Disrupted growth plates and progressive deformities in osteogenesis imperfecta as a result of the substitution of glycine 585 by valine in the $\alpha 2(I)$ chain of type I collagen

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Abstract

The skeleton of a child with osteogenesis imperfecta type III, resulting from the substitution of glycine 586 by valine in the triple helical domain of the $\alpha 2(I)$ chain of type I collagen, was severely porotic but contained lamellar bone and Haversian systems. From early childhood, structural failure of the bone resulted in the disruption of growth plates, progressive bone deformities, and severe growth retardation.

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Key words: osteogenesis imperfecta; disrupted growth plates; progressive deformity.

Osteogenesis imperfecta (OI) is a heterogeneous disorder of the type I collagen containing tissues such as bone, dentine, sclera, and ligaments.¹ The Sillence classification includes four main types of which the progressively deforming type III form (OI-III) is the focus of this report.²

Dominant negative mutations of COL1A1, which encodes the $\alpha 1(I)$ chains, and of COL1A2, which encodes the $\alpha 2(I)$ chain of type I collagen, have been identified in cases of OI-III.³ Most of the mutations produce substitutions of glycine residues within one of the 338 Gly-X-Y triplets that make up the triple helical domains of the $\alpha 1(I)$ or $\alpha 2(I)$ chains of type I collagen. Glycine substitutions by valine in the $\alpha 2(I)$ chain have been identified in only a few cases of OI. A case (OI-50) of progressively deforming OI was previously shown to be the result of the heterozygous substitution of Gly 586 by Val in the $\alpha 2(I)$ chain.⁴ In this paper, we describe the clinical, radiographical, pathological, and further biochemical studies of this baby with progressively deforming OI.

Case report

CLINICAL HISTORY

The female proband, OI-50, was the first child of unrelated, normal 26 year old parents. Her birth weight at 40 weeks' gestation was 2954 g (20th centile). She had numerous fresh and healing fractures. The clinical features indicated that she had OI-III.¹

Her musculoskeletal and general development were prospectively recorded over

14 years (fig 1). Her longitudinal growth and weight were less than the third centile throughout growth. Her sclerae faded to a slightly bluish tinge at 14 years of age. She had severe dentinogenesis imperfecta of her primary and secondary dentition. During the study period she did not develop basilar compression.⁵ She had mild conductive hearing loss.

Her motor development was delayed because of osteopenia, muscular weakness, ligamentous laxity, and recurrent subtrochanteric fractures of the femora. Tibial fractures were uncommon despite increasing tibial kyphosis throughout childhood (fig 1). She developed severe bowing of her upper arms and forearms over the first few years of life which persisted throughout childhood (fig 1). Her chest became progressively more barrel shaped over the first few years of life. She rapidly developed a rigid thoracic scoliosis which showed slow progression.

Figure 1 Clinical appearance at 4 years of age.

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Figure 2 Radiograph of the left humerus and elbow at 6 years of age. The humerus, radius, and ulna are bowed and slender. Retarded growth of the sclerotic medial half of the proximal humeral growth plate increased the varus deformity of the humeral neck.

RADIOGRAPHICAL FEATURES

Radiographs shortly after birth showed generalised osteopenia with thinning of the cortices and numerous fractures. The calvarium was thin and the midface and mandible were hypoplastic. The many hundreds of wormian bones in the cranial sutures persisted for many years.

At birth, the diaphyses of her long bones were either widened or of normal width. They progressively narrowed and bowed over the following two years. In addition, most of the growth plates of the long bones showed premature disruption from 2 years of age and complete growth arrest by 8 to 10 years of age (figs 2, 3, and 4).^{6,7} As a neonate, the vertebral bodies were of normal height and the spine showed a minor postural scoliosis. Generalised platyspondyly with expansion of the intervertebral discs developed over the following two years. She developed a fixed thoracic kyphoscoliosis that progressed slowly throughout childhood. The vertebral growth plates were also disrupted.

PATHOLOGICAL FEATURES

Histology of femoral cortical bone at 4 years of age and tibial cortical bone at 10 years of age showed thinning of the cortices when compared to age matched controls. Haversian systems and lamellar bone were present but the osteocytes were more closely spaced and the nuclei of the osteocytes were abnormally large when compared to age matched controls

Figure 3 Radiograph of the pelvis and legs at 1 month of age. The shafts of the femora and tibiae are short and broad. There is a healing fracture of the proximal right femur with a severe varus deformity.

(fig 5). The lacunae of the Haversian systems were abnormally large.

Ultrastructure of the skin showed no abnormalities of the collagen fibrils. The fibrils had a normal periodicity and were round and of normal size distribution in cross section.

Figure 4 Radiograph of the legs at 6 years of age shows that the femora and tibiae are slender. The distal femoral growth plate has broken into pieces that produce abnormal areas of ossification in the metaphysis and epiphysis. The tibia is kyphotic and the proximal tibial growth plate shows similar changes to the distal femoral growth plate.

Figure 5 Tibial cortical bone at 10 years of age. The cortex is thin but contains Haversian systems and lamellar bone. It is covered by thick periosteum (P). Haematoxylin and eosin stain.

Figure 6 Electrophoresis of type I collagens extracted from OI-50 dermis and bone. The collagens are stained with Coomassie Brilliant Blue. Lane 1, normal dermis; lane 2, OI-50 dermis; lane 3, normal bone; lane 4, OI-50 bone. The $\alpha 1(I)$ and $\alpha 2(I)$ chains of type I collagen and their $\beta 11$, $\beta 12$, and $\beta 22$ dimers are shown. They migrate more slowly than their normal counterparts. The $\alpha 1(III)_3$ chains of type III collagen and the $\alpha 1(V)$ and $\alpha 2(V)$ chains of type V collagen are also shown. The open arrow head indicates a pepsin artefact that is present in the normal and OI samples.

BIOCHEMICAL FEATURES

Pepsin solubilised collagen was prepared from the dermis and bone using previously described methods.⁴ The collagens were resolved into their component chains by electrophoresis on 5% polyacrylamide gels containing sodium dodecyl sulphate.⁴ The protein chains were stained with Coomassie Brilliant Blue. The $\alpha 1(I)$ and $\alpha 2(I)$ chains of type I collagen from dermis and bone migrated slowly on electrophoresis (fig 6). No other abnormalities were identified.

Discussion

Progressive deformities and premature growth plate arrests accounted for the progressive shortening of the trunk and limbs in the proband. Disruption of the growth plates, owing to inadequate support from the osteopenic metaphyseal and epiphyseal bone, produced a radiographic appearance called popcorn calcification.⁶⁷ The latter changes were evident at 2 years of age and progressively worsened with complete growth arrest at 8 to 10 years of age. Popcorn calcification is a characteristic feature of OI-III.²

There are few reports of bone histology in OI-III. Woven bone was observed, without evidence of lamellar bone or Haversian systems, in OI-III owing to substitutions of Gly 427 by Arg and Gly 1006 by Ala in the $\alpha 2(I)$ chain.^{8,9} In the latter cases, who were aged approximately 2 years, there was a paucity of mineralised bone. As the osteoid seams were thin it was likely that the rate of new bone formation was impaired. Bone from age matched samples showed a coherent structure with regular striations because of a well organised lamellar plywood-like arrangement of the mineralised collagen fibrils.^{8,10} In the present case, the cortical and trabecular bone contained well ordered lamellar bone and Haversian systems without woven bone. However, the cortices were thin and the lamellar bone structures were not as compact as in normal bone. The finding of similar lamellar bone structures at 4 and at 10 years of age indicates that an ordered lamellar arrangement of collagen fibrils, rather than a disorganised woven arrangement, was present in the proband's bone from early childhood. The dermal collagen fibrils were also well formed. In contrast, Gly substitutions by Val at residues 973 and 1006 of the $\alpha 1(I)$ chain produce severe osteopenia with woven bone, thin dermal collagen fibrils, and perinatal lethal OI-II.¹¹

The dermis and bone contained the normal types of collagen found in these tissues. There was no evidence of increased amounts of type III and V collagens in bone as has been observed in OI-II bone.¹² The slow electrophoretic migrations of the $\alpha 1(I)$ and $\alpha 2(I)$ chains of type I collagen were probably the result of overmodification of lysine residues towards the amino-terminus of the substitution, as shown previously by peptide mapping.⁴ The overmodifications involve the single mutant $\alpha 2(I)$ chain as well as the two normal $\alpha 1(I)$ chains that make up each mutant type I collagen molecule. As the proband was heterozygous for the mutation we expected that at least half of the type I collagen molecules would contain normal chains with normal electrophoretic migrations. However, all of the monomeric and dimeric $\alpha 1(I)$ and $\alpha 2(I)$ chains migrated slowly indicating that the normal and mutant molecules were both overmodified. Previous studies did not find evidence of a second mutation in the $\alpha 1(I)$ or $\alpha 2(I)$ chains.⁴ Similar electrophoretic findings have been observed in other heterozygous cases of OI.¹²

Glycine substitutions by valine in Gly-X-Y triplets of the $\alpha 2(I)$ chain have also been

identified at residues 319, 544, 676, and 892.^{13,14} The substitutions at residues 319 and 892 produce perinatal lethal OI-II while those at residues 544 and 676 produce OI-IV. Another substitution of Gly 586 by Val in an unrelated subject also produced progressively deforming OI-III (Gomez-Lira, personal communication). Gly to Val substitutions in the $\alpha 1(I)$ chain produce mild OI-I at residue 85 and perinatal lethal OI-II at residues 256, 565, 637, 802, 973, and 1006.¹⁵⁻²⁰ As with other glycine substitutions of the $\alpha 2(I)$ and $\alpha 1(I)$ chains, there is a need to define further the factors that determine the nature and severity of the structural, functional, and growth abnormalities of the extracellular matrix and the consequent severity of the clinical OI phenotype.

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