Aspectos genéticos y moleculares de la talla baja idiopática

Genetic and molecular aspects of idiopathic short stature

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• Idiopathic short stature is a condition in which the height of the individual is more than 2 SDS below the mean height for a given age, sex and population, in whom no identifiable disorder is present.

• It can be subcategorized into familial and non-familial ISS, and according to pubertal delay.

• It should be differentiated from dysmorphic syndromes, skeletal dysplasias, small for gestational age, and systemic and endocrine diseases.

• ISS is the diagnostic group that remains after excluding known conditions in short children.

Wit JM, Clayton PE, Rogol AD, Savage MO, Saenger PH, Cohen P.
• Growth hormone - insulin-like growth factor I (GH - IGF-I) axis - *GHR, IGF-ALS*……

• *SHOX*

• *NPR2*

• *NPPC (CNP)*

• *ACAN*
• Located on the pseudoautosomal region 1 (PAR1) of the X and Y chromosomes

• Encodes a homeodomain transcription factor

• Expressed in chondrocytes of the human growth plate

• Involved in determining stature in humans
Madelung deformity

Langer mesomelic dysplasia (LMD)
(-9 to 6.2 SD)

Léri-Weill dyschondrosteosis (LWD)
(-4.6 to +0.6 SD)\(^1\)

Idiopathic short stature (ISS)
(< -2 SD)\(^2\)

Turner syndrome

Trisomy X

Klinefelter syndrome

XYY syndrome

12 30 Atif SHOX region
Mutations (1/3)

(Belin et al, 1998; Shears et al, 1998)

Tel

SHOX

Cent

Complete SHOX deletions (2/3)

(Rao et al, 1997; Ellison et al, 1997)

- ~ 50% of LWD cases
- ~ 80% of LMD cases
- ~ 2.5% of ISS cases
Three enhancers identified downstream of SHOX

Fukami et al 2006, Am j Hum Genet 78:1768-8

Sabherwal et al, 2007; Hum Mol Genet 16(2):210-222
Three enhancers located upstream of SHOX

**Upstream enhancer deletions**
Benito-Sanz et al, Eur J Hum Genet 2012

**Upstream enhancer duplications**
Benito-Sanz et al, J Clin Endocrinol Metab 2011
Recurrent deletion in 30 patients (19 LWD & 11 ISS)
Array CGH

log2 ratio

Chr Y

~47.5 kb

700549:748093

Tel - T G C T C T C C A A C A G T A T C T C C C G A T - Cent

Deletion 47543 bp

LINE (L1P12)/no repetitive sequence

Recurrent deletion – NHEJ/unknown mechanism

(NCBI36/hg18)
Chromosome Conformation Capture (3C) analysis of ECRs in Chicken Limb Embryos

Luciferase assays in U2OS cells

Recurrent ~47.5 kb enhancer deletion

Identification of 4th downstream SHOX enhancer – ECR1
TABLE IV. The 17 Patients With a 47.5 kb Deletion 160 kb 3’ of the \( SHOX \) Gene, Showing the Genotype (Mut, mutation; PM, point mutation) and Phenotype (Phe, Phenotype; Hgt, Height in cm; SD, Standard Deviation of Height From the National Mean; Meso, Mesomelia; Mad, Madelung Deformity; Bilat, Bilateral) of the Proband and Both Parents Where Available, Plus the Country of Referral for All Probands

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No mutation has been identified in this region in 126 possible LWD with no SHOX defect.

But virtually all enhancer deletions include the ZED enhancer, i.e., not only the recurrent 47.5 kb deletion.
PAR1-SHOX

(-9 to 6.2 SD) (-4.6 to +0.6 SD) (< -2 SD) (-2 to +2 SD) (+>2 SD)

Langer mesomelic dysplasia (LMD)
Léri-Weill dyschondrosteosis (LWD)
Idiopathic short stature (ISS)
Normal stature
Trisomy X
Klinefelter syndrome
XYY syndrome
Partial *SHOX* duplications and small duplications of CNE9 enhancer are highly penetrant in ISS and LWD.
Normal

SHOX deficiency

No SHOX deficiency if complete duplication in tandem or on another chromosome
Pseudoautosomal region 1 (PAR1)

**Upstream enhancer duplication**
Benito-Sanz et al, J Clin Endocrinol Metab 2011

**Downstream enhancer deletions (Region 2)**
Benito-Sanz et al, Am J Hum Genet 2005

**Upstream enhancer deletions (Region 3)**
Benito-Sanz et al, Eur J Hum Genet 2012

**Non-pathogenic CNVs**

**Recurrent ~47.5 kb enhancer deletion**

**Partial and complete SHOX deletions**
Benito-Sanz et al, Am J Hum Genet 2006
Benito-Sanz et al, J Hum Genet 2016

**Partial and complete SHOX duplications**
Benito-Sanz et al, J Clin Endocrinol Metab 2011

**Downstream enhancer duplication**
Hirschfeldova et al, 2011

**Region 4 Del/Dup**
Tsuchiya et al, 2012
Bunyan et al, 2014
Benito-Sanz et al, unpub
Craniosynostosis + others
Idiopathic short stature

Plasma membrane

FGFR3

RAS

RAF

PRKG2

MEK1/2

ERK1/2

Transcription

ACAN

FGFR3

NPPB

Léri-Weill dyschondrosteosis (LWD)
Langer mesomelic dysplasia (LMD)

Noonan syndrome

CNP>>>BNP

CNP>BNP

NPRB

NPRC


NPR2
NPR2 → Acromesomelic dysplasia, type Maroteaux (<5 SDS)

NPR2
NPR2 → 3% Idiopathic short stature (ISS)

NPR2
NPR2* → Extremely tall stature with
• Homodimer receptor.
• Union with its ligand, CNP causes a post-receptor signalling cascade.
• Expressed in cartilage, growth plate, brain, hypothalamus.
Detected 8 variants in *NPR2* en 9 patients; 7 LWD and 2 ISS.
Se han detectado un total de 8 variantes en NPR2 en 9 pacientes; 7 DLW y 2 TBI.

**A**

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**Cellular localization – ability to transport to ER**

A: DAPI, ER, NPR-B, MERGE

B: ER, ER, ER, ER, ER, ER, ER, ER, ER
• Seven mutations were confirmed to be pathogenic.

• Approx 3% of suspected LWD cases had a pathogenic NPR2 mutation. No ISS patient had a mutation.

• Similar phenotype to those referred for SHOX screening but without the Madelung deformity:
  • Short stature with limb shortening
  • Height:armspan < 0.965
  • Brachydactyly
  • Frontal bossing
  • Muscular hypertrophy
  • High-arched palate
Craniosynostosis + others

Idiopathic short stature

~3% Disproportionate short stature
~3% Idiopathic short stature (ISS)

Noonan syndrome

Léri-Weill dyschondrosteosis (LWD) ~ 70%
Langer mesomelic dysplasia (LMD) ~100%
Cohort:
697 patients:
• 357 with disproportionate short stature and mild skeletal defects
• 340 with ISS

Methods:
• Whole exome sequencing
• HRM & Sanger sequencing of candidate genes
Family 1: c.349C>G (p.Arg117Gly)

Family 2: c.55G>T (p.Gly119Cys)

HRM WES Sanger

Variants in patient II.1 (SNVs and In/dels) (n = 55,081)
Located in exons or splice site (n = 7,207)
Low frequency in public database MAF <0.001 (n = 963)
Absent in our internal database (n = 350)
Absent in her brother (II.2) and present in her father (I.1) (n = 71)
Predicted to be deleterious (nonsynonymous) or to be loss of function (n = 15)
Height = SDS

Proportionate short stature and small hands

Hand length (SDS)

Family 1
III.1 (M) II.5 (M) II.4 (N) I.3 (N) I.4 (M) II.6 (M) III.2 (N)

-3.7 -4.5 -1.1 -4.2** -5.2 -6.0 -1.4
Short stature and short hands
abnormality" (Ibab/Ibab) with CNP p.Arg117Gly mutation

Ibab/Ibab mice – 60% smaller than WT

CNP-22 rescues phenotype

Hmz and htz Ibab mice have a reduction in the
Both CNP variants lose their capacity to synthesize cGMP in the homozygous state, and a reduction in the heterozygous state, therefore confirming their pathogenicity.
Proportionate short stature and small hands

Bone age

Anastraz

T2

rhGH

M/N

-3.96 SDS

7.5y

16y

1.95

2.36

1.48

-4.54

-3.70

-1.95

-1.48

M/N
Craniosynostosis + others  

Idiopathic short stature  

0.6% Short stature & small hands

Idiopathic short stature (ISS)

~3% Disproportionate short stature

Léri-Weill dyschondrosteosis (LWD)

Langer mesomelic dysplasia (LMD)
• Spondyloepiphyseal dysplasia, Kimberley type (SEDK) – AD

• Familial osteochondritis dissecans – AD

• Spondyloepimetaphyseal dysplasia, aggrecan type (SEMD) - AR

Tompson et al, 2004;
14 mild to moderate short stature and brachydactyly (missense, nonsense and frameshift). Those with osteoarthritis and/or discopathy.

2 Spondyloepiphyseal dysplasia, Kimberley type (SEDK)
Median height SDS in children was -2.91 (range -4.30/-1.86 SDS)
Median height SDS in adults was -3.77 (range -5.40 SDS/-1.79 SDS)
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<th>SGA</th>
<th>Range of height (SD)</th>
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<th>Adults</th>
<th>Advanced BA in children</th>
<th>Frontal bossing</th>
<th>Flat nasal bridge</th>
<th>Mid facial hypoplasia</th>
<th>Brachydactyly</th>
<th>Short thumbs and short first metacarpals</th>
<th>Broad great toes</th>
<th>Hyperlordosis</th>
<th>Hip anomalies</th>
<th>Mild osteochondral defects</th>
<th>Knee defects</th>
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Craniosynostosis + others
Idiopathic short stature (ISS)

Disproportionate short stature
Idiopathic short stature (ISS)

Léri-Weill dyschondrosteosis (LWD)
Langer mesomelic dysplasia (LMD)
Idiopathic short stature is a condition in which the height of the individual is more than 2 SDS below the mean height for a given age, sex and population, in whom no identifiable disorder is present.

It can be subcategorized into familial and non-familial ISS, and according to pubertal delay.

Many patients with ISS are actually often the mild forms of various skeletal dysplasias, short stature secondary to a small birth size (small for gestational age, SGA), and systemic and endocrine diseases.

ISS is the diagnostic group that remains after excluding known conditions in short children.

Wit JM, Clayton PE, Rogol AD, Savage MO, Saenger PH, Cohen P.
Group members
Sara Benito-Sanz
Miriam Aza-Carmona
Alfonso Hisado-Oliva
Lucia Sentchordi-Montañé
Jimena Barraza-García
Carlos Rivera-Pedroza
Alba Ruzafa-Martín
María Rodríguez-Zabala
Beatriz Paumard-Hernández
David Medino
Carolina de la Torre