constants used to obtain the acid-base data contained in Tables 1, 2, and 3. Such inaccuracies, however, did not seriously compromise loci placement in the completed nomogram since distinct linear and curvilinear interrelationships between and within buffer base scales fostered loci placement with reasonable precision.

CONCLUSIONS

Acid-base nomograms or other procedures designed for use with human blood produce inaccurate results when used to estimate the buffer base or base excess concentration of porcine blood. Procedures specifically designed for porcine blood must be used.

The inaccuracies are due primarily to the higher plasma bicarbonate and pH values and secondarily to the lower plasma protein concentration and slightly higher temperature that characterize normal porcine as compared to normal human blood.

RECOMMENDATIONS

Blood acid-base alignment nomograms for other common laboratory animals should be constructed. A prime candidate in this regard is the mongrel dog which has blood acid-base characteristics which are distinctly different from those of humans.

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Series of Programs for Construction of Nomogram

APPENDIX A

A programable calculator, Texas Instruments TI-59 with PC-100 printer, greatly expedites the acquisition of data needed for construction of a buffer base alignment nomogram. The following series of programs are designed to meet this need.

1. Logarithm Generator

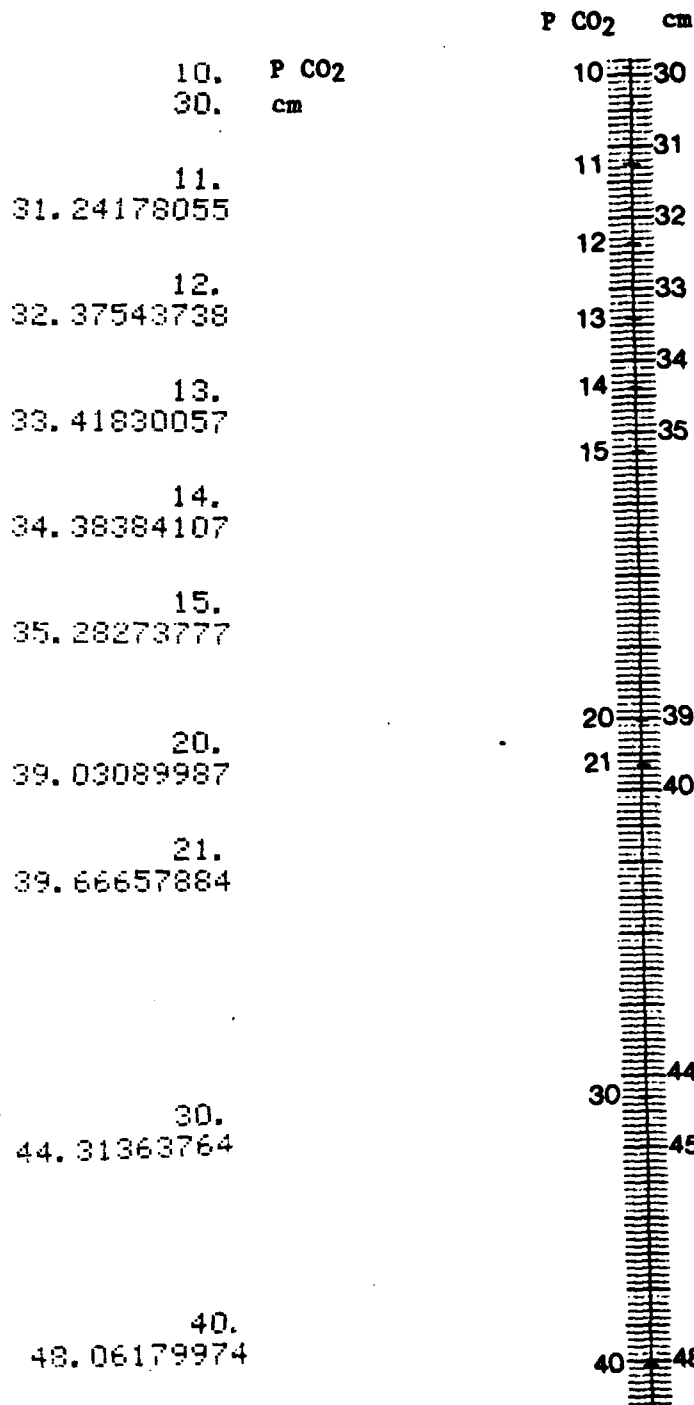
This program generates logarithms of $P\text{ CO}_2$ and $[\text{HCO}_3^-]$ values which are subsequently plotted as vertical scales on either side of linear graph paper. By means of the Henderson-Hasselbalch equation, these scales are used subsequently to plot an intermediate pH scale.

a. Program listing

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
000	76	LBL	
001	11	A	Initiate program
002	91	R/S	Enter starting $P\text{ CO}_2$ or $[\text{HCO}_3^-]$
003	42	STD	
004	01	01	
005	76	LBL	
006	12	B	
007	43	RCL	
008	01	01	
009	99	PRT	Print $P\text{ CO}_2$ or $[\text{HCO}_3^-]$ value
010	28	LDG	
011	65	X	
012	03	3	Length of log cycle, cm*
013	00	0	
014	95	=	
015	99	PRT	Print scale point, cm
016	01	1	
017	44	SUM	Add 1 to previous $P\text{ CO}_2$ or $[\text{HCO}_3^-]$
018	01	01	
019	98	ADV	
020	61	GTD	
021	12	B	Recycle to new value
022	91	R/S	

*may be altered as desired

b. Program readout and scale plot



2. Plasma Protein Anion Concentration

The equation of Van Slyke et al (1), modified for protein concentrations expressed as g/l, is used to calculate anion concentrations.

$$[\text{Pr}^-]_p = 0.125(\text{A}^-)(\text{pH}_p - 5.16) + 0.077(\text{G}^-)(\text{pH}_p - 4.89)$$

where $[\text{Pr}^-]_p$ = total protein anion, A^- = albumin, and G^- = globulin.

a. Program listing

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
000	76	LBL	
001	11	A	Initiate program
002	47	CMS	
003	25	CLR	
004	91	R/S	Enter A^- , g/l
005	99	PRT	
006	42	STD	
007	01	01	
008	91	R/S	Enter G^- , g/l
009	99	PRT	
010	42	STD	
011	02	02	
012	06	6	
013	93	.	Starting pH_p
014	08	8	
015	42	STD	
016	03	03	
017	98	ADV	
018	98	ADV	
019	76	LBL	
020	12	B	
021	93	.	
022	01	1	
023	02	2	
024	05	5	
025	65	*	
026	43	RCL	
027	01	01	
028	65	*	
029	53	(
030	43	RCL	
031	03	03	
032	99	PRT	Print pH_p
033	75	-	

Program listing (cont)

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
034	05	5	
035	93	.	
036	01	1	
037	06	6	
038	54)	
039	95	=	
040	99	FRT	Print $[A^-]_p$, mEq/l
041	42	STD	
042	04	04	
043	93	.	
044	00	0	
045	07	7	
046	07	7	
047	65	x	
048	43	RCL	
049	02	02	
050	65	x	
051	53	<	
052	43	RCL	
053	03	03	
054	75	-	
055	04	4	
056	93	.	
057	08	8	
058	09	9	
059	54)	
060	95	=	
061	99	FRT	Print $[G^-]_p$, mEq/l
062	44	SUM	
063	04	04	
064	43	RCL	
065	04	04	
066	99	FRT	Print $[Pr^-]_p$, mEq/l
067	93	.	
068	01	1	Add 1.0 to pH_p
069	44	SUM	
070	03	03	
071	98	ADV	
072	61	GTD	Recycle to next pH_p value
073	12	B	
074	91	R/S	

b. Program readout

25.4
32.3

Plasma albumin, g/l
Plasma globulin, g/l

6.8
5.207
4.750361
9.957361

pH_p
[A⁻]_p, mEq/l
[G⁻]_p, mEq/l
[Pr⁻]_p, mEq/l

6.9
5.5245
4.999071
10.523571

7.
5.842
5.247781
11.089781

7.1
6.1595
5.496491
11.655991

7.2
6.477
5.745201
12.222201

7.3
6.7945
5.993911
12.788411

7.4
7.112
6.242621
13.354621

3. Erythrocyte Protein Anion Concentration

Erythrocytic protein anion concentration, almost entirely hemoglobin, is calculated by the equation of Dill et al (2):

$$[\text{Hb}^-]_e = 20[-0.5(\text{pH}_e)^2 + 10.625(\text{pH}_e) - 48.46]$$

where $[\text{Hb}^-]_e$ = mEq hemoglobin anion/l erythrocytes and pH_e = erythrocyte pH reported by Dill et al (2), but corrected to 38 C by subtracting 0.016 unit from the 37 C value (3).

a. Program listing

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
000	76	LBL	
001	11	A	Initiate program
002	47	CMS	
003	25	CLR	
004	91	R/S	Enter and print pH_p
005	99	PRT	
006	91	R/S	Enter and print pH_e
007	99	PRT	
008	42	STD	
009	01	01	
010	02	2	
011	00	0	
012	65	X	
013	53	(
014	93	.	
015	05	5	
016	94	+/-	
017	65	X	
018	43	RCL	
019	01	01	
020	33	X ²	
021	85	+	
022	01	1	
023	00	0	
024	93	.	
025	06	6	
026	02	2	
027	05	5	
028	65	X	
029	43	RCL	
030	01	01	

Program listing (cont)

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
031	75	-	
032	04	4	
033	08	8	
034	93	.	
035	04	4	
036	06	6	
037	54	>	
038	95	=	
039	99	PRT	Print $[\text{Hb}^-]_e$
040	98	ADV	
041	61	GTO	Recycle for next pH_e
042	11	A	
043	91	R/S	

b. Program readout

6.8	pH_p
6.737	pH_e
8.54081	$[\text{Hb}^-]_e$

6.9
6.814
14.46904

7.
6.891
20.27869

4. Protein Anion Concentration of Blood

The protein anion concentration of whole blood is equal to the sum of the fractional contributions of plasma and erythrocytic protein anions. The following equation describes this relationship:

$$[\text{Pr}^-]_b = [\text{Pr}^-]_p(1-v_e) + [\text{Hb}^-]_e v_e$$

where $[\text{Pr}^-]_b$ = whole blood protein anion and v_e = the erythrocyte fraction (essentially the hematocrit) of the blood sample.

a. Program listing

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
000	76	LBL	
001	11	R	Initiate program
002	47	CMS	
003	25	CLR	
004	91	R/S	Enter and print pH_p
005	99	PRT	
006	91	R/S	Enter $[\text{Pr}^-]_p$
007	99	PRT	
008	42	STO	
009	01	01	
010	91	R/S	Enter $[\text{Hb}^-]_e$
011	99	PRT	
012	42	STO	
013	02	02	
014	98	ADV	
015	76	LBL	
016	12	B	
017	43	RCL	
018	01	01	
019	65	*	
020	53	(
021	01	1	
022	75	-	
023	43	RCL	
024	03	03	
025	99	PRT	Print V_e
026	54)	
027	95	=	
028	99	PRT	Print $[\text{Pr}^-]_p(1-V_e)$
029	42	STO	
030	04	04	
031	43	RCL	
032	02	02	
033	65	*	
034	43	RCL	
035	03	03	
036	95	=	
037	99	PRT	Print $[\text{Hb}^-]_e V_e$
038	85	+	
039	43	RCL	
040	04	04	
041	95	=	
042	99	PRT	Print $[\text{Pr}^-]_b$
043	93	.	
044	01	1	Add 0.1 to V_e

Program listing (cont)

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
045	44	SUN	
046	03	03	
047	98	ADV	
048	61	GTD	Recycle to new V_e
049	12	B	
050	91	R/S	

b. Program readout

7.	pH_p
11.09	
20.28	
0.	V_e
11.09	$[Pr^-]_p (1-V_e)$
0.	$[Hb^-]_e V_e$
11.09	$[Pr^-]_b$
0.1	
9.981	
2.028	
12.009	
0.2	
8.872	
4.056	
12.928	

5. Ratio, F , of Plasma CO_2 Content to Blood CO_2 Content

Whole blood CO_2 content is comprised of a fraction attributable to plasma and a fraction attributable to erythrocytes. Van Slyke and Sendroy (4) described this relationship by the following equation:

$$F = 1/(V_e(d-1)+1)$$

where, at a given pH_p , $d = CO_2$ content/l of erythrocytes divided by CO_2 content/l of plasma.^P

a. Program listing

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
000	76	LBL	
001	11	A	Initiate program
002	47	CMS	
003	25	CLR	
004	91	R/S	Enter and print pH_p
005	99	PRT	
006	91	R/S	Enter and print d
007	99	PRT	
008	42	STD	
009	01	01	
010	98	ADV	
011	76	LBL	
012	12	B	
013	01	1	
014	55	+	
015	53	(
016	43	RCL	
017	02	02	
018	99	PRT	Print V_e
019	65	x	
020	53	(
021	43	RCL	
022	01	01	
023	75	-	
024	01	1	
025	54)	
026	85	+	
027	01	1	
028	54)	
029	95	=	
030	99	PRT	Print F ratio
031	93	.	
032	01	1	Add 0.1 to V_e
033	44	SUM	
034	02	02	
035	98	ADV	
036	61	GTD	
037	12	B	Recycle to next V_e
038	91	R/S	

b. Program readout

7.1	pH _p
0.672	d
0.	V _e
1.	F
0.1	
1.033912324	
0.2	
1.070205479	
0.3	
1.109139308	
0.4	
1.151012891	

6. Determination of P CO₂ and [HCO₃⁻] Coordinates for Plotting Buffer Base Loci

Pairs of P CO₂ and [HCO₃⁻]_p values, used to delineate the position of buffer base loci in the Henderson-Hasselbalch alignment nomogram, are calculated by a sequence of equations originally described by Singer and Hastings (5). In the program listing, [BB]_b refers to buffer base in mEq/l, S to CO₂ solubility in plasma or blood, (H-K)_p to the antilog of pH_p-pK'_p.

a. Program listing

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
000	76	LBL	
001	11	A	Initiate program
002	25	CLR	
003	47	ONS	
004	91	R/S	Enter, store, and print V _e
005	42	STD	
006	01	01	
007	99	PRT	
008	91	R/S	Enter, store, and print [BB] _b
009	42	STD	

Program listing (cont)

<u>Step</u>	<u>Code</u>	<u>Key</u>	<u>Operational Function</u>
010	02	02	
011	99	PRT	
012	91	R/S	Enter, store, and print S
013	42	STD	
014	03	03	
015	99	PRT	
016	98	ADV	
017	76	LBL	
018	12	B	
019	91	R/S	Enter and print pH _p
020	99	PRT	
021	91	R/S	Enter, store, and print (H-K) _p
022	42	STD	
023	04	04	
024	99	PRT	
025	43	RCL	
026	02	02	
027	75	-	
028	91	R/S	Enter and print [Pr ⁻] _b
029	99	PRT	
030	95	=	
031	42	STD	
032	05	05	
033	91	R/S	Enter and print F ratio
034	99	PRT	
035	42	STD	
036	06	06	
037	65	X	
038	43	RCL	
039	05	05	
040	98	ADV	
041	55	+	
042	93	.	
043	00	0	
044	03	3	
045	00	0	
046	01	1	
047	42	STD	
048	07	07	
049	55	+	
050	43	RCL	
051	04	04	
052	95	=	
053	42	STD	

Program listing (cont)

Step	Code	Key	Operational Function
054	08	08	
055	65	X	
056	43	RCL	
057	03	03	
058	85	+	
059	43	RCL	
060	05	05	
061	99	PRT	Print $[\text{HCO}_3]_b$
062	95	=	
063	65	X	
064	43	RCL	
065	06	06	
066	95	=	
067	42	STD	
068	09	09	
069	55	+	
070	43	RCL	
071	07	07	
072	55	+	
073	53	(
074	01	1	
075	85	+	
076	43	RCL	
077	04	04	
078	54)	
079	95	=	
080	99	PRT	Print computed P CO ₂
081	65	X	
082	43	RCL	
083	07	07	
084	95	=	
085	42	STD	
086	10	10	
087	43	RCL	
088	09	09	
089	75	-	
090	43	RCL	
091	10	10	
092	95	=	
093	99	PRT	Print computed $[\text{HCO}_3]_p$
094	98	ADV	Recycle for next calculation of
095	98	ADV	P CO ₂ and $[\text{HCO}_3]_p$ pair at same V_e ,
096	98	ADV	$[\text{BB}]_b$, and S, but new pH, (H-K) _p ,
097	61	GTO	$[\text{Pr}^-]_b$, and F ratios.
098	12	B	
099	91	R/S	

b. Program readout

0.2	v_e
35.	$[BB]_b$
0.0292	$S_{0.2}$
7.2	pH_p
12.68	$(H-K)_p$
15.95	$[Pr^-]_b$
1.076	F
19.05	$[HCO_3^-]_b$
53.87789844	P CO ₂
20.56046974	$[HCO_3^-]_p$
7.5	
26.112	
20.17	
1.097	
14.83	
20.74762116	
18.3070027	

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