

# Correction of the anion gap for albumin in order to detect occult tissue anions in shock

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**Background:** It is believed that hypoalbuminaemia confounds interpretation of the anion gap (AG) unless corrected for serum albumin in critically ill children with shock.

**Aim:** To compare the ability of the AG and the albumin corrected anion gap (CAG) to detect the presence of occult tissue anions.

**Methods:** Prospective observational study in children with shock in a 22 bed multidisciplinary paediatric intensive care unit of a university children's hospital. Blood was sampled at admission and at 24 hours, for acid-base parameters, serum albumin, and electrolytes. Occult tissue anions (lactate + truly "unmeasured" anions) were calculated from the strong ion gap. The anion gap ( $(\text{Na} + \text{K}) - (\text{Cl} + \text{bicarbonate})$ ) was corrected for serum albumin using the equation of Figge:  $\text{AG} + (0.25 \times (44 - \text{albumin}))$ . Occult tissue anions (TA) predicted by the anion gap were calculated by  $(\text{anion gap} - 15 \text{ mEq/l})$ . Optimal cut off values of anion gap were compared by means of receiver operating characteristic (ROC) curves. Ninety three sets of data from 55 children (median age 7 months, median weight 4.9 kg) were analysed. Data are expressed as mean (SD), and mean bias (limits of agreement).

**Results:** The incidence of hypoalbuminaemia was 76% ( $n = 42/55$ ). Mean serum albumin was 25 g/l (SD 8). Mean AG was 15.0 mEq/l (SD 6.1), compared to the CAG of 19.9 mEq/l (SD 6.6). Mean TA was 10.2 mmol/l (SD 6.3). The AG underestimated TA with mean bias 10.2 mmol/l (4.1-16.1), compared to the CAG, mean bias 5.3 mmol/l (0.4-10.2). A clinically significant increase of TA  $>5$  mmol/l was present in 83% ( $n = 77/93$ ) of samples, of which the AG detected 48% ( $n = 36/77$ ), and the CAG 87% ( $n = 67/77$ ). Post hoc ROC analysis revealed optimal cut off values for detection of TA  $>5$  mmol/l to be AG  $>10$  mEq/l, and CAG  $>15.5$  mEq/l.

**Conclusion:** Hypoalbuminaemia is common in critically ill children with shock, and is associated with a low observed anion gap that may fail to detect clinically significant amounts of lactate and other occult tissue anions. We suggest that the albumin corrected anion gap should be calculated to screen for occult tissue anions in these children.

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It has been shown that hypoalbuminaemia is associated with metabolic alkalosis, an effect that is readily explained using strong ion theory.<sup>1-4</sup> It follows that coexisting hypoalbuminaemia may confound interpretation of conventional acid-base parameters by underestimating the degree of acidosis.<sup>1-5</sup> The anion gap, the difference between the major serum cations and anions, is commonly used to estimate the presence of excess inorganic and organic "unmeasured" anions. Since the normal anion gap is largely a result of the charge on albumin, the observed anion gap may fail to detect increased concentrations of lactate or "unmeasured" anions, such as ketones, pyruvate, or pyroglutamic acid, if the serum albumin is low.<sup>1-6</sup> This finding has lead previous authors to derive an equation to correct for the artificially low observed anion gap in the presence of hypoalbuminaemia.<sup>7-11</sup>

Nevertheless, the conventional uncorrected anion gap is still widely used to detect occult or "unmeasured" anions in children, since there is little published data to alert paediatricians to the pitfalls of this practice. (Note that lactate might also be termed an "unmeasured" anion in centres where it is not a routine investigation. In order to avoid confusion we shall refer to the sum of lactate and the truly "unmeasured" anions as occult tissue anions.<sup>12</sup>) While critically ill children may develop lactic acidosis because of tissue hypoxia-ischaemia, among other causes, they are also likely to be hypoalbuminaemic.<sup>13-14</sup> Since a raised lactate is associated with increased mortality in these children, early detection and awareness may be important.<sup>13</sup> Therefore, it is of some concern that, in the very group of patients in whom we wish to detect

occult tissue anions such as lactate, the uncorrected anion gap may fail to reveal them.

Whereas the anion gap estimates these occult tissue anions, the amount of lactate and "unmeasured" anions may be calculated with greater precision using equations derived from strong ion theory.<sup>3-4</sup> Although the strong ion equations are too cumbersome to use in clinical practice, it is possible to use them to calculate the underestimation of occult tissue anions by the uncorrected anion gap. We may also determine whether correcting the anion gap for the albumin concentration, using the equation of Figge, reliably detects a clinically significant increase of occult tissue anions in these children.<sup>11</sup> This correction might then be applied in clinical practice, without the need to calculate strong ion equations.

We present a comparison of the uncorrected anion gap (AG) and the albumin corrected anion gap (CAG) for the detection of occult tissue anions in critically ill children with shock.

## METHODS

This prospective observational study was set in the 22 bed multidisciplinary paediatric intensive care unit (PICU) of a university children's hospital. Children admitted to the PICU

**Abbreviations:** AG, anion gap; BE, base excess; CAG, albumin corrected anion gap; INR, international normalised ratio; PICU, paediatric intensive care unit; ROC, receiver operating characteristic; SB, standard bicarbonate; SID, strong ion difference; SIG, strong ion gap; TA, tissue anions

**Table 1** Acid-base parameters, albumin, tissue anions (TA), uncorrected (AG) and corrected (CAG) anion gap, and predicted tissue anions

| Parameter                    | Mean  | SD   | Range        |
|------------------------------|-------|------|--------------|
| pH                           | 7.25  | 0.20 | 6.70 to 7.62 |
| BE (mmol/l)                  | -11.4 | 8.4  | -30 to 5.3   |
| SB (mmol/l)                  | 15.6  | 6.3  | 2.6 to 28.1  |
| Albumin (g/l)                | 25    | 8.0  | 7 to 42      |
| Lactate (mmol/l)             | 5.2   | 4.8  | 0.4 to 20.1  |
| "Unmeasured" anions (mmol/l) | 5.0   | 3.6  | -1.1 to 14.5 |
| SIG calculated TA (mmol/l)   | 10.2  | 6.3  | 0.8 to 32.5  |
| AG (mEq/l)                   | 15.0  | 6.1  | 5.8 to 37.3  |
| CAG (mEq/l)                  | 19.9  | 6.6  | 9.0 to 42.8  |
| AG predicted TA (mmol/l)     | 0.0   | 6.1  | -9.2 to 22.3 |
| CAG predicted TA (mmol/l)    | 4.9   | 6.6  | -6.0 to 27.8 |

with shock requiring either additional fluid resuscitation or inotropic support, were eligible for enrolment with the informed consent of their parents. Shock was defined as hypotension (blood pressure lower than 2 standard deviations from the age appropriate mean), or poor peripheral perfusion (absent peripheral pulses or capillary refill time >4 seconds).<sup>15</sup> Children admitted following trauma, cardiac surgery, or with an inherited metabolic disease were excluded. The institutional research ethics committee approved the study protocol.

Ringer's lactate is the routine resuscitation fluid for children with hypovolaemia and shock at this institution, in preference to albumin based solutions, normal (0.9%) saline, or synthetic colloid. It is not routine practice to correct hypoalbuminaemia by infusion of a concentrated albumin solution.

Blood was sampled on admission to PICU, 24 hours after admission, and at intervals determined by the patient's clinical condition, for determination of: arterial pH, base excess (BE), standard bicarbonate (SB), serum lactate, serum albumin, and serum electrolytes. A maximum of two samples per patient were included for analysis. Acid-base parameters were measured and derived using a Radiometer ABL 520 blood gas analyser (Copenhagen, Denmark). Serum electrolytes were measured by a Beckman Synchron CX3 analyser, using ion specific electrodes (Berlin, Germany). Serum albumin and lactate were measured by a Beckman Synchron CX5 analyser, using the enzymatic method (Berlin, Germany).

Hypoalbuminaemia was defined as <33 g/l.<sup>15</sup> Protein energy malnutrition was defined as weight below 60% of expected weight for age, or weight below 3rd centile for age with the presence of oedema. Liver dysfunction was defined as an increase of hepatic enzymes, or international normalised ratio (INR) in the absence of a disseminated intravascular coagulopathy, of more than twice the upper limit of normal.

The AG was calculated using the formula  $(Na + K) - (Cl + HCO_3)$ , whereby potassium was considered a "measured", rather than "unmeasured" cation.<sup>11,16</sup> The upper limit of normal of the AG calculated by this formula was defined as 15 mEq/l, based on 2 standard deviations from the mean in studies of "normal" populations which used modern ion specific electrode techniques for the measurement of sodium and chloride, and including an upward adjustment of 4 mEq/l for potassium where necessary.<sup>17-21</sup> It should be noted that this limit is lower than that historically derived by flame photometry and colorimetry.<sup>16,17,22-24</sup> The CAG was calculated using the formula of Figge.<sup>11</sup>

The strong ion difference (SID) and strong ion gap (SIG) were calculated using the standard formulae reported previously.<sup>4</sup> "Unmeasured" anions were derived from the SIG. Occult tissue anions predicted by the SIG were calculated from the serum lactate plus "unmeasured" anions.<sup>12</sup> See Appendix 1 for formulae.

Hyperlactataemia was defined as serum lactate >2 mmol/l.<sup>13,25</sup> Raised "unmeasured" anions were defined as >3 mmol/l.<sup>12</sup> A

clinically significant increase of occult tissue anions was defined as >5 mmol/l.<sup>12</sup>

Occult tissue anions predicted by the anion gap (both AG and CAG) were calculated from the anion gap minus 15 mEq/l. This method assumes that no lactate or "unmeasured" anions occurred in the serum of the "normal" populations from whom this upper limit of 15 mEq/l was obtained. Since the serum lactate of normal individuals may vary between zero and 2 mmol/l, and unmeasured anions between zero and 3 mmol/l, there is inherent potential for underestimation of occult tissue anions, but importantly, this effect is common to both the AG and CAG.<sup>12,25,26</sup>

In order to further define the optimal cut off value of anion gap that should be used for detection of clinically significant occult tissue anions in this study population, post hoc analysis of receiver operating characteristic (ROC) curves was performed for the AG, and CAG, for TA >5 mmol/l.

Non-parametric demographic data were reported as median (range), and analysed using the Mann-Whitney test for continuous data. Parametric data were reported as mean (SD) for continuous data, and analysed using the  $\chi^2$  test for categorical data. Differences between techniques were reported as mean bias and limits of agreement (2 SD).

Complete data for calculation of tissue anions from the strong ion difference were available in 55 children, median age 7 months (range 0.1-144), weight 4.9 kg (range 1.8-30). Thirty eight children (69%) had blood sampled both at admission and at 24 hours, such that 93 samples were available for analysis.

## RESULTS

Forty four of the 55 children (80%) were admitted with shock associated with local or systemic sepsis. Other diagnoses included viral myocarditis (n = 4), meningoencephalitis (n = 2), as well as iron poisoning, hepatitis, aortic stenosis, hypoplastic left heart syndrome, and near drowning (all n = 1).

The incidence of hypoalbuminaemia was 76% (n = 42/55). The incidence of liver dysfunction was 22% (n = 12/55), and the incidence of protein energy malnutrition was 7% (n = 4/55). The serum albumin concentration was lower in children with liver dysfunction, median 23 g/l (range 9-33) compared to 25 g/l (range 7-42), and in children with protein energy malnutrition, median 17 g/l (range 15-22) compared to 25 g/l (7-42) (both p < 0.001).

Table 1 shows acid-base, albumin, tissue anion, and anion gap data. The mean AG was 15.0 (SD 6.1) mEq/l, compared to the CAG of 19.9 (SD 6.6) mEq/l (mean bias 4.9 mEq/l, limits of agreement 0.9-8.8 mEq/l).

Occult tissue anions predicted by the AG were 0.0 (SD 6.1) mmol/l, compared to 4.9 (SD 6.6) mmol/l predicted by the CAG (mean bias and limits of agreement the same as for anion gap above).

**Table 2** Relation between an elevated (>15 mEq/l) uncorrected (AG), corrected (CAG) anion gap, and tissue anions (TA), n (%)

|               | TA <2 mmol/l | TA 2–5 mmol/l | TA >5 mmol/l | Total     |
|---------------|--------------|---------------|--------------|-----------|
| AG <15 mEq/l  | 5 (100%)     | 11 (100%)     | 40 (52%)     | 56 (60%)  |
| AG >15 mEq/l  | 0 (0%)       | 0 (0%)        | 37 (48%)     | 37 (40%)  |
| CAG <15 mEq/l | 5 (100%)     | 7 (64%)       | 10 (13%)     | 22 (24%)  |
| CAG >15 mEq/l | 0 (0%)       | 4 (36%)       | 67 (87%)     | 71 (76%)  |
| Total         | 5 (5%)       | 11 (12%)      | 77 (83%)     | 93 (100%) |

Occult tissue anions calculated from the SIG were 10.2 (SD 6.3) mmol/l. It follows that the AG under predicted the amount of tissue anions with mean bias 10.2 mmol/l (limits of agreement 4.1–16.1 mmol/l). The CAG also under predicted tissue anions, with mean bias 5.3 mmol/l (limits of agreement of 0.4–10.2 mmol/l).

Table 2 shows the relations between an increased (>15 mEq/l) AG, CAG, and occult tissue anion concentrations of <2 mmol/l, 2–5 mmol/l, and >5 mmol/l respectively.

A clinically significant increase of occult tissue anions (>5 mmol/l) was present in 83% (n = 77/93) of samples. The AG detected a clinically significant increase of tissue anions in 48% (n = 36/77) (that is, sensitivity 48%, specificity 100%, and negative predictive value 27%), compared to the CAG, which detected a clinically significant increase of tissue anions in 87% (n = 67/77) (that is, sensitivity 87%, specificity 75%, and negative predictive value 55%) (p = 0.002).

ROC curve analysis of the AG and CAG for the detection of TA >5 mmol/l shows that both parameters are equally useful, with area under the ROC curve of 0.93 (95% CI 0.87 to 0.98), and 0.91 (95% CI 0.85 to 0.97), respectively. However, the optimal cut off value to be used for the detection of TA >5 mEq/l was an AG >10 mEq/l (sensitivity 94%, specificity 81%, and likelihood ratio = 4.9), compared to a CAG >15.5 mEq/l (sensitivity 87%, specificity 81%, and likelihood ratio = 4.9).

## DISCUSSION

We have shown that in this population of critically ill children with shock, metabolic acidosis, and hyperlactataemia, the incidence of hypoalbuminaemia is greater than 75%. This finding lends support to the hypothesis that critically ill children are likely to have both increased tissue anions and hypoalbuminaemia simultaneously.<sup>1–14</sup> In other words, a factor that confounds interpretation of the anion gap is present in the very group of patients in whom we most wish to apply it.

Furthermore, the anion gap underpredicted the amount of occult tissue anions by 10 mmol/l, and failed to detect the presence of a clinically significant increase of tissue anions >5 mmol/l in more than 50% of cases. Since both lactate and “unmeasured” anions have been associated with severity of illness and increased mortality, failure to detect such an increase of occult tissue anions might have adverse consequences for the patient.<sup>13–16, 27</sup>

However, it is gratifying that correction of the anion gap for albumin underestimated occult tissue anions to a lesser degree, and revealed clinically significant increase of these anions in more than 80% of cases. This finding is of clinical importance, since calculation of the albumin corrected anion gap (anion gap + [0.25 × (44 – albumin)]) is relatively simple, and may be performed at the bedside.<sup>11</sup>

Clinical application of the anion gap hinges on three key issues. Firstly, the upper limit of normal depends on whether potassium is included in the equation. Many authors classify potassium as an “unmeasured” cation, and use the equation (Na) – (Cl + HCO<sub>3</sub>).<sup>11, 16–28</sup> Although serum potassium varies only within a narrow range compatible with life (charge 4

mEq/l), there is no real justification for this practice since potassium concentrations are readily obtainable. Ultimately, it does not matter which equation is used, provided that the reference limit is adjusted accordingly for the inclusion, or exclusion, of the charge on potassium.<sup>11, 16</sup>

Secondly, the reference limit should be compatible with the technique used to measure serum electrolytes in that laboratory. Historically, the upper reference limit still quoted by some current texts is that of 16 mEq/l, using (Na) – (Cl + HCO<sub>3</sub>), which equates to 20 mEq/l using (Na + K) – (Cl + HCO<sub>3</sub>).<sup>16, 23–29</sup> However, these reference ranges were obtained during the 1970s using flame photometry and colorimetry techniques, which underestimate chloride compared to the modern ion specific electrode techniques that have superseded them.<sup>17</sup> Therefore, since the reference range for serum chloride has risen, the upper limit of the anion gap (mean + 2SD) has fallen correspondingly, to an average of 11 mEq/l excluding potassium or 15 mEq/l including potassium.<sup>17–20</sup>

Thirdly, the anion gap should be interpreted in the context of other pathophysiological derangements in the individual patient. We have shown that children with shock are frequently hypoalbuminaemic, and that this results in a lowered anion gap that severely underestimates the presence of occult tissue anions. We have shown that in order to detect the presence of tissue anions in children with shock, either the upper limit of “normal” might be lowered to 10 mEq/l, or the anion gap should be corrected for the serum albumin concentration.

It would appear from the ROC analysis that a CAG of approximately 15 mEq/l (including potassium) would be the optimal value—that is, the best combination of sensitivity and specificity, to detect occult tissue anions. Yet the negative predictive value of a CAG >15 mEq/l was only 55%. It follows that many shocked children with a normal CAG may still have clinically significant tissue anions, since even the corrected anion gap underestimated the total amount of tissue anions by a considerable margin.

This finding may be due partly to the inherent underestimation of the technique, in that occult tissue anions present “within” the normal anion gap are not accounted for. Interpretation of the anion gap, and the strong ion gap, may also be confounded by the presence of excess “unmeasured” cations, such as paraproteins and cationic drugs, which decrease the observed anion gap.<sup>11</sup> We should also consider the influence of pH. While correction of the anion gap for albumin takes into account the charge on protein at that pH, the equation makes use of the linear relation between albumin and charge shown in critically ill adults whose pH ranged from 7.09 to 7.65.<sup>11, 30</sup> However, at the extremes of metabolic acidosis seen in these children with shock, the relation between pH and charge on protein may not be linear.

Our study population had a higher incidence of protein energy malnutrition (7%) than might be expected in most “developed world” intensive care units. Although these children had a lower albumin concentration than children without evidence of malnutrition, the high incidence of

hypoalbuminaemia in the entire study group suggests that this finding is a function of critical illness, rather than nutritional status, as has been found previously in both children and adults.<sup>14</sup> These findings should therefore be applicable to all critically ill children.

We suggest that the albumin concentration should be measured in all critically ill children with shock. In centres where the serum lactate is not routinely measured, the albumin corrected anion gap should be calculated to screen for the presence of lactate and other occult tissue anions.

### Conclusion

Hypoalbuminaemia is common in critically ill children with shock, and is associated with an artificially low observed anion gap that may fail to detect the presence of lactate and other occult tissue anions. Correction of the anion gap for serum albumin will reveal a clinically significant increase of these anions in the majority of cases.

### APPENDIX 1: FORMULAE<sup>4 11</sup>

Strong ion difference (apparent) (SIDa) = (Na + K + Mg + Ca) – (Cl + lactate)

Strong ion difference (effective) (SIDE) =  $(1000 \times 2.46E-11 \times pCO_2 / 10^{-pH}) + [\text{albumin} \times (0.123 \times pH - 0.631)] + [PO_4 \times (0.309 \times pH - 0.469)]$

Strong ion gap (SIG) = "unmeasured" anions (UA) = SIDa – SIDE

Uncorrected anion gap (AG) = (Na + K) – (Cl + HCO<sub>3</sub>)

Albumin corrected anion gap (CAG) = AG +  $[0.25 \times (44 - \text{albumin})]$

Tissue anions (TA) = lactate + UA

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### REFERENCES

- 1 **Fencel V**, Jabor A, Kazda A, *et al*. Diagnosis of metabolic acid-base disturbances in critically ill patients. *Am J Respir Crit Care Med* 2000;**162**:2246–51.
- 2 **McAuliffe JJ**, Lind LJ, Leith DE, *et al*. Hypoproteinemic alkalosis. *Am J Med* 1986;**81**:86–90.
- 3 **Stewart PA**. Modern quantitative acid-base chemistry. *Can J Physiol Pharmacol* 1983;**61**:1444–61.
- 4 **Kellum JA**, Kramer DJ, Pinsky MR. Strong ion gap: a methodology for exploring unexplained anions. *J Crit Care* 1995;**10**:51–5.
- 5 **Constable PD**. Clinical assessment of acid-base status. Strong ion difference theory. *Vet Clin North Am Food Anim Pract* 1999;**15**:447–71.
- 6 **Figge J**, Rossing TH, Fencel V. The role of serum proteins in acid-base equilibria. *J Lab Clin Med* 1991;**117**:453–67.
- 7 **Carvounis CP**, Feinfeld DA. A simple estimate of the effect of the serum albumin level on the anion gap. *Am J Nephrol* 2000;**20**:369–72.
- 8 **Sheth KJ**, Kher KK. Anion gap in nephrotic syndrome. *Int J Pediatr Nephrol* 1984;**5**:89–92.
- 9 **Lolekha PH**, Lolekha S. value of the anion gap in clinical diagnosis and laboratory evaluation. *Clin Chem* 1983;**29**:279–83.
- 10 **Nanji AA**, Campbell DJ, Pudek MR. Decreased anion gap associated with hypoalbuminemia and polyclonal gammopathy. *JAMA* 1981;**246**:859–60.
- 11 **Figge J**, Jabor A, Kazda A, *et al*. Anion gap and hypoalbuminaemia. *Crit Care Med* 1998;**26**:1807–10.
- 12 **Durward A**, Skellett S, Mayer A, *et al*. The value of the chloride:sodium ratio in differentiating the aetiology of metabolic acidosis. *Intensive Care Med* 2001;**27**:828–35.
- 13 **Hatherill M**, McIntyre AG, Wattie M, *et al*. Early hyperlactataemia in critically ill children. *Intensive Care Med* 2000;**26**:314–18.
- 14 **Balasubramanyan N**, Havens PL, Hoffman GM. Unmeasured anions identified by the Fencel-Stewart method predict mortality better than base excess, anion gap, and lactate in patients in the pediatric intensive care unit. *Crit Care Med* 1999;**27**:1577–81.
- 15 **Shann F**. *Drug doses*. Melbourne: Collective Pty Ltd, 1998.
- 16 **Emmett M**, Narins RG. Clinical use of the anion gap. *Medicine* 1977;**56**:38–54.
- 17 **Winter SD**, Pearson R, Gabow PA, *et al*. The fall of the serum anion gap. *Arch Intern Med* 1990;**150**:311–13.
- 18 **Singh RN**, Singh NC, Hutchinson J, *et al*. Lower anion gap increases sensitivity in predicting elevated lactate. *Clin Intensive Care* 1994;**5**:221–4.
- 19 **Jurado RL**, Del Rio C, Nassar G, *et al*. Low anion gap. *South Med J* 1998;**91**:624–9.
- 20 **Lolekha PH**, Vanavanan S, Teerakarnjana N, *et al*. Reference ranges of electrolyte and anion gap on the Beckman E4A, Beckman Synchron CX5, Nova CRT, and Nova Stat Profile Ultra. *Clin Chim Acta* 2001;**307**:87–93.
- 21 **Roberts WL**, Johnson RD. The serum anion gap. Has the reference interval really fallen? *Arch Pathol Lab Med* 1997;**121**:568–72.
- 22 **Witte DL**, Rodgers JL, Barrett DA. The anion gap: its use in quality control. *Clin Chem* 1976;**22**:643–6.
- 23 **Frohlich J**, Adam W, Golbey MJ, *et al*. Decreased anion gap associated with monoclonal and pseudomonoclonal gammopathy. *Can Med Assoc J* 1976;**114**:231–2.
- 24 **Thomas DW**, Pain RW, Duncan BM. The anion gap. *Lancet* 1973;**2**:848–9.
- 25 **Mizock BA**, Falk JL. Lactic acidosis in critical illness. *Crit Care Med* 1992;**20**:80–93.
- 26 **Wilkes P**. Hypoproteinemia, strong-ion difference, and acid-base status in critically ill patients. *J Appl Physiol* 1998;**84**:1740–8.
- 27 **Weil MH**, Afifi AA. Experimental and clinical studies on lactate and pyruvate as indicators of the severity of acute circulatory failure (shock). *Circulation* 1970;**41**:989–1001.
- 28 **Oh MS**, Carroll HJ. The anion gap. *N Engl J Med* 1977;**297**:814–17.
- 29 **Nicholson JF**, Pesce MA. Reference ranges for laboratory tests and procedures. In: Behrman RE, Kliegman RM, Jensen HB, eds. *Nelson textbook of pediatrics*, 16th edn. Philadelphia: WB Saunders Company, 2000:2190.
- 30 **Figge J**, Mydosh T, Fencel V. Serum proteins and acid-base equilibria: a follow-up. *J Lab Clin Med* 1992;**120**:713–19.