Plasma alkaline phosphatase activity in rickets of prematurity

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SUMMARY Plasma alkaline phosphatase activity was measured in 349 infants aged between 5 and 10 days to establish a normal range for different gestational ages. Significant differences were observed between term and preterm infants, the highest values being associated with the shortest lengths of gestation. Plasma calcium, phosphate, and alkaline phosphatase activity were measured sequentially in 51 preterm infants <1500 g at birth. A significant correlation was found between raised plasma alkaline phosphatase activity and radiological changes of osteoporosis, metaphyseal change, and periosteal reaction. Plasma alkaline phosphatase appears to be of value in screening for and monitoring rickets of prematurity.

The predisposition of the preterm infant to rickets has been known for many years,¹ and is being increasingly found among extremely low birthweight infants.²⁻⁵ The early detection and treatment of rickets may have important implications for linear growth of the preterm infant.⁶ Florid changes in the x-ray film occur late in the disease and although photonabsorptiometry⁷ can detect early demineralisation, the technique is not widely available. We report the value of plasma alkaline phosphatase activity as a sensitive and early indicator of rickets of prematurity.

Patients and methods

To establish a normal range for infants of differing gestational ages, plasma alkaline phosphatase activity was measured in 349 infants aged between 5 and 10 days. This timing was chosen as after parturition the placental isoenzyme disappears from plasma with a half-life of 3 or 4 days.⁸ The gestational age of these infants ranged from 26 to 40 weeks. Plasma alkaline phosphatase was measured by the method of Morgenstern *et al.*⁹

Fifty-one preterm infants weighing less than 1500 g at birth who were admitted to the regional intensive care unit during the period October 1978 to October 1980 were studied sequentially. Feeding was by the enteral or parenteral route. Plasma calcium, phosphate, and alkaline phosphatase levels were measured weekly during their stay in hospital, and subsequently at 2-4 weekly intervals as outpatients until

the alkaline phosphatase activity was below 500 U/l. Plasma alkaline phosphatase activity greater than 500 U/l was taken as an indication that isoenzyme studies were necessary.⁸

Radiological examination was not performed routinely, but 42 babies had x-ray films taken on clinical indication. On completion of the study all x-ray films were assessed independently of clinical and biochemical data by a paediatric radiologist. Generally chest x-ray films were analysed, but many infants had abdominal x-rays which included the spine and pelvis on the film, while x-ray films of the skull and wrist were examined if available. The radiographic features analysed were bone mineralisation, the presence or absence of metaphyseal changes, and periosteal reaction. The degree of bone demineralisation was arbitrarily graded 1-3 according to severity. Osteoporosis was detected most readily in the ribs and scapulae. Metaphyseal changes; cupping, fraying, and expansion were noted in the humeri. The biochemical and radiological data were correlated statistically by Kendall's rank correlation for osteoporosis, by Wilcoxon's rank sum for metaphyseal change, and by Fisher's exact test for periosteal reaction.

During the study 400 IU of vitamin D2 a day were given to enterally-fed infants, and 100 IU/kg a day intravenously to parenterally-fed infants. If the plasma alkaline phosphatase activity exceeded 500 U/l, the vitamin D2 supplement was increased to 1000 IU a day. At values above 1000 U/l vitamin D2 was replaced by an oral preparation of 1, α -

hydroxycholecalciferol (1 α) 0.05 μ g/kg a day. In the event of failure to respond to the latter 1,25-dihydroxycholecalciferol (Rocaltrol) was prescribed in a dosage of 0.05 μ g/kg a day.

Results

The variation in plasma alkaline phosphatase activity in relation to gestational age at birth in 349 infants is shown in Table 1. Significant differences (Student's *t* test) in plasma alkaline phosphatase activity were observed between term and preterm infants.

The 51 infants studied sequentially were divided into 4 groups according to their maximum plasma alkaline phosphatase activity during the period of the study (Table 2). In patients with plasma alkaline phosphatase values greater than 500 U/l (71%), including infants with cholestasis, the predominant isoenzyme was of bone origin. The groups of infants

with higher plasma alkaline phosphatase values (groups 3 and 4) were of lower mean birthweight and gestation, and had longer periods of parenteral nutrition than infants with lower peak alkaline phosphatase values (groups 1 and 2). No significant differences were noted in plasma phosphate levels, and plasma calcium was significantly lower only in the most severely affected infants (group 4).

In infants developing radiological rickets the highest and most sustained maximal plasma alkaline phosphatase values occurred in the more severe cases. However, the maximal value and the period of increased plasma alkaline phosphatase values were modified by treatment. Of 7 infants with peak alkaline phosphatase values greater than 1000 U/l treated with 1, α -hydroxycholecalciferol (mean duration 13 days), 6 showed a satisfactory response with a decline in plasma alkaline phosphatase activity from a mean value of 1221 U/l $\pm$ 125 (before treatment) to 751 U/l $\pm$ 83.3 (after treatment). In the 7th infant,

Table 1 *Plasma alkaline phosphatase variations in relation to gestational age*

Alkaline phosphatase activity (U/l)	Number in group	Gestation (weeks)	Gestation (weeks)					
			26+27	28+29	30+31	32+33	34+35	36
319.54 $\pm$ 142.31	13	26+27		NS	NS	<0.02	<0.001	<0.001
291.81 $\pm$ 86.49	26	28+29			NS	NS	<0.001	<0.001
281.15 $\pm$ 84.49	46	30+31				NS	<0.001	<0.001
253.98 $\pm$ 71.85	53	32+33					NS	<0.001
236.30 $\pm$ 61.78	103	34+35						<0.01
207.16 $\pm$ 59.52	48	36						<0.001
164.1 $\pm$ 67.8	60	38-41						<0.001
	349							

Alkaline phosphatase activity expressed as mean $\pm$ 1 SD.

Table 2 *Classification of 51 preterm <1500 g infants according to maximum plasma alkaline phosphatase activity*

	Group				P value
	1 (n=4)	2 (n=11)	3 (n=29)	4 (n=7)	
Peak alkaline phosphatase (U/l)	<300	300-500	500-1000	>1000	2 v 4
Birthweight (kg)	1.41 $\pm$ 0.09	1.34 $\pm$ 0.13	1.26 $\pm$ 0.15	1.13 $\pm$ 0.25	<0.05 2 v 3
Gestation (weeks)	31.3 $\pm$ 1.3	32.2 $\pm$ 2.0	29.9 $\pm$ 2.0	29.9 $\pm$ 1.9	<0.01 2 v 4 <0.005
Plasma calcium (mmol/l)	2.33 $\pm$ 0.06	2.45 $\pm$ 0.15	2.39 $\pm$ 0.24	2.02 $\pm$ 0.35	3 v 4 <0.01
Plasma phosphate (mmol/l)	2.10 $\pm$ 0.32	2.01 $\pm$ 0.32	1.88 $\pm$ 0.34	1.71 $\pm$ 0.37	NS 2 v 4 <0.05
Day that maximum alkaline phosphatase reached	20 $\pm$ 15	28 $\pm$ 12	51 $\pm$ 33	60 $\pm$ 38	2 v 3 <0.001 1 v 4 <0.05
Duration of parenteral nutrition (days)	7.5 $\pm$ 5.8	7.8 $\pm$ 4.1	29.4 $\pm$ 20.5	35.1 $\pm$ 18.4	2 v 3 <0.001

Conversion: SI to traditional units—calcium: 1 mmol/l $\approx$ 4 mg/100 ml; phosphate 1 mmol/l $\approx$ 3.1 mg/100 ml.

Table 3 Correlations between plasma alkaline phosphatase and radiological bone changes in preterm low birthweight (<1500 g) infants

Peak alkaline phosphatase (U/l)	<300 (n=4)	300-500 (n=11)	500-1000 (n=29)	>1000 (n=7)	P value
Mineralisation					
Normal mineralisation	2	8	1	0	
?Osteoporosis/normal	1	1	1	0	
Osteoporosis					
Grade 1	0	1	7	0	
Grade 2	0	0	10	4	<0.001
Grade 3	0	0	3	3	
Metaphyseal change	0	0	4	4	<0.01
Periosteal reaction	0	0	0	2	<0.024
No x-ray film	1	1	7	0	

who had appreciable cholestasis, the plasma alkaline phosphatase activity continued to rise despite 1, α -hydroxycholecalciferol. After 21 days of treatment with 1,25-dihydroxycholecalciferol, the plasma alkaline phosphatase activity declined from 1940 to 995 U/l.

Correlation between biochemical and radiological findings are shown in Table 3. The degree of osteoporosis, presence of metaphyseal changes, and periosteal reaction were all significantly related to maximum alkaline phosphatase activity. All patients with metaphyseal changes had maximum alkaline phosphatase activity greater than 750 U/l. Periosteal reactions were seen only if alkaline phosphatase activity exceeded 1000 U/l. All infants with abnormal wrist x-ray films had changes in the humeri. Maximal peak alkaline phosphatase activity generally preceded maximum radiological change by 2 to 4 weeks.

Discussion

Variations in plasma alkaline phosphatase activity with gestational age were found in a group of 349 infants aged between 5 and 10 days, with a downwards trend between 26 and 40 weeks' gestation. Significant differences were noted between term and preterm infants ($P<0.001$). This variation may reflect the measurement of different alkaline phosphatase isoenzyme activities at different gestational ages.

In the 51 infants studied sequentially, the radiological changes of osteoporosis, metaphyseal change, and periosteal reaction were all significantly related to peak alkaline phosphatase activity.

The classical changes which occur in children with rickets have been well described.¹⁰ The main features are rarefaction, cupping, and fraying of the metaphyses together with loss of the provisional zone of calcification. Periosteal reactions and fractures through demineralised bone may occur. In preterm infants a diffuse demineralisation of the

skeleton particularly affecting the ribs, spine, skull, and scapula often with super added fractures has been noted.^{11 12} In the infants studied radiological changes of osteoporosis correlated with biochemical evidence of rickets. In contrast Eek *et al.*¹³ regarded diffuse osteoporosis, resolving without treatment as a normal phenomenon in the preterm infant. Periosteal reaction has been described in preterm infants during both active³ and healing¹⁰ rickets. However Malmberg¹⁴ described periosteal reaction unassociated with rickets in preterm infants. Periosteal reactions were noted in only 2 of our patients, both of whom had peak alkaline phosphatase levels in excess of 1000 U/l.

Plasma calcium and phosphate values did not appear to be good indicators of rickets in these infants. Plasma calcium fell significantly only in the most severely affected group, and no significant differences were noted in plasma phosphate values. Plasma calcium values are likely to be particularly misleading in parenterally-fed infants.

Inadequate calcium input is one of the factors implicated in the aetiology of rickets of prematurity. Fetal calcium accumulation during the last trimester is high.¹⁵ Pre-eclampsia which impairs placental calcium transfer¹⁶ is noted more often among mothers of preterm infants with rickets.¹⁷ Rapid postnatal growth demands a large calcium input which is not met by breast milk or most modified milk formulae. The parenteral regimen used during the period of the study provided 1-1.4 mmol/kg per 24 hours of calcium at a fluid intake of 150-200 ml/kg a day. This contrasts with an intake of 1.6-2.2 mmol/kg a day on a similar intake of a modified formulae but the latter would be incompletely absorbed.¹⁵ Thus postnatally neither parenteral nor enteral feeding provides sufficient calcium to allow intrauterine accretion rates of calcium to be achieved. Calcium supplementation of the preterm infant has been shown to improve bone mineralisation.⁷

Disordered vitamin D metabolism has also been

implicated in the development of rickets in preterm infants. Preterm infants seem unable to increase plasma 25-hydroxycholecalciferol levels until 38–42 weeks' postconceptual age despite intravenous supplementation with vitamin D.¹⁸ Thus impaired 25-hydroxylation of vitamin D may contribute to the development of rickets. Low values of 25-hydroxycholecalciferol have previously been noted in patients with cholestasis. Our cholestatic infant had a value of 2.1 nmol (normal 10–20 nmol) and his rickets did not respond until 1, 25-dihydroxycholecalciferol was given.

It has also been considered that activation of vitamin D to 1, 25-dihydroxycholecalciferol in the immature kidney may be impaired and thus lead to the development of rickets. In 6 of the 7 infants in whom alkaline phosphatase activity exceeded 1000 U/l a decrease in alkaline phosphatase activity and radiological healing of rickets occurred only after the introduction of 1, α -hydroxycholecalciferol.

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