

EVALUATION OF ERROR IN MEASUREMENT OF pH OF BLOOD AT ROOM TEMPERATURE

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BLOOD pH values obtained in vitro vary as inverse functions of the temperature of the specimen. These variations are sufficiently great that values determined on blood at room temperature must be corrected to have clinical significance. Ideally, measurements of blood pH should be made at body temperature, and the modern manufacture of instruments is now rapidly providing the means for such measurements. But many determinations are still made at room temperature, and then are corrected to yield what is presumed to be true in vivo pH values.

Rosenthal¹ as well as Craig, Lange, Oberman, and Carson² studied the effect of temperature on blood pH, and in each study a formula was derived that purported to allow accurate corrections to be made. These formulas are shown in *Table 1* in which pH_{37} and pH_{38} are the values at 37 and 38 C., respectively, and pH_t is the value observed at room temperature. Objections have been raised to

Table 1.—Correction formulas for pH of human blood measured at room temperature

Rosenthal ¹	$pH_{38} = pH_t (38_t) 0.0147$
Craig and associates ²	$pH_{37} = pH_t (37_t) 0.0149$

this practice of measuring blood pH at room temperature and then correcting the observed values by means of one of the formulas. The objections are based on the possible variations in the responses of specimens from different persons to changes in temperature.¹⁻³ The total number of persons from whom specimens were obtained for examination in two independent studies^{1,2} was only 46, which probably is insufficient to appraise the full potential error of the technic.

The work presented here involves blood specimens from 243 patients. The findings confirm the observations of other workers that individual variations occur, and furthermore they indicate the probable numerical error in correcting the pH of blood measured at room temperature.

Materials and Reagents

The blood specimens from 243 patients in the Cleveland Clinic Hospital were examined over an eight-month period. Two pH meters were used in this study. One was a Cambridge Research Model* with a micro glass electrode of the con-

**Cambridge Instrument Co., 3732 Grand Central Terminal, New York, New York.*

denser type having a capacity of 0.4 ml. This instrument was used to measure blood at room temperature. The complete assembly is shown in *Figure 1*. The

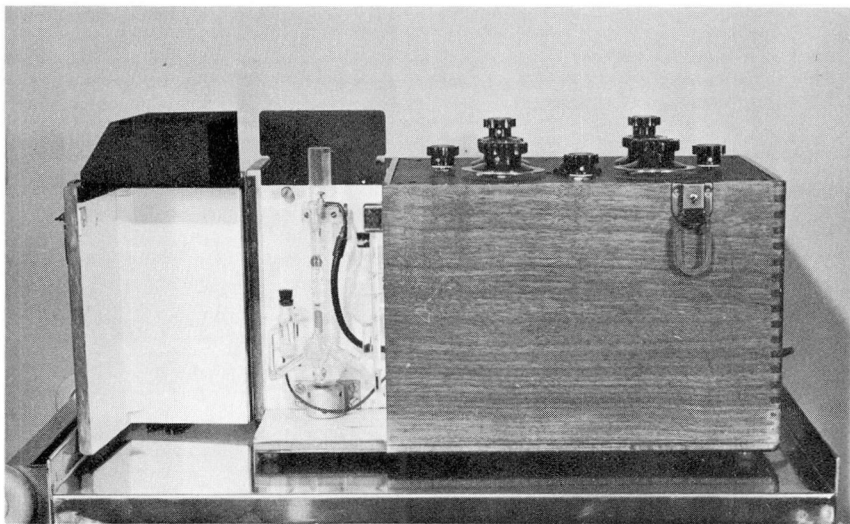


Fig. 1. Photograph of Cambridge Research Model pH meter.

other pH meter was a Metrohm Precision Compensator E322* using a Metrohm Blood Electrode, type EA 520, with a capacity of 0.05 ml. A Haake thermostat, type "F," was used to circulate water at 37.5 C. through the jacket of the glass electrode. Both instruments were standardized with phosphate buffer (0.025 M, pH 6.86 at 25 C., pH 6.84 at 37 C.) immediately before each series of measurements was made. *Figure 2* shows the complete apparatus.

Procedure

One glass bead, 1 ml. of mineral oil,† 2 drops of heparin sodium,‡ and 4 drops of 10 per cent sodium fluoride were placed in the barrel of each syringe. The plunger was fitted into the barrel, and with the tip pointed upward the excess oil was expelled; the tip was then wiped off and the syringe was capped. With the tourniquet in place, from 5 to 10 ml. of blood was withdrawn from the antecubital vein. The syringe was capped and the blood was mixed with the reagents. The measurements of pH were made directly from the syringe usually within one hour of collection. In rare instances, the sample was kept in the syringe and was stored in a refrigerator until it could be examined, at which time it was allowed to come to room temperature before the measurements were made.

*C. A. Brinkman & Co., Inc., 115 Cutter Mill Road, Great Neck, Long Island, New York.

†Standard Oil Company of Ohio, light No. 135, NF grade.

‡Heparin sodium, 1000 USP units per milliliter, Organon, Inc., Orange, N. J.

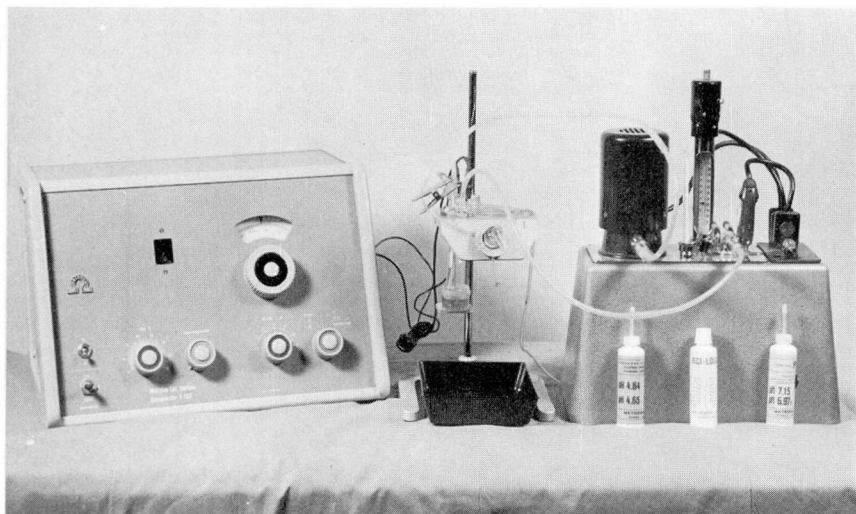


Fig. 2. Photograph of Metrohm pH meter with Sanz blood electrode and Haake thermostat.

Results

The deviations of the corrected blood pH values, from those on the same specimen at 37.5 C. are summarized in *Table 2*. *Figure 3* presents the findings graphically, and indicates that the deviations were almost equally distributed on either side of the zero point.

Table 2.—*Deviations in pH units of 243 human blood specimens: measured at room temperature and corrected to 38 C.,* measured at 37.5 C.*

Deviations, pH unit	Specimens		Cumulative percentage
	Number	Percentage of total	
±0.00	41	16.9	16.9
±0.01	84	34.5	51.5
±0.02	53	21.8	73.2
±0.03	39	16.0	89.3
±0.04	20	8.2	97.5
±0.05	3	1.2	98.7
±0.06	1	0.4	98.9
±0.07	1	0.4	99.0
±0.08	1	0.4	99.4

*Corrections made with Rosenthal's¹ formula (*Table 1*).

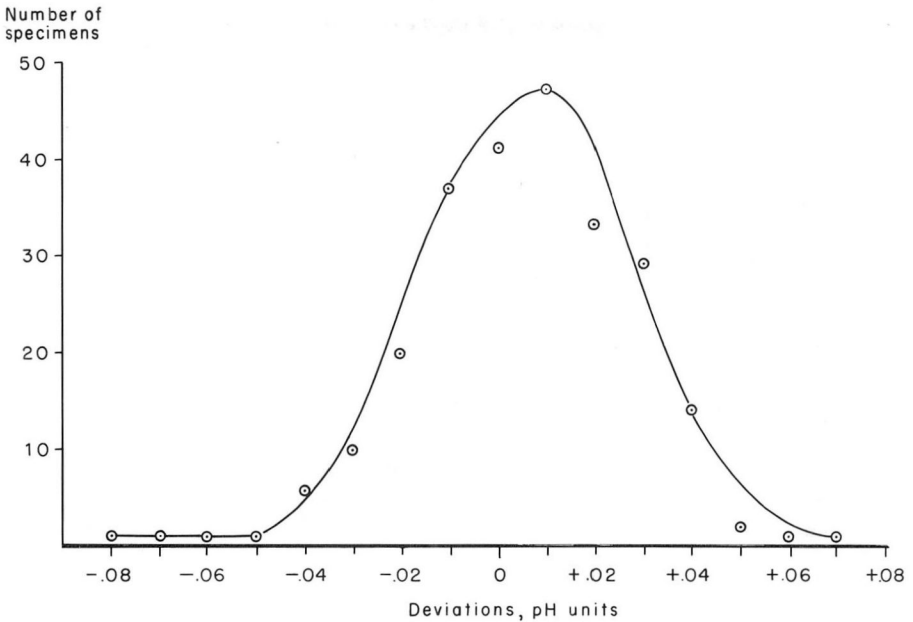


Fig. 3. Graph showing the deviations in pH units of the corrected blood pH values. Note that deviations are almost equally distributed on either side of zero.

Discussion

In Rosenthal's study,¹ pH values of blood specimens, from humans, measured at room temperature were compared with values on the same specimens at 38 C. His analyses were performed with semi micro glass electrodes. Three samples were determined simultaneously in three identical glass electrodes. The electrical potential of each glass electrode was measured with respect to the same calomel reference electrode in the same amplifying circuit. One aliquot of each specimen was maintained at 18 C., one at 38 C., and the third at some intermediate temperature. Rosenthal¹ found that the pH of blood was an inverse linear function of the temperature over the range from 38 to 18 C. He further observed that the slope of the line relating blood pH to temperature exhibited little variation among specimens when the initial values at 38 C. were from pH 7.25 to 7.45. From his data he calculated the mean temperature coefficient to be 0.0147 pH unit per degree, with a standard deviation of 0.006.

Craig and his associates² used Cambridge Research Model pH meters with condenser glass electrodes. The pH values of 27 blood specimens were compared under two sets of conditions. In the first study, each of 18 specimens was divided into two portions. One pH meter and one portion of the specimen were equilibrated at 37 C. and the portion of the specimen was measured. After the meter had cooled

to room temperature, it was used to measure the other portion of specimen. In order to exclude the possibility that the delay in reading one portion of the sample might have affected the pH, a second procedure was employed. One pH meter was equilibrated at 37 C. and the other at room temperature. Nine additional specimens were measured almost simultaneously with the two instruments. The temperature coefficient obtained in both studies was 0.0149 pH unit per degree with a standard deviation of 0.00273. Although this coefficient is essentially the same as Rosenthal's,¹ the standard deviation is somewhat greater. Craig and associates² called attention to the fact that of the 52 human specimens examined by Rosenthal,¹ 34 were from the same person; this factor may have reflected an unusually small standard deviation. Craig and associates² further stated that on the basis of their own coefficient and standard deviation, the maximal error in measuring the pH of any given blood specimen probably would not exceed 0.05 pH unit.

Since the over-all inaccuracy of the Cambridge pH meter⁴ is about 0.02 of a pH unit, and the absolute accuracy of the Metrohm instrument⁵ is 0.01 pH unit, the extent of the discrepancy of the measurement from purely instrumental causes could be as high as 0.03 unit if the errors were additive. It was assumed that when the two measurements were discrepant, the one at 37.5 C. was probably the more accurate of the two. Deviations from the result at 37.5 C. that exceed 0.03 unit can therefore logically be assigned to individual variations in the specimens from the average.

It is seen that errors of 0.05 pH unit or even higher may be incurred when measurements of pH are made at room temperature and the results are corrected by a standard formula. These errors may be either plus or minus, thereby doubling the uncertainty. For the greatest accuracy, pH measurements of blood should be made at the patient's actual body temperature. When this is not feasible, a satisfactory approach to the ideal is to measure the specimen at the usual average body temperature, 37.5 C. When measurements are made at room temperature, the possibility of large errors should be recognized, even when correction factors are applied.

Summary

In 243 patients, pH values of blood, measured at 37.5 C., were compared with the corresponding values determined at room temperature and were corrected by means of Rosenthal's¹ formula. Discrepancies of 0.05 pH unit and even higher were observed between the two sets of values. As much as 0.03 unit may be due solely to instrumental variation. It was assumed that the values at 37.5 C. were more nearly correct than those at room temperature and that deviations in excess of 0.03 pH units were errors due to variations in specimens.

Ideally, blood pH measurements should be made at the body temperature of the patient, but practically, measurements made at some standard temperature in the normal range, such as 37.5 C., are usually satisfactory. If this cannot be done, it should be recognized that errors of considerable magnitude may occur.

Acknowledgment

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References

1. Rosenthal, T. B.: Effect of temperature on pH of blood and plasma in vitro. *J. Biol. Chem.* 173: 25-30, 1948.
2. Craig, F. A.; Lange, K.; Oberman, J., and Carson, S.: Simple accurate method of blood pH determination for clinical use. *Arch. Biochem. & Biophys.* 38: 357, 1952.
3. Sendroy, J.: Problems in measurement of pH of blood and other biological fluids. *In* Symposium on pH measurement, Special Technical Publication No. 190. Philadelphia: American Society for Testing Materials, 1959, 110 p.
4. Instruction for the Use and Care of Research Model, pH Meter, New York: Cambridge Instrument Company, Inc., 11 p.
5. Operating Instructions, Metrohm Precision Compensator. Herisau, Switzerland, 1959.