Effects of perinatal asphyxia and myoglobinuria on development of acute, neonatal renal failure

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SUMMARY Thirty four consecutive neonates with birth asphyxia or respiratory problems were examined in the first week of life to clarify the relation between neonatal myoglobinuria and acute renal failure. Investigations included determination of creatinine clearance, fractional sodium excretion, and N-acetyl-β-D glucosaminidase index as an indicator of tubular injury. The infants' gestational ages ranged from 29 to 41 weeks (mean 36 weeks). Fifteen infants did not have myoglobinuria on the first day of life (group A); myoglobinuria was mild in eight infants (group B) and severe in eleven (group C). Two infants in group B and seven in group C developed acute renal failure (47%). Ten infants in group C (91%) had severe asphyxia, five of whom (45%) also suffered neonatal seizures and intracranial haemorrhage. We suggest that myoglobin derived from muscle breakdown in asphyxiated infants may lead to acute renal failure secondary to a reduction in renal blood flow, or to tubular damage.

Myoglobinuria has been associated with acute renal failure.[1][2] Although the exact mechanism of the renal damage is not well established, nephrotoxicity, tubular obstruction,[1] and alterations in renal perfusion and vascular resistance[2] have been suggested as pathogenic mechanisms. Rhabdomyolysis and subsequent myoglobinuria, however, have rarely been reported in neonates.[3] Massive breakdown of muscle tissue can lead to myoglobinuria, which may result in acute renal failure.[2] Fetal and neonatal asphyxia have been considered the main causes of transient renal impairment or acute renal failure in neonates.[4] Asphyxia is known to be the major cause of reduced renal blood flow because of increased renal vascular resistance, and if it is prolonged, the renal parenchyma are frequently damaged. Recently, Haftel *et al*[3] reported the relation between myoglobinuria and acute renal failure in one newborn infant and suggested that myoglobinuric renal failure was associated with anoxia and sepsis. The present study was undertaken to clarify the association between myoglobinuria and acute renal failure in asphyxiated neonates.

Patients and methods

Renal function and urine myoglobin values were investigated in 34 newborn infants—21 asphyxiated infants, seven with respiratory distress syndrome, and six with transient tachypnoea of the newborn. None had a congenital heart disease or congenital renal abnormality. Gestational age, assessed according to Dubowitz *et al*,[5] ranged from 29 to 41 weeks and birthweight ranged from 1480 to 3720 g. Informed consent was obtained from all the parents. All infants were cared for in the neonatal intensive care unit at this hospital according to established protocol.

Eleven infants, five with respiratory distress syndrome and six with asphyxia, were ventilated with the Bournes BP 2001 respirator; and 10, six with transient tachypnoea of the newborn, two with respiratory distress syndrome, and two with asphyxia, required continuous positive airway pressure. Fractional inspired oxygen was adjusted to maintain a transcutaneous oxygen tension of between 6·6 and 10·6 kPa. Continuous positive airway pressure was applied at a pressure of 3 to 4 cm H_2O, assisted ventilation with 3 to 5 cm H_2O pressure, and intermittent mandatory ventilation was given 20 to 40 times per minute. Six asphyxiated infants, four with respiratory distress syndrome and two with transient tachypnoea, underwent phototherapy and had their serum haematocrit and urine volume monitored every eight hours to ensure adequate hydration. The blood pressure of all infants was measured in the right arm by Doppler ultrasound. There were no severe episodes of

hypotension or hypertension. No infants were given any nephrotoxic agents such as aminoglycoside, tolazoline, or indomethacin. The infants were given a 5% glucose solution until the occurrence of diuresis: it was infused at a rate of 50 to 60 ml/kg on day 1, 60 to 70 ml/kg on day 2, and 70 to 80 ml/kg on day 3. Urine myoglobin was tested on the first and third day of life in collected urine samples. Initial laboratory tests—serum creatinine concentration, white blood count, creatinine phosphokinase activity, and blood gas analysis—were made on hospital admission. Kidney function tests, including creatinine clearance and fractional sodium excretion, and determination of urinary N-acetyl-β-D glucosaminidase as an indicator of tubular injury, were undertaken at ages 1, 2, 3, 5, and 7 days. Urine was collected for 18 to 24 hours each day by using a urine collecting bag (Atom). A blood sample was obtained at the midpoint of the creatinine clearance study. Serum was separated and frozen until analysed within two weeks of collection, during which period no change was noted in the above parameters.

Myoglobin was analysed by radioimmunoassay,[6] and was expressed as the myoglobin concentration (ng/ml) and the myoglobin to urinary creatinine ratio (mg/gCr). Normal concentrations of urine myoglobin, determined in 10 healthy newborn infants, were always below 5 ng/ml or 0·03 mg/gCr.

Serum and urine creatinine concentrations were analysed by using a creatinine kinetic method[7] (Wako). Urine N-acetyl-β-D glucosaminidase activity was measured by spectrophotometric assay (Shionogi).[8] Flamephotometry was used to analyse urinary and serum sodium concentrations.

The diagnosis of renal failure was made when urine output was less than 1 ml/kg per hour over a period of 30 hours, and the serum creatinine was greater than 1·8 mg/dl (159 μmol/l). Duration of oliguria was defined as the period during which urine output was less than 1 ml/kg per hour. Creatinine clearance was expressed as the rate to body weight (ml/min per kg) as reported by Coulthard *et al.*[9] N-acetyl-β-D glucosaminidase activity was expressed as the NAG index (U/gCr).

Comparisons were made using the Ryan method and statistical significance was set at $P < 0.05$. Data are presented as mean (SD).

Results

Urinary myoglobin was positive in 19 of 34 cases on the first day of life. It ranged from 10 to 4300 ng/ml or 0·05 to 71·6 mg/gCr. Urine myoglobin lessened considerably on day 3, however, and was positive in only eight infants ranging from 7 to 120 ng/ml or

0·05 to 26·6 mg/gCr. Because this range of values was so wide the 34 infants were divided into three groups. Group B comprised infants with mild myoglobinuria (urine myoglobin ranging from 5 to 500 ng/ml, mean (SD) 100 (115) ng/ml, or from 0·05 to 0·7 mg/gCr mean (SD) 0·29 (0·26)) and Group C infants with severe myoglobinuria (urine myoglobin over 500 ng/ml, mean (SD) 2120 (1618) or over 1·0 mg/gCr, mean (SD) 20·37 (28·24)). Fifteen infants in whom urine myoglobin was negative served as the control (group A).

Table 1 gives the clinical findings and diagnoses in the three groups of infants. Ten of 11 (91%) infants in group C were severely asphyxiated compared with two of eight in group B (25%). Five (45%) patients in group C had neonatal seizures and intracranial haemorrhages compared with two cases of neonatal seizures and one of intracranial haemorrhage in eight infants in group B. Renal failure occurred in two of eight children in group B (25%) and in seven of 11 in group C (64%).

Gestational age, birthweight, Apgar scores at one and five minutes, serum creatinine, white blood count, creatine phosphokinase, blood gas analysis, duration of oliguria, and urine myoglobin values on admission are given in Table 2. The Apgar scores at one minute were lower in groups B and C than in group A. Five minute Apgar scores in groups B and C were also lower than those of group A, and there was a significant difference between groups B and C. White cell count, creatine phosphokinase, and urine myoglobin were also significantly higher in group C than in groups B and A. Base excess was lower in group C than in groups B and A. Duration of oliguria was longer in group C than in groups B and A.

Serum creatinine, creatinine clearance, fractional sodium excretion, and NAG index during the first week are given in Table 3. In week 1 serum creatinine, fractional sodium excretion, and NAG index were higher in group C than in group A, and

Table 1 *Clinical findings and diagnoses in infants without myoglobinuria (group A) with mild myoglobinuria (group B), and with severe myoglobinuria (group C)*

	Group A (n=15)	Group B (n=8)	Group C (n=11)
Asphyxia			
Mild (≥7 at 5 min Apgar score)	2	6	1
Severe (≤6 at 5 min Apgar score)	0	2	10
Neonatal seizure	0	2	5
Intracranial haemorrhage	0	1	5
Meconium aspiration syndrome	1	2	2
Respiratory distress syndrome	3	3	1
Transient tachypnoea of newborn	6	0	0
Acute renal failure	0	2	7

Table 2   *Clinical details and laboratory findings on hospital admission in infants without myoglobin (group A), those with mild myoglobinuria (group B), and those with severe myoglobinuria (group C). Values mean (SD)*

	Group A (n=15)	Group B (n=8)	Group C (n=11)
Boys:girls	8:7	4:4	6:5
Gestational age (wks)	36 (3·2)	35·8 (4·3)	38 (3·8)
Birthweight (g)	2372 (434)	2508 (803)	2634 (614)
Apgar score:			
1 min	7·6 (2·1)	4 (2·1)***	3·2 (2·1)***
5 mins	8·3 (1)	7 (0·7)*	4·5 (2·1)***
Serum creatinine (μmol/l)	69 (10·8)	81·8 (15·2)	100·5 (22·1)***
White blood count (×10⁹/l)	13·8 (3·8)	16·5 (4·1)	24·1 (5·5)***
Creatine phosphokinase (IU/l)	323 (98)	810 (518)	3008 (2425)***
Blood gas analysis			
pH	7·25 (0·07)	7·26 (0·1)	7·23 (0·11)
Base excess (mmol/l)	−5·1 (2·9)	−4·28 (5·3)	−10·9 (4·3)**
Duration of oliguria (hrs)	15·3 (5)	18 (6·7)	31·2 (16·4)***
Urine myoglobin on day 1 (ng/ml)	3·7 (0·8)	122·5 (113·7)	2034·5 (476·1)***

*P<0·05; **P<0·002; ***P<0·001.
Significance of group B v group C—5 min Apgar score P<0·002; white blood count P<0·002; creatine phosphokinase P<0·005; base excess P<0·005; oliguria P<0·02; urine myoglobin P<0·001.

Table 3   *Renal function during first week of life in infants without myoglobinuria (group A), those with mild myoglobinuria (group B), and those with severe myoglobinuria (group C). Values mean (SD)*

	Group A	Group B	Group C
Serum creatinine (μmol/l)			
1st day	69·0 (10·8)	81·8 (15·2)	100·5 (22·1)*****
3rd day	93·8 (18·8)	98·4 (14·3)	139·9 (34·7)*****
7th day	58·9 (11·0)	75·2 (10·8)	124·7 (124·4)*
GFR(Ccr) (ml/min/kg)			
3rd day	0·67 (0·18)	0·60 (0·25)	0·35 (0·21)*****
5th day	0·73 (0·16)	0·63 (0·14)	0·49 (0·24)***
7th day	0·91 (0·14)	0·83 (0·20)	0·68 (0·34)*
FENa (%)			
3rd day	2·3 (1·6)	2·9 (1·6)	1·31 (10·0)*****
5th day	1·8 (1·6)	3·3 (2·2)	14·7 (22·1)*
7th day	1·2 (1·1)	1·6 (1·1)	12·4 (22·3)*
NAG index (NAG/gCr)			
3rd day	58 (43)	102 (87)	236 (200)****
5th day	58 (27)	76 (93)	241 (298)**
7th day	44 (26)	83 (75)	168 (196)**
Urine output (ml/kg/hr)			
1st day	0·70 (0·33)	0·67 (0·47)	0·39 (0·29)*
2nd day	2·50 (1·30)	2·0 (0·95)	1·53 (0·96)*
3rd day	3·89 (1·01)	3·19 (0·85)	2·84 (1·36)*
Water intake (ml/kg/day)			
1st day	60·9 (9·8)	52·6 (6·6)*	52·2 (4·3)**
2nd day	70·1 (12·1)	62·6 (7·6)	60·5 (8·1)*
3rd day	79·4 (13·4)	77·0 (16·1)	74·3 (14·2)

*P<0·05; **P<0·02; ***P<0·01; ****P<0·002; *****P<0·001.
Significance of group B v group C—1st day serum creatinine P<0·05; 3rd day serum creatinine P<0·002; 3rd day FENa P<0·002; 3rd day GFR P<0·02; 3rd day NAG P<0·05; GFR=glomerular filtration rate; FENa=fractional sodium excretion; NAG index=N-acetyl-β-D glucosaminidase (U/gCr).

creatinine clearance was lower in group C than in group A. Serum creatinine concentrations in group C during days 1 and 3 were higher than those of group B. Creatinine clearance in group C on day 3 was lower than that of group B. Fractional sodium excretion and NAG index on day 3 were higher in group C than in group B. The urine output in group C was lower than that of group A on days 1 to 3,

while fluid intake in group C was lower than that of group A during the same period.

Discussion

This study shows that there is a strong association between myoglobinuria, birth asphyxia, and the development of acute renal failure. In every case,

myoglobinuria was found in asphyxiated infants, and its severity was related to the severity of asphyxia. Serum creatinine and creatine phosphokinase values were significantly high in infants with severe myoglobinuria. Since severe asphyxia is known to cause tissue damage as a consequence of tissue hypoxia or hypoperfusion, the most likely explanation for the above association is that muscle tissue is also similarly injured with a resultant release of creatinine and creatine phosphokinase. Although the white cell count was also high in group C, it seems likely that this was associated with the severity of asphyxia because there was no relation between the white cell count and infection. In addition, the infants in group C had complications such as neonatal seizures and intracranial haemorrhages, which seemed to exacerbate myoglobinuria as a result of decreasing peripheral hypoperfusion. Since creatinine clearance, fractional sodium excretion, and NAG index, used as parameters of renal function and renal tubular injury were also abnormal in group C infants, decreased glomerular filtrating ability and tubular cell damage seemed to occur in these infants. All cases of acute renal failure were also seen in the myoglobinuric infants. Haftel et al,[3] in a case report, showed that myoglobin pigment is itself toxic to the kidneys, causing a spectrum of diseases from minimal tubular damage to fulminant severe acute tubular necrosis. The heme pigments are non-nephrotoxic in themselves,[10-12] but they may act as nephrotoxic agents, producing renal damage through an alteration in renal vascular resistance or tubular obstruction when renal ischaemia or dehydration coexist. Oken et al[13] speculated that in the rat model of glycerol-induced acute renal failure, myoglobinuric acute renal failure might be marked by a sharp reduction in the renal blood flow because there was no increase in the intratubular pressure. Recently, Howard et al[14] described 10 patients with acute haemoglobinuric-myoglobinuric renal failure in whom fractional sodium excretion was less than 1% in the oliguric phase, and concluded that redistribution of renal blood flow was an important factor in the development of renal failure. The renin-angiotensin system, alterations in prostaglandin production, and arginine vasopressin have been associated with renal vasoconstriction in acute myoglobinuric renal failure. In the current study, however, specific investigations were not undertaken into these aspects, and will be the subject of future study.

Nephrotoxicity of myoglobin has been evaluated in numerous studies[10-12 15 16] which have shown that volume depletion and acidosis are necessary for myoglobinuric renal failure to occur. Chedru et al[17]

showed that long term saline loading before the induction of myohaemoglobinuria seemed to prevent acute renal failure by abolishing sustained renal cortical ischaemia. Neonates, however, are not well equipped to maintain water and electrolyte balance. Since the fluid overload may increase the risk of hyponatraemia leading to brain oedema and bronchopulmonary dysplasia in infants with respiratory distress syndrome,[18] it may be difficult to increase the water intake to prevent the occurrence of acute myoglobinuric renal failure. Asphyxia in neonates is known to be the major cause of reduced renal blood flow.[19-22] Since these infants were frequently found to have myoglobinuria in our study, it may act as a factor which further reduces the renal blood flow. From these observations, it seems likely that acute renal failure may occur more easily in asphyxiated infants with myoglobinuria than in those without myoglobinuria.

On the basis of these results, it is interesting to speculate that in severely asphyxiated neonates, myoglobinuria also serves as one of the factors in acute renal failure when combined with the direct action of asphyxia.

Further studies are necessary to clarify the causal relation between myoglobinuria and acute renal failure, as such a combination is frequently observed in asphyxiated infants.

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