A COMPARATIVE STUDY OF ACID-BASE DETERMINATION USING THE ASTRUP MICRO-TECHNIQUE

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Acid-base determinations are an important and established aid to modern clinical medicine. Severinghaus[1] said of Astrup's ultra-micro method 'This approach greatly simplifies this entire problem of acid-base balance for clinical medicine and as far as I can see, for research work too'. Serial acid-base determinations are particularly important in the management of newborn infants suffering from the idiopathic respiratory distress syndrome. The Neonatal Respiratory Unit at Groote Schuur Hospital recently acquired an Astrup apparatus and this method has now replaced the older technique of Van Slyke blood gas analysis, which we had used in conjunction with a Metrohm pH meter. The Astrup method has the advantage that arterialized capillary blood can be used. After warming the extremity, collection of blood samples and acid-base determination take only 10 minutes.

A comparative study was carried out between the Astrup micro-method and our previous method. The aim was to determine our experimental error when using the Astrup apparatus and to assess the comparability of the 2 methods. The Astrup micro-method will be used exclusively in the cases to be studied in future.

Determinations were carried out as a part of routine laboratory procedure. No attempt was made to improve the accuracy by taking special precautions which would not be practicable under normal laboratory conditions. A wide range of acid-base disturbances was chosen such as would be encountered in our investigation of newborn infants.

APPARATUS

Method 1

(a) *The Metrohm pH meter E300* (scale divisions = 0·02 pH units) with the Metrohm suction microglass electrode using about 100 µl. of blood.

The pH meter and electrode jacket were kept at a constant temperature by a circulating water bath with built-in thermostat set at 38°C.

(b) *The Van Slyke manometric apparatus* for determination of whole blood total CO_2 content using 0·2 ml. of whole blood.[2]

Plasma total CO_2 content corrected for pH and oxygen saturation was calculated with the Van Slyke and Sendroy line chart.[3] Oxygen saturation was determined from the oxygen content and oxygen capacity (Hb. × 1·34 volumes % in adults and 1·26 volumes % in neonates).[4] The plasma bicarbonate, PCO_2 and base excess were read from a nomogram.[5]

Method 2

(a) *The Radiometer pH meter 27* (scale divisions = 0·01 pH units) with the Radiometer suction microglass electrode using about 20 µl. blood.

(b) *The Astrup micro-tonometer* for equilibration by the Astrup method.[6-9]

A circulating water thermostat kept both the electrode and tonometer at 38°C and Centigrade thermometers inserted into the system at both these points checked the temperature. Corrections were made for unsaturation and results were plotted on the nomogram provided to obtain PCO_2, actual bicarbonate and base excess. The CO_2 content of the gases used for equilibration were analysed with the Scholander micro-gas analyser (SE = 0·047%).

Method

Blood samples of 3 ml. were collected anaerobically after filling the dead space of a syringe with heparin. The syringe was then capped and kept on ice. Samples were either umbilical cord blood, venous blood or arterial blood. All determinations were carried out in duplicate and were completed within ¼-hour of collection.

Results

A wide variation in acid-base status was found in the patients studied. The range was as follows:

pH = 7·105 to 7·547;
O_2 = 4·9 to 23·0 volumes %;
HCO_3 = 14·6 to 25·2 mEq./l.;
PCO_2 = 20 to 75 mm.Hg;
Base excess = −9·8 to +1·9 mEq./l.

The standard error of all determinations was within acceptable limits. No significant difference was found between the 2 methods for any of the values, with the exception of the pH (Table I and Figures 1, 2 and 3).

DISCUSSION

On examination of the data it was found that in 13 of the 14 readings the Radiometer pH was higher than that of the Metrohm. The mean difference was 0·009 units. Sendroy *et al.*[11] found that the glass electrode read 0·01 - 0·02 pH units lower than the hydrogen electrode when using whole blood. This difference did not appear when plasma was used. Severinghaus *et al.*[12] resolved the problem by adding 0·01 to all whole blood pH readings when the PCO_2 was to be calculated, and Siggaard-Andersen[13] came to the same conclusion.

Jenny *et al.*[14] reported that solid particles in pastes and suspensions have a lowering effect on the observed pH and ascribed it to the effect these particles (in our case the red blood cells) have on the diffusion of ions at the boundary between the KCl and the unknown suspension. Siggaard-Andersen, however, ascribed the lower pH readings to the precipitation and haemolysis of the red blood cells at the junction between the blood and the saturated KCl.[13]

Although both electrodes used in the study were glass, these results raise the suspicion that the observed difference of 0·009 pH units may have been due to the following structural differences in the 2 systems:

(*a*) The Radiometer micro-electrode lies horizontally, thus reducing the danger of error due to sedimentation and diffusion whereas the Metrohm has a vertical electrode.

(*b*) The inner bore of the Metrohm pH meter electrode capillary is more than 3 times that of the Radiometer. A larger surface of blood will, therefore, be presented to the saturated KCl and more haemolysis will take place.

Another important difference is the lack of a built-in thermometer to check the thermostat setting in the

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TABLE I. COMPARISON OF DIFFERENT METHODS OF STUDYING ACID—BASE STATUS

Type blood	O_2 vol. % V.Slyke	Total CO_2 blood mEq./l. V.Slyke	pH		HCO_3 mEq./l.		PCO_2 mm.Hg		Base excess mEq./l.	
			Metrohm	Radiometer	V.Slyke and Metrohm	Astrup method	V.Slyke and Metrohm	Astrup method	V.Slyke and Metrohm	Astrup method
Cord	5·98	19·35	7·143	7·150	20·5	20·6	62·0	60·3	—9·0	—8·5
	5·84	19·20	7·160	7·139	20·5	20·2	62·6	60·3	—9·8	—8·9
Arterial	19·45	16·20	7·547	7·534	19·2	21·0	23·4	22·8	—1·5	—1·8
	19·60	15·90	7·515	7·536	19·2	20·2	23·5	21·5	—1·4	—2·8
Arterial	20·80	17·15	7·520	7·545	20·3	20·5	25·0	24·5	—0·5	—1·0
	20·60	16·90	7·530	7·545	20·8		25·7		0·0	
Arterial	16·40	12·70	7·482	7·490	14·6	16·7	20·0	21·9	—6·5	—5·0
	16·70	13·10	7·465	7·494	15·3	16·7	21·6	22·1	—6·3	—5·2
Arterial	16·60	13·25	7·475	7·484	15·0	17·25	21·2	23·8	—6·5	—5·8
	16·40	13·40	7·470	7·483	15·3		21·7		—6·3	
Cord	11·47	20·40	7·105	7·140	22·0	19·2	73·0	81·0	—9·5	—5·4
	11·60	20·45	7·105	7·150	21·6	17·5	75·0	76·0	—10·0	—7·4
Venous	4·90	22·50	7·315	7·315	24·6	24·4	51·0	49·5	—1·0	—0·8
	5·25	22·70	7·315	7·320	24·3		49·6		—1·2	
Arterial	22·70	19·00	7·500	7·510	22·0	22·4	28·9	28·8	+0·7	+1·8
	22·60	17·95	7·500	7·510	22·7	21·4	29·8	27·6	+1·9	+0·9
Arterial	19·10	18·90	7·465	7·471	22·2	20·4	30·7	28·7	—0·3	—0·8
	19·70	18·20	7·465	7·465	22·7	20·0	32·3	28·5	+0·3	—1·4
Arterial	22·50	16·37	7·380	7·386	19·0	20·3	33·5	35·0	—4·6	—3·4
	23·00	16·30	7·375	7·377	18·6	20·5	33·4	36·0	—5·0	—3·4
Arterial	17·75	19·05	7·380	7·380	22·0	20·2	38·5	35·6	—2·0	—3·6
	16·85		7·390	7·380	22·0		38·4		—2·0	
Venous	11·95	22·30	7·394	7·394	25·0	24·8	43·0	42·4	+0·5	+0·4
	12·30	21·50	7·375	7·394	25·2	24·5	44·2	41·6	+0·6	+0·3
Cord	7·65	20·40	7·245	7·265	22·2	24·2	53·0	53·5	—5·0	—3·6
	7·75	20·24	7·250	7·275	22·4		55·0		—5·0	
Arterial	16·05	13·75	7·499	7·506	14·9	15·4	20·0	20·2	—6·5	—7·2
	16·20	13·60	7·493	7·493	15·6	15·7	20·7	20·5	—5·0	—6·7
Mean difference between duplicate determinations	0·298	0·317	0·009	0·006						
Standard error	0·005	0·008	0·003	0·001						
p†			$p \leqslant 0{\cdot}3$							
Mean difference between the two methods			0·009		1·136		1·28		0·99	
Standard error			0·013		1·420		1·53		1·14	
p*			0·02		0·9		0·2		0·9	

p† refers to the probability that the difference between the two means is due to chance.
p* refers to the probability that the mean difference between the two methods is different from zero.

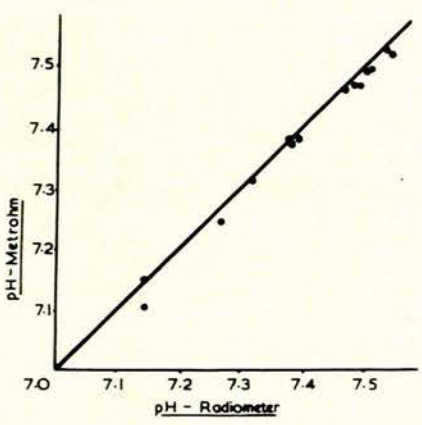

Fig. 1

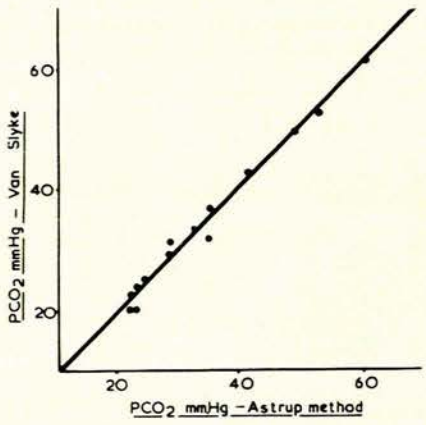

Fig. 2

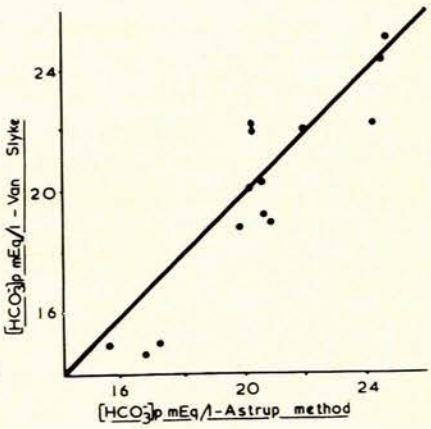

Fig. 3

Fig. 1. Scatter diagram showing comparison of pH between method 1 and method 2. *Fig. 2.* Scatter diagram showing comparison of PCO_2 between method 1 and method 2. *Fig. 3.* Scatter diagram showing comparison of $[HCO_3^-]$ p between method 1 and method 2.

Metrohm pH system. An increase in temperature of 1 °C will cause a decrease of 0·0147 in pH in whole blood.[13] Indeed, the correction of pH and PCO_2 for the actual temperature of the patient has been stressed by various authors.[10,11]

An error of 0·01 in the pH will cause a 2·25% error in the PCO_2.[12] Even after correction of the Metrohm pH readings our standard error in the PCO_2 remained unchanged. This is a reflection of the experimental error in the plasma bicarbonate measurement by the 2 methods, as well as the limitations inherent in the reading of nomograms. These and other sources of error have been fully discussed by Severinghaus *et al.*,[12] Berglund *et al.*[16] and Siggaard-Andersen.[9,13]

SUMMARY

Acid-base determinations were carried out on 14 blood samples, using 2 methods. The standard error was within acceptable limits and the results were comparable, despite the wide range in acid-base status.[17] The Metrohm pH meter

consistently gave a lower reading. Possible factors for this difference are discussed.

We consider that the Astrup micro-method fulfils all the requirements of acid-base determination stated by Wynn[18]: 'It is not entirely accuracy we want—it is reproducibility, reliability, speed and convenience'.

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