ACQUIRED BONE DISEASE IN SMALL PRETERM INFANTS — A POTENTIALLY FATAL DISORDER

by

J. F. T. GLASGOW
P. S. THOMAS
B. G. McCLURE

BETTER neonatal care has improved survival of infants less than 1.0 kg birth weight (Stewart et al, 1977). One of the challenges in managing very low birth weight (VLBW) babies is that of providing optimal nutrition (Shaw, 1973), given the constraints upon absorption (Grand, Watkins and Torti, 1976) and subsequent metabolism (Rosenberg and Scriver, 1974). Our aim is to highlight the development of bone disease in these infants, despite prophylaxis with vitamins, including Vitamin D and other essential trace elements.

PATIENTS AND METHODS

Clinical details

Fourteen VLBW infants have been recognised with acquired bone disease, 13 since 1974 and one in 1969. Five mothers had ante-partum haemorrhage or severe pre-eclampsia, four had multiple pregnancy (one triplets) and six others went spontaneously into premature labour. Gestational maturity at birth varied from 27 to 32 weeks. This was measured according to Dubowitz, Dubowitz and Goldberg (1970) and confirmed in eight infants by radiological assessment of the degree of calcification of the molar teeth cusps (Kuhns, Sherman and Poznanski, 1972). Birth weights were 0.64 to 1.45 kg. Six infants fell belowe the tenth percentile for gestational maturity (Gairdner and Pearson, 1971). Two babies were asphyxiated at birth, while three others developed idiopathic respiratory distress syndrome. Patient 8 had severe rhesus disease.

Each of the infants had received vitamin supplements from the first week of life. Patients 3 and 6 were given Vitamin D 10 μg, patient 2, 20μg and the rest, 30 μg daily. Patients 2, 3 and 6 were nourished on evaporated milk (Carnation), patients 8 and 13 on SMA Gold Cap and the rest on Cow & Gate Premium. The copper content of reconstituted feeds was 35-40 μg/dl and the daily intake about 50-60 μg/kg therefore. None of the expectant mothers had been nutritionally deprived and although no formal dietary assesment was undertaken, all except the mother of patient 4 received dietetic counselling.

This paper was presented at the Royal Society of Medicine (Paediatric Section) meeting in the Royal Belfast Hospital for Sick Children on Saturday, 1st July, 1978.

Bone disease was recognised between 5 and 12 weeks after birth. In eight infants this took the form of gradually increasing dyspnoea with sub-costal recession and was associated with apnoeic episodes, cyanosis and bradycardia. In patient 13 this appeared to be associated with a viral bronchiolitis. The lighter the infant the more severe their illness and the two who died were lightest of all. Six others were asymptomatic and were detected between 1975 and 1977, because of increased surveillance of VLBW infants.

Clinical features of rickets were few. An abnormally large or full anterior fontanelle was noted in two and in only one of these was there obvious expansion of the wrists. None had evidence of a rachitic rosary, although early radiological changes of rickets were already present.

LABORATORY RESULTS

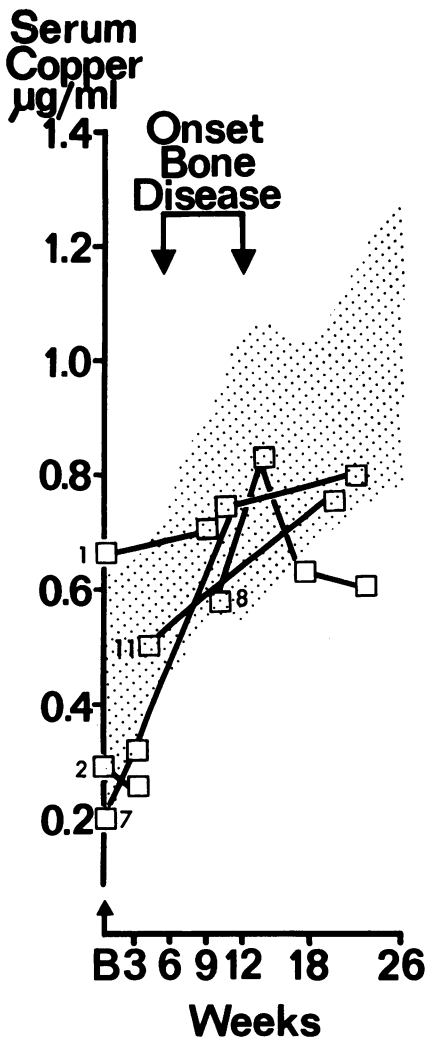
Tests were carried out at the time radiological bone disease was found. Serum alkaline phosphatase activity showed a consistent increase (Table 1). Serum

TABLE 1: BIOCHEMICAL DETAILS

Patient	*Total Maturity weeks* [a]	*Alkaline Phosphatase u/l* [b]	*Calcium mmol/l*	*Magnesium mmol/l*	*Phosphate mmol/l*	*25-OHD$_3$* [c] *ng/ml*	*Urinary aminoacids*
1	35	297	2.48	0.74	2.50	30.05	Threonine increased
2	36	47 KAU[d]	1.40	1.13	2.40	19.5	Tyrosine increased
3	37	55 KAU	1.80	0.57	1.30	ND[e]	Generalised increase
4	38	235	2.29	0.96	2.17	ND	Threonine increased
5	35	41 KAU	2.00	0.93	2.50	ND	Threonine increased
6	44	60 KAU	1.68	0.85	1.20	ND	Generalised increase
7	37	67 KAU	2.24	0.80	2.30	ND	Threonine increased
8	37	700	1.67	ND	1.70	16.5	Generalised increase
9	34	43 KAU	2.10	0.73	2.20	ND	Normal
10	35	338	2.39	ND	2.02	ND	Normal
11	38	250	2.25	ND	1.70	34.5	Normal
12	36	230	2.26	0.72	1.60	ND	ND
13	39	438	2.19	ND	1.80	17.5	Normal
14	37	295	1.85	0.75	1.80	20.0	Normal

a Gestational maturity plus post-natal age at onset of bone disease.
b Alkaline phosphatase – normal range at this age 56-190 u/l.
c 25-OHD$_3$ – lower limits in adults for January, March, June and September are 16, 12, 18 and 25 ng/ml, respectively.
d King Armstrong Units.
e Not determined.

calcium and inorganic phosphate levels were variable. Three infants had biochemical evidence of secondary hyperparathyroidism. Patient 8 had a serum parathormone concentration of 1.8 μg/l (range in normal adults 0.3-0.73 μg/l); this decreased to normal over four weeks following treatment with 0.1 μg/kg/day of 1 α-hydroxyvitamin D (1α-OHD_3) (Glasgow and Reid, 1977). Two others had hypophosphataemia and generalised aminoaciduria which were corrected



Serial copper determinations in the sera of five infants shown in relation to the range (M ± SD) for normal preterm infants (stippled) of mean gestational age 33 weeks.

following larger doses of Vitamin D. Dr. Angela Fairney, St. Mary's Hospital Medical School, London, using a modification of the method of Haddad and Chyu (Haddad and Chyu, 1971) kindly measured 25-hydroxyvitamin D_3 (25-OHD_3) for us. The plasma concentration in four babies were within or just above the normal range in adults according to season.

Serum copper concentrations were measured by atomic absorption spectrophotometry (McMaster, 1977). The levels are shown in relation to normal values for preterm infants whose gestational age at birth was 33 weeks (Halliday, 1977). In four infants the copper levels fell within the normal range at the time bone disease was diagnosed (Fig). Although none of the infants was frankly icteric, four were subclinically jaundiced, three of whom had a mildly elevated direct-reacting bilirubin level. Plasma proteins, full blood count and red cell morphology were normal in each case.

RADIOLOGY

Marked skeletal demineralisation affecting the skull and axial skeleton in particular, was present in all babies (Table 2). In the skull the fine details of the inner ear bones were clearly etched and the sutures poorly defined. More pronounced skeletal changes were found in those with respiratory symptoms. Four of these infants had a fine layer of periosteal new bone along the inferior aspect of the mandible, in two of whom this extended onto the ascending rami (Thomas and Glasgow, 1978).

Pulmonary abnormalities reflecting symptomatic bone disease were present in eight infants. In seven of these the lungs were markedly over-inflated with linear strands radiating out from the hili and small patches of consolidation and collapse. The pattern was such that an initial diagnosis of Mikity-Wilson syndrome was suspected and this was supported in patient 3 by the presence of apparent cystic changes in the right lower zone (Glasgow and Thomas, 1977). Patients 7, 9 and 14 with asymptomatic bone disease, had cardiomegaly and pulmonary plethora due to intracardiac left to right shunts.

The findings in the long bones were less pronounced, although in the majority rachitic changes of variable degree were present at the wrist. These consisted of demineralisation, loss of clarity and slight cupping of the metaphyses of the lower radius and ulna. In only four infants was there definite metaphyseal splaying. Flaring of the anterior ends of the ribs was present in six children. Rickets were seen at the inferior scapular angles in patients 3 and 6.

Fractures were present in five patients; infants 5, 6, and 14 had several fractures of the lateral arcs of the ribs on the left side, and patient 3 had multiple rib and long bone fractures in various stages of healing.

TABLE 2: RADIOLOGICAL FINDINGS IN RELATION TO AGE OF ONSET OF DISEASE

Patient	*Age at onset bone disease (wk)*	*Skeletal changes*	*Lung changes*
1	6	D,R. P	O,L,C,
2	6	D,R, P	O,L,C,
3	9	D,R,F,P	O,L,C,Cy
4	9	D,R	O,L,C,
5	5	D,R,F	O,L,C,
6	12	D,R,F	C
7	7	D,R	
8	10	D,R	O,L,C,
9	6	D,R	
10	5	D,R, P	
11	10	D,R,F	
12	7	D,R	
13	10	D,R	O,L,C
14	7	D,R,F	

D, demineralisation; R, rickets; F, fractures; P, periosteal new bone; O, over-inflation; L, linear strands; C, collapse/consolidation; Cy, cysts.

MANAGEMENT

Vitamin D therapy was given in daily doses varying between 20-125 μg to Nos. 1, 2, 4-7 and 9-12; one patient received two injections of 500 μg at an interval of three weeks. Over the subsequent 3-5 weeks as skeletal healing took place in the symptomatic infants, respiration gradually returned to normal. In number 8 the level of 25-OHD_3, initially 16.5 ng/ml, increased threefold following therapy with 0.2 μg of 1α-OHD_3 daily for three weeks (Glasgow and Reid, 1977). In numbers 1 and 2 however, the respiratory difficulties became particularly marked and the apnoeic episodes frequent and protracted leading to profound bradycardia. Each was managed in a Greogry Head Box with constant positive airways pressure. This resulted in an initial improvement (number 1) but progressive ventilatory failure with increasingly frequent apnoea led to her demise at nine weeks of age. Patient 2 died at the same age following intestinal obstruction due to paralytic ileus (Glasgow and Thomas, 1977).

Necropsy (*Patient* 1)

The body weighed 2.0 kg. Abnormal findings were confined to the lungs and bones. The lungs showed generalised thickening of alveolar walls with increase in interstitial fibrous tissue. There were small patches of bronchopneumonia and intra-alveolar fibrosis.

Bone (right femur) was examined by Dr. H. A. Ellis, Royal Victoria Infirmary, Newcastle upon Tyne. Both decalcified and plastic embedded, undecalcified sections were examined. Endochondral ossification was normal. The blood vessels and connective tissue invading the mineralised cartilage were excessive and there was an increase in chondroclastic and osteoclastic activity. The trabeculae were fewer than normal and those at the periphery were not being incorporated into cortical bone. The metaphyseal cortex was thin because resorption was still occurring. Formation of osteoid seemed to be proceeding normally. There was considerable woven osteoid around the diaphysis, and some reduction in lamellar bone formation. No definite mineralisation defect was present. The overall picture was that of osteopaenia somewhat reminiscent of Vitamin C, copper or manganese deficiency.

Necropsy findings in patient 2 have been published previously (Glasgow and Thomas, 1977).

DISCUSSION

Fourteen VLBW infants are described with acquired metabolic bone disease. All but one were seen from 1974 to 1977 in a high risk Maternity Hospital where the annual delivery rate is about 2,500; in addition approximately 170 babies are admitted annually from peripheral hospitals, of which about one-third are preterm.

Because of improved perinatal care and reduced neonatal mortality, the occurrence of bone disease may be increasing in infants of this gestational maturity. It seems likely however, that it often goes unrecognised because of the paucity of clinical signs. Hence diagnosis depends largely upon knowledge of its existence, coupled with careful radiological and laboratory surveillance. X-ray findings of demineralisation and rickets are uniformly present, although not always of marked degree. Biochemical features usually consist of an elevated alkaline phosphatase activity with more variable reductions of inorganic phosphate, calcium or magnesium. The combination of aminoaciduria and hypophosphataemia indicate hyperparathyroidism (Fraser, Kooh and Scriver, 1967). Raised levels of parathormone were documented in one patient and this responds to specific therapy with drugs, such as 1α-OHD_3, which correct the abnormality in calcium metabolism and heal bone disease (Glasgow and Reid, 1977).

Pure Vitamin D deficiency seems unlikely to be the entire explanation in every patient. The vitamin intake was satisfactory; in only two infants was this less than $20\mu g$ daily. The serum levels of 25-OHD_3 from four of our babies and in two similar infants of Davies, Hughes and Moore (1978) were within the normal range and, although this does not exclude a disorder of Vitamin D metabolism, it suggests that larger doses of Vitamin D are unlikely to be prophylactic, or the use of 25-OHD_3, therapeutic. Whether a defect is present in renal 1α-hydroxylation must await further study, nonetheless we have found treatment with 1α-OHD_3 to be efficacious (Glasgow and Reid, 1977).

An infant born very prematurely is already nutritionally deprived, since it is during the last trimester that the fetus acquires much of its mineral content, be it iron, copper or calcium (80 per cent) (McCance and Widdowson, 1961). In those with disturbed placental function as in six of our infants, calcium transport to the baby may be impaired (Khattab and Forfar, 1971). After birth accumulation of nutrients by the VLBW infant is often sub-optimal. Calcium assimilation, for example, is only about 40 per cent of that during late gestation (Shaw, 1976). Therefore such babies acquire about one-twelfth of the term infant's calcium load.

With regard to Vitamin D, the work of Hillman et al (1977) showed no difference in cord blood 25-OHD_3 levels between preterm and mature babies. None, however, was less than 1,500 g. Earlier work by this group tended to show that absorption of 25-OHD_3 by preterm infants was normal, but that 25-hydroxylation was probably impaired (Hillman and Haddad, 1975). The finding of normal 25-OHD_3 values in four babies however, would not support this view. Furthermore the absence of an obvious calcification abnormality on bone histology has led us to seek other aetiological factors which may account for the disorder.

Assuming the histological findings in the femur to be representative of the skeleton as a whole, they suggested a defect in bone matrix. On the other hand there was no radiological support for defects like scurvy or osteogenesis imperfecta. The idea that a trace element, such as copper, may be deficient in VLBW infants with bone disease has been suggested by Griscom, Craig and Neuhauser (1971). The hepatic copper level in their one baby who died was abnormally low and in another infant respiratory difficulty, similar to that in six of our patients, was thought radiologically to resemble the Mikity-Wilson syndrome. Moreover, Hambidge (1976) states that copper deficiency in prematures may be associated with osteoporosis, costochondral beading, muscular hypotonia and apnoea and it is now known their copper retention is low (Dauncey, Shaw and Urman, 1977). Copper deficiency however, was clearly not present in four of our infants by comparison with the range defined in normal premature babies (Halliday, 1977). In any event, about half of our patients had radiologically changes closely resembling those associated with classical nutritional rickets, which contrasts with the findings of Griscom et al (1971). None of our babies showed any haematological or medullary changes, apart from the mild anaemia of prematurity, while those with proven copper deficiency have sideroblastic anaemia with cytochemical and cytological abnormalities in the erythroid precursors associated with myeloid hypolasia (Al-Rashid and Spangler, 1971; Ashkenazi et al, 1973).

We have been impressed by the proportion of these infants born in the winter months of the past three years, a period during which all infants at risk, especially those less than 1.0 kg, have been carefully scrutinized. During winter, solar radiation, the prime source of Vitamin D, is less than in summer (Coblentz, 1947) and this results in significantly lower levels of 25-OHD_3 in the adult (McLaughlin et al, 1974). Since maternal blood concentrations largely determine those in the infant's circulation at birth (Rosen et al, 1974), winter born babies are likely to have lower 25-OHD_3 levels. It may be noteworthy therefore that only three of our 12 infants were born in summer or autumn. On the other hand

an abnormality in 1α-hydroxylation would not be expected to conform to a seasonal expression.

It would appear in the current state of knowledge, that no single explanation can account for all the clinical and laboratory abnormalities in each infant. Various facets of the syndrome may be caused by several nutritional deficiencies, some of which influence the expression of others. Nonetheless, an abnormality in renal 1α-hydroxylation is under further investigation.

Finally, our most severely affected babies each had lung changes which were similar to those described in the early stages of the Mikity-Wilson syndrome (Mikity, Hodgman and Tatter, 1967). While the pathogenesis of this disorder is not yet understood, it may be significant that the population at risk, age of onset and clinical presentation, early X-ray changes and natural history of that condition are identical to those of our infants with a disorder primarily of the musculo-skeletal system (Glasgow and Thomas, 1977). Pulmonary function studies in infants with this potentially fatal disorder should be carried out.

SUMMARY

Fourteen very low birth weight infants, 12 born since early 1974, developed bone disease during the first three months of life, in spite of apparently adequate nutrition including prophylaxis with Vitamin D. In eight the bone disease, possibly linked to muscle weakness, was associated with secondary respiratory distress and apnoea, in six of whom the radiological features closely resembled those seen in the syndrome of pulmonary dysmaturity (Mikity-Wilson). The infants who were lightest of all died at nine weeks of age. Blood biochemistry was variable but the plasma 25-hydroxyvitamin D, in four infants, was within the normal range indicating that pure Vitamin D deficiency is not aetiological, that 25-OHD_3 is unlikely to be therapeutic and suggesting that an abnormality of renal 1αhydroxylation may be present. Greater scrutiny of the overall population is urged, and in particular, infants of mothers with placetal insufficiency, multiple pregnancies of those born in the winter months, since it is these infants who seem most to be at risk of developing bone disease.

ACKNOWLEDGMENTS

Thanks are due to Professor I. J. Carré and Dr. W. A. B. Campbell for permission to publish details of patients under their care.

REFERENCES

AL-RASHID, R. A. and SPANGLER, J. (1971). Neonatal copper deficiency. *New England Journal of Medicine*, **285**, 841.

ASHKENAZI, A., LEVIN, S., DJALDETTI, M., FISHEL, E. and BENVENISTI, D. (1973). The syndrome of neonatal copper deficiency. *Pediatrics*, **52**, 525.

COBLENTZ, W. W. (1947). Measurements of biologically effective ultra-violet, solar and sky radiation in Washington DC, 1941-46. *Bulletin of the Meteorological Society*, **28**, 465.

DAUNCEY, M. J., SHAW, J. C. L., URMAN, J. (1977). The absorption and retention of magnesium, zinc, and copper by low birth weight infants fed pasteurised human breast milk. *Pediatric Research*, **11**, 991.

DAVIES, D. P., HUGHES, C. A. and MOORE, J. R. (1978). Rickets in preterm infants. *Archives of Disease in Childhood*, **53**, 88.

DUBOWITZ, L. M. S., DUBOWITZ, V. and GOLDBERG, C. (1970). Clinical assessment of gestational age in the newborn infant. *Journal of Pediatrics*, **77**, 1.

FRASER, D., KOOH, S. W. and SCRIVER, C. R. (1967). Hyperparathyroidism as the cause of hyperaminoaciduria and phosphaturia in human Vitamin D deficiency. *Pediatric Research*, **1**, 425.

GAIRDNER, D. and PEARSON, J. (1971). A growth chart for premature and other infants. *Archives of Disease in Childhood*, **46**, 783.

GLASGOW, J. F. T. and REID, M. McC. (1971). 1 α-hydroxyvitamin D in nutritional rickets. *Lancet*, **2**, 302.

GLASGOW, J. F. T. and THOMAS, P. S. (1977). Rachitic respiratory distress in small preterm infants. *Archives of Disease in Childhood*, **52**, 268.

GRAND, R. J., WATKINS, J. B. and TORTI, F. M. (1976). Development of the human gastrointestinal tract. *Gastroenterology*, **70**, 790.

GRISCOM, N. T., CRAIG, J. N. and NEUHAUSER, E. B. D. (1971). Systemic bone disease developing in small premature infants. *Pediatrics*, **48**, 883.

HADDAD, J. G. and CHYU, K. J. (1971). Competitive protein-binding assay for 25-hydroxycholecalciferol. *Journal of Clinical Endocrinology and Metaboloism*, **33**, 992.

HALLIDAY, H. L. (1977). personal communication.

HAMBIDGE, K. M. (1976). The importance of trace elements in infant nutrition. *Current Medical Research and Opinion*, 4, Suppl. 1, 44.

HILLMAN, L. S. and HADDAD, J. G. (1975). Perinatal Vitamin D metabolism II. Serial 25-hydroxyvitamin D concentrations in sera of term and premature Infants. *Journal of Pediatrics*, **86**, 928.

HILLMAN, L. S., ROJANASATHIT, S., SLATOPOLSKY, E. and HADDAD, J. G. (1977). Serial measurements of serum calcium, magnesium, parathyroid hormone, calcitonin, and 25-hydroxyvitamin D in premature and term infants during the first week of life. *Pediatric Research*, **11**, 739.

KHATTAB, A. K. and FORFAR, J. O. (1971). The interrelationship between calcium, phosphorus and glucose levels in mother and infant in conditions commonly associated with 'placental insufficiency'. *Biology of the Neonate*, **18**, 1.

KUHNS, L. R., SHERMAN, M. P. and POZNANSKI, A. K. (1972). Determination of neonatal maturation on the chest radiograph. *Radiology*, **102**, 597.

MCCANCE, R. A. and WIDDOWSON, E. M. (1961). Mineral metabolism in the fetus and newborn. *British Medical Bulletin*, **17**, 132.

MCMASTER, D. (1977). Personal communication.

MCLAUGHLIN, M., RAGGATT, P. R., FAIRNEY, A., BROWN, D. J., LESTER, E. and WILLIS, M. R. (1974). Seasonal variation in serum 25-hydroxy-cholecalciferol in healthy people. *Lancet*, **1**, 536.

MIKITY, V., HODGMAN, J. E. and TATTER, D. (1967). The radiological findings in delayed pulmonary maturation in premature infants. *Progress in Pediatric Radiology*, **1**, 149.

ROSEN, J. F., ROGINSKY, M., NATHENSON, G. and FINBERG, L. (1974). 25-hydroxyvitamin D. Plasma levels in mothers and their premature infants with neonatal hypocalcemia. *American Journal of Diseases of Children*, **127**, 220.

ROSENBERG, L. E. and SCRIVER, C. R. (1974). Diseases of Metabolism, p. 465. *Saunders, Philadelphia.*

SHAW, J. C. L. (1973). Nutritional Problems in a Changing World, p. 127. *Applied Science Publishers, London.*

SHAW, J. C. L. (1976). Evidence for defective skeletal mineralisation in low birthweight infants. The absorption of calcium and fat. *Pediatrics*, **57**, 16.

STEWART, A. L., TURCAN, D. M., RAWLINGS, G. and REYNOLDS, E. O. R. (1977). Prognosis for infants weighing 1000 g or less at birth. *Archives of Disease in Childhood*, **52**, 97.

THOMAS, P. S. and GLASGOW, J. F. T. (1978). The "mandibular mantle" – a sign of rickets in very low birthweight infants. *British Journal of Radiology*, **51**, 93.