REVIEW

Unexplained fractures in infancy: looking for fragile bones

Nick Bishop, Alan Sprigg, Ann Dalton

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A fracture occurs when the force exerted on a bone exceeds the ability of the bone to absorb the force by deforming. Fractures in children are common—approximately one third of children will have a fracture by 16 years of age, with more boys experiencing fracture than girls.¹ This differentiation in fracture risk is apparent from 2 years of age. Before the age of 2 years, fracture incidence is equal and occurs at a rate of approximately 80/10 000 person years. For the UK, therefore, approximately 4800 infants will have a clinically evident fracture before their first birthday each year.

Some long-bone fractures may occur at birth² in association with events such as shoulder dystocia³; skull fractures may occur during forceps⁴ and ventouse delivery.⁵ Some may (uncommonly) occur as a result of clearly defined trauma such as road accidents.⁶ Most, however, fall into the “unexplained” category. This article reviews our current approach to identifying bone disease in the infant presenting with more than one unexplained fractures, and discusses the recognised disease processes that result in increased bone fragility.

The history should include inquiry into specific areas as listed in the box. The two most frequently recognised underlying disease processes causing bone fragility in infancy are metabolic bone disease of prematurity⁷ and osteogenesis imperfecta, and directed questioning is appropriate for these conditions. For premature infants, the features commonly associated with fracture are delivery at <28 weeks of gestation, necrotising enterocolitis, late (>30 days) establishment of full enteral feeds, conjugated hyperbilirubinaemia, chronic lung disease, and use of furosemide.^{8–9} For a proportion of infants with osteogenesis imperfecta, there will be a family history either of osteogenesis imperfecta itself or of features that suggest osteogenesis imperfecta. The other elements of the history relating to the possibility of non-accidental injury should of course be applied in every case.

Clinical examination, in addition to documenting signs of concern with respect to non-accidental injury, should include assessment of the musculo-skeletal system, looking specifically for the features detailed in the box. Gross bowing of long bones is uncommon in type I osteogenesis imperfecta. Blue sclerae are present in many healthy infants and children without any associated bone disease. Deep blue sclerae persisting beyond 6 months of age should be regarded as potentially significant; Bauze *et al*¹⁰ found that blue sclerae persisted in all their mildly affected cases. However, many children with a moderately severe form of osteogenesis imperfecta, type IV osteogenesis imperfecta, have white sclerae.¹¹ It is certainly worth examining siblings and parents who may show

additional features of osteogenesis imperfecta (see box). It is easier to show ligamentous laxity in this older group than in infants.

PATHOPHYSIOLOGY OF BONE DISEASES PRESENTING WITH FRACTURES IN INFANCY

Metabolic bone disease of prematurity

Metabolic bone disease of prematurity (also known as osteopenia of prematurity and preterm rickets) is seen in the UK mainly in infants born at <28 weeks of gestation.⁷ Fractures occur typically at an age of at least 10 weeks and usually stop before the age of 6 months (uncorrected for gestation).

Three reviews of fractures occurring during the first year of life in premature infants^{8–12} found that rib fractures often remain undetected and are only discovered on x rays taken for other reasons. Hence, the true incidence of fractures in infants born prematurely remains unknown.

Koo *et al*⁹ reported a prospective study of 78 infants, weighing <1500 g and 23–26 weeks of gestation at birth, carried out during the mid to late 1980s in Cincinnati, Ohio, USA. Infants were examined for clinical signs of rickets and had x rays of both wrists and forearms at 1, 3, 6, 9 and 12 months of age. There were also x rays taken for clinically suspected fractures and for other clinical reasons. Skeletal surveys were not undertaken. By day 88, fractures along with rickets were present in 12 of the 78 infants, and a further 7 infants had fractures without any signs of rickets on x ray; clinical suspicion of fracture was recorded in only 3 of the 19 infants. The smaller the infant at birth, the higher the risk of radiological abnormality. Almost 30% of all the fractures were rib fractures. Affected infants took longer to establish full enteral feeds, required longer periods in oxygen and in hospital overall, received longer periods of furosemide, and were also more likely to have had physiotherapy.

Amir *et al*⁸ reported 12 of 973 premature infants surviving for >6 months in a Tel Aviv hospital from 1977 to 1984 with fractures during their time in the neonatal unit; in all but one, the birth weight was <1500 g. The overall incidence in the <1500 g group was 2.1%. Three infants died subsequently. The fractures occurred from ages 24 to 160 days. All the rib fractures occurred silently, and were diagnosed only on “routine” x ray—that is, full skeletal surveys were not undertaken. The authors identified four risk factors associated with fracture development: cholestatic jaundice, prolonged intravenous nutrition (>3 weeks), bronchopulmonary dysplasia

Abbreviations: DXA, dual energy x ray absorptiometry; FEVR, familial exudative vitreoretinopathy; IJO, idiopathic juvenile osteoporosis

See end of article for authors' affiliations

Correspondence to: Dr N Bishop, Academic Unit of Child Health, University of Sheffield, Sheffield Children's Hospital, Sheffield S10 2TH, UK; n.j.bishop@sheffield.ac.uk

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Summary of bone fracture during infancy

Features of bone disease in infancy—history

- Age at first fracture
- Number of fractures, sites and timing
- Apparent causation: mechanism and force
- Associated features: swelling and pain
- Infantile hernia
- Family history of recurrent fractures with minor trauma, dislocations, hernia, early-onset deafness, dentinogenesis imperfecta (discoloured, translucent teeth, cracking or chipping of teeth), late walking in siblings, osteoporosis in older family members (especially when apparent before the age of 50 years), eye disease (retinopathy, early blindness)
- Pregnancy/delivery/neonatal course: prematurity, gestation, birth weight, necrotising enterocolitis, time to establish full enteral feeds, conjugated hyperbilirubinaemia, use of furosemide, chronic lung disease and metabolic bone disease of prematurity

Features of bone disease in infancy—examination (apply to the infant and any available family members)

- Brachycephaly/plagiocephaly/dolicocephaly
- Large anterior fontanelle/sutural diastasis without hydrocephalus
- Craniotabes
- Blue sclerae: scleral hue variable in infancy but blue sclerae persist in cases of mild osteogenesis imperfecta
- Harrison's sulcus
- Translucent skin
- Ligamentous laxity: use the Beighton scale for children and adults. In infants, check whether the knees and elbows extend beyond 180° and whether the thumbs can be apposed to the forearm.
- Hernia(s)
- Bowing deformity of limbs
- Easy bruising is reported in some cases of osteogenesis imperfecta, but is not a universal finding
- Dentinogenesis imperfecta (translucent teeth that chip or crack easily, and may wear excessively, more so in primary dentition) may not be clinically apparent.⁴⁷

(chronic lung disease), and prolonged diuretic treatment with furosemide (>2 weeks). Only one of the 12 infants had none of the above risk factors. However, the number of infants overall with these risk factors was not recorded.

A third retrospective study by Dabezies *et al*¹² identified fractures in 26 (10.5%) of 247 infants over a 42-month period in the early 1990s, with the same risk factors and similar timing for fracture as in the other two studies.

None of these studies undertook skeletal surveys; the preponderance of rib fractures probably reflects the frequency of chest x rays as opposed to other radiographs. Longer periods in oxygen might simply be a proxy for inactivity. In addition to the plethora of literature on the effect of different calcium and phosphate supplementation regimens on bone mass accretion during the first 3–6 months of life, a clear effect of physical exercise and stress on bone mass accretion is evident.¹³ Lack of physical activity, a more common problem in the smallest, sickest infants, will probably contribute to reduced bone mass

acquisition and hence compromise bone strength. However, both Koo and Dabezies identified physiotherapy as being associated with fracture in their studies. Prolonged intravenous nutrition and failure to establish full oral feeds quickly indicate a continuing poor supply of mineral substrates necessary for bone formation and mineralisation. Furosemide is a diuretic that unfortunately promotes the loss of calcium in the urine, adding to the deficit and increasing the activity of homeostatic mechanisms designed to keep the serum calcium concentration in the normal range. These mechanisms result in increased bone turnover and release of calcium from the skeleton. The increased bone turnover increases bone fragility because the bone turnover sequence involves initial removal of bone followed by its replacement.

A case-control study of fracture frequency in children born preterm up to age 5 years¹⁴ showed no increase in fracture risk for such children overall, but an increase in risk up to the age of 2 years. All children in the latter group were born at <32 weeks of gestation.

Metabolic bone disease in preterm infants is a self-limiting disease. Growth slows postnatally and the supply of mineral substrate improves, so that the needs of the skeleton for calcium and phosphate to mineralise newly formed bone are more consistently met and the skeletal envelope is "filled in". Bone mass rises consistently until it is similar to that of infants born at term by the age of 2 years¹⁵; this fits with the observation that the increased fracture risk for children born prematurely seems most apparent in the first 2 years of life.

Severe rickets in term infants

Rickets is increasingly reported in the developed world.¹⁶ Vitamin D deficiency is most common in infants wholly breastfed and whose mothers are of non-white Caucasian origin. Vitamin D deficiency in a term infant sufficient to result in bone weakening and fracture will be accompanied by unequivocal radiological and biochemical evidence of deranged skeletal structure and homeostasis. Vitamin D deficiency is extremely rare in formula-fed infants and should lead to investigations for malabsorption.

Osteogenesis imperfecta

Type I collagen is the most abundant protein in bone, and is composed of a heterotrimer of two type I collagen α -1 protein chains and one type I collagen α -2 protein chain. The three chains coil tightly around one another to form a molecule that self-assembles into fibrils, creating the template on which mineral is deposited to make bone material. Osteogenesis imperfecta is caused in 90% of cases by mutations in the type I collagen genes *COL1A1* and *COL1A2*, which encode type I collagen α -1 and α -2 protein chains, respectively.¹⁷ The spectrum of the disease is wide and now encompasses seven distinct types; the more recently described types V,¹⁸ VI¹⁹ and "rhizomelic"²⁰ are not due to type I collagen mutations and are associated with more severe forms of bone disease.

Of the traditional four types of osteogenesis imperfecta, type II (lethal) and type III (severe, progressively deforming) are readily diagnosed on clinical and radiological grounds.¹¹ Infants with type I (mild) and type IV (moderate to severe) osteogenesis imperfecta may be difficult to differentiate from normal infants. Type I infants typically have mutations in the *COL1A1* or *COL1A2* genes that result in reduced type I collagen α -1 and α -2 chains, respectively.²¹ This means that although most of the type I collagen protein produced is normal, the total amount of protein is reduced. The effect is to reduce the overall bone mass within the skeletal envelope. In addition, bone biopsies in older children with type I disease indicate that the cortex is thinner and more porous, and the trabecular bone, a honeycomb-like structure that provides internal bracing at the ends of long

bones, is reduced in amount and is less well interconnected than in healthy bone.²² Overall, tubular bones in children with osteogenesis imperfecta are narrower, adding to their propensity to fracture. Those with type IV osteogenesis imperfecta have more pronounced changes of a similar nature. Some cases of type IV are also due to null alleles, presumably with additional influences from other genes of importance to bone health modifying the phenotype to increase the severity.²¹ Others have mutations in the *COL1A1* gene leading to the production of an abnormal protein, which then interferes with the three-dimensional arrangement of the collagen molecules.

How common is osteogenesis imperfecta in cases of unexplained fracture in infancy? A study by Marlowe *et al*²³ showed that of 262 infants and children up to the age of 3 years with unexplained fractures referred for genetic testing for osteogenesis imperfecta to the major US centre in Seattle, 11 definitely had mutations in the type I collagen genes, and mutations could not be excluded in a further 11. Of the 11 who were definitely affected, 6 were identified as likely to have osteogenesis imperfecta on clinical grounds, 3 were thought not to have osteogenesis imperfecta, and inadequate information was given in the remaining two cases. Of the 11 in whom osteogenesis imperfecta could not be excluded in the laboratory, none was thought to have osteogenesis imperfecta. Of the remaining 240 cases not found to have genetic changes indicating osteogenesis imperfecta, four were thought to have osteogenesis imperfecta on the basis of a blue scleral hue. This study was clearly biased in terms of the selection of candidates for testing, but nevertheless suggests that a proportion of cases of unexplained fracture is due to type I collagen mutations.

Failure to continue to fracture after removal from a potentially abusive environment should not be taken as unequivocal evidence of the lack of an underlying bone disease. The natural history of fractures in mild cases of osteogenesis imperfecta is often episodic in nature, with long periods where no fracture may occur. By contrast, if a child continues to experience fracture in care, this suggests either a more severe form of underlying bone fragility or abuse on the part of the new carers.

Cole-Carpenter syndrome

Infants with this disorder have ocular proptosis, usually present from birth, and develop hydrocephalus and fractures during the first 12 months of life.²⁴ On plain x ray, the bones appear osteopenic. The genetic origin of this syndrome is unknown, and the number of reported cases is very small. Another syndrome of microcephaly, cataracts and fractures in infancy was reported in 1978,²⁵ but no further cases have emerged.

Bruck syndrome

Infants with this disorder are born with joint contractures and bone fragility. There are two distinct forms. In one, the genetic defect has been identified in the *PLOD2* gene,²⁶ resulting in a failure of hydroxylation of lysine residues in collagen 1 telopeptides. The second locus is 17p12, but the gene is as yet unidentified.

Familial osteoporosis

Osteoporosis may run in families. Recently, a group in Toronto identified 3 of 20 children with idiopathic juvenile osteoporosis (IJO) as having heterozygotic defects in the gene *LRP5*.²⁷ *LRP5* has been the topic of much interest over recent years. It is a receptor in the canonical wnt-signalling pathway.²⁸ Activation of the pathway increases bone formation by upregulating the growth of preosteoblasts and the activity of differentiated osteoblasts. Activating mutations in *LRP5* are associated with a high bone mass phenotype, with no fractures reported in family members carrying the mutation.²⁹ Homozygotic loss-of-function mutations result in the osteoporosis pseudoglioma

syndrome, with <50 cases reported worldwide.³⁰ This disorder is characterised by a progressively deforming bone disease with multiple fractures, low bone mass and reduced stature, similar in some respects to type III osteogenesis imperfecta. Additionally, patients having eye disease (pseudoglioma) brought about by failure of vascularisation of the peripheral retina, with hyperplasia of the vitreous, corneal opacity and secondary glaucoma are also described. The wnt-signalling pathway through *LRP5* also has the function of patterning the peripheral retinal vasculature during embryogenesis.³¹

Apart from being associated with vertebral crush fractures and metaphyseal fractures in children with the clinical diagnosis of IJO, heterozygotic defects in *LRP5* are associated with familial exudative vitreoretinopathy (FEVR). Two children in the series reported by Toomes had fractures starting in infancy.³¹ *LRP5* heterozygosity produces an eye disease that is similar in some respects to retinopathy of prematurity, and that is associated with hyperpermeable blood vessels, neovascularisation, retinal folds and exudates in some individuals. Both the eye and bone features are variable in *LRP5* heterozygotes; some family members may have reduced bone mass and no eye signs, whereas some have eye signs and no fractures.

It is unclear whether *LRP5* heterozygosity is associated with metaphyseal fractures and retinal haemorrhages occurring together in infancy; to suggest that it would be purely speculative. The appearance of the peripheral retina and pattern of haemorrhages is likely to be important in distinguishing between FEVR and retinal haemorrhage due to non-accidental injury.

No information is available at present regarding the frequency with which *LRP5* mutations occur. Clinically apparent IJO is much less common than osteogenesis imperfecta; the estimate of IJO incidence is approximately 1 in 100 000, with osteogenesis imperfecta being 1 in 10–15 000. FEVR is similarly very rare.³¹ There have been no reports as yet of *LRP5* mutations in infants with unexplained fractures, with or without retinal haemorrhage. This is clearly a matter of great interest but also of great uncertainty at the present time.

Other conditions

There are several rare bone diseases that can cause fractures in infancy. These include panostotic fibrous dysplasia/McCune-Albright syndrome, osteopetrosis, infantile severe hypophosphatasia, congenital insensitivity to pain with anhidrosis, congenital rickets, and congenital cytomegalovirus infection. All these disorders have radiologically apparent bone disease and should not present diagnostic difficulty. Infants with congenital disorders affecting nerve and muscle function may also have slim bones that are mechanically inadequate. The accompanying clinical features in such cases should alert the clinician. Having a bone disease or fragile bones does not preclude the possibility of non-accidental injury, however. In disorders in which bone strength is diminished, the pattern of fractures is typically of a series of individual fractures over time. It is unusual to see multiple rib fractures of the same age in a child with mild osteogenesis imperfecta or bone disease of prematurity without a clear explanation.

Pain

All fractures are painful, whether in children with normal bones or in those with bone disorders. Fracture-related pain is likely to recur when the affected site is disturbed in any way, and will probably be more intense and persist longer when the affected area is not splinted by surrounding structures. Thus, rib fractures in particular may go unsuspected by both parents and clinical staff, as previously indicated, but fractures of the mid-shaft of a long bone will be associated with protective disuse.

INVESTIGATIONS

Plain film radiography

Plain film radiographs are required for assessing structural changes in long bones, ribs, skull and spine. Radiography provides data in both suspected non-accidental injury and suspected bone dysplasia. In addition to imaging of a fractured limb at presentation, a full skeletal survey must be performed. This requires an agreed protocol with the radiology department and dedicated radiographers with a paediatric interest. The survey should be performed to include all long bones, frontal and both oblique views of the chest, two views of the skull, the lateral whole spine and views of both hands and feet. The British Society of Paediatric Radiology has established agreed standards for skeletal imaging in suspected non-accidental injury (www.bspr.org.uk). As soon as a skeletal survey has been performed, an experienced radiologist interested in paediatric imaging should review it. A report should be issued or arrangements made for a referral opinion. Acute rib fractures may not be detectable on the initial chest film, and in suspected non-accidental injury, a single follow-up chest film is indicated 2–3 weeks after original presentation to exclude rib fractures that become evident as they heal. Each injury shown radiographically should be considered in its clinical context with the history of mechanism of injury and any features that suggest an underlying bone disorder before confirming non-accidental injury.

Skull *x* rays are suggested even if a computed tomography of the head has been performed, as it is accepted that computed tomography imaging may miss skull fractures that are detectable on plain radiographs. Skull *x* rays may also show wormian bones. Cremin *et al*³² observed an excess of mosaic-like bones (>10, measuring at least 4×6 mm) within the sutures in 81 cases with osteogenesis imperfecta and not in 500 “normal” individuals, of whom 39 were children. The relationship between their occurrence and osteogenesis imperfecta type was not specified. The lack of skull vault ossification in severe cases may mean they are initially invisible; delayed films (6 months) may show them. Such mosaic bones are not uncommon in normal infants, but are then usually restricted to the lambdoid sutures. The cortical thickness of long bones can be estimated from plain radiographs. Overtubulation may be seen in osteogenesis imperfecta. It results in bones that have a smaller transverse diameter than is appropriate for their length. Cortical thickening and bone deformity may be the result of previous fracture.

Radiologically apparent osteopenia (sometimes reported by radiologists) implies that there is less attenuation of the *x* ray beam, due to a reduction in the total amount of calcium in bone tissue. The older radiology literature^{33–34} describes studies indicating that a 20–40% loss of mineralised bone mass is needed before osteopenia is radiologically detectable, and that this value changes according to the site assessed. The converse is clearly that it is difficult to detect losses of up to 20% of mineralised bone tissue, and the percentage loss of tissue that occurs before becoming radiologically evident may be higher at some sites in the skeleton.

Osteopenia can result from demineralisation (as in rickets) or loss of bone material (as in osteogenesis imperfecta). Although necessarily less precise than dual-energy *x* ray absorptiometry (DXA) in quantifying bone mineral density, it does provide the radiologist with an initial indication of the likelihood of an underlying bone disorder. However, quantitation of bone mass by DXA has not been shown to be helpful in infancy in discriminating those with bone disease from normal infants.³⁵ Normative data are lacking for infants <2 years of age; the data available from manufacturers relate to the older pencil beam DXA instruments and not to the current generation of fan beam devices. In older children with mild osteogenesis imperfecta,

bone mass in both the lumbar spine and total body is often within the normal range (own unpublished data); however, these mildly affected children will frequently have other radiologically apparent features such as a change in bone shape, particularly in the vertebrae and femurs, which may be difficult to discern in infancy.

Dating fractures is the province of experienced paediatric radiologists. Patterns of fracture may suggest, in some instances, inflicted injury. Fractures in infants born prematurely tend to be in the diaphyses of long bones and the ribs, as indicated previously. Skull fractures are rarely reported, but have not been sought systematically. Fractures can be found in almost any site or combination of sites in osteogenesis imperfecta. However, metaphyseal fractures are rare in both osteogenesis imperfecta and ex-premature infants, and multiple fractures in infants with osteogenesis imperfecta are usually accompanied by some degree of bony deformity.

Plain film radiography is not the final arbiter of bone fragility in infancy; as with the other forms of investigation discussed here, it is a part of the overall approach to discriminating between a diagnosis of bone fragility and one of non-accidental injury.

Biochemical markers of bone disease

Bone markers have been studied in infants and children with osteogenesis imperfecta³⁶ and with metabolic bone disease of prematurity.³⁷ Where inadequate bone mineralisation due to rapid growth (rickets of prematurity) or vitamin D deficiency is suspected to contribute to the problem, the measurement of fasting serum 25-hydroxyvitamin D, parathyroid hormone, alkaline phosphatase, calcium and phosphate should be undertaken. It is not clear whether reduced serum levels of 25-hydroxyvitamin D and increased parathyroid hormone are associated with an increased fracture risk in the absence of radiologically apparent osteopenia. Fasting phosphate is reduced and alkaline phosphatase activity increased in cases of active rickets; such changes may also be seen in bone disease of prematurity.

Bone turnover markers are increased in children with osteogenesis imperfecta³⁶ and are also increased after fracture. It is unclear exactly how long the serum alkaline phosphatase level remains raised, but activity should reflect both new bone formation at the fracture site and remodelling of callus. Once that is complete, serum alkaline phosphatase should return to a value within the normal range. The ranges for bone formation and resorption markers in healthy infants are wide.³⁸ In a study of bone markers in older children, markers reflecting the production of type I collagen were reduced in more than half the children with osteogenesis imperfecta who were tested,³⁹ but there is no evidence that the tests would be discriminatory in individual cases or in infants. However, persistently raised values of formation and resorption markers should alert the clinician to the possibility of underlying bone disease with increased bone turnover.

Postmortem testing

This can be undertaken at the radiological, tissue and molecular levels. DNA and skin samples can be taken after death and treated as indicated below. Investigation at tissue level for osteogenesis imperfecta is possible in terms of looking directly at bone. Bone biopsy has been used as a research tool principally in Montreal,^{40–41} and to assess bone architecture before starting bisphosphonate treatment. As stated above, there are clear architectural differences in the bones of children who are normal and in those with type I osteogenesis imperfecta, but it is not clear when these first emerge. It is technically difficult to take biopsies of sufficient quality before

the age of 1 year in living children, and hence applicability in this setting is likely to be limited to postmortem examination.

Molecular diagnosis of bone disease

There are three routes to laboratory diagnosis: biochemical analysis of collagen species, mutation analysis of RNA and mutation analysis of DNA. The first two require skin fibroblasts and hence a skin biopsy, which is often thought to be unacceptable, particularly in infants in whom no surgical procedure is required for treatment, and when there is an alternative testing strategy available.

Where fibroblasts are available, biochemical analysis involves isotopic labelling of protein in the skin fibroblasts, isolation of protein including collagen species, and gel electrophoresis (sodium dodecyl sulphate–polyacrylamide gel electrophoresis). Abnormalities can be either quantitative or qualitative. Specificity is high, but up to 20% of abnormalities may be missed.^{42–43} Mutation analysis of *COL1A1* and *COL1A2* from fibroblast RNA can then be carried out. Using message-based techniques, about 15% of samples from individuals with clinically apparent osteogenesis imperfecta will be recorded as negative; as many as half of the remainder can be detected as having mutations by direct sequencing.⁴²

Mutation analysis of DNA for *COL1A1* and *COL1A2* can be conducted by scanning methodologies such as conformation-sensitive capillary electrophoresis, or by direct sequencing. However, neither will detect other mutational mechanisms such as large deletions/duplications of DNA, although these are likely to be rare (<3%) and if the mutation has occurred in another gene regulating bone formation, it will be missed. However, large deletions/duplications can be detected by RNA analysis from a skin biopsy.

No comprehensive large study has been published comparing the positive predictive value of biochemical versus molecular analysis. It is likely that the two methods complement each other,⁴⁴ with molecular analysis showing a high positive predictive value for mutations in *COL1A1* or *COL1A2*, whereas biochemical analysis may show abnormalities in collagen potentially arising from mutations in other genes in the bone formation pathway, as well as large deletions/duplications in *COL1A1* or *COL1A2*. Other genes are likely to be involved in <10% of all cases. The sensitivity of mutation scanning methodologies versus direct sequencing is variable, although extensive experience in other disorders indicates that recent scanning methodologies such as conformation-sensitive capillary electrophoresis are very sensitive. However, the final arbiter of the presence of a mutation is direct sequencing, and the advantage of only requiring a blood sample for analysis is important.

Molecular diagnostic testing for LRP5 mutations currently remains a research tool.

When should genetic testing be requested?

The incidence of osteogenesis imperfecta in the general population is 1 in 10–20 000.^{45–46} NHS funding for molecular testing has been recommended by the UK Genetics Testing Network (UKGTN) for cases where there is a clinical diagnosis of types II and IV, with further clarification of referral pathways currently being sought for types I and III. If a diagnosis of osteogenesis imperfecta of any type is made, referral to the local clinical genetics service is appropriate. Diagnostic, predictive and prenatal testing in the family will then be considered in the light of the clinical need and the circumstances of the family.

In addition, in unequivocal cases of osteogenesis imperfecta, referral to a specialist centre that has experience in the long-term management of the condition should be made (see Appendix) so that optimum management can be put in place. Severely affected infants need input from a specialist multi-disciplinary team, but even those apparently mildly affected

initially may go on to develop vertebral crush fractures and need medical intervention. Where a clinician suspects that osteogenesis imperfecta is present, on the basis of the clinical and/or family history and examination or imaging findings described in this paper, genetic testing may assist in reaching a diagnosis, but the cost is substantial (see Appendix) and needs to be justified by a considerable effect on clinical management.

Where the only indication is unexplained or inadequately explained fractures in the absence of the clinical features of osteogenesis imperfecta, the situation is more complex, and there are currently no clear guidelines regarding suspected non-accidental injury. Consideration must be given to any possible explanation offered, the level of proof required (eg, family or criminal court) and the effect any result will have.

In such situations, a positive test result provides evidence of underlying bone fragility, but determining the degree of force likely to give rise to fractures when such fragility exists is essentially impossible. When there are only one or two fractures, occurring together or at different times, it is easier to believe that the fractures could have occurred with normal handling and without the parents being immediately aware of the problem. Where there are many fractures, without accompanying bony deformity, or with additional extraskelatal injury, it is more difficult to believe that the injuries have occurred accidentally, even allowing for the underlying bone fragility. These are judgements that are rightly the province of the courts, but consideration should be given in each case to the appropriateness of testing.

If skin biopsy samples are available through clinical interventions for treatment such as surgery or through postmortem examination, biochemical and molecular testing is as complete a test as possible. Taking a skin biopsy simply for diagnostic purposes is more problematic; clinicians have refused to take a blood sample for diagnostic purposes in the belief that this constitutes further abuse to an already abused child. A negative result, which will never be completely definitive, must be considered in the context of the full clinical and social history.

Properly conducted prospective studies are needed to clarify the exact place of genetic testing in these circumstances.

SUMMARY

Clinical history and examination supplemented by radiology are currently the mainstays of detection of bone disease in infants presenting with one or more unexplained fractures. Osteogenesis imperfecta and prematurity are the commonest established causes of bone fragility leading to fracture in infancy, but these are infrequent compared with non-accidental injury. Mutations in LRP5 are associated with both eye disease and fractures, but the extent to which such mutations contribute to unexplained fractures in infancy is entirely unclear. The role of genetic testing in discriminating bone disease from non-accidental injury in infants with unexplained fractures still needs to be properly evaluated in an appropriate population-based study.

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Authors' affiliations

Nick Bishop, Academic Unit of Child Health, University of Sheffield, Sheffield Children's Hospital, Sheffield, UK

Alan Sprigg, Sheffield Children's NHS Foundation Trust, Sheffield Children's Hospital, Sheffield, UK

Ann Dalton, Sheffield Molecular Genetics Service, Sheffield Children's NHS Foundation Trust, Sheffield Children's Hospital, Sheffield, UK

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APPENDIX

WHERE GENETIC TESTING IS PERFORMED IN THE UK

Testing for osteogenesis imperfecta is available from Sheffield Molecular Genetics Service. For diagnostic testing the cost is approximately £1560 for both the type I collagen genes and includes sequencing of both genes and a clinical report. The turnaround time is generally 8 weeks. For medico-legal cases the cost depends on what is required, particularly turnaround time, reporting requirements and legal processes such as attendance at court. For details contact Ann Dalton, Sheffield Molecular Genetic Service: Ann.dalton@sch.nhs.uk. Further development of the service is ongoing including dosage analysis for COL1A1 and COL1A2, analysis of other genes and biochemical analysis of collagen species.

Centres providing multidisciplinary services for children with metabolic bone disease (Clinical leads)

Belfast, Musgrave Park—Catherine Duffy
Birmingham Children's Hospital—Nick Shaw
Bristol Children's Hospital—Christine Burren
Cardiff, University Hospital—John Gregory
Glasgow Yorkhill—Faisal Ahmed
Great Ormond Street Hospital—Catherine De Vile (OI only), Caroline Brain
Manchester, St Mary's Hospital—Zulf Mughal
Royal London Hospital—Jeremy Allgrove
Sheffield Children's Hospital—Nick Bishop

Other useful resources

UK Brittle Bone Society: www.brittlebone.org
US Osteogenesis Imperfecta Foundation: www.oif.org
Testing of collagen protein in the US and some additional information around their approaches to deciding if testing is indicated:
www.pathology.washington.edu/clinical/collagen/guidelines/osteogenesis.php