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Effect of Heparin or Saline Dilution of Blood on P_{CO_2} and pH

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ABSTRACT

The effect of different dilutions with heparin solutions or saline on blood P_{CO_2}, pH and standard bicarbonate was investigated. Blood was first equilibrated to give about 40 or 60 mmHg P_{CO_2}. The solutions were in equilibrium with room air. The effect on blood P_{CO_2} etc. could be fully explained by the dilution with a medium having a much lower P_{CO_2}. Thus, correction of the heparin solution to pH 7.40 and P_{CO_2} 40 mmHg eliminated the effect on P_{CO_2}, pH and standard bicarbonate. With ordinary procedure for blood heparinization (about 2% dilution) the effect is practically negligible.

INTRODUCTION

It has earlier been reported that dilution of blood with anticoagulant solution or saline gives a lower carbon dioxide tension while pH is not significantly affected. Siggaard Andersen (5) found a fall in P_{CO_2} of 16% when blood was diluted by 12–13% with saline. When adding dry heparin to concentrations of 2, 4 and 10 mg/ml, Siggaard Andersen found that the average effect of 1 mg heparin per ml blood was +0.1 mmHg P_{CO_2}. In contrast, Bradley (1) reported a 28% fall in P_{CO_2} at the same dilution. He also suggested that the reason for this discrepancy was methodological.

In routine analysis of blood gases and acid–base status, either 1% heparin solution or the same solution diluted by 70% with saline (3+7 parts) is most commonly used. The effect of such dilution of blood sample appears to be of practical importance and was therefore re-examined.

METHODS

Blood was collected into dry heparinized test tubes (Vitrum AB, Stockholm) and was equilibrated with two different gas mixtures, A and B, calculated to give 40–42 and 55–60 mmHg P_{CO_2}, respectively, and in both cases about 70 mmHg P_{CO_2}. For the equilibration, a Laué tonometer (3) combined with Instrumentation Laboratory Tonometer Bath (model 137) was used. For the transfer of equilibrated blood to the analyzer, 1 ml glass syringes were used for undiluted blood and 5 ml or 10 ml glass syringes (BD Yale) for diluted blood. For dilution, one of the following solutions was used:

I	1% commercial heparin in distilled water	P_{CO_2}	3 mmHg,	pH 7.6
II	1% commercial heparin, diluted with saline	P_{CO_2}	3	pH 7.2
III	1% heparin in distilled water (pH-corrected by Vitrum AB)	P_{CO_2}	3	pH 7.40
IV	1% heparin in distilled water	P_{CO_2}	40	pH 6.8
V	1% heparin in distilled water	P_{CO_2}	40	pH 7.4
VI	5% commercial heparin in distilled water	P_{CO_2}	3	pH 6.9
VII	saline (0.9% NaCl)	P_{CO_2}	3	pH 6.0

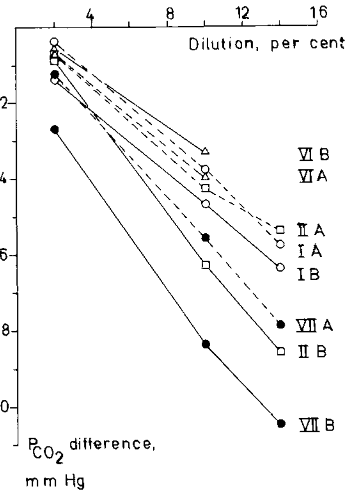
The heparin Vitrum is a sodium salt of heparin, dissolved in distilled water. The 1% and 5% solutions and the 1% pH-corrected solutions of heparin were kept in glass bottles covered with rubber caps, which were removed during sampling, i.e. giving access to room air (P_{CO_2} about 1 mmHg). All other solutions were kept in glass syringes with metal caps.

In part of the series, the 5 ml and 10 ml syringes were weighed empty, with diluent (V_D) and with diluent+blood (V_D+V_B). At 20°C the density of blood was 1.058 and of 1% heparin solution in distilled water was 1.002 but this difference was negligible when per cent volume dilution was calculated. Dilution was calculated as V_D in per cent of V_D+V_B.

Analyses of pH and P_{CO_2}, performed on Radiometer instruments BMS 3+PHM 72a (digital meter), were carried out immediately after the sampling into a syringe. The undiluted equilibrated blood was analyzed at the beginning and at the end, sometimes also in the middle, of each series of 4 to 6 diluted samples. For 30 series the coefficient of variation was 1% and the standard error of a single determination was 0.46 mmHg.

RESULTS

As is shown in Fig. 1, a dilution of about 2% with either one of the solutions (I, II, VI) did not consid-



erably affect the carbon dioxide tension, the fall in P_{CO_2} always being less than 2 mmHg. The fall was between 1.0 and 1.9% at a P_{CO_2} of about 40 mmHg, and 1.1–2.4% at a P_{CO_2} of about 60 mmHg. The same result was obtained with solution III. Dilution with saline showed a slightly greater effect with an average fall of 3.2 and 4.6% at the two P_{CO_2} levels respectively. The effect was more pronounced at dilutions of 10% and 14% and could be explained by the dilution with a medium having a much lower CO_2 tension than the blood sample. The effect could therefore be predicted by means of the degree of dilution. For all solutions, except saline, the observed carbon dioxide tension of diluted blood samples corresponded to the predicted value within 2 mmHg, as can be seen in Table I. Saline, however, gave a somewhat greater difference. At a CO_2 tension of about 60 mmHg, the average difference between the observed P_{CO_2} of a sample diluted by 13–14% and the predicted P_{CO_2} was 3.3 mmHg, the observed value being too low. In a few cases, the dilution was determined precisely by both weight and density correction, but this procedure did not significantly influence the precision of the ordinary P_{CO_2} prediction, the difference being less than 0.5 mmHg.

Dilution by about 14% with 1% heparin solution gave a fall in P_{CO_2} of 14.4 and 11.7%, respectively, at the lower and higher CO_2 tensions. For samples diluted to the same degree with saline, the decrease in P_{CO_2} was 19.7% and 19.2%, respectively. The latter values would correspond to the correction factors (*f*) of 1.245 and 1.238 when calculated (diluted $P_{CO_2} \times f$=undiluted P_{CO_2}) according to Bradley (1). At 15% dilution with saline and P_{CO_2} about 40 mmHg Bradley reported a fall in P_{CO_2} of 32%, corresponding to a correction factor of 1.480. Siggaard Andersen (5) found a 16% lower P_{CO_2} when the blood sample was diluted 12–13% with saline.

The effect of dilution of pH and standard bicarbonate (st.bic.) is much smaller than on P_{CO_2}. At 2% dilution there was no change in st.bic. for any of the solutions used. In the present investigation, a 14% dilution with saline resulted in a rise in pH of 0.005 and 0.008 units. In st.bic. a fall of 2 mE/l (9.3%) and

Table I. *Difference between observed and calculated effects of dilution on blood carbon dioxide tension, mmHg. Dilution procedure I–VII as described in Methods*

Figures are mean values of 5 experiments each

Diluent and per cent dilution (%)		Observed value before–after dilution		Calculated value after dilution		Difference±S.E.D. between observed and calculated value after dilution	
		Gas A	Gas B	Gas A	Gas B	Gas A	Gas B
I	10	39.7–35.9	57.7–53.0	36.0	52.2	−0.1±0.19	+0.8±0.39
	14	40.2–34.4	54.9–48.5	35.0	47.7	−0.6±0.32	+0.8±0.16
II	10	40.2–35.9	54.6–48.2	36.5	49.4	−0.6±0.20	−1.2±0.30
	14	40.0–34.6	54.5–45.9	34.8	47.3	−0.2±0.07	−1.4±0.12
III	10	40.8–37.0	57.6–51.8	37.0	52.1	0 ±0.25	−0.3±0.48
IV	10	40.3–39.1	55.6–54.8	40.3	54.1	−1.2±0.32	+0.7±0.13
V	10	40.3–41.0	54.5–54.0	40.3	53.1	+0.7±0.20	+0.9±0.10
VI	10	43.5–39.5	54.7–51.4	39.5	49.5	0 ±0.16	+1.9±0.30
VII	10	39.8–34.2	58.4–50.0	36.1	52.9	−1.9±0.30	−2.9±0.72
	14	40.2–32.3	55.1–44.5	35.0	47.8	−2.7±0.21	−3.3±0.62

3 mE/l (12.4%) was found at the lower and higher CO_2 tensions respectively. When blood was diluted with solution II the fall in st.bic. was similar, i.e.—2.0 and 2.5 mmE/1. Dilution of blood with solution V did not affect the value of standard bicarbonate.

COMMENT

The results of the present investigation show that the effect of diluent must be considered in blood for gas analysis and acid–base status. According to Jorpes (2) 2 mg heparin is a sufficient amount of anticoagulant to approximately 50 ml blood. At 2% dilution, i.e. adding 1% heparin in the syringe dead space volume (0.2 ml) to 10 ml blood, the effect on pH, P_{CO_2} and standard bicarbonate is minimal and the values are unnecessary to correct. The present results correspond well with the results found by Siggaard Andersen (5).

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