Clinical Vignette

Ghosal Hematodiaphyseal Dysplasia: An Unusual but Easy-to-Diagnose Genetic Cause of Anemia

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Introduction

Ghosal hematodiaphyseal dysplasia (GHDD) also known as Ghosal syndrome (OMIM# 231095) is a rare autosomal recessive disorder associated with skeletal changes in the form of increased bone density and predominant diaphyseal involvement, and hypoplastic anemia (Ghosal et al., 1988). It is caused by biallelic variants in the *TBXAS1* (OMIM*274180) gene. As the hematological abnormalities respond to corticosteroid therapy, early and accurate diagnosis helps in effective control of the disease course. We report here a 12 years-old male patient from a consanguineous family referred for evaluation of chronic anemia, whose clinical and radiographic findings were suggestive of the diagnosis of GHDD and molecular genetic testing revealed a homozygous nonsynonymous known variant in the *TBXAS1* gene.

Patient details

A 12-years-old boy, the third offspring of third-degree consanguineous parents, was referred for evaluation of chronic anemia detected at around 6 years of age. He had received six transfusions of packed red blood cells (PRBC) between 6 to 12 years of age. There was no history of abdominal distension, jaundice, gall stones, bleeding manifestations, or recurrent infections and fever. No specific symptoms were noted in early childhood. The developmental milestones were age-appropriate but the scholastic performance was below average. The two elder female siblings aged 18 years and 15 years, and both parents were normal. There was no history of similar illness or any other known genetic disease in other family members. On examination, his height and weight were 146 cm and 36 kgs respectively (both corresponding to around 30th centile for age), and head circumference was 55 cm. He had severe pallor, but no icterus, edema, or lymphadenopathy. Craniofacial dysmorphic features noted in the child included dolichocephaly, frontal prominence, telecanthus, bilateral proptosis, depressed nasal bridge with bulbous tip of nose, thick lips, and retrognathia (Figure 1). There was no hepatosplenomegaly and rest of the systemic examination was also normal.

Hemogram revealed normocytic normochromic anemia (hemoglobin: 6.9 g/dL; red blood cell count: $2.62 \times 10^6/\mu L$) with normal total leucocyte count (5600/cu mm) and normal platelet count (3×10^5/cu mm). Bone marrow studies (aspiration and biopsy) showed evidence of marrow hypocellularity. Skeletal radiographs showed mild increase in bone density, widening of the diploic spaces of the skull, sclerosis of the base of the skull, and mild diaphyseal widening and cortical hyperostosis of the femoral and tibial bones bilaterally (Figure 2). Ophthalmological evaluation revealed bilateral pseudo-proptosis with lagophthalmos. The salient findings of hypoplastic anemia and skeletal dysplastic changes suggested the diagnosis of Ghosal hematodiaphyseal dysplasia. Though the clinical findings also suggested the possibility of pycnodysostosis, the skeletal radiographic findings were not in favour of the diagnosis.

Next generation sequencing-based Exome sequencing was performed to look for variants in the *TBXAS1* gene (which has 17 exons of which 13 are coding exons) and to rule out variants in other genes associated with diaphyseal dysplasia and sclerosing bone disorders (including

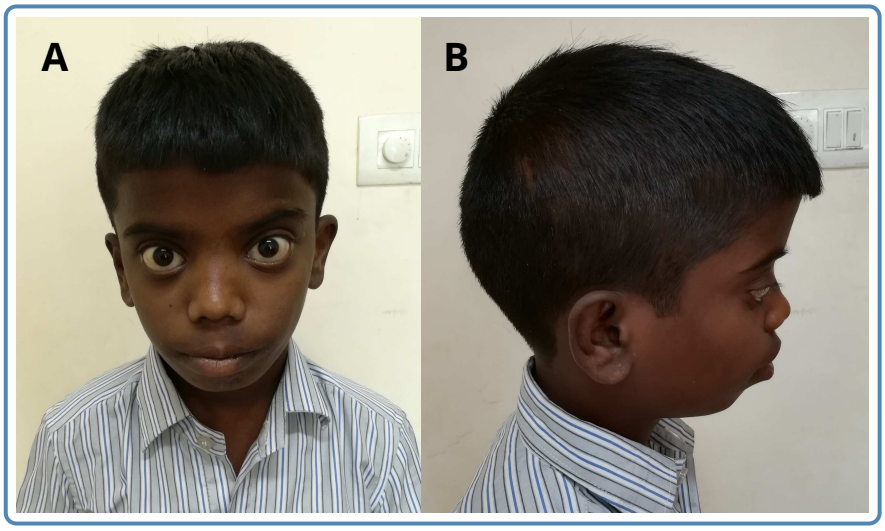


Figure 1 Photographs of the patient showing the craniofacial dysmorphic features. A. Frontal view showing telecanthus, bilateral proptosis, depressed nasal bridge, bulbous nose and thick lips. B. Lateral view showing dolichocephaly, depressed nasal bridge and retrognathia.

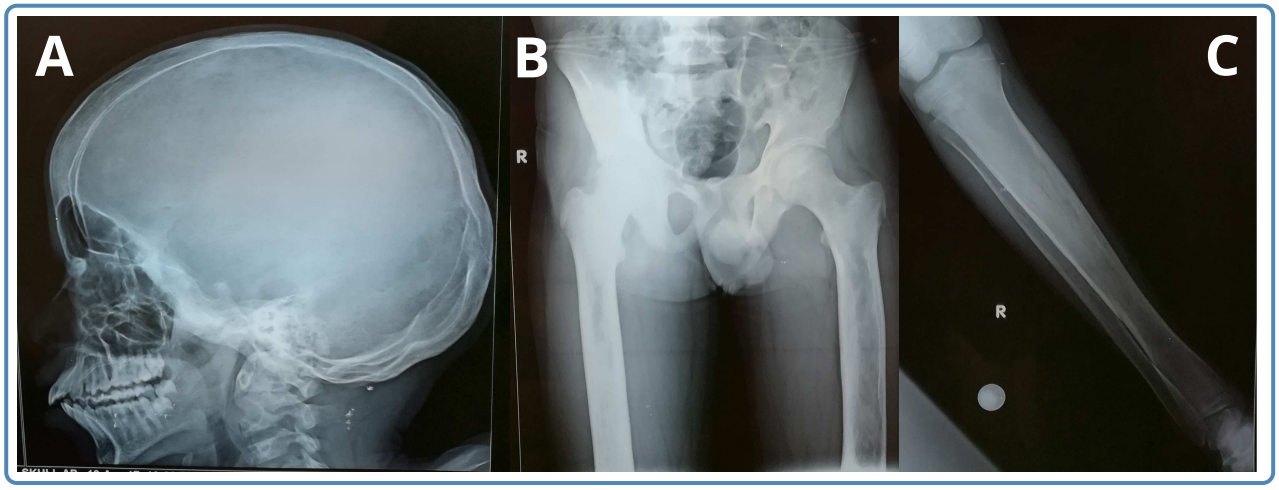


Figure 2 Skeletal radiographs of the patient. A. Lateral view of the skull showing the widening of the diploic spaces and sclerosis of the base of the skull. B. & C. Anteroposterior view of the pelvis, both femurs and the right tibia & fibula showing the increased bone density, diaphyseal widening and cortical hyperostosis.

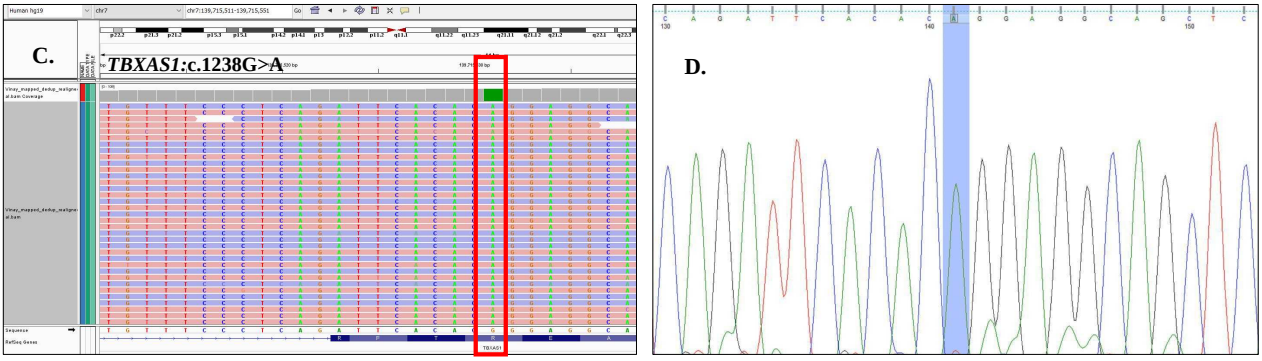
Camurati-Engelmann disease-associated *TGFB1* gene) and genes associated with bone marrow hypocellularity. A homozygous non-synonymous variant c.1238G>A (p.Arg413Gln) was identified in exon 15 of the *TBXAS1* gene (transcript id ENST00000263552; NM_001130966) (Figure 3). This variant has been previously reported in other patients of South Asian origin with Ghosal syndrome, and is listed in HGMD (http://www.hgmd.cf.ac.uk/ac/) and ClinVar (https://www.ncbi.nlm.nih.gov/clinvar/). The variant has minor allele frequencies of 0.001 and 0.00006 in the 1000 Genomes (https://www.internationalgenome.org/1000-genomes-browsers/) and gnomAD (https://gnomad.broadinstitute.org/) databases, respectively. It is classified as a 'likely pathogenic' variant, as per the American College of Medical Genetics and Genomics and Association for Molecular Pathology (ACMG/AMP) guidelines

A. Variant Interpretation

Gene (Transcript)	Location	Variant	Zygosity	Classification*	Disease (OMIM)	Inheritance
TBXAS1 (NM_001130966)	Exon 15	c.1238G>A (p. Arg413Gln)	Homozygous	Likely Pathogenic	GHOSAL HEMATODIAPHYSEAL DYSPLASIA; GHDD (# 231095)	Autosomal recessive

B. Database Information

1000 Genomes	gnomAD	dbSNP	In-house exome database†	HGMD	ClinVar	Mutation Taster	CADD score
0.001	0.00006369	rs199422117	Absent	CM080565	Variation 11889	Disease causing	32



*Evidence for classification of the variant as 'likely pathogenic' as per the ACMG/AMP 2015 guidelines: PM2 + PP3 + PP4 + PP5

†In-house exome database contains data from >1000 Whole exomes previously analyzed in our laboratory

Figure 3 A. Interpretation of the likely pathogenic variant identified in *TBXAS1* gene B. Information available in various population databases about the variant C. Integrative Genomics Viewer (IGV) image of the homozygous variant identified in the *TBXAS1* gene (NM_001130966: c.1238G>A) in the patient. D. Sanger sequence chromatogram of the patient showing the same homozygous variant.

(Richards et al., 2015). It has a CADD score (https://cadd.gs.washington.edu/) of 32 and is predicted to be disease-causing/ damaging by the Mutation Taster (http://www.mutationtaster.org/) and SIFT (https://sift.bii.a-star.edu.sg/) *in-silico* pathogenicity prediction programs. The presence of the homozygous variant was validated further through targeted PCR amplification and Sanger sequencing of exon 15 of the *TBXAS1* gene in the patient's DNA (Figure 3D), and both parents were confirmed to be heterozygous carriers of the same. No other significant gene variants matching the phenotype were detected.

The patient was thus confirmed to have Ghosal hematodiaphyseal dysplasia. Corticosteroid therapy was advised, but the patient was subsequently lost to follow-up and the response to therapy therefore could not be assessed.

Discussion

Ghosal hematodiaphyseal dysplasia was first reported by Ghosal et al. (1988) in five children with moderate to severe anemia and diaphyseal dysplasia. Cormier-Daire and co-workers identified the *TBXAS1* gene to be associated with this syndrome (Genevieve et al., 2008). *TBXAS1* codes for the thromboxane synthase enzyme, which is a component of the arachidonic acid cascade that produces thromboxane A2 (TXA2). Both the enzyme and TXA2 modulate expression of osteoprotegerin and RANKL (receptor activator of nuclear factor kappa-*B* ligand) which are involved in osteoclast-mediated bone resorption (Genevieve et al., 2008).

Only 13 mutations have been reported in the *TBXAS1* gene till date in the Human Gene Mutation Database (http://www.hgmd.cf.ac.uk/ac/).

The variant detected in our patient has been previously reported in other patients of South Asian origin, including a family of Pakistani origin (Genevieve et al., 2008) and a 3 years 9 months old Indian boy with severe anemia, hepatosplenomegaly and diaphyseal dysplasia (Jeevan et al., 2016). Our patient had chronic severe anemia, but had less severe skeletal findings and did not have visceromegaly.

Though a rare disorder, it is relatively easy to diagnose, as it has the typical combination of skeletal dysplastic changes (increased bone density, diaphyseal dysplasia, wide diaphyseal medullary cavities, cortical hyperostosis, metaphyseal changes) and hematological abnormalities (anemia, thrombocytopenia, less often leucopenia, hypocellular marrow, myelofibrosis). Molecular confirmation is essential to rule out other disorders with overlapping phenotypes particularly Camurati-Engelmann disease.

It is important to establish the diagnosis early and accurately, because the hematological abnormalities and skeletal changes have been found to respond very well to corticosteroid therapy and patients on low-dose long term maintenance therapy have been reported to maintain normal hemoglobin levels, with no requirement for blood transfusions (John et al., 2015).

This case report reiterates the importance of deep clinical phenotyping in the diagnosis and management of genetic disorders.

References

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