Bone Status in a Patient with Insulin-Like Growth Factor-1 Receptor Deletion Syndrome: Bone Quality and Structure Evaluation Using Dual-Energy X-Ray Absorptiometry, Peripheral Quantitative Computed Tomography, and Quantitative Ultrasoundography

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Haploinsufficiency of the insulin-like growth factor (IGF)-1 receptor (IGF1R) gene is a rare, probably under-diagnosed, cause of short stature. However, the effects of IGF1R haploinsufficiency on glucose metabolism, bone status, and metabolism have rarely been investigated. We report the case of a patient referred to our center at the age of 18 months for short stature, failure to thrive, and Silver–Russell-like phenotype. Genetic analysis did not show hypomethylation of the 11p15.5 region or uniparental disomy of chromosome 7. Growth hormone (GH) stimulation tests revealed GH deficiency, whereas IGF-1 was 248 ng/mL. r-hGH treatment showed only a slight improvement (from −4.4 to −3.5 SDS). At 10 years of age, the child was re-evaluated: CGH-array identified a het erotyzogous de novo 4.92 Mb deletion in 15q26.2, including the IGF1R gene. Dual-energy X-ray absorptiometry showed a normal bone mineral density z-score, while peripheral quantitative computed tomography revealed reduced cortical and increased trabecular elements. A phalangeal bone quantitative ultrasonography showed significantly reduced amplitude-dependent speed of sound and bone transmission time values. The changes in bone architecture, quality, and metabolism in heterozygous IGF1R deletion patients, support the hypothesis that IGF-1 can be a key factor in bone modeling and accrual.

Keywords: insulin-like growth factor-I receptor, insulin-like growth factor-I, bone metabolism, quantitative ultrasonography, peripheral quantitative computed tomography

INTRODUCTION

Insulin-like growth factor (IGF)-1 (IGF-1) and 2 (IGF-2) are major regulators of cell growth, proliferation, and death (1). IGF plays an important role in several tissues through the IGF receptor type 1 (IGF1R), a heterotetramer (αβ2) with intrinsic tyrosine kinase activity (1). Insulin-like growth factor-1 is a crucial factor in bone formation and remodeling via its actions on both osteoblasts and osteoclasts, and it is involved in the development of peak bone density during growth and bone loss in senile osteoporosis (2, 3). Furthermore, IGF-1 regulates glucose metabolism.
metabolism by modulating insulin sensitivity and secretion (4); it is also expressed in the central nervous system and is essential for brain development (5, 6).

Haploinsufficiency of the gene encoding the IGF1R may be a rare and under-diagnosed cause of short stature (7, 8). Both heterozygous deletion and point-inactivating mutations of the gene can lower IGF1R mRNA and protein expression, with partial IGF-1 resistance (7, 8).

Different clinical phenotypes, including microcephaly, mental retardation, and facial dysmorphic features, were observed in patients with IGF1R deletion, but these phenotypes are uncommon in IGF1R mutations (4, 6–21). Some of the phenotypical features described in the patients are likely to be attributable to the absence of one copy of the IGF1R gene, but other findings are linked to the haploinsufficiency of other genes included in the deletion (7, 14, 22). Heterozygous missense and nonsense mutations of IGF1R show similar effects on growth and development. However, IGF1R mutations and deletions seem to differ in vitro, probably because of a dominant-negative effect of the mutation, which could decrease the number of fully functional receptors by up to 25%, whereas haploinsufficiency would theoretically lead to a 50% reduction (15).

The first IGF1R mutation was described by Abuzzahab et al. These authors reported the case of two patients with a history of intrauterine growth restriction (IUGR), poor postnatal growth, and biochemical features of IGF-1 resistance (9). One patient was a compound heterozygote for point mutations in exon 2 of the IGF1R gene and the other carried a nonsense mutation (Arg59stop) of the same gene (9).

Walenkamp et al. described the case of a girl carrying a terminal 15.q26.2 → qter deletion. She displayed growth retardation, microcephaly, short stature, and elevated IGF-1 levels. She was treated with growth hormone (GH) with a good growth response and her final height was within the normal range (14).

Various aspects of IGF1R defects have been analyzed to date, but the effects of IGF1R haploinsufficiency on bone status and metabolism have not been reported.

So, in this report, we describe a female patient with a terminal deletion of chromosome 15, involving the IGF1R gene, who has been treated with r-hGH from the age of 4.5 years with a long-term follow-up. In this patient, we evaluated glucose metabolism and bone metabolism and status using dual-energy X-ray absorptiometry (DXA), peripheral quantitative computed tomography (pQCT), and quantitative ultrasonography (QUS).

As per institutional and national guidelines, no ethics approval was needed. Written informed consent was obtained from the parents before publication of this case report and any accompanying images.

**CASE REPORT**

The proband was a toddler referred to the Paediatric Auxoendocrinology Unit of Meyer Children’s University Hospital of Florence for short stature and failure to thrive. The patient was the first child of healthy, unrelated parents and was born after a 36-week gestation, complicated by unexplained (IUGR), by cesarean section. No behavioral or dietary problems affected the proposita’s mother during pregnancy. The Apgar score was 7-9. Birth weight was 1,930 g (-1.46 SD), length 41.6 cm (-2.07 SD), and head circumference 29.2 cm (-2.16 SD).

She was breastfed, but did not catch up after birth, showing failure to thrive and suffering gastroesophageal reflux symptoms from the first months of life. Her psychomotor development was slightly delayed: she sat upright at 7 months, began to walk independently at 16 months, and began to use language at 17 months.

At 18 months of age, she was referred to our Hospital: at physical examination, she showed dysmorphic features suggestive of Silver–Russell Syndrome: a triangular face, with a prominent forehead and micrognathia, clinodactyly V, inferior limb asymmetry (near 1.5 cm), and multiple hyperpigmented lesions on the body. Her length was 67 cm (-4.27 SD), weight 6.600 kg (-3.77 SD), and head circumference 44.5 cm (-2.00 SD). Her mother’s height was 169 cm (1.09 SD) and the height of her father was 176 cm (-0.16 SD); consequently, her target height was 166.5 cm (0.67 SD). Screening blood tests, including celiac, disease serological markers, and thyroid function, were normal; basal IGF-1 was in the upper normal range (248 µg/L; 97th percentile 250 ng/mL).

Bone age was delayed: 10 months at 18 months of age. She showed a normal female karyotype (46, XX), whereas the methylation study of the 11p15.5 region and the evaluation of uniparental disomy of chromosome 7 were both negative.

At the age of 4 years and 4 months, she was newly evaluated for a persistent and significant growth delay [length was 81.5 cm (-5.11 SD), weight 8.860 kg (-6.33 SD), and body mass index (BMI) 13.31 (-1.83 SD)].

An endocrine work-up was performed: free-thyroxin [(FT₄) 1.32 ng/dL, n.v. 0.86–2.12 ng/dL], thyroid-stimulating hormone [(TSH) 3.03 µUI/dL, n.v. 0.4–4.0 µUI/dL], cortisol (11.34 µg/dL, n.v. 5–25 µg/dL), adrenocorticotropic hormone [(ACTH) 27 ng/L, n.v. 09–52 ng/L], glucose (78 mg/dL, n.v. 55–110 mg/dL), and prolactin [(PRL) 127 µU/ml], were in the normal range. The electrolyte, venous blood gas, hemoglobin, total protein, serum albumin, coagulation profile, calcium, and phosphorous were also normal. The anti-tissue transglutaminase (TTG) test was negative.

Arginine [basal (GH) 1.99, peak 8.92 ng/mL] and clonidine (basal GH 0.48, peak 6.92 ng/mL) stimulation tests disclosed a GH deficiency; IGF-1 level was 243 µg/L. Bone age was significantly delayed: 1 year, 10 months at 4 years, 4 months of chronological age. r-hGH treatment was started at a dosage of 0.23 mg/kg/week; the auxological follow-up showed a slight improvement in the first year of r-hGH treatment (from -5.11 to -3.5 SD) (Figure 1).

During r-hGH therapy: IGF-1 was persistently between the 90th and 97th percentile for age and sex, with glycemia, FT₄, TSH, basal glycemia, basal insulin, and glycosylated hemoglobin (HbA1c) in normal ranges.

Therefore, in light of the unsatisfactory response to r-hGH treatment, a re-evaluation was performed at the age of 10 years and 10 months: height was 124.7 cm (-2.82 SDS), weight 21.900 kg (-2.93 SDS), BMI 14.08 (-2.14 SDS), and pubertal evaluation was B1 PH1 AH1. A re-testing of GH secretion confirmed low values of GH after arginine (basal GH 1.31, peak 7.76 ng/mL) and clonidine (basal GH 1.11, peak 7.23 ng/mL).
testing; the IGF-I level was 371 µg/L. Bone age was still significantly delayed: 7 years, 11 months at 10 years, 10 months of chronological age. A new extensive endocrine work-up gave normal results. The TTG test was negative. Since the presence of HbA1c was in the upper limit of normality, we performed an oral glucose tolerance test that disclosed a reduced glucose tolerance: fasting glucose was 83 mg/dL, 2-h glucose 181 mg/dL, fasting insulin 2.19 µU/mL, peak 54.2 µU/mL. 2-h insulin 54.0 µU/mL. The patient showed low leptin level (0.5 ng/mL, n.v. 1.0–12.0 ng/mL). At 11 years old, her intelligence quotient (IQ) was 108, even though the performance IQ was 85 and she exhibited some behavioral abnormalities.

**Genetic Analysis**

At 10 years and 10 months of age, CGH-array analysis was performed using the Agilent Human Genome CGH Microarray Kit 60 K (Agilent Technologies, Santa Clara, CA, USA). The CGH-array revealed a heterozygous deletion of chromosome 15, comprising 4.942 Mbp of the terminal part of its long arm (15q26.2-q26.3) involving several genes, such as IGF1R, ADAMTS17 (A Disintegrin-Like and Metalloproteinase with Thrombospondin Type 1 Motif, 17), CERS3 (Ceramide Synthase 3), ALDH1A3 (Aldehyde Dehydrogenase 1 Family, Member A3), and Chondroitin Sulfate Synthase 1 (CHSY1) ([Figure 2; Table 1](#)). The deletion was not present in the parents.
TABLE 1 | Genes involved in the 15q26.2-q26.3 deletion of the patient.

| Gene    | Possible effects on bone structure and quality                                                                 |
|---------|---------------------------------------------------------------------------------------------------------------|
| IGF1R   | Decreased bone quality, impaired cortical density, and increased trabecular density                           |
| ADAMTS17| BDNA. Homozygous mutation in the ADAMTS17 gene caused Weill–Marchesani-like syndrome                           |
| CERS3   | BDNA. Homozygous mutation in the CERS3 gene caused a form of congenital ichthyosis                              |
| ALDH1A3 | BDNA. Homozygous mutation in the ALDH1A3 gene caused a form of microphthalmia                                  |
| CHSY1   | BDNA. Mice lacking Chsy1 display chondrodisplasia and decreased bone density                                   |

**IGF1R**, insulin-like growth factor I receptor; **ADAMTS17**, a disintegrin-like and metalloproteinase with thrombospondin type 1 motif, 17; **CERS3**, ceramide synthase 3; **ALDH1A3**, aldehyde dehydrogenase 1 family, member A3; **CHSY1**, chondroitin sulfate synthase 1; **BDNA**, bone data not available.

**Bone Density and Structure Evaluation**

At the age of 10 years and 11 months, the patient underwent an evaluation of bone metabolism, density, and structure. Bone mineral density (BMD, g/cm²) was measured by DXA at the lumbar spine (L2–L4) (Delphi-A System, Hologic, Inc., Waltham, MA, USA) and expressed as z-scores. To estimate the volumetric density (bone mineral apparent density or BMAD), we used the formula of Kröger et al. (23). The BMD z-score, corrected for height, was 0.67; BMD at the lumbar spine was 0.631 g/cm² and the bone mineral content was 22.27 g.

Furthermore, we performed a pQCT of the left (non-dominant) radius at sites 4 and 66% using a Norland-Stratec XCT 3000 scanner (Stratec Medical, Pforzheim, Germany). As for the growth retardation of the patient, all bone size-dependent parameters (Total, Cortical, and MuscleCSA) were corrected for height (24, 25). We disclosed an imbalance between the trabecular and cortical bone, with an augmented trabecular component (318.4 mg/cm³, z-score 3.8) and a very low cortical density (727.8 mg/cm³, z-score −6.9) in relation to the age. The proband showed a normal total density value (321.3 mg/cm³, z-score 0.7) and a significantly reduced bone area for muscle area (31.2 mm², z-score −4.0) and for height (28.9 mm², z-score −4.1). The SSI polar (62.5 mm³, z-score −2.2) was significantly reduced. Fat and muscle components were also poorly represented (304 mm², z-score −1.8; and 1,376 mm², z-score −1.9, respectively) (Figures 3A–H).

Finally, the bone quality status was evaluated with a DBM Sonic 1200 device (IGEA Bone Profiler, Carpi, Italy) (24).
FIGURE 3 | Cross-sectional evaluation of trabecular bone mineral density (TrabBMD) (A), cortical bone mineral density (CrtBMD) (B), total density corrected for age (C), bone area corrected for height (D), muscle cross-sectional area (MuscleCSA) corrected for height (E), bone area corrected for MuscleCSA (F), fat cross-sectional area (FatCSA) corrected for height (G), and density-weighted polar section modulus (SSIp) (H). The gray squares in the panels (A,B,C,H) represent the bone age-adjusted values.
The evaluation showed a very low amplitude-dependent speed of sound (AD-SoS, 1.791 m/s; z-score ~3.85) and bone transmission time (BTT, 0.78 μs; z-score ~2.18) values. Since bone size could also influence QUS parameters (26), we created a height-adjusted z-score for AD-SoS.

The study of the bone metabolism showed a low 25(OH) vitamin D [25(OH)D] level (14.3 ng/mL, n.v. >20 ng/mL) and a moderately high parathyroid hormone level (51 pg/mL; n.v. <43 pg/mL). Total protein, serum albumin, calcium, and phosphorous levels were normal; however, osteocalcin (34.3 mg/ml; n.v. 55–135 mg/ml), bone alkaline phosphatase (30 IU/L; n.v. 39.4–346.1 IU/L), and urinary deoxypyridinoline concentrations (23.45 nM/mM creatinine; n.v. 30.3–54.7 nM/mM creatinine) were lower than reference values for sex and age.

DISCUSSION

We reported the case of a heterozygous, de novo DIsCUssIoN were lower than reference values for sex and age. (23.45 nM/mM creatinine; n.v. 30.3–54.7 nM/mM creatinine) 39.4–346.1 IU/L), and urinary deoxypyridinoline concentrations (23.45 nM/mM creatinine; n.v. 30.3–54.7 nM/mM creatinine) were lower than reference values for sex and age.

We reported the case of a heterozygous, de novo IGF-1 Receptor Deletion and Bone Status...
Furthermore, IGF-1 also exerts an anabolic action on muscle, increasing the protein synthesis and decreasing the protein breakdown (41, 42). In fact, IGF-1 elicits skeletal muscle cell proliferation and myocytes differentiation (42). These data may explain the poorly represented muscle component in our patient.

Finally, another interesting aspect is the role of the IGF-system on glucose metabolism and the possible effects of IGF1R haploinsufficiency on carbohydrate homeostasis, as shown by our case report. The IGF1R gene has sequence homology with the insulin receptor gene; both are transmembrane tyrosine kinase receptors. IGF-I also has structural homology with pro-insulin and has insulin-like metabolic effects, while GH has some effects that are antagonistic to those of insulin (43). IGF-1 is important in maintaining beta-cell mass by stimulating their proliferation and can enhance peripheral insulin sensitivity (44). Mohn et al. studied four members of a family carrying a novel nonsense mutation of the IGF1R gene. The defect was associated with a variable impairment of prenatal and postnatal growth. The authors also reported alterations in carbohydrate metabolism, ranging from normal glucose tolerance in the presence of insulin resistance to IGT and fasting hyperglycemia in association with both insulin resistance and impaired beta-cell function (4).

In conclusions, our study showed the presence of changes in bone architecture, quality, and metabolism in a heterozygous IGF1R deletion patient. The changes in bone metabolism due to the lack of action of IGF-1, a key in bone modeling and accrual, as occurring in a heterozygous IGF1R deletion patient, can be well evaluated through three different techniques (DXA, p-QCT, and QUS) assessing its effects on bone density and quality.

**AUTHOR CONTRIBUTIONS**

PP carried out the endocrinological evaluation, conceived the study, and participated in its design. EL carried out the clinical genetic diagnosis, conceived the study, and participated in its design. LC and AV participated in the endocrinological evaluation and in the design of the study. MM participated in the endocrinological evaluation and in the coordination of the study. SS carried out the endocrinological evaluation, conceived the study, and participated in its design and coordination. All authors have read and approved the final manuscript.

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