Original Article

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Pathogenesis of Metabolic Acidosis in Preterm Infants

Acid Base Balance Section

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ABSTRACT

Objective: To determine how the balance between mineral base, and carbonic and organic acids is altered to cause metabolic acidosis in preterm infants.

Study Design: Mineral balance and arterial blood measurements of 3 groups of preterm infants given 5 different Total Parenteral Nutrition (TPN) regimens were analyzed. Mineral base was measured as the difference between mineral cations and anions. Organic acid was measured as the difference between mineral base, bicarbonate and protein anion.

Results: The degree of metabolic acidosis measured as base excess, was determined by deviation in both mineral base and organic acid from normal.

Sodium minus chloride balance determined change in arterial blood mineral base. TPN containing more chloride than sodium caused mineral acidosis with low mineral base, whereas TPN containing more sodium than chloride caused mineral alkalosis with high mineral base. Lactic, organic and carbonic acidosis all increased mineral base. Arterial blood organic acid was determined by:

1. Glomerular filtration rate: Low rates after delivery caused high organic acid that fell as GFR improved.

2. TPN non metabolized organic acid content: Gluconate and sulphate caused organic acidosis by accumulating in blood and mineral acidosis by urine excretion resulting in mineral base loss.

3. Rate of protein catabolism: Increased protein catabolism from TPN providing only 25 kcal/g amino acids or from dexamethasone caused organic acidosis.

Conclusion: Metabolic acidosis was caused by high organic acid, resulting from low glomerular filtration rates in the first 1-2 weeks, exacerbated by TPN containing gluconate or sulphate or only 25 kcal/g amino acids. Renal bicarbonate wasting could not account for metabolic acidosis.

Key Words: Prematurity, Total parenteral nutrition, Acidosis, Metabolism, Organic acid

INTRODUCTION

At present "renal bicarbonate wasting" provides the textbook explanation for otherwise unexplained metabolic acidosis in preterm infants. In blood, carbon dioxide is mainly in the form of bicarbonate kept in a constant ratio with 1/20th the amount of carbonic acid. The lungs eliminate large amounts of carbon dioxide, but insignificant amounts are excreted via the kidneys. So how could "renal bicarbonate wasting" be significant in the face of massive "pulmonary bicarbonate wasting"?

Studies since 1970 have found low urine net hydrogen ion excretion after delivery in preterm compared to term infants [1], which then increases by 4 weeks following the time course of their metabolic acidosis [2]. Since then low renal hydrogen ion secretion in preterm babies has been assumed to cause "renal bicarbonate wasting". However urine bicarbonate is determined by urine organic acid, mineral base and ammonium concentrations, not merely the rate of hydrogen ion secretion. In one of these groups of preterm infants [3], the development of metabolic acidosis was clearly shown to be due to chloride retention exceeding sodium retention for the first 14 days, but this has not become recognized as the cause. Reports of metabolic acidosis in infants given Total Parenteral Nutrition (TPN) [4, 5] also showed that the tendency to cause acidosis was related to how much more chloride than sodium TPN regimens contain, but once again this has not been recognized as the cause.

The previous article describes how to measure mineral base, carbonic and organic acids in blood and the principles governing the balance between them [6]. This is the original acid base theory, based on the work of Lawrence Henderson, Donald van Slyke, Alfred Shohl and others up to 1928. It was validated by three prospective cohort studies of mineral balance and arterial blood gas measurements in preterm infants receiving TPN. This article demonstrates that the composition of parenteral fluids, renal function and the rate of protein catabolism affect mineral base and organic acids accounting for the acid base status of these infants. The following two ar-

ticles examine the relation between blood gas measurements and outcome following perinatal asphyxia and the inverse physiological relation between carbonic and lactic acids [7, 8].

METHODS

Three cohorts of preterm infants requiring (TPN) were prospectively studied in Brighton, England during 1986; in Wellington, New Zealand during 1993 and in New Plymouth, New Zealand from 1996 to 2001. The equilibrium between mineral base, and carbonic and organic acids was investigated from blood and urine measurements made as part of the clinical care of these infants. During these periods all preterm infants needing parenteral fluid therapy because of respiratory or feeding problems were studied. Over the three study periods, five different TPN regimens were given and their compositions are shown in [Table/Fig-1].

First cohort [9] In 1986, arterial blood gases, mineral base, protein, urea and creatinine were measured, all on the same sample of blood, obtained on admission, when 10% glucose was started as maintenance fluid, at 48 hours, when TPN was commenced, and at 7 days after birth. At the same time the infants were weighed and between these periods all urine was collected and analyzed for mineral base, urea and creatinine concentrations. Over the same periods the volumes of all parenteral fluids, including fresh frozen plasma and sodium bicarbonate, were recorded. No milk was given during these balance periods, which were stopped when milk feeds could be started. Samples of TPN and fresh frozen plasma were analyzed for mineral base concentrations.

Second cohort In 1993, arterial blood gases, lactate, mineral base, protein, urea and creatinine were measured, all on the same sample of blood, obtained at 7 days after birth. At about the same time, a urine sample was obtained and analyzed without delay for mineral base, creatinine and pH. Gluconate was measured on frozen aliquots of blood and urine samples at a later date using a commercial kit. The rate of TPN administration at the time was recorded and a record kept of all sodium bicarbonate administered since delivery.

Third cohort Between 1996 and 2001, arterial blood gases, lactate, mineral base, protein, urea and creatinine were measured, all on the same sample of blood, obtained from infants whenever clinically indicated. Urine samples were obtained daily or less frequently and analyzed without delay for mineral base, urea, creatinine and pH. TPN was started on day 1 and records were kept of volumes of TPN, other parenteral fluids and milk given until the infants were fully milk fed. Measurements of creatinine production made in the first cohort [9] were used to measure urine mineral, urea or gluconate excretion in mmol/kg/day in the second and third cohorts as follows:

Mineral excretion=[Mineral]urine/ [Creatinine]urine x Creatinine production

Creatinine production= -2.07+2.34 x gestational age (weeks) µmol/kg/day

Sulphate in TPN cannot be metabolized and must be excreted in urine. The increase in plasma sulphate in mmol/L produced by sulphate in TPN regimens was measured as follows:

Increase in plasma SO_4= TPN SO_4 intake (mmol/kg/day)/Glomerular filtration rate

Glomerular Filtration Rate (GFR)= Creatinine production/ [Creatinine] plasma L/kg/day

GFR was measured like this when plasma creatinine was no longer increasing after delivery. When plasma creatinine was increasing soon after delivery, GFR was measured using paired plasma creatinine measurements as previously described [9]. These simple and readily available methods produce measurements that are consistent with inulin infusion measurements of GFR in preterm infants [10, 11]. GFR was measured in L/kg/day because weight is the best standard for glomerular filtration in the newborn [10], and because this time period allows the filtration rate of a substance to be compared with its excretion rate over 24 hours. Preterm babies with low GFR of 0.5 L/kg/day filter their entire extracellular fluid volume only once a day.

Urine urea nitrogen rather than urine total nitrogen was measured. Urea nitrogen balance overestimates total nitrogen balance, because it does not include urine losses of non-urea nitrogen. However, under stable conditions, urea excretion equals urea production, which measures the rate of protein catabolism. Urea filtration rate, calculated by multiplying plasma urea by GFR, was compared with urea excretion, measured from urine samples obtained within 6 hours of the blood samples.

Results were expressed as mean ± sample Standard Deviation (SD) and analysis performed with standard parametric tests, linear, logarithmic, power and polynomial regression analysis.

RESULTS

1. TPN composition influences arterial plasma mineral base and organic acid

Infants receiving the five different TPN regimens were of comparable birth weight and gestational age, and had comparable intakes of TPN on day 7, as shown in [Table/Fig-2]. Here their arterial blood gas results on day 7 are compared, together with their urine pH and need for sodium bicarbonate while on TPN.

The theory of acid base balance predicted that two factors determine how TPN composition influences acid base balance

a. The difference between TPN sodium and chloride influences mineral base:

Infants receiving the 1986 TPN regimens, that had considerably more chloride than sodium, developed mineral acidosis, with low mineral base, whereas infants receiving 1996 TPN, that had more sodium than chloride, de-

veloped mineral alkalosis, with high mineral base. The 1986 TPN regimen with the greater excess of chloride, regimen X, tended to cause worse mineral acidosis, although this difference was not significant.

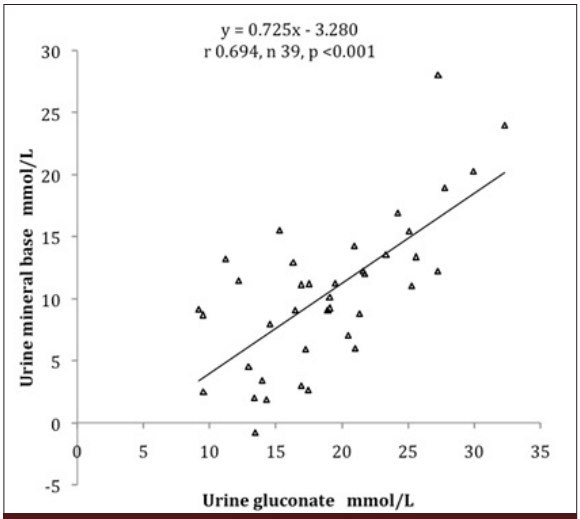
b. The TPN concentration of non metabolized organic acids influences organic acid

Infants receiving 1993 TPN developed more severe mineral acidosis, which needed more sodium bicarbonate to correct, than infants on the 1986 TPN regimens, despite 1993 TPN having only 5 mmol/L more chloride than sodium. The theory of acid base balance predicted that this would be caused by gluconate in TPN not being fully metabolized.

On 1993 TPN, plasma gluconate was 1.7 ± 0.3 mmol/L and 67.2 ± 9.9% of administered gluconate was excreted in urine, with the rate of gluconate filtration equaling the rate of gluconate excretion. Gluconate was poorly metabolized and along with sulphate caused metabolic acidosis, partly by accumulating in blood as organic acid and partly by being excreted in urine along with mineral base, as shown in [Table/Fig-2].

Year	TPN	1986	1986	1993	1996	1998
TPN regimen		Regimen X	Regimen Y	High gluconate	Standard	High calcium
Glucose	g/L	93	93	100	100	125
Amino acids	g/L	19.24	19.24	19.24	27.17	24.5
Nitrogen	g/L	2.74	2.74	2.74	3.84	3.5
Sodium	mmol/L	35.8	35.8	35	25	39
Potassium	mmol/L	23.3	23.3	21.7	25	25
Calcium	mmol/L	9.48	9.48	9.79	10	17.5
Magnesium	mmol/L	1.40	1.40	1.96	2.42	2
Chloride	mmol/L	58	49.2	40	20.83	39
Phosphate	mmol/L	7.26	11.63	11.76	10	19.5
Gluconate	mmol/L	6.4	6.4	19.58	10	0
Sulphate	mmol/L	0	0	1.96	1.6	0
Acetate	mmol/L	0	0	0	27.83	25
Sodium Chloride	mmol/L	-22.2	-13.4	-5	4.17	0

[Table/Fig-1]: The compositions of the five TPN regimens studied



[Table/Fig-2]: Gluconate was accompanied by mineral base in urine samples on day 7.5±1.0 days from 39 infants, birth weight 914±167 g, gestation 27.4±1.4 weeks, receiving 123±27 mL/kg/day of 1993 TPN
Charge on phosphate was corrected for urine pH in calculating mineral base

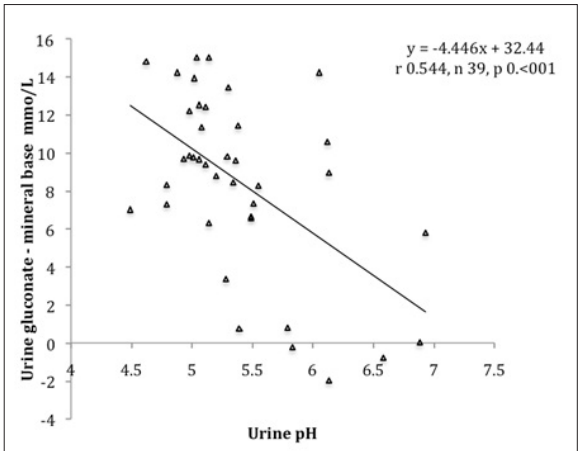
Year	TPN	1986 X	1986 Y	1993	1996	1998
Infants	n	8	13	27	16	22
Birth weight	g	968 ± 182	997 ± 213	941 ± 178	1107 ± 160	1034 ± 270
Gestation	weeks	27.4 ± 1.3	27.2 ± 2.1	27.4 ± 1.7	28.1 ± 1.9	27.2 ± 1.9
Postnatal age	days	6.9 ± 0.4	6.8 ± 0.6	7.5 ± 1.2	6.7 ± 1.3	6.8 ± 0.8
TPN intake	mL/kg/day	122 ± 20	114 ± 20	116 ± 22	108 ± 11	120 ± 17
Lipid intake	mL/kg/day	7.9 ± 3.6	6.9 ± 3.0	9.4 ± 1.5	10.0 ± 1.3	12.0 ± 1.8
NaHCO3 intake	mmol/kg	1.86 ± 2.58	1.72 ± 2.08	3.17 ± 2.51	0	0
Arterial blood pH		7.264 ± 0.081	7.271 ± 0.060	7.285 ± 0.043	7.410 ± 0.066	7.361 ± 0.066
PCO2	mmHg	44.7 ± 10.7	44.1 ± 6.4	41.3 ± 9.9	45.8 ± 9.7	50.3 ± 8.2
HCO3	mmol/L	19.3 ± 2.4	19.7 ± 2.7	18.7 ± 3.1	28.3 ± 3.4	27.8 ± 2.5
Base excess	mmol/L	-7.1 ± 2.2	-6.5 ± 3.0	-7.1 ± 2.0	3.4 ± 3.0	1.4 ± 2.8
Mineral base	mmol/L	34.8 ± 3.2	36.5 ± 3.1	37.4 ± 3.4	49.1 ± 2.6	42.2 ± 3.0
Mineral base deviation	mmol/L	-4.3 ± 2.8	-2.6 ± 3.1	-2.0 ± 3.8	9.6 ± 3.2	3.7 ± 2.5
Organic acid	mmol/L	5.2 ± 2.1	6.3 ± 1.2	7.9 ± 2.6	9.2 ± 2.7	4.2 ± 2.4
Lactate	mmol/L			1.8 ± 0.7	1.5 ± 0.5	1.59 ± 0.56
Gluconate	mmol/L			1.7 ± 0.3		0
Sulphate increase	mmol/L	0	0	0.53 ± 0.12	0.38 ± 0.09	0
GFR	L/kg/day	0.73 ± 0.23	0.67 ± 0.15	0.91 ± 0.27	0.94 ± 0.23	0.90 ± 0.27
Urine pH				5.4 ± 0.6	6.8 ± 1	7.0 ± 0.9

[Table/Fig-3]: Arterial blood acid base results on day 7 from infants below 32 weeks gestation and below 1500 grams birth weight on 5 different TPN regimens

This depleted mineral base causing mineral acidosis, as shown in [Table/Fig-3]. In response infants acidified their urine to reduce the loss of mineral base with gluconate, as shown in [Table/Fig-4].

On 1996 TPN, organic acid was higher than expected from the gluconate and sulphate content of the TPN, but was balanced by high mineral base. Measurements confirmed that acetate was fully metabolized with negligible plasma levels. As presented below, high rates of protein catabolism, caused by the regimen's low energy to protein ratio of 25 kcal/g amino acids, accounted for higher than expected organic acid on 1996 TPN.

The 1998 TPN, with equal sodium and chloride concentrations and no sulphate or gluconate, was designed to cause no acid base disturbance. This regimen provided 30kcal/g amino acids, which did not increase protein catabolism. Infants on 1998 TPN had significant lung problems producing respiratory acidosis, appropriately compensated for by mild mineral alkalosis with normal organic acid.



[Table/Fig-4]: Loss of mineral base with gluconate was decreased in acid urine

2. Extracellular mineral base balance determines change in arterial plasma mineral base

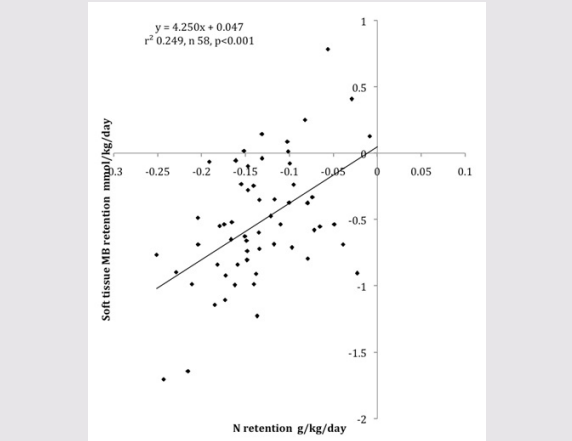
When examining the relation between mineral base balance and change in arterial plasma mineral base, it is important to remember that arterial mineral base measures concentration not amount and so change in concentration caused by fluid changes must first be corrected for, either by change in weight or by change in ECF volume [9].

Change in plasma MB = Plasma MB Final x Weight Final-Plasma MB Initial corrected for weight Weight Initial

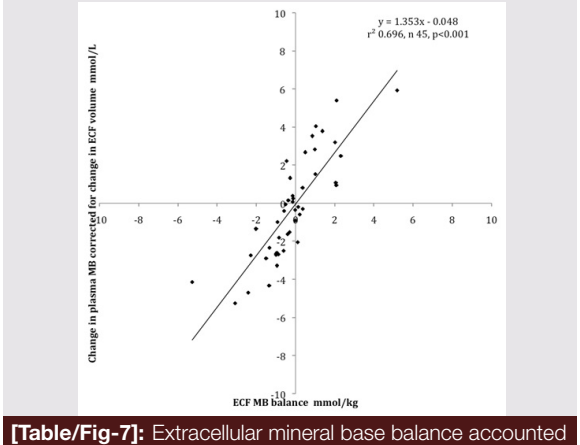
Change in plasma MB = Plasma MB Final x ECF volume Final corrected for ECF volume - Plasma MB Initial x ECF volume Initial

Postnatal age		Admission to 48 hours	48 hours to 5-7 days	p
Parenteral fluid		10% glucose	1986 TPN	
Infants	n	58	31	
Birth weight	g	1369 ± 520	1176 ± 390	
Gestational age	weeks	29.5 ± 2.9	28.2 ± 2.3	
MB intake	mmol/kg/day	0.610 ± 0.930	0.815 ± 0.439	NS
MB urine	mmol/kg/day	0.875 ± 0.625	1.256 ± 0.926	<0.05
MB bone retention	mmol/kg/day	0.101 ± 0.103	0.537 ± 0.223	<0.001
MB soft tissue retention	mmol/kg/day	-0.521 ± 0.463	0.156 ± 0.487	<0.001
Nitrogen retention	g/kg/day	-0.1339 ± 0.0545	0.0939 ± 0.0908	<0.001
ECF MB balance	mmol/kg/day	0.156 ± 1.255	-1.134 ± 0.665	<0.001

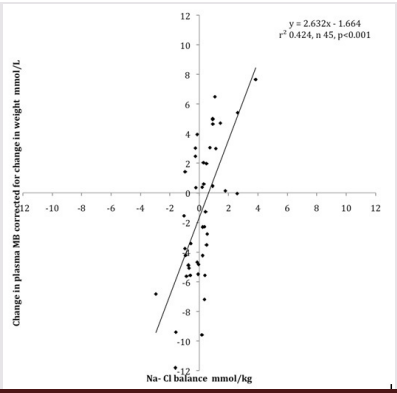
[Table/Fig-5]: Comparison of mineral base and nitrogen balances on 1986 TPN regimen X and Y from 48 hours to 7 days with those on 10% glucose from admission to 48 hours



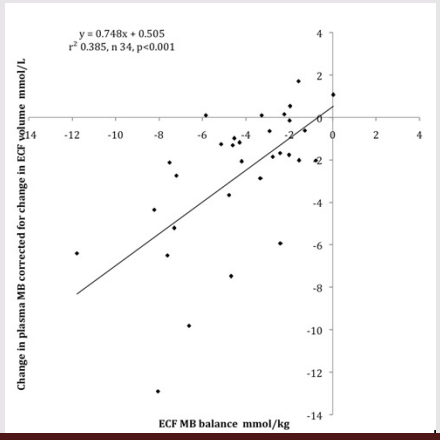
[Table/Fig-6]: Soft tissue mineral base production\ correlated with nitrogen production from admission to 48 hours on 10% glucose



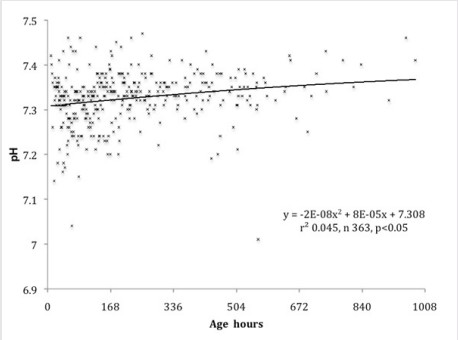
[Table/Fig-7]: Extracellular mineral base balance accounted for change in arterial plasma mineral base corrected for change in extracellular fluid volume from admission to 48 hours on 10% glucose



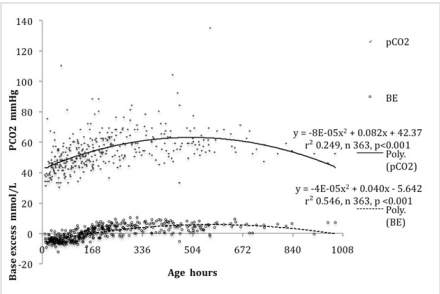
[Table/Fig-8]: Sodium minus chloride balance accounted for change in arterial plasma mineral base corrected for change in weight from admission to 48 hours on 10% glucose



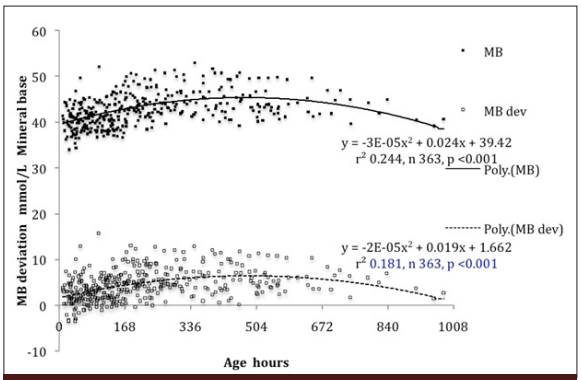
[Table/Fig-9]: Extracellular mineral base balance accounted for change in arterial plasma mineral base corrected for change in extracellular fluid volume from 48 hours to 7 days on 1986 TPN



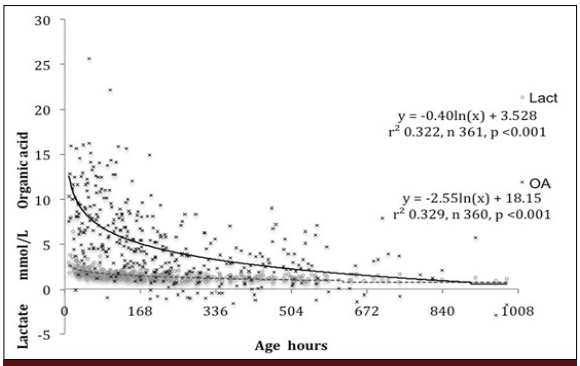
[Table/Fig-10]: Arterial blood pH in 20 infants below 30 weeks gestation on 1998 TPN during first 6 weeks



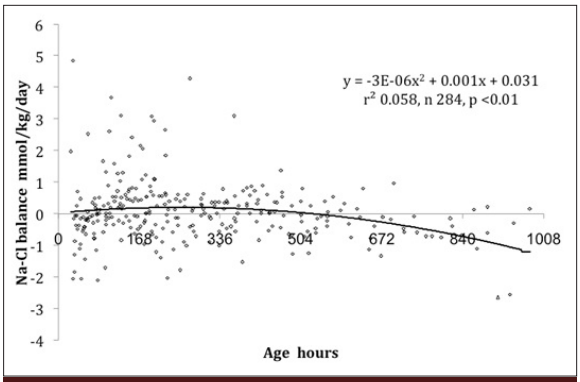
[Table/Fig-11]: Arterial blood PCO2 and base excess in 20 infants below 30 weeks gestation on 1998 TPN during first 6 weeks



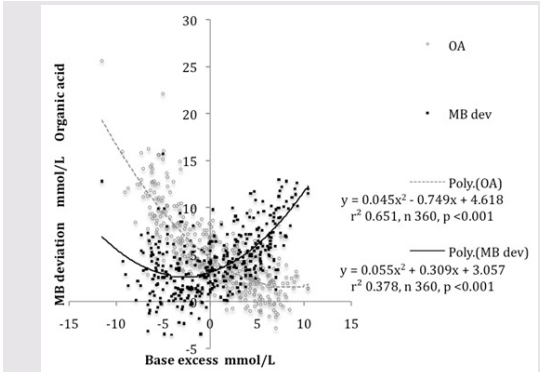
[Table/Fig-12]: Arterial blood mineral base and mineral base deviation in 20 infants below 30 weeks gestation on 1998 TPN during first 6 weeks



[Table/Fig-13]: Arterial blood lactic and organic acids in 20 infants below 30 weeks gestation on 1998 TPN during first 6 weeks



[Table/Fig-14]: Sodium minus chloride balance accounted for change in arterial blood mineral base deviation in 20 infants below 30 weeks gestation on 1998 TPN during first 6 weeks



[Table/Fig-15]: Arterial base excess was determined by mineral base deviation and organic acid in 20 infants below 30 weeks gestation on 1998 TPN

a. Admission to 48 hours on 10% glucose in 1986

Infants receiving 10% glucose only for the first 48 hours developed negative nitrogen balance, as shown in [Table/Fig-5]. Potassium exceeded phosphate production resulting in mineral base production from cells into extracellular fluid, as shown in [Table/Fig-6].

[Table/Fig-7] shows that extracellular mineral base balance accounted very well for change in arterial mineral base corrected for change in ECF volume. A similar highly significant relationship was found between sodium minus chloride balance and change in arterial mineral base corrected for change in weight, as shown in [Table/Fig-8]. In the latter but not the former relationship all the parameters are independent of each other. However the former relationship takes into account mineral base retention in bone and production from soft tissue and corrects arterial mineral base better for fluid changes.

b. 48 hours to 7 days on 1986 TPN

Mineral base and nitrogen balances on 1986 TPN are shown in [Table/Fig-5]. This regimen produced only modest rates of protein retention with on average 1.66 mmol of soft tissue mineral base retained with every gram of nitrogen. Bone mineral base retention was also low on this regimen.

1986 TPN caused negative extracellular mineral base balance, which produced mineral acidosis, as shown in [Table/Fig-9].

c. 1998 TPN

Arterial blood acid base measurements during the first 6 weeks from the 20 infants below 30 weeks gestation receiving 1998 TPN are shown in [Table/Fig-10] - pH, [Table/Fig-11] - PCO2 and base excess, [Table/Fig-12] mineral base and mineral base deviation and [Table/Fig-13] lactic and organic acids.

These babies were mildly acidotic, particularly for the first 2 weeks [Table/Fig-10]. During the 1st week they had metabolic acidosis [Table/Fig-11], caused by elevated organic but not lactic acids, which gradually declined to normal by the 2nd week [Table/Fig-13]. This was followed by respiratory acidosis, particularly during the 2nd to 4th weeks, caused by most having significant respiratory problems [Table/Fig-11]. Their initial organic acidosis was compensated for by increased mineral base, which rose further to compensate for their later respiratory acidosis [Table/Fig-12]. Concomitant sodium minus chloride balances demonstrating the retention of sodium in excess of chloride accounted well for the increase in mineral base, as shown in [Table/Fig-14]. Furthermore, as their respiratory acidosis resolved, mineral base declined, produced by the appropriate retention of chloride in excess of sodium. Mineral base increased to compensate for both organic and respiratory acidosis, as shown in [Table/Fig-15].

Mineral base and nitrogen balances of infants below 30 weeks gestation on 1996 and 1998 TPN are compared in [Table/Fig-16]. Bone mineral base retention on 1998 TPN was more than twice that on 1996 TPN. Urea excretion was increased on 1996 TPN resulting in lower nitrogen retention than on 1998 TPN. Nitrogen retention on 1998 TPN correlated well with potassium, soft tissue phosphate and magnesium retention, with on average 4.48 mmol of potassium, 2.06 mmol of soft tissue phosphate and 0.46 mmol of magnesium retained with each gram of nitrogen [10]. This is equivalent to 1.69 mmol of soft tissue mineral base retention with every gram of nitrogen, similar to 1.64 mmol from the regression equation:

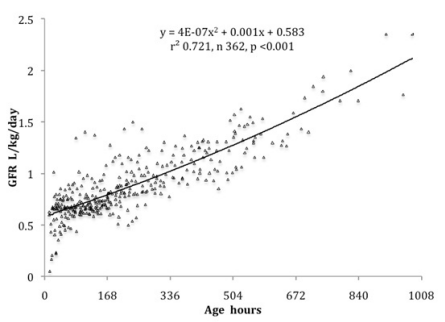
Soft tissue MB retention mmol/kg/day = 1.932 N retention g/kg/day-0.289 r 0.290 n 306.

Soft tissue mineral base retention was affected by phosphate metabolism, as shown in [Table/Fig-16]. 1996 TPN was phosphate deficient and produced lower soft tissue phosphate retention [10] and therefore higher soft tissue mineral base retention than 1998 TPN.

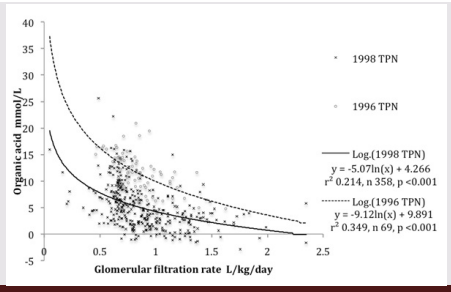
Seven infants on 1998 TPN were treated with dexamethasone for chronic lung disease. This increased protein catabolism and urine phosphate [12], thereby decreasing soft tissue phosphate retention and increasing soft tissue mineral base retention. Treatment with dexamethasone allowed greater TPN intakes that prevented nitrogen retention from falling.

		1996 TPN no steroids	p%	1998 TPN no steroids	p%	1998 TPN dexamethasone
Infants	n	10		20		7
Birth weight	g	1078 ± 176	ns	998 ± 273	ns	941 ± 240
Gestational age	weeks	27.0 ± 1.2	ns	26.6 ± 1.6	ns	26.3 ± 1.6
Balance studies	n	37		254		54
TPN intake	mL/kg/day	99.2 ± 20.1	<0.001	119.2 ± 19.6	<0.001	129.0 ± 11.1
17.7% Lipid intake	mL/kg/day	8.3 ± 2.7	<0.001	11.6 ± 2.3	<0.001	13.4 ± 1.3
Urine urea nitrogen	g/kg/day	0.125 ± 0.030	<0.001	0.076± 0.028	<0.001	0.103 ± 0.042
Nitrogen retention	g/kg/day	0.253 ± 0.077	<0.001	0.339± 0.072	ns	0.346 ± 0.055
Soft tissue MB retention	mmol/kg/day	0.604 ± 0.423	<0.01	0.395± 0.461	<0.001	0.738 ± 0.519
Bone MB retention	mmol/kg/day	0.841 ± 0.154	<0.001	1.862± 0.313	<0.001	2.017 ± 0.193

[Table/Fig-16]: Comparison of mineral base and nitrogen balances on 1998 TPN with those on 1996 TPN and those on 1998 TPN with dexamethasone in infants less than 30 weeks gestation



[Table/Fig-17]: Glomerular filtration rate in 20 infants below 30 weeks gestation on 1998 TPN during first 6 weeks



[Table/Fig-18]: Glomerular filtration rate determined arterial organic acid in below 30 weeks gestation infants on 1998 and 1996 TPN

3. Low glomerular filtration rates increase organic acid

The effect of glomerular filtration on organic acid was examined in arterial blood samples from below 30 weeks gestation infants on 1998 and 1996 TPN. Infants given 1998 TPN had high organic acid in the first few days [Table/Fig-13], which gradually fell as their low GFR improved, as shown in [Table/Fig-17]. Halving GFR roughly doubled organic acid, both on 1998 TPN with normal organic acid and 1996 TPN with organic acid twice as high, as shown in [Table/Fig-18]. This is in keeping with organic acids as a rule being freely filtered through glomeruli and neither reabsorbed nor secreted by renal tubules.

Low glomerular filtration rates increased arterial lactate to a similar extent to organic acid, but with no difference between 1998 and 1996 TPN, as shown in [Table/Fig-19].

4. High rates of protein catabolism increase organic acid

Preterm infants given 1996 TPN had higher arterial blood organic acid [Table/Fig-3] than could be accounted for by the regimen's gluconate and sulphate content [Table/Fig-1]. Protein catabolism, measured as urea excretion, was also significantly higher on 1996 TPN, which provided only 25kcal/g amino acids, compared to 30kcal/g amino acids in 1998 TPN [Table/Fig-16].

To investigate further how protein catabolism affects organic acid, blood urea filtration was used as a guide to urine urea excretion, as shown in [Table/Fig-20]. Fractional urea excretion varied with urine concentration, approaching 100% in dilute urine and below 50% in concentrated urine, as shown in [Table/Fig-21]. Urea filtration therefore overestimated urea excretion when urine was concentrated.

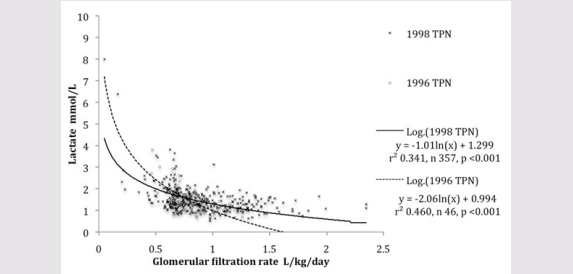
Arterial blood gases taken between 8 and 168 hours of delivery from the 207 preterm infants without infection or congenital abnormalities in the third cohort were compared according to the composition of parenteral fluids given at the time. Their mean birth weight was 2020 ± 673 g and gestation 32.4 ± 2.9 weeks.

Urea filtration, arterial blood organic acid and mineral base deviation, during the first week, are shown in [Table/Fig-22], [Table/Fig-23], [Table/Fig-24] respectively. On 10% glucose, urea filtration gradually declined during the first week as infants adapted to the absence of protein intake [Table/Fig-22]. On 1998 TPN urea filtration remained constant during the first week, as it did on 1996 TPN but at 1.36 times the rate [Table/Fig-22]. Organic acid was similar on 10% glucose and 1998 TPN, but was twice as high on 1996 TPN [Table/Fig-23]. High lactate found in some infants on 10% glucose promptly fell to normal soon after TPN was started but apart from this there were no differences in lactate between 10% glucose, 1996 and 1998 TPN. Mineral base deviation increased on 1996 TPN but changed little on 1998 TPN and 10% glucose [Table/Fig-24]. Organic acid was inversely related to GFR on: 10% glucose OA = -3.64ln(GFR) + 7.369 n 336 r 0.295 p <0.001 1998 TPN OA = -4.22ln(GFR) + 5.802 n 355 r 0.352 p <0.001 1996 TPN OA = -3.05ln(GFR) + 11.53 n 77 r 0.285 p <0.05.

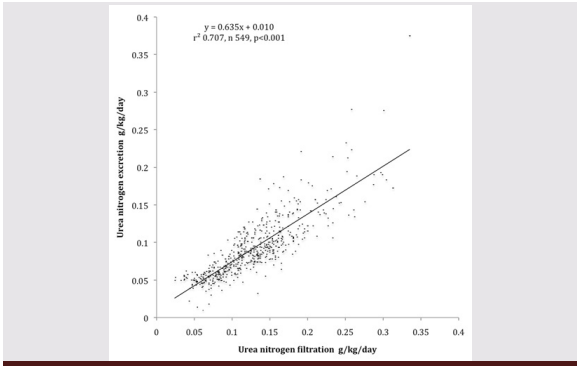
Dexamethasone, given to 7 below 30 weeks gestation infants on 1998 TPN, was started after day 10. In addition to increasing their protein catabolism [Table/Fig-16], dexamethasone also affected their arterial blood gases when compared with gases off dexamethasone from below 30 weeks gestation infants after day 10, as shown in [Table/Fig-25].

On dexamethasone, elevated arterial blood carbonic acid (PCO2) fell, with concomitant reductions in mineral base. Urea filtration increased 1.65 times and organic acid was 1.76 times higher, despite the significantly higher GFR of the samples obtained at an older age on dexamethasone. This increase in organic acid was greater than could be accounted for by the reduction in carbonic acid on dexamethasone [8].

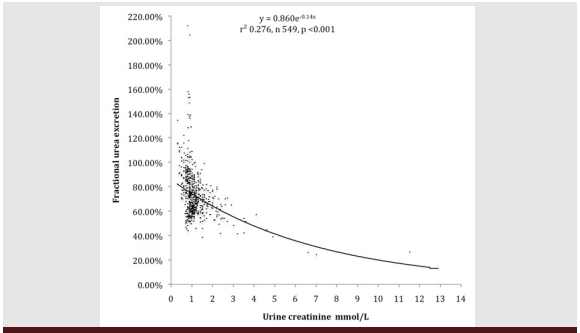
The high rates of protein catabolism produced by 1996 TPN, together with its sulphate and gluconate content, caused organic acid to increase up to around 20 mmol/L during the first few days when GFR was low, as shown in [Table/Fig-18]. Of the 6 blood samples with organic acid around 20mmol/L in [Table/Fig-18], 4 were consistent measurements between 48 and 96 hours in one baby on 1996 TPN. This baby later developed otherwise unexplained periventricular leucomalacia. The other 2 were from two babies on 1998 TPN, but these high levels were inconsistent with low organic acid levels before and afterwards in these infants.



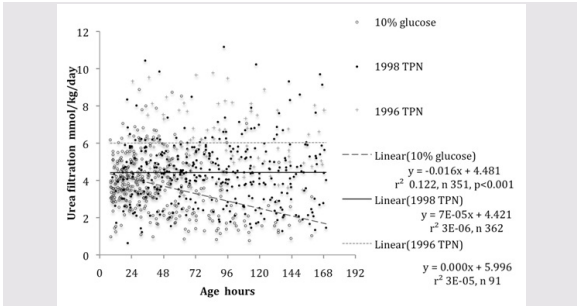
[Table/Fig-19]: Glomerular filtration rate determined arterial lactate in below 30 weeks gestation infants on 1998 and 1996 TPN



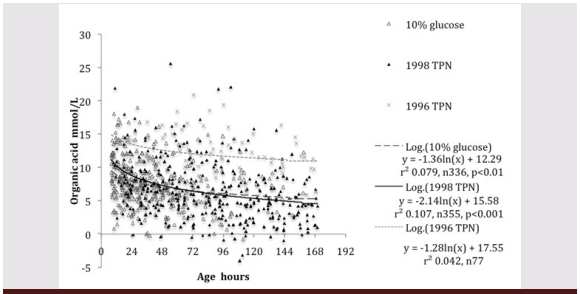
[Table/Fig-20]: Urine urea nitrogen excretion correlated well with the glomerular filtration rate of urea nitrogen measured on blood samples obtained within 6 hours of urine samples



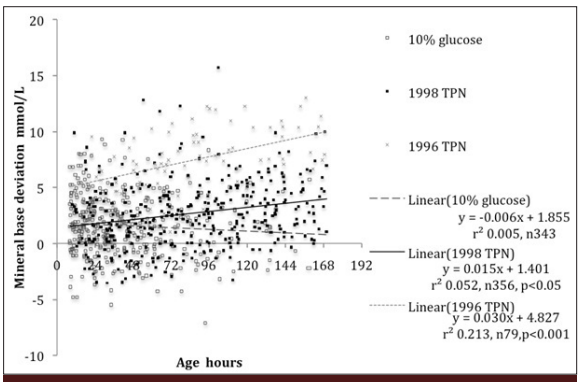
[Table/Fig-21]: Fractional urea excretion decreased as urine became more concentrated



[Table/Fig-22]: The composition of parenteral fluids affected blood urea filtration during the first week in 207 preterm infants



[Table/Fig-23]: The composition of parenteral fluids affectedarterial blood organic acid during the first week in 207 preterm infants



[Table/Fig-24]: The composition of parenteral fluids affected arterial blood mineral base deviation during the first week in 207 preterm infants

Dexamethasone		No	Yes	p %
Infants	n	16	7	
Birth weight	g	952 ± 255	941 ± 240	ns
Gestation	weeks	26.4 ± 1.4	26.3 ± 1.6	ns
Arterial blood samples	n	132	72	
Age after day 9	days	18.4 ± 6.7	22.7 ± 10.2	<0.001
pH		7.340 ± 0.055	7.369 ± 0.047	<0.001
PCO2	mmHg	60.5 ± 12.1	49.1 ± 7.9	<0.001
Base excess	mmol/L	4.3 ± 3.1	1.5 ± 2.2	<0.001
Mineral base	mmol/L	44.7 ± 3.2	42.6 ± 2.6	<0.001
Mineral base deviation	mmol/L	5.8 ± 3.0	3.7 ± 2.5	<0.001
Organic acid	mmol/L	2.5 ± 2.4	4.4 ± 2.9	<0.001
Lactate	mmol/L	1.18 ± 0.31	1.27 ± 0.32	ns
GFR	L/kg/day	1.181 ± 0.306	1.326 ± 0.334	<0.01
Urea filtration	mmol/kg/day	3.496 ± 1.168	5.767 ± 2.644	<0.001

[Table/Fig-25]: The effect of dexamethasone on arterial blood gases obtained from below 30 weeks gestation infants after day 10

DISCUSSION

Thirty years ago the benefits to neonatal care provided by antenatal steroid therapy, surfactant therapy and effective CPAP had yet to be realized. At that time very preterm babies frequently had major acid base and electrolyte disturbances caused by severe respiratory problems. They often needed high positive pressure ventilation compromising circulatory and renal function.

The 1986 study set out to determine how changes in fluid and mineral balance alter blood gas and electrolytes in sick preterm infants requiring parenteral fluids [9]. To achieve this accurate urine collections and recordings of all fluid intakes were made between timed blood and weight measurements on admission, at 48 hours and at 7 days or earlier if milk feeds were started.

The many prolonged urine collections produced a good way to predict creatinine production from gestational age. This allowed mineral balance studies to be made from spot urine samples and records of TPN intake once a baby was stable, forming the basis of the later studies.

Nutrients are delivered at a continuous rate on TPN and spot urine samples show little variation in N, Ca, PO4 and Mg excretion, which can therefore be estimated accurately, but wide variation in Na, K and Cl excretion, which cannot be estimated with the same accuracy.

The composition of TPN affects mineral base and organic acid

Using the theory of net base balance, these mineral balance studies clearly showed that the composition of TPN affects acid base balance in preterm babies in three ways.

First that excess of chloride over sodium in TPN causes mineral acidosis, with low mineral base, whereas excess of sodium over chloride causes mineral alkalosis, with high mineral base.

Second that the presence of organic acids that are not fully metabolized in TPN, such as gluconate and sulphate, cause metabolic acidosis, partly by accumulating in ECF as organic acids and partly by increasing urine mineral base excretion causing mineral acidosis.

Third that a low TPN energy to protein ratio of 25 kcal/g amino acids increases protein catabolism in order to maintain energy requirements, which in turn causes organic acidosis, presumably by increasing sulphate.

Pathogenesis of metabolic acidosis

In these three cohorts of preterm infants, the worst metabolic acidosis developed in infants given 1993 TPN. These babies produced markedly acidic urine with a mean pH of 5.4. At this pH and their mean PCO2 of 41 mmHg, urine bicarbonate averaged 0.3 mmol/L, hardly sufficient to produce "renal bicarbonate wasting".

By measuring all acids and bases, these studies demonstrate that metabolic acidosis in preterm infants is in fact caused by high organic acid, resulting from low GFR in the first 1-2 weeks, exacerbated by less than ideal TPN composition as discussed above. Along with their low GFR these infants have reduced renal hydrogen ion secretion compared to older infants [1, 2], but, provided they receive TPN of the ideal composition, are able to produce the normal renal response of increasing mineral base, and hence base excess to compensate for respiratory acidosis. Other studies of how TPN formulation affects the acid base state of preterm infants have measured only a few of the acid base components and have therefore failed to demonstrate these findings [13].

Some units are currently recommending that TPN for very preterm babies should have a high protein content, over 30g/L amino acids, but with only up to 10% glucose [14]. Such high protein content may make up for its energy deficiency while providing enough for growth. A recent report on infants receiving such TPN described increases in blood urea in proportion to the amount of protein given, but no significant differences in bicarbonate [15]. This does not exclude serious organic acidosis from increased protein catabolism, which on 1996 TPN was completely masked by mineral alkalosis producing normal bicarbonate and base excess. Of particular concern was the subsequent development of otherwise unexplained periventricular leucomalacia in the VLBW infant on 1996 TPN with the highest organic acid, which from 48 to 96 hours was around 20 mmol/L, the level associated with disability following perinatal asphyxia [7].

High protein TPN regimens increase protein catabolism, which increases plasma sulphate and hence organic acid, and also amino acids [16] and probably ammonia [17] to levels that could be neurotoxic. Measurements of all of them are urgently needed to ensure that such high protein TPN regimens are safe. There are virtually no sulphate measurements in preterm infants and no methods readily available. In 1927, as a part of his gigantic contribution to acid base knowledge, Donald van Slyke described an accurate micromethod for measuring sulphate and iodate [18].

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