7th JOINT MEETING
In pediatria e medicina dell'adolescenza
Sobre pediatría y medicina de la adolescencia
21-22-23-24 OTTOBRE 2015 CATANZARO ITALIA

EFFICACIA A LUNGO TERMINE DEL TRATTAMENTO CON GH NEI BAMBINI CON DEFICIT DEL GENE SHOX

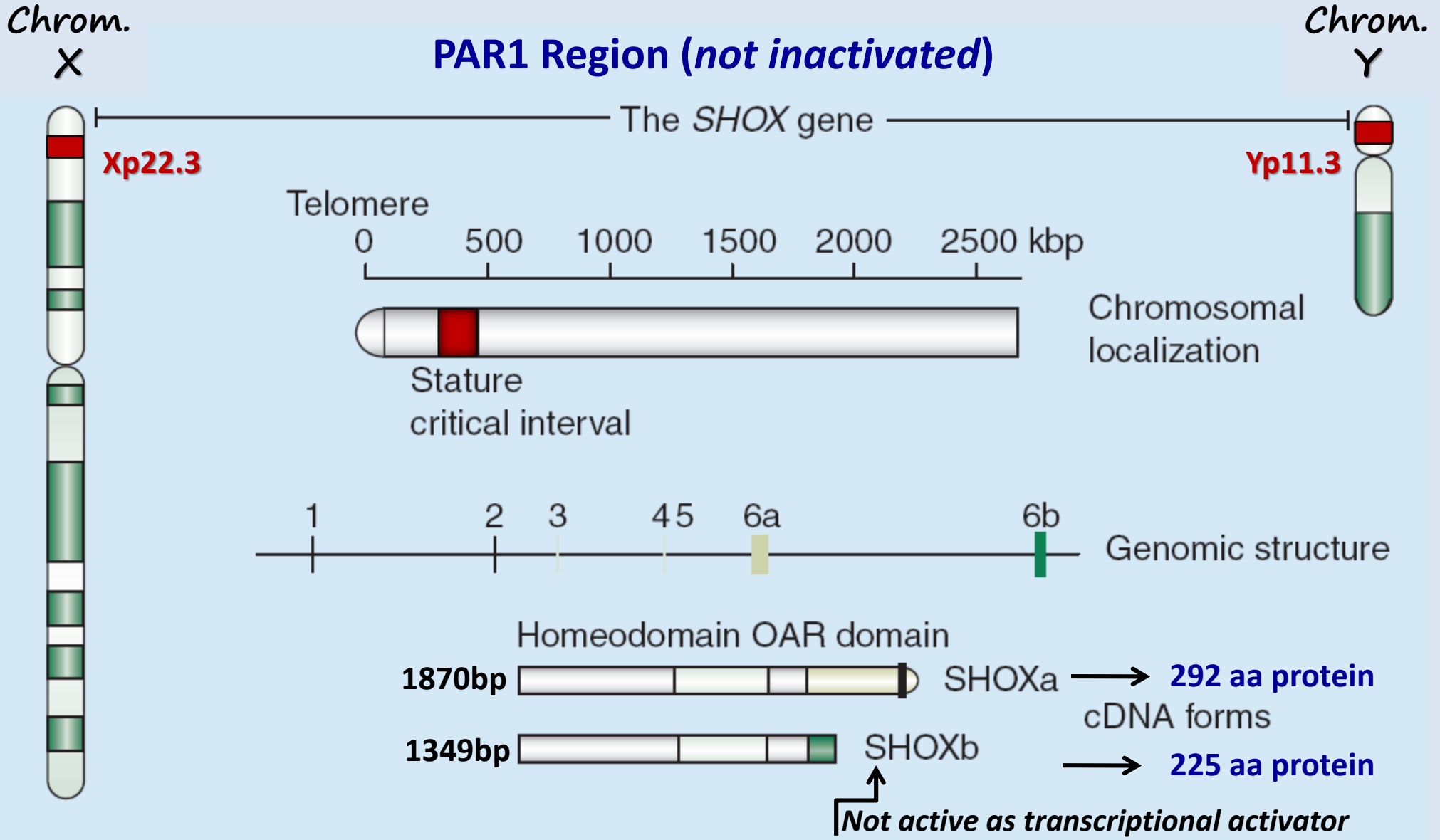
S. Bertelloni

Azienda Ospedaliero Universitaria Pisana

SST Servizio Sanitario della Toscana

SIRPed Società Italiana di Ricerca Pediatrica

SHOX* GENE: SCHEMATIC REPRESENTATION



*Short stature Homeobox on X chromosome

(Iughetti et al, Expert Rev Endocrinol Metab. 4: 2009)

SHOX EXPRESSION & ACTIONS

Shox gene:

- is expressed in the ulna, radius, elbow and wrist, the equivalent bones in the leg & in the first and second pharyngeal arches.

SHOX protein:

- functions as a repressor of growth plate fusion & skeletal maturation in the distal limbs;
- counteracts the skeletal maturing effects of gonadal steroids (estrogens).

(Leka et al, Hormones 2006; Bider HRP, 2011)

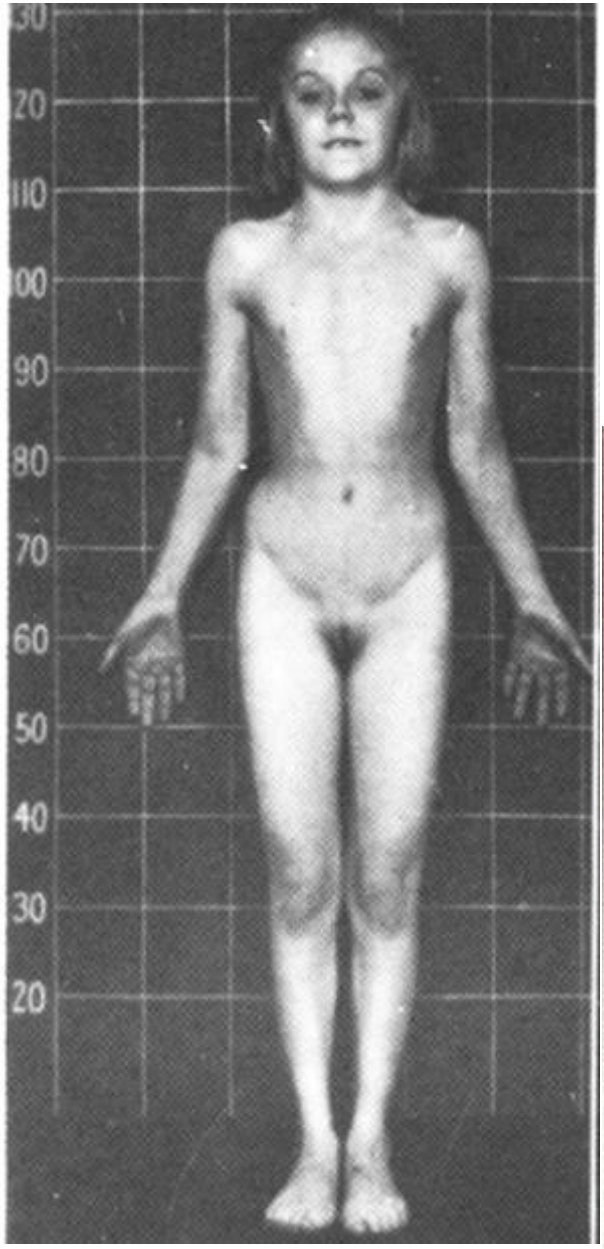
SHOX Gene & Short Stature: Objectives

- To define SHOX gene structure & function.
- To characterize clinical disorders related SHOX gene deficiency (*haploinsufficiency*).
- To discuss the effects of GH treatment on linear growth in SHOX gene deficiency.

SHOX deficiency: Associated Clinical Disorders

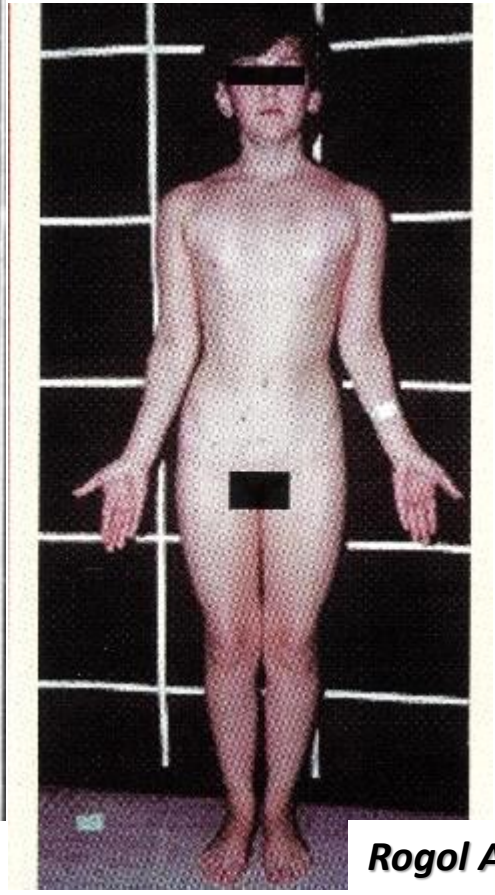
- Chromosomal disorders:
  - Turner syndrome (45,X and variants; female phenotype);
  - 45,X male (SRY+; male phenotype);
  - 45,X/46,XY mosaicism (male or ambiguous phenotypes).
- Gene disorders:
  - Langer mesomelic dysplasia (LMD) [biallelic deficiency (homozygous or compound heterozygous)];
  - Leri-Weill dyschondrosteosis (LWD) (monoallelic deficiency);
  - "Idiopathic" short stature (monoallelic deficiency).

TURNER'S SYNDROME: CLINICAL FEATURES



Ruth

- Age, yrs: 16.9
- Height, cm: 130
- Height age, yrs: 8.5
- Bone age, yrs: 14.5

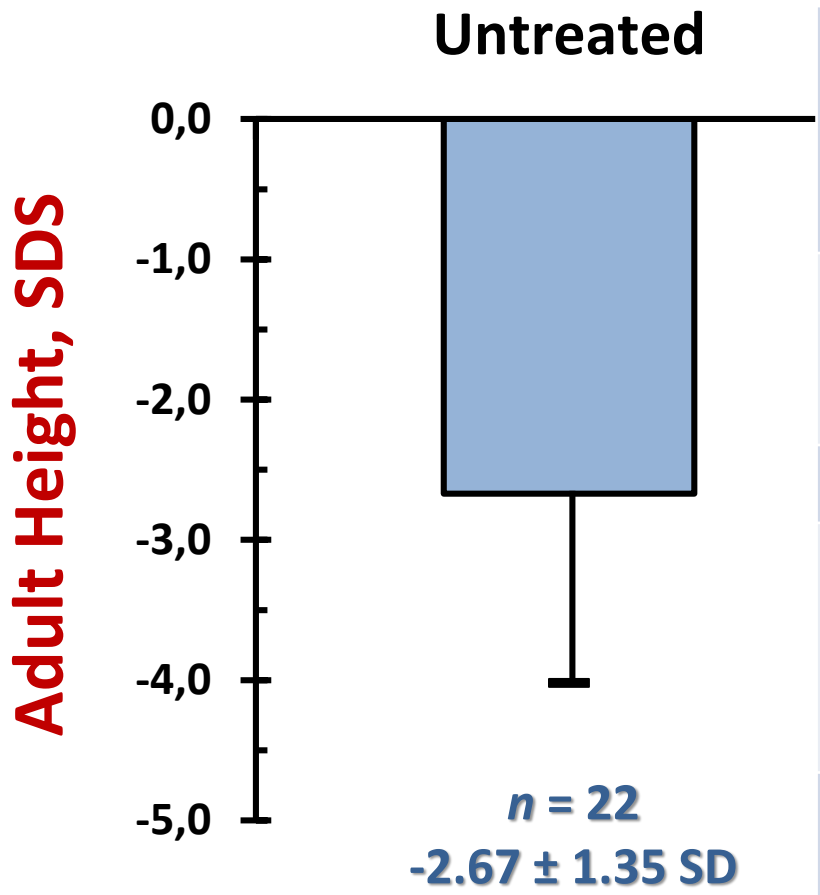


Rogol A, 1996

Feature*	Frequency (%)
SHORT STATURE	98
Pubertal failure	95
Micrognathia	60
Cubitus valgus	47
Low posterior hairline	42
Short neck	40
High arched palate	38
Short fourth (fifth)metacarpal	37
Multiple naevi	25
Webbed neck	25
Lymphedema (hands, feet)	22
Nail dysplasia	13
Scoliosis	11
Madelung deformity	7

**Clin.Ped/Lab. Gen. JPEM 2003 (n = 58)*

Statural Growth of Males with 45,X/46,XY Mosaicism: A Review*



	MPH	AH, cm	AH, SD	Phenotype
Kojima *et al.* (2007)	—	153.0	**-3.5**	Hypodpadias
	—	150.0	**-4.1**	Hypospadias
	—	155.0	**-3.1**	Hypospadias
	—	150.0	**-4.1**	Hypospadias
Layman *et a*l. (2009)	—	144.1	**-4.5**	Turner stigma
	—	170.0	**-1.0**	**Normal male**
	—	167.0	**-1.4**	**Normal male**
Lara et al. (2008)	175.8	158.4	**-2.1**	**Normal male**
Tosson *et al.* (2010)	188.2	167.4	**-1.3**	Hypospadias
	185.2	165.5	**-1.6**	**Normal male**
	169.7	149.7	**-3.7**	**Soft testes**
	180.7	158.2	**-2.6**	**Small testes**
Martinerie *et al.* (2012)	—	154.0	**-3,5**	Hypospadias
	—	146.0	**-4.0**	Hypospadias
	—	152.0	**-3.8**	Hypospadias
	—	160.0	**-2.5**	Hypospadias
	—	171.0	**-0.7**	Hypospadias
Personal observations	172.5	162.5	**-2.1**	Amb. genitalia
	175.5	153.0	**-3.6**	Amb. genitalia

*Raw SD data were drawn from authors' published data or calculated from absolute values (in cm) according to male sex and country of origin.

(Sex. Develop. 2015, in press)

Sex Assignment in Babies with 45,X/46,XY Mosaicism

EuroDSD

SEVENTH FRAMEWORK PROGRAMME

EuroDSD is a Collaborative Project funded by the European Commission under the 7th Framework Programme

	Male	Female	Male, %[a]
Pre-1990			
n	17	40	30
EMS, median (range)	5 (0–12)	0[b] (0–6)	
1990–1999			
n	31	53	37
EMS, median (range)	6 (0–12)	0[b] (0–6)	
Post-1999			
n	54	37	59
EMS, median (range)	6 (0–12)	0[b] (0–6)	

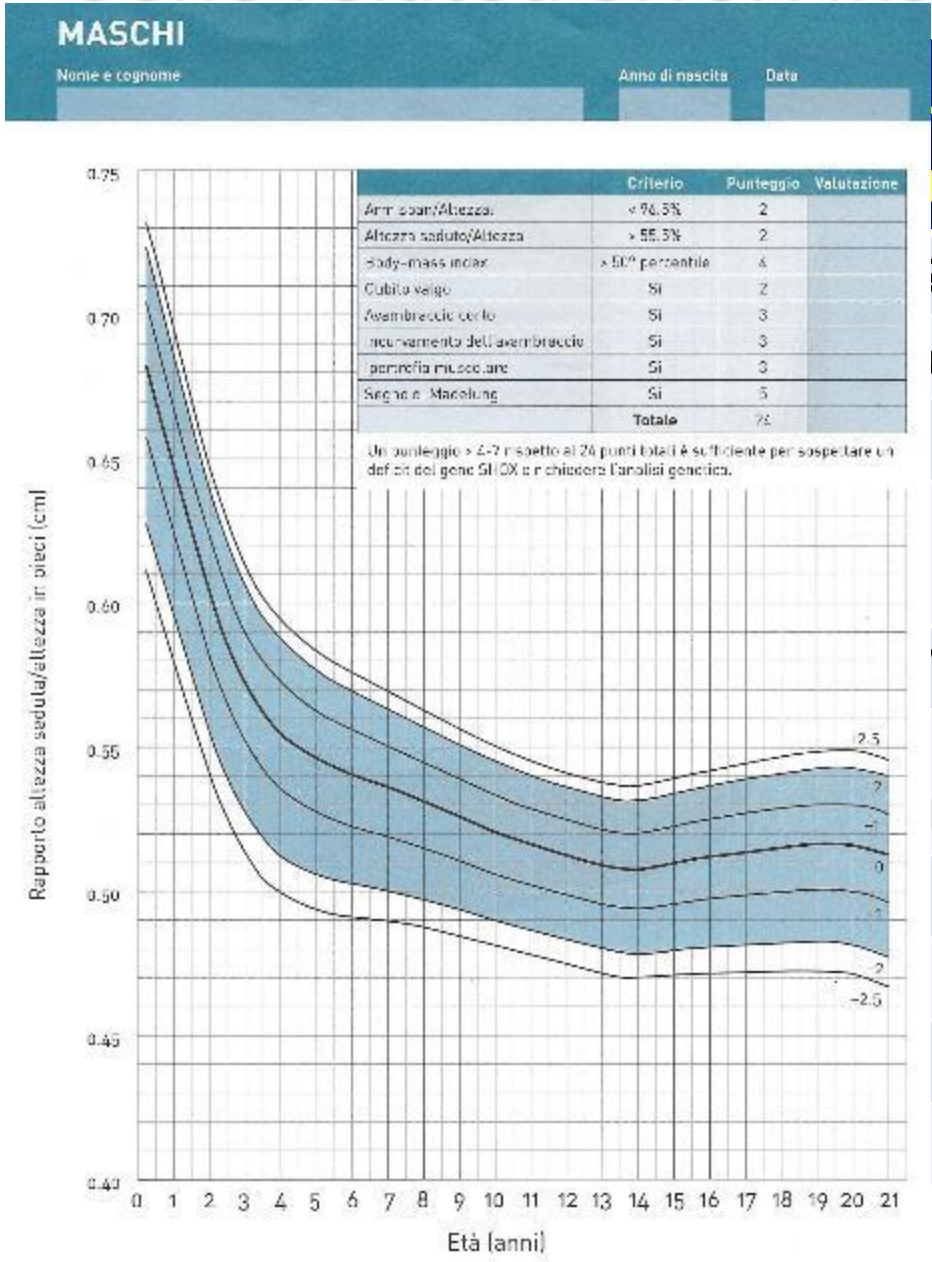
EMS = external masculinization score

Pediatrics 2014; 134:e710–e715

SHOX deficiency: Associated Clinical Disorders

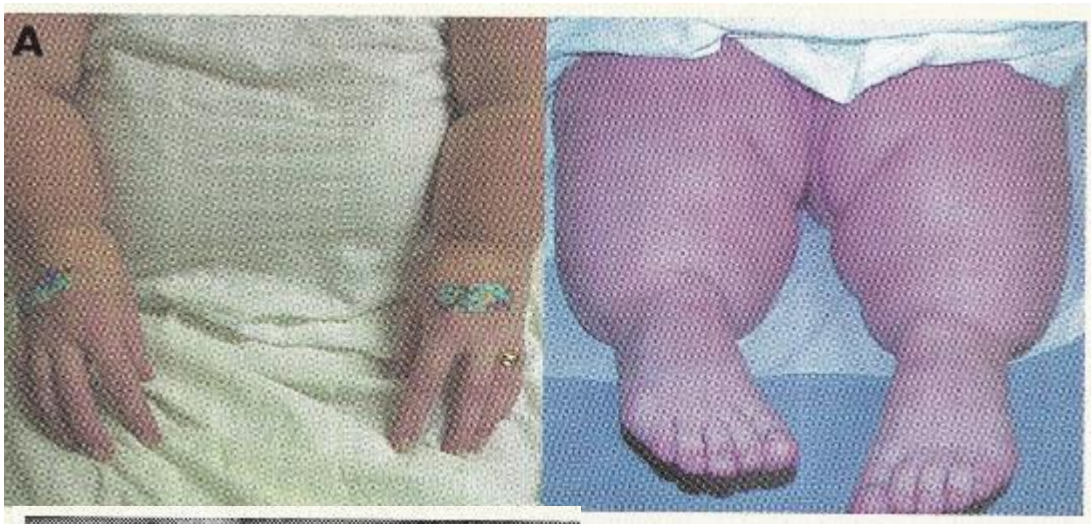
- Chromosomal disorders:
  - Turner syndrome (X0, female phenotype);
  - 45,X male (SRY+; male phenotype);
  - 45,X/46,XY mosaicism (male, female or ambiguous phenotype).
- Gene disorders:
  - Langer mesomelic dysplasia (LMD) [biallelic deficiency (homozygous or compound heterozygous)];
  - Leri-Weill dyschondrosteosis (LWD) (monoallelic deficiency);
  - "Idiopathic" short stature (monoallelic deficiency).

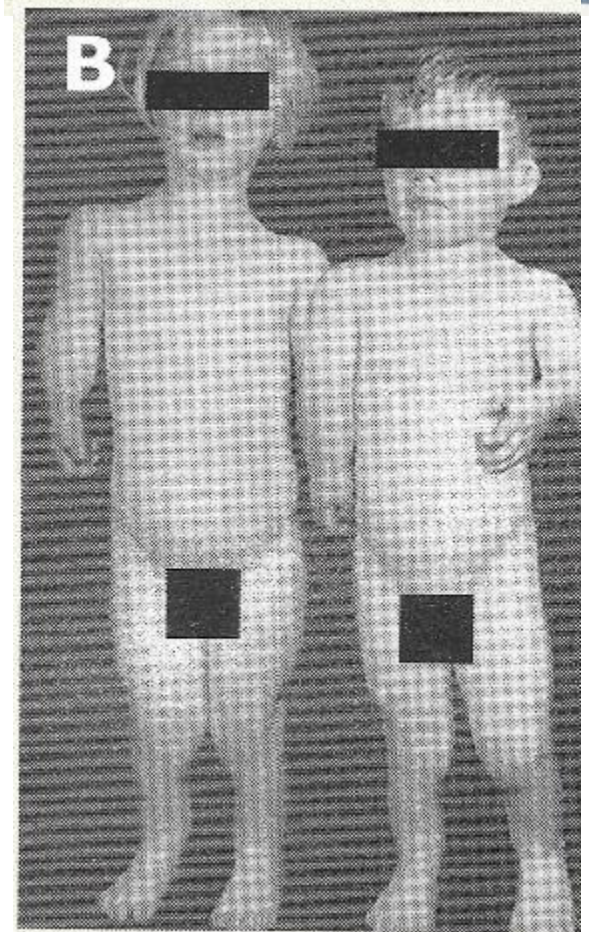
Gene related SHOX Insufficiency: Clinical Disorders



Langer's Mesomelic Dwarfism	Léri-Weill Dyschondrosteosis
? (~30 pts)	1: 2000 – 1: 4000
us deficiency (100%)	Haploinsufficiency (60-90%)
omal dominant	Autosomal dominant
-6 SDS	F: 145 cm (-2.1 → -3.9 SDS) M: 155 cm (-2.0 → -3.0 SDS)
Standing H > 2 SDS	Sitting H/Standing H > 2 SDS
Severe imbs > forearms)	Present (variable)
Severe	Present (variable)
Severe	Present (females > males)
	Present (females > males)
?	Present (mainly males)

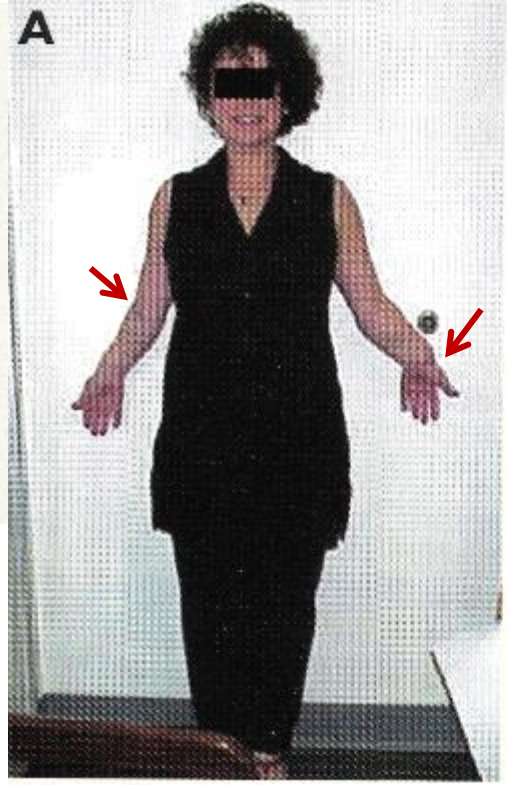
Leka et al., Hormones 2006; 5: 107-118

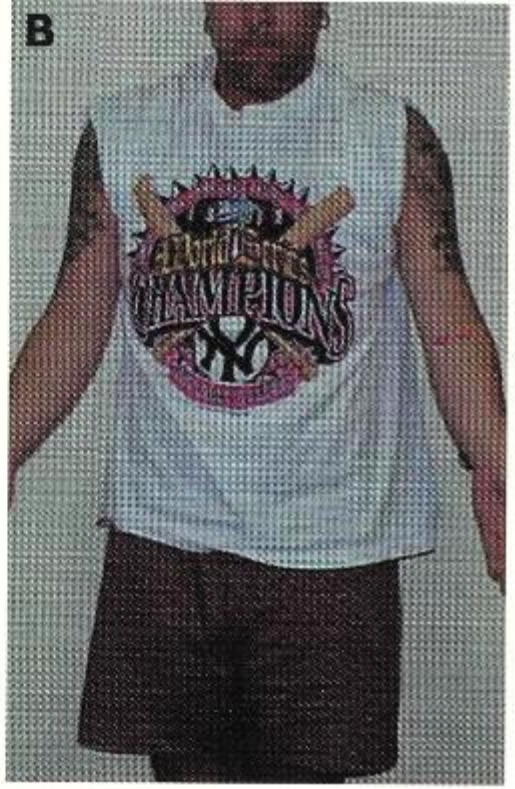




Children with Langer Mesomelic Dwarfism (homozygous SHOX deficiency)

Observations Judith Ross & Blockley Lawrie

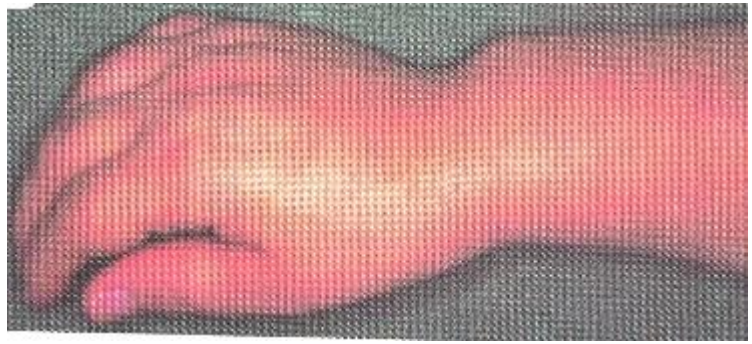




141.5 cm
-3.6 SDS

167.1 cm
-1.6 SDS

Adults with Léri-Weill dyschondrosteosis (SHOX haploinsufficiency)

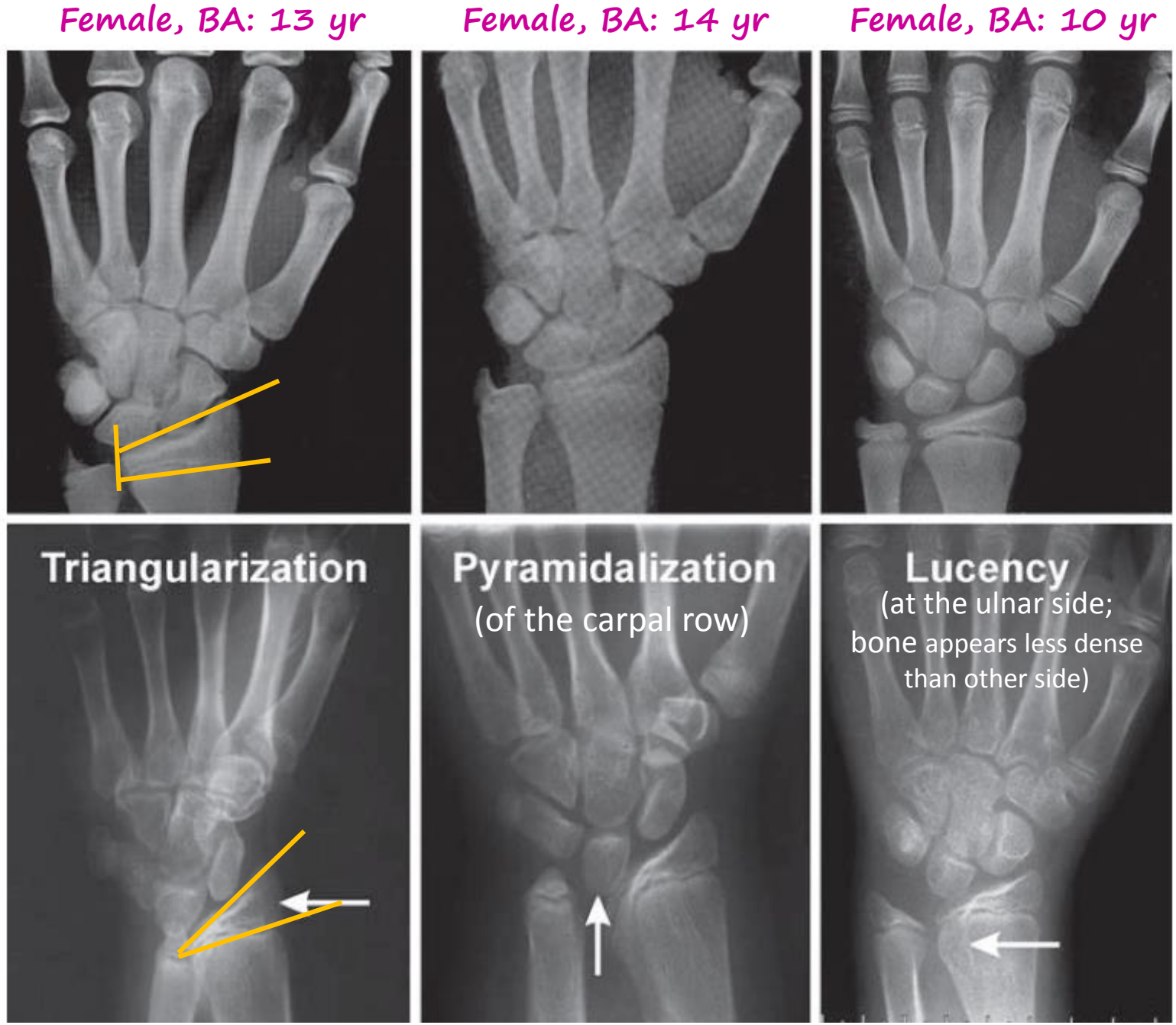


Observations Judith Ross

SHOX Deficiency (2): RX Features

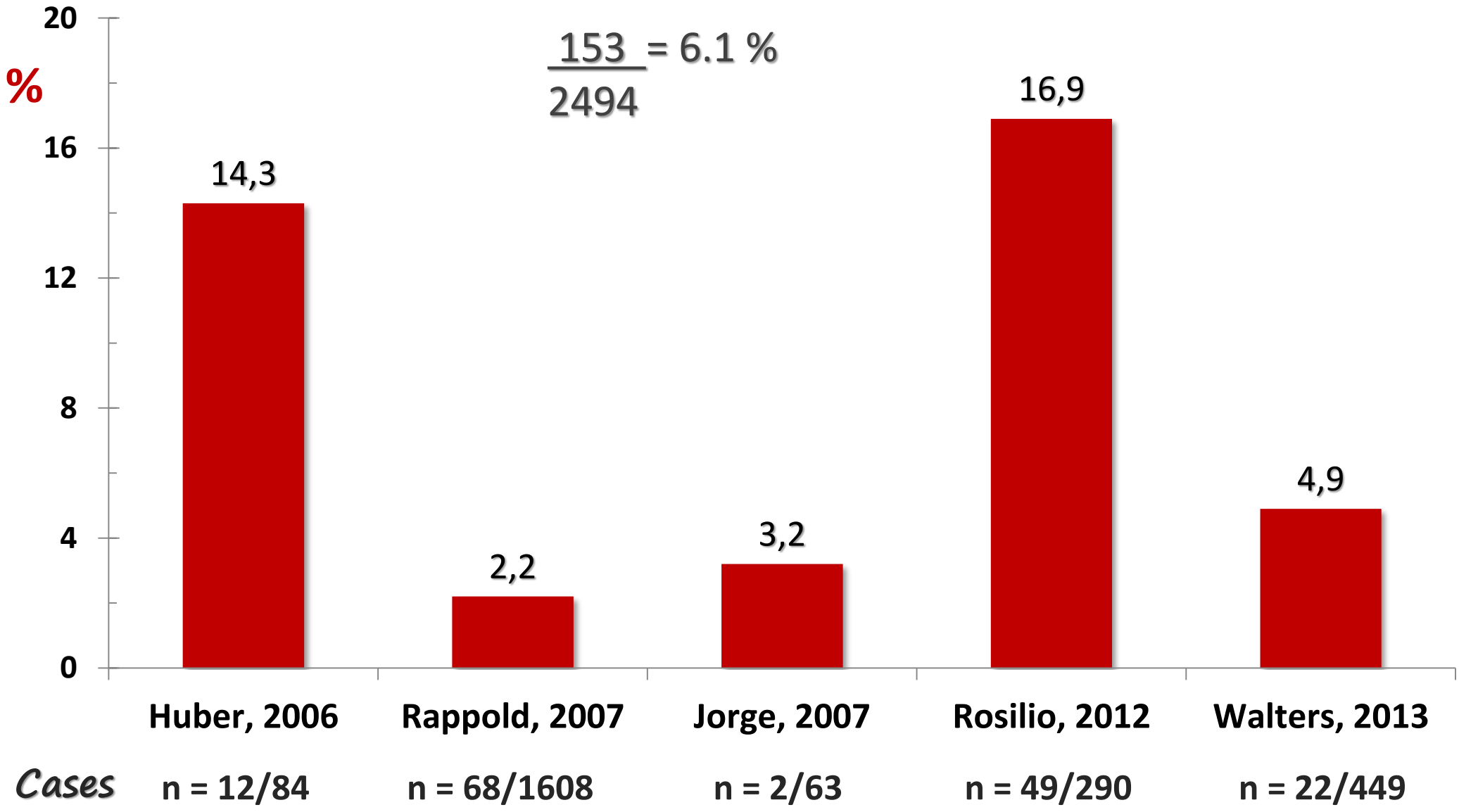
Normal hand X-rays

Leri-Weill syndrome

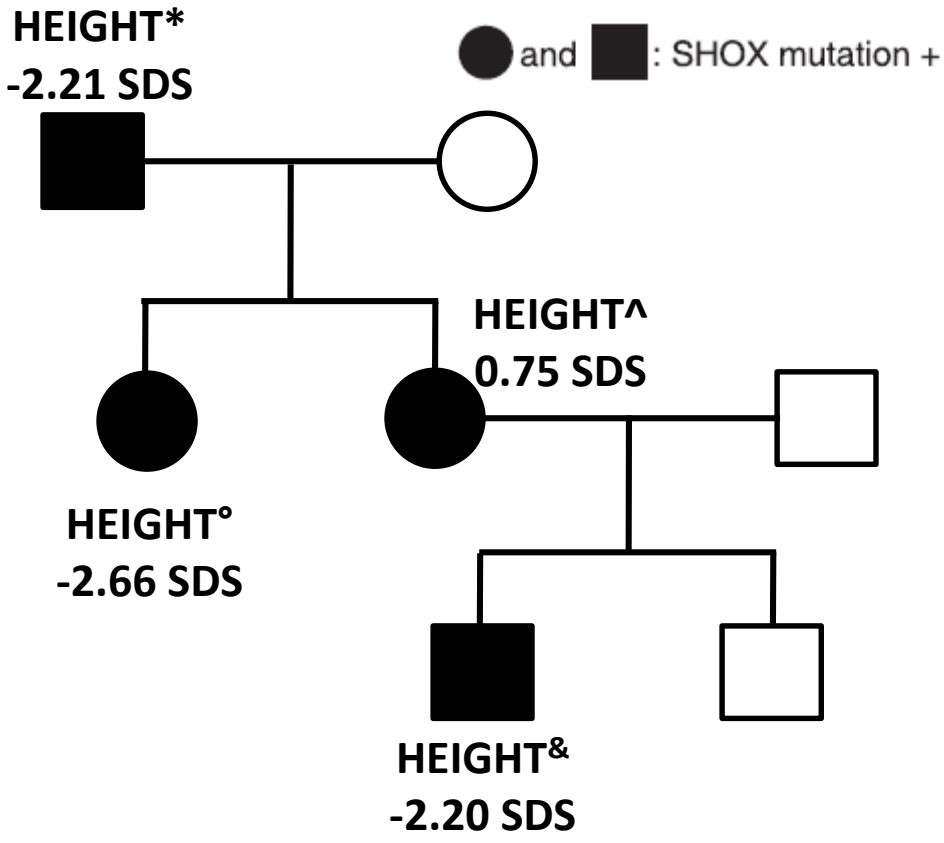


(Binder, HRP 2011)

“Idiopathic” Short Stature & SHOX Deficiency



SHOX DEFICIENCY: CLINICAL VARIABILITY



*Madelung deformity, mesomelia (arms)

°Madelung deformity (severe), cubitus valgus, short metacarpals

^Madelung deformity (mild)

&<u>Only short stature</u> (age 4 yrs)

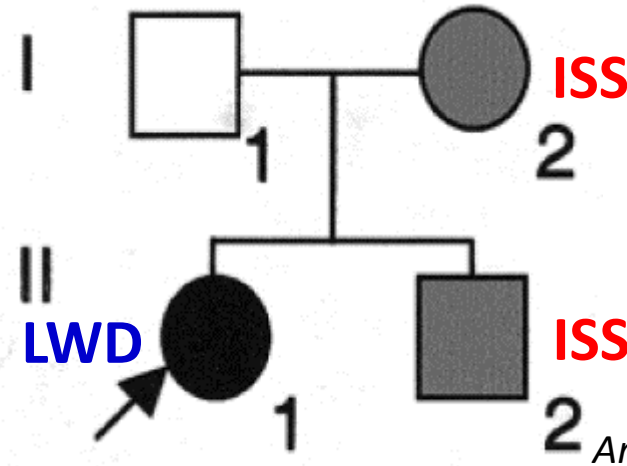
AOUP - Pisa

Clinical characteristics of children and adults with molecular *SHOX* defects

	Children	Adults	*P*-value
Male : female	7 : 7	8 : 9	ns
Height SDS	− 1·9 ± 0·9	− 2·9 ± 0·8	< 0·01
Height SDS in male	− 2·2 ± 0·9	− 2·9 ± 0·6	ns
Height SDS in female	− 1·7 ± 1·1	− 3·0 ± 0·9	< 0·05
Height SDS > −2	36%	6%	ns
Sitting height/height SDS for age	+3·3 ± 0·6	+3·1 ± 1·3	ns
Sitting height/height SDS > +2	100%	82%	ns
Presence of Madelung deformity	50%	47%	ns

ns, Not significant.

*Jorge et al, Clin Endocrinol (2007) **66:** 130–135*



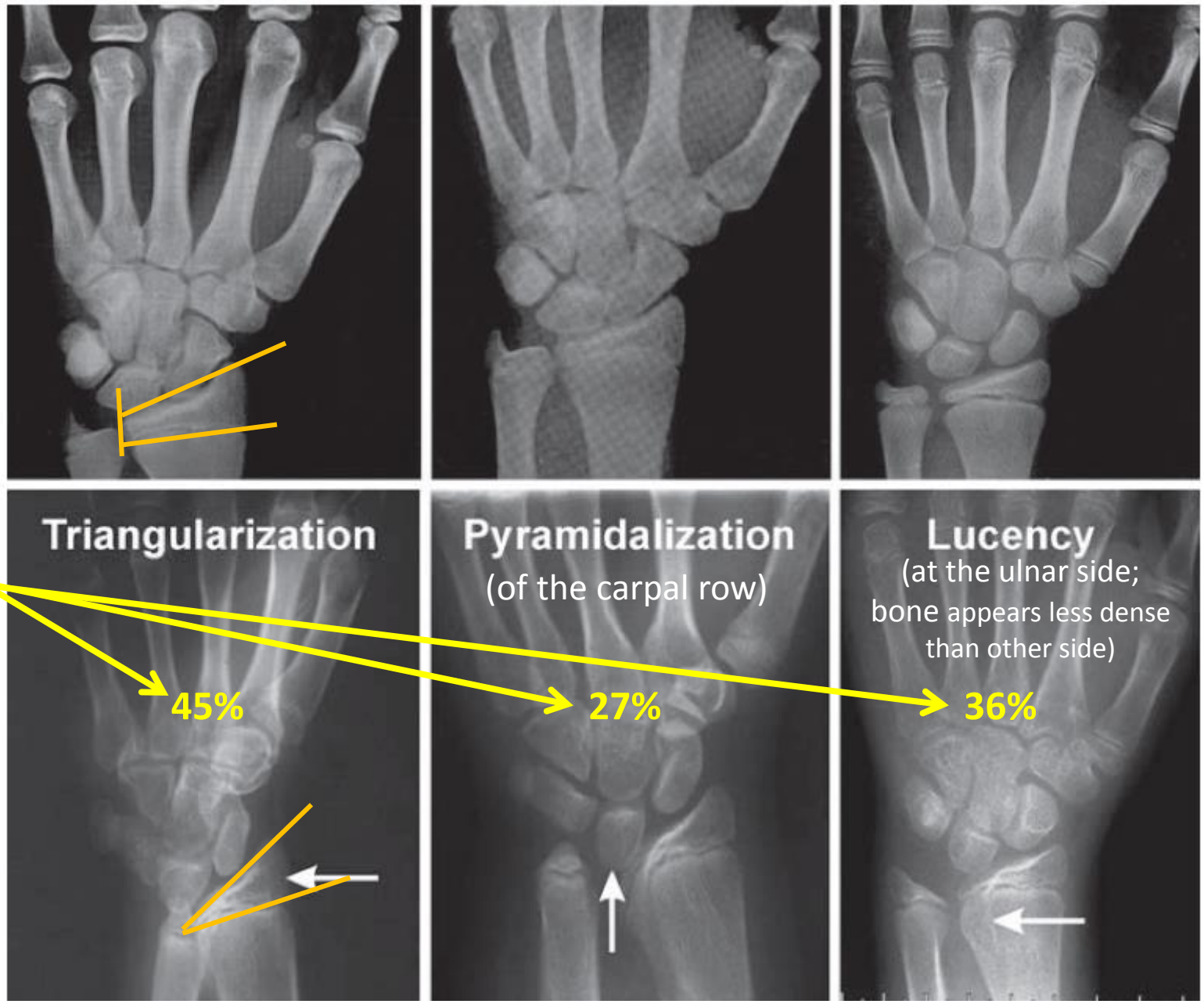
Benito-Sanz et al, Am. J. Hum. Genet. 2005

SHOX Deficiency: RX Features Pittfalls

Female, BA: 13 yr | Female, BA: 14 yr | Female, BA: 10 yr

Normal hand X-rays

Wolters et al. (*HRP 2013*)
n = 22 short children with SHOX haploinsufficiency
NB = none of children < 8 yr had any radiologic signs



Leri-Weill syndrome

(Binder, HRP 2011)

SHOX Deficiency: Genetic Heterogenicity

Author, yr	Chen *et al.*, 2009		Rosilio *et al.*, 2013	Walters *et al.*, 2013	*AOUP-Biol. Molecolare
	ISS	LWS	ISS + LWS + DSS	ISS	ISS
Patient Population	36/740 (4.9%)	38/67 (56.7%)	149/537 (27.7%)	22/449 (4.9%)	15/141 (15%)
Complete gene deletions	63.8%	42.1%	40.5%	14%	6%
Intragenic small deletions				—	—
Point mutations	13.9%	23.7%	18.4%	68%	40%
Enhancer deletions (downstream)	22.2%	34.2%	41.1%	14%	49%
Duplications	—	—	—	4%	14%

ISS = idiopathic short stature; LWS = Léry-Weill syndrome; DSS = disproportionate short stature

*2011-2015

SHOX Gene & Short Stature: Objectives

- To define SHOX gene structure & function.
- To characterize clinical disorders related SHOX gene deficiency (*haploinsufficiency*).
- To discuss the effects of GH treatment on linear growth in SHOX gene deficiency.

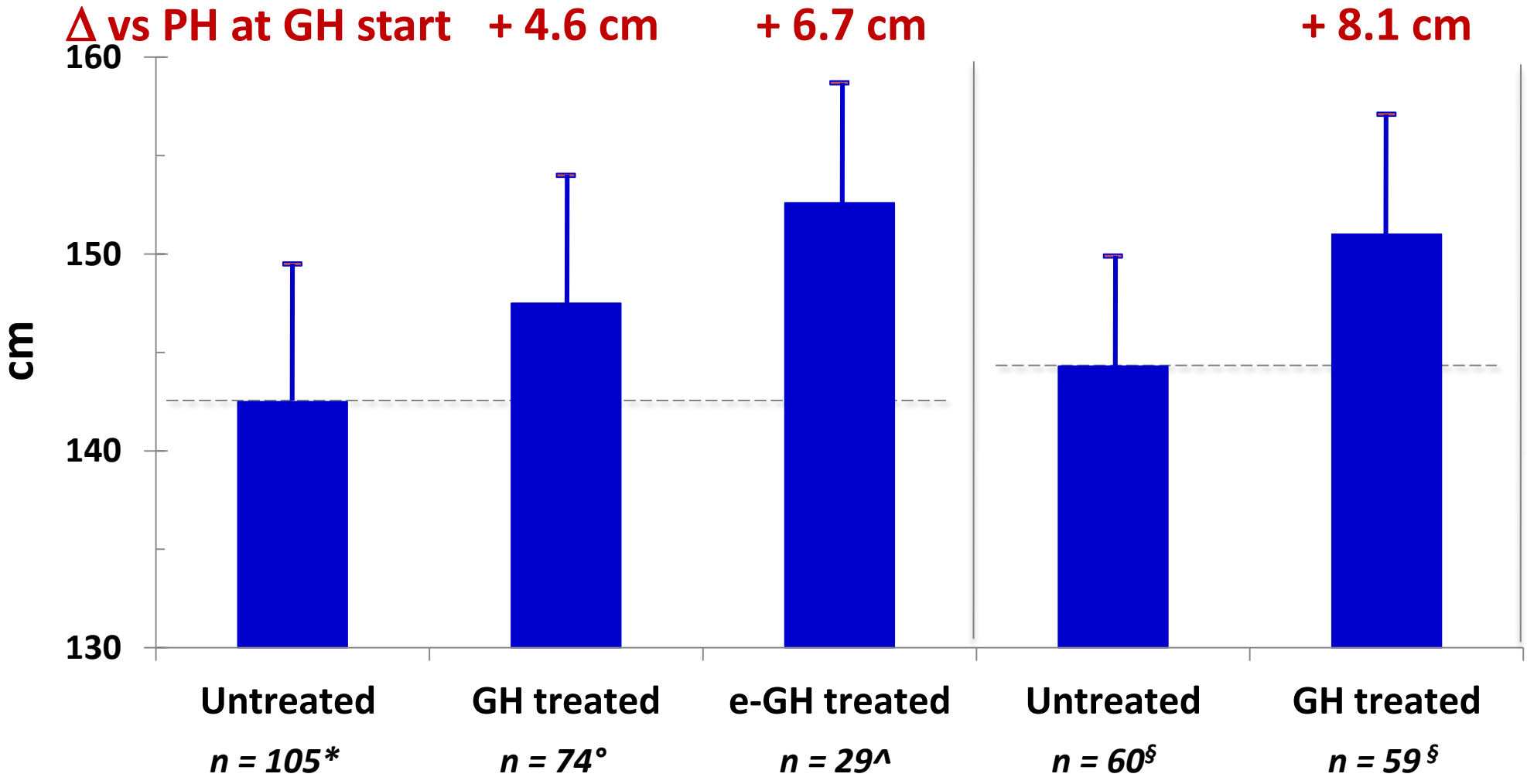
*La prescrizione **(di GH)** a carico del SSN, su diagnosi e piano terapeutico di centri specializzati, Università, Aziende Ospedaliere, Aziende Sanitarie, IRCCS, individuati dalle Regioni e dalle Province autonome di Trento e Bolzano, è limitata alle seguenti condizioni:*

- bassa statura da deficit di GH, …
- **sindrome di Turner citogeneticamente dimostrata;**
- deficit staturale nell'insufficienza renale cronica;
- soggetti affetti dalla sindrome di Prader Willi, geneticamente dimostrata, …
- **soggetti con alterata funzione del gene SHOX, geneticamente dimostrata;**
- bambini nati piccoli per l'età gestazionale...

Roma, 19-06-2014

Il Direttore Generale
Luca Pani

Turner Syndrome: Final Height in Untreated and GH Treated *(0.33 mg/kg/wk)* Italian Patients

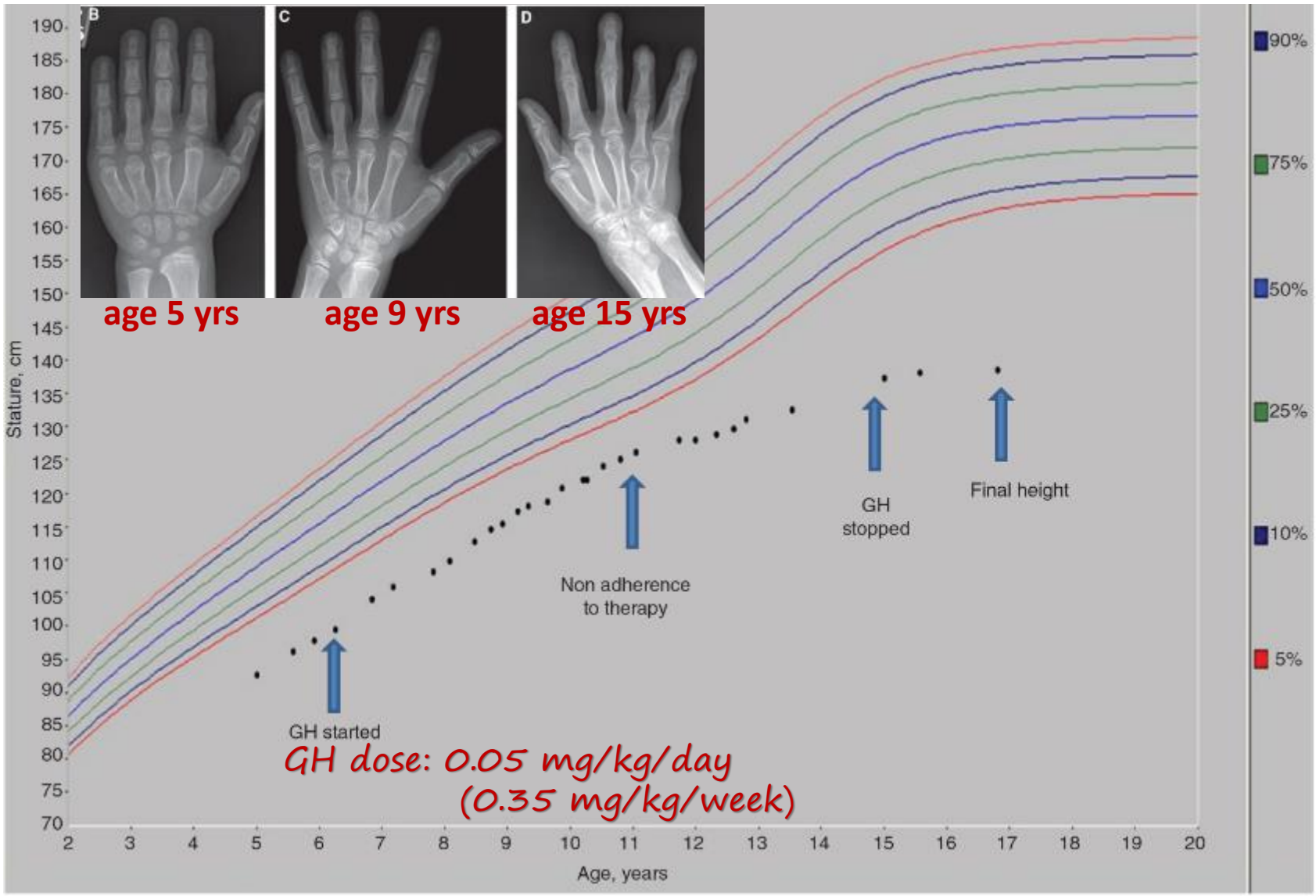


*Bernasconi et al: Italian multicentric study; Acta Paediatr 1994; °Cacciari et al: Italian multicentric study; JCEM 1999
^Wasniewska et al: early treated (< 6 yrs): PAH at 5 yrs of therapy, Italian multicentric study EJE 2004
§Pasquino et al: single centre study, JEI 2005

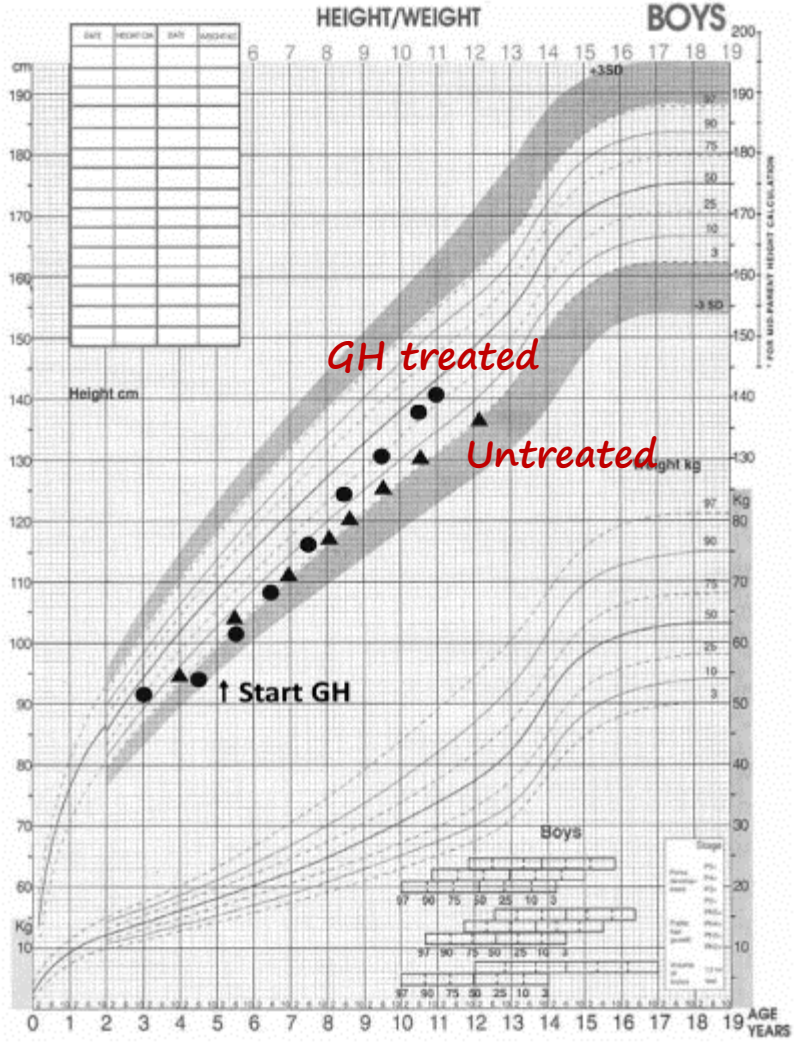
Monica Grover, Shannon French* and Parvin Yazdani

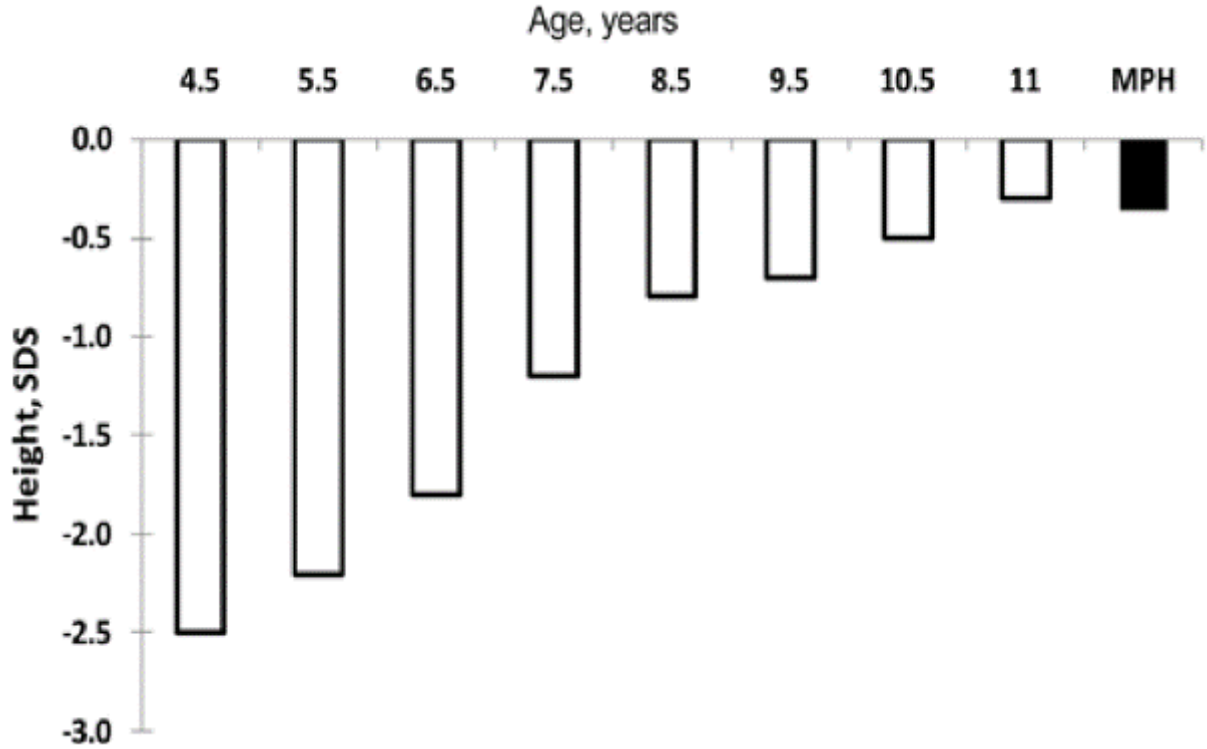
Case report: long-term follow-up of a 45,X male with *SHOX* haploinsufficiency

J Pediatr Endocr Met 2015; 28(7-8): 937–941



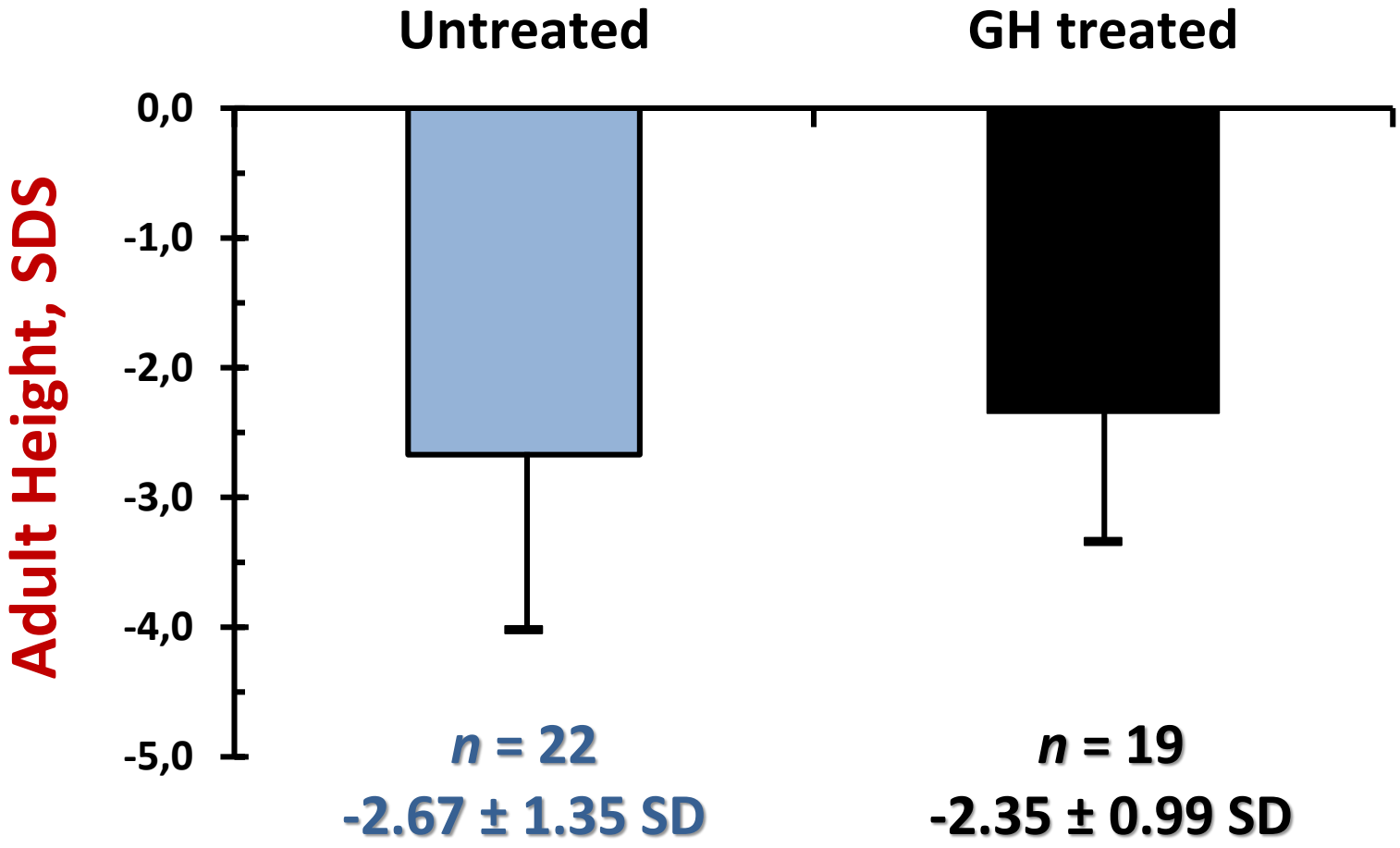
Long-term growth hormone treatment in a boy with 45,X/46,X,idic(Yp) mixed gonadal dysgenesis: comparison with growth pattern of an untreated patient





HORMONES 2015, 14(1):142-147

Statural Growth of Males with 45,X/46,XY Mosaicism
A Review*



*Raw SD data were drawn from authors' published data or calculated from absolute values (in cm) according to male sex and country of origin.

(Sex Dev 2015, in press)

*La prescrizione **(di GH)** a carico del SSN, su diagnosi e piano terapeutico di centri specializzati, Università, Aziende Ospedaliere, Aziende Sanitarie, IRCCS, individuati dalle Regioni e dalle Province autonome di Trento e Bolzano, è limitata alle seguenti condizioni:*

- bassa statura da deficit di GH, …
- sindrome di Turner citogeneticamente dimostrata;
- deficit staturale nell'insufficienza renale cronica;
- soggetti affetti dalla sindrome di Prader Willi, geneticamente dimostrata, …
- **soggetti con alterata funzione del gene SHOX, geneticamente dimostrata;**
- bambini nati piccoli per l'età gestazionale...

Roma, 19-06-2014

Il Direttore Generale
Luca Pani

GH Treatable Short Stature: Main Causes

Disease	Estimated incidence*	Note
SHOX deficiency (gene mutations)	1: 1.000 1: 2.000	—
Growth hormone deficiency	1: 4.000	—
Prader-Willi syndrome	1: 15.000 1: 20.000	—
Turner syndrome	1: 5.000	1: 2.500 Females
End stage renal disease	9-12: 10^6	Age related population

*In general population

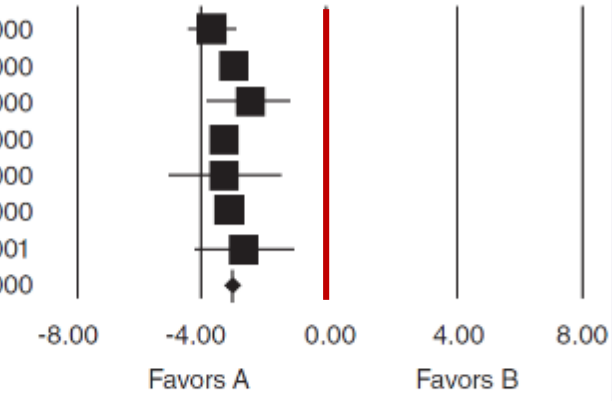
GH treatment in SHOX gene deficiency: A meta-analysis

A

Study (year)	Difference in means	Standard error	Variance	Lower limit	Upper limit	Z-value	p-value
Ogata (2001) [14]	-3.730	0.379	0.144	-4.473	-2.987	-9.842	0.000
Munns (2003) [13]	-3.000	0.010	0.000	-3.020	-2.980	-300.000	0.000
Binder (2004) [5]	-2.530	0.685	0.469	-3.873	-1.187	-3.693	0.000
Blum (2007) [12]	-3.300	0.200	0.040	-3.692	-2.908	-16.500	0.000
Blum (2009) [19]	-3.300	0.900	0.810	-5.064	-1.536	-3.667	0.000
Urakami (2009) [18]	-3.150	0.212	0.045	-3.566	-2.734	-14.858	0.000
Salmon-Musial (2011) [20]	-2.700	0.800	0.640	-4.268	-1.132	-3.375	0.001
	-3.080	0.082	0.007	-3.243	-2.923	-37.770	0.000

Statistics for each study

Difference in mean and 95% CI

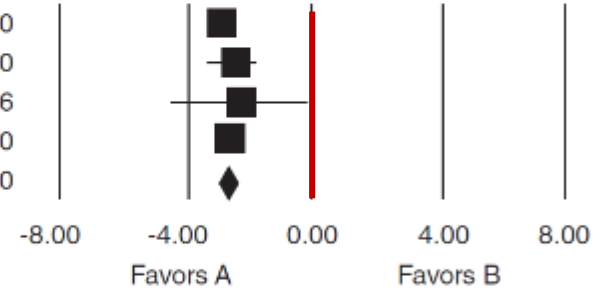


B

Study (year)	Difference in means	Standard error	Variance	Lower limit	Upper limit	Z-value	p-value
Ogata (2001) [14]	-2.950	0.212	0.045	-3.366	-2.534	-13.915	0.000
Munns (2003) [13]	-2.500	0.424	0.180	-3.331	-1.669	-5.896	0.000
Binder (2004) [5]	-2.330	1.109	1.230	-1.504	-0.156	-2.101	0.036
Blum (2007) [12]	-2.600	0.200	0.040	-2.992	-2.208	-13.000	0.000
	-2.731	0.137	0.019	-2.998	-2.463	-19.997	0.000

Statistics for each study

Difference in mean and 95% CI

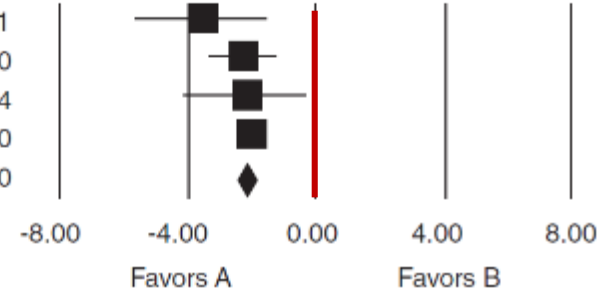


C

Study (year)	Difference in means	Standard error	Variance	Lower limit	Upper limit	Z-value	p-value
Ogata (2001) [14]	-3.650	1.061	1.126	-5.730	-1.570	-3.440	0.001
Munns (2003) [13]	-2.300	0.566	0.320	-3.409	-1.191	-4.064	0.000
Binder (2004) [5]	-2.230	0.988	0.976	-4.166	-0.294	-2.257	0.024
Blum (2007) [12]	-2.100	0.200	0.040	-2.492	-1.708	-10.500	0.000
	-2.171	0.182	0.033	-2.529	-1.813	-11.898	0.000

Statistics for each study

Difference in mean and 95% CI



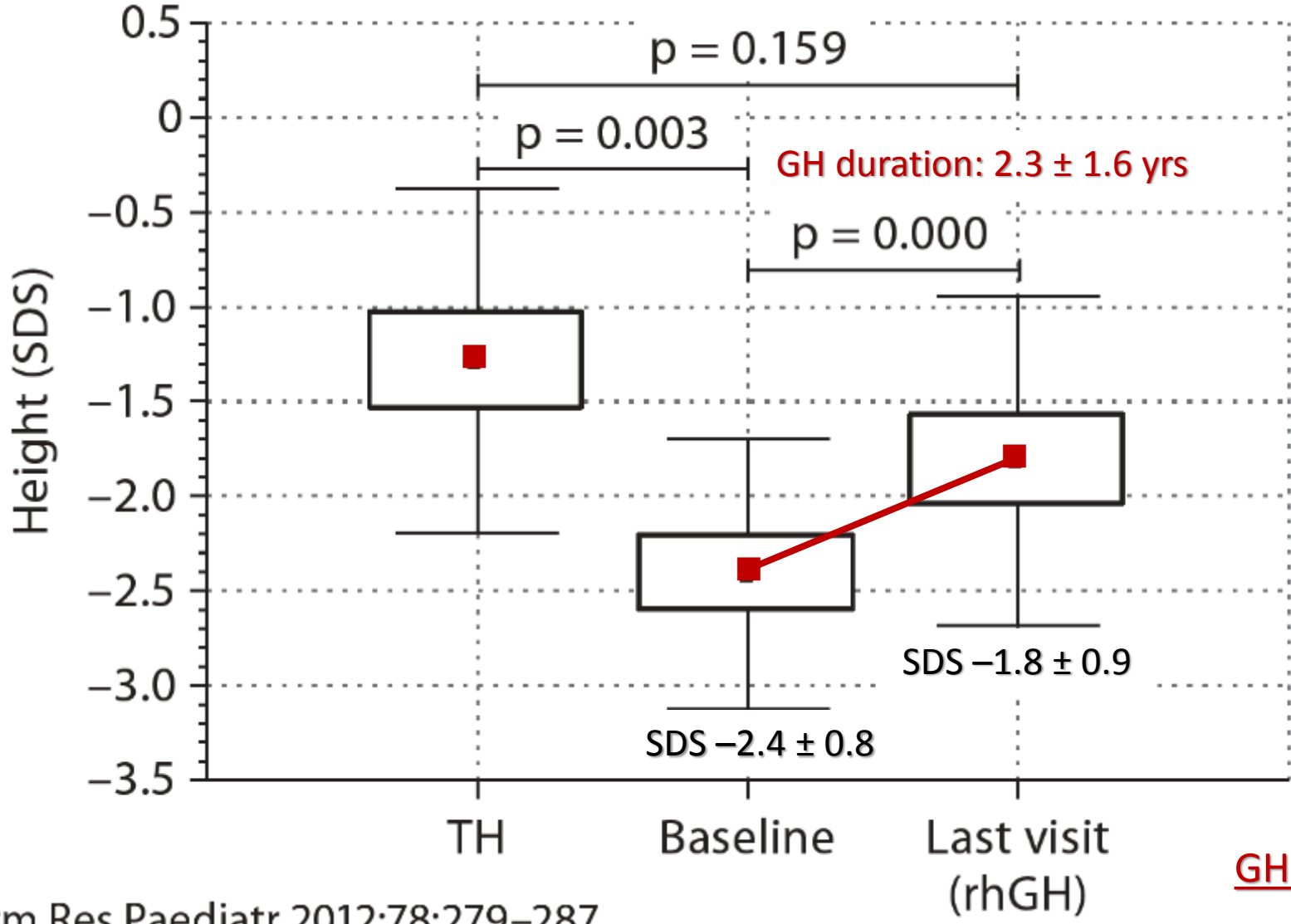
rhGH progressively improved height deficit from baseline to 24 months.

The major catch-up growth was after 12 months.

Massart et al., Pharmacogenomics: 2013

SHOX Deficiency & Efficacy of GH Therapy

(n = 15 children; 9 females; age 9.7 ± 2.9 years)



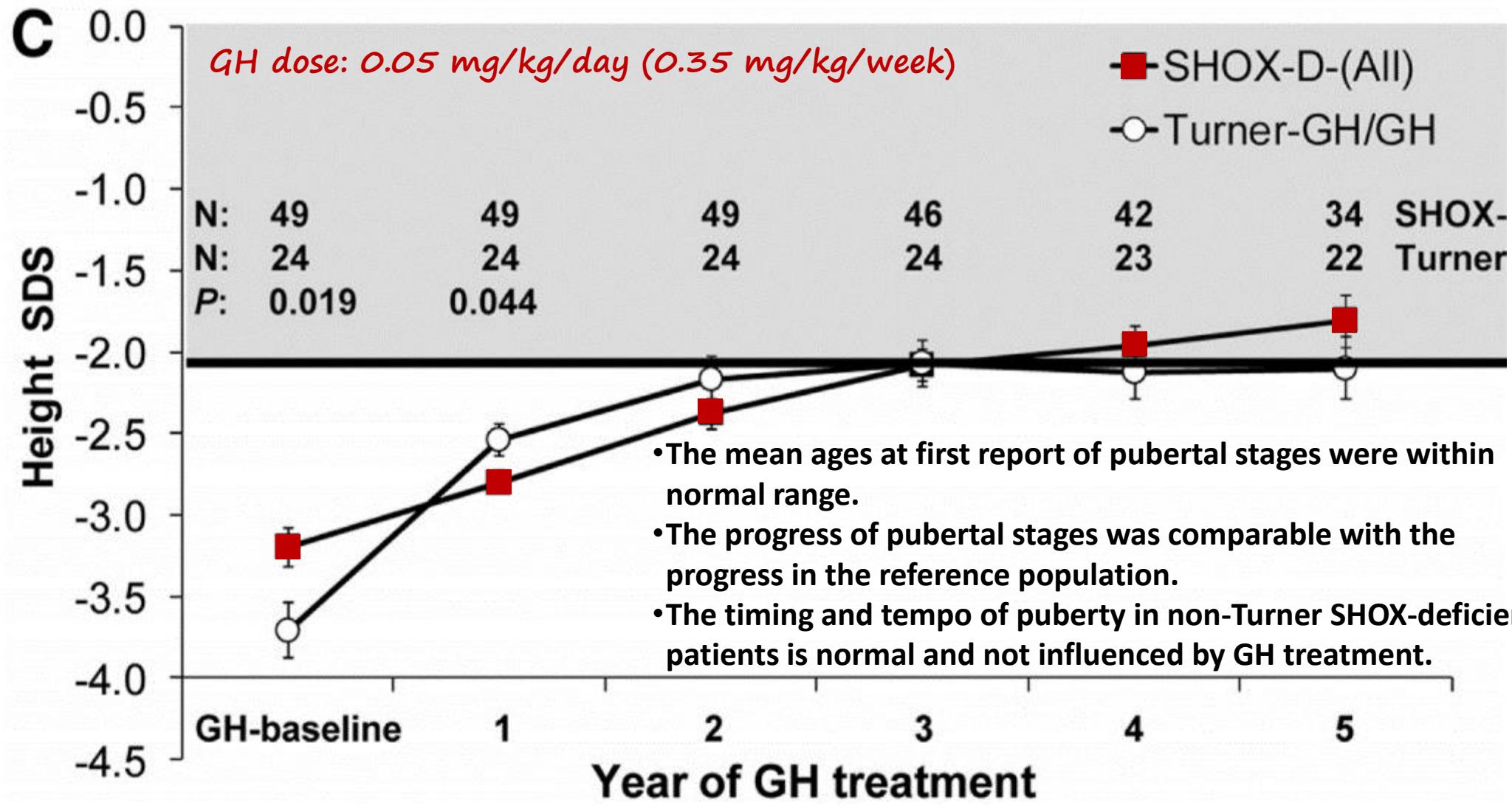
Collaborative study:

- Modena (coord.)
- Bologna
- Bolzano
- Fano
- Genova
- Palermo
- Parma
- Pisa

GH dose: 0.273 ± 0.053 mg/kg/week

Horm Res Paediatr 2012;78:279–287

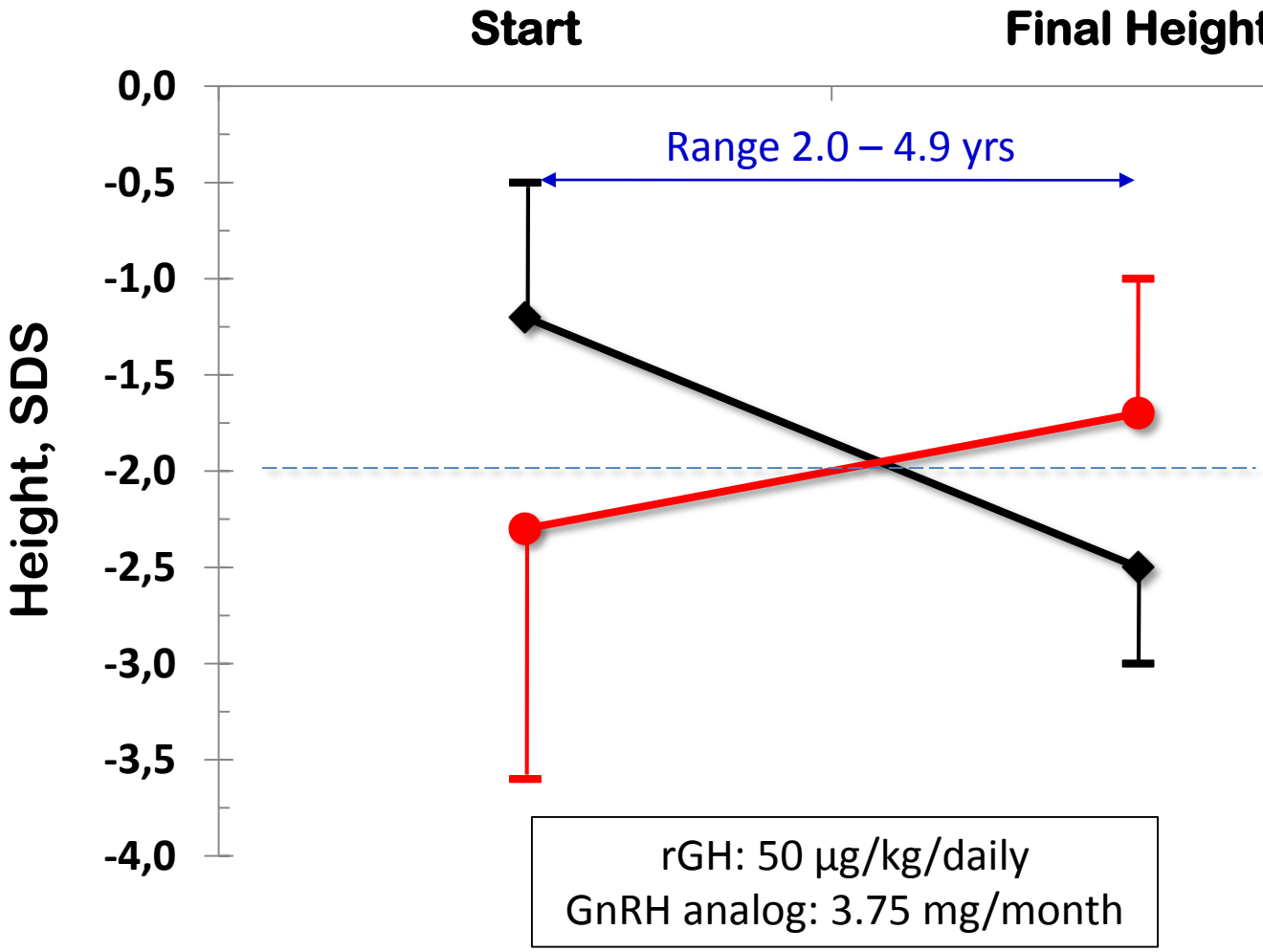
Final Height after GH Treatment in SHOX Deficiency & TurnerSyndrome



Blum et al, J Clin Endocrinol Metab 98: E1383–E1392, 2013

Short Stature due to SHOX Deficiency

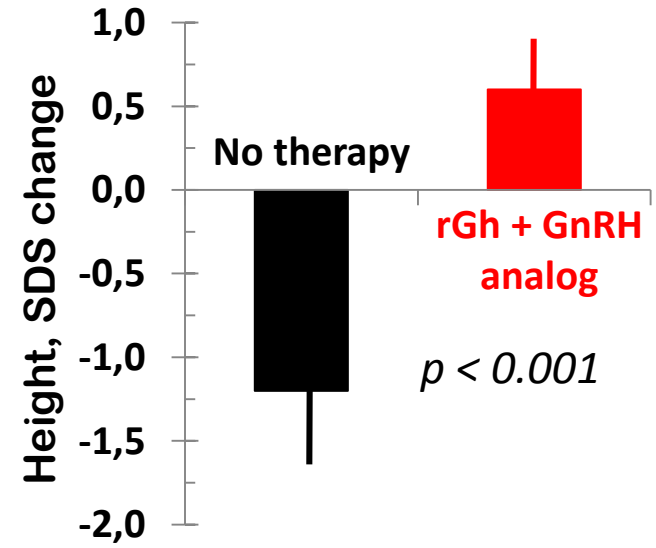
Effect of rGH + GnRH analog on Final Height



rGH: 50 µg/kg/daily
GnRH analog: 3.75 mg/month

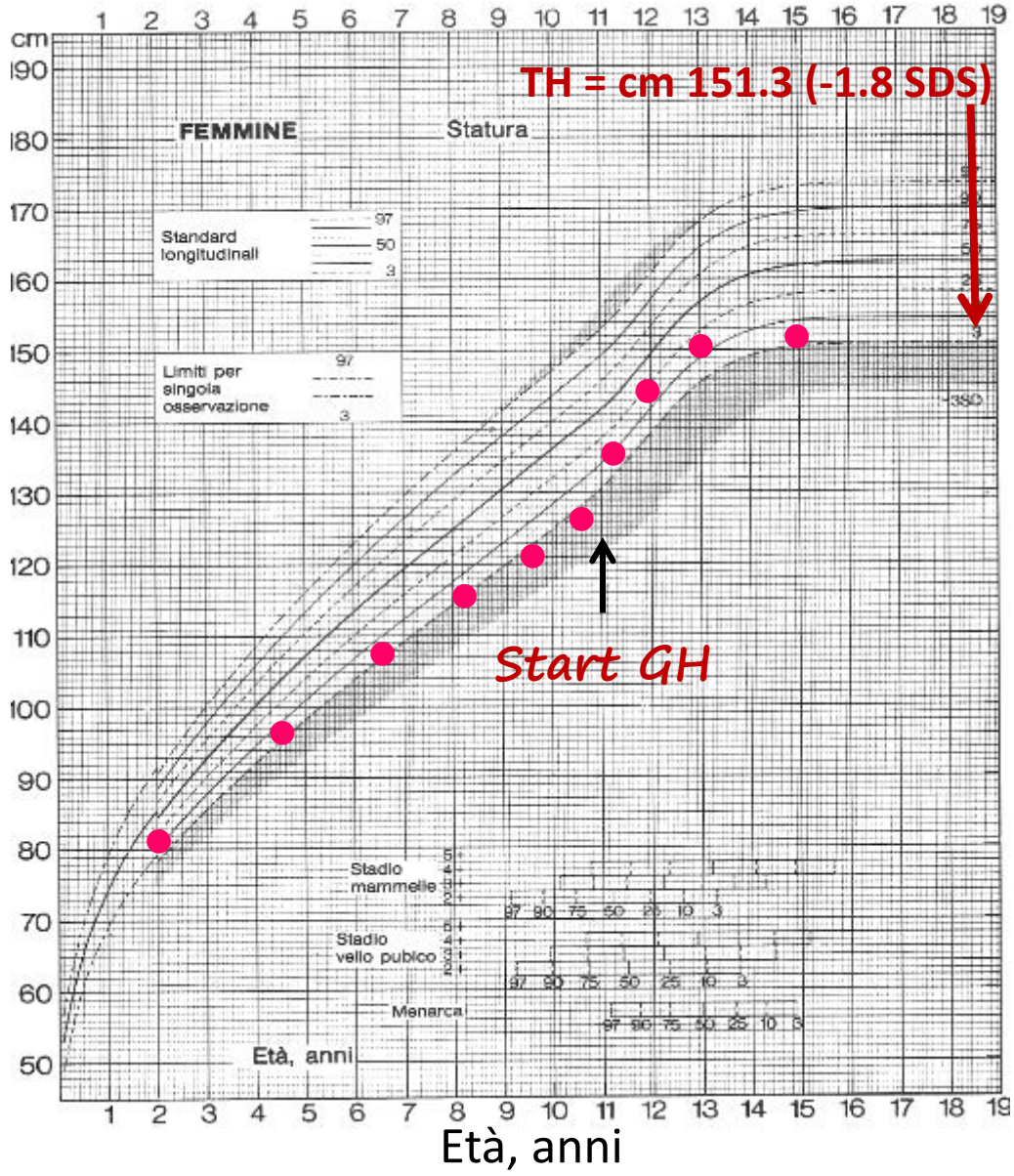
◆**No therapy**
n = 5,
age at start 11.4±1.4 yrs

● **rGh + GnRH analog**
n = 5,
age at start 11.8±2.1 yrs



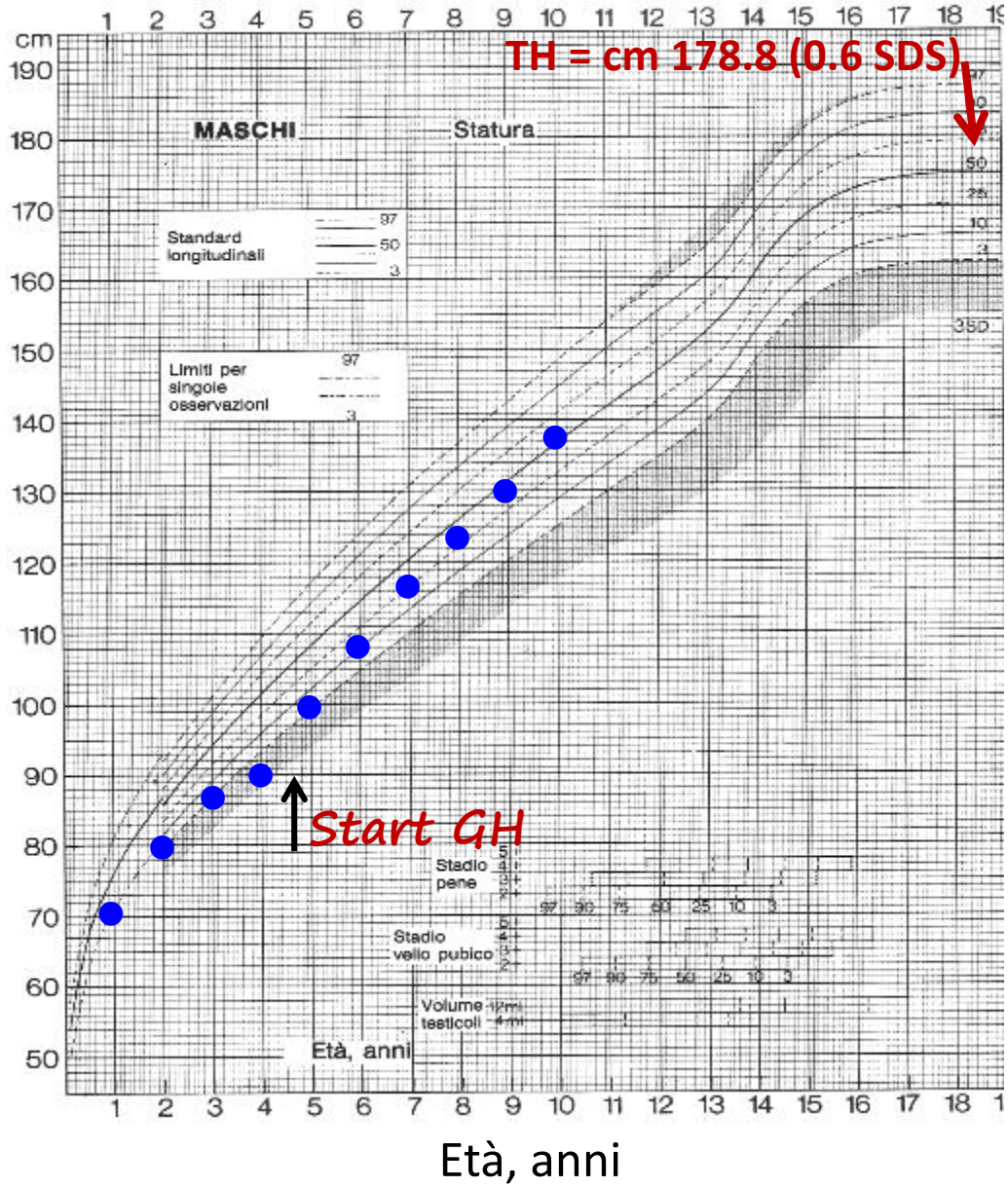
Scalco et al, JCEM 2010

SHOX Deficiency: Effect of Age at Start GH



Età, anni

SHOX mutation c.205C>T (exon 2)



Età, anni

SHOX mutation c.486+25G>A (intron 3)

GH treatment in SHOX Gene Haploinsufficiency

Safety

- No adverse effects of rGH treatment were observed.
- A tendency to disproportionate growth (*lower extremities < trunk*) has been rarely reported.
- The number of reported SHOX deficient children treated with GH is relatively few, so further long-term surveillance studies involving larger patient population is required.

Terapia con GH nel Deficit del Gene SHOX

Conclusioni (1)

- Il deficit del gene SHOX (*aploinsufficienza* per cause cromosomiche o geniche) è una causa importante di bassa statura.
- Il trattamento con GH nel deficit del gene SHOX da cause cromosomiche determina:
  - mediamente buoni risultati in termini di statura adulta nella sindrome di Turner, ma variabili in base a diverse componenti cliniche (*compreso centro di trattamento*);
  - scarsi risultati a lungo termine nei maschi 45,X o con mosaicismo 45,X/46,XY [*migliore definizione degli schemi terapeutici? coinvolgimento di altri geni localizzati sul cromosoma Y? prescrizione (non citati in nota 39*)?].

Terapia con GH nel Deficit del Gene SHOX

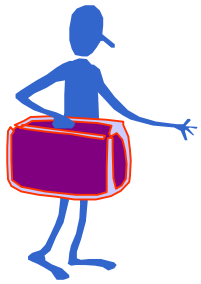
Conclusioni (2)

- Il trattamento con GH nel deficit del gene SHOX da cause geniche determina incoraggianti risultati a lungo termine sulla statura adulta (*attualmente paragonabili a quelli ottenibili nella sindrome di Turner*).
- Rimane la necessità di trials:
  - a lungo termine su campioni più ampi;
  - maggiormente omogenei dal punto di vista auxologico (*compresa l'età di inizio del trattamento*);
  - in grado di meglio definire la risposta accrescitiva in pubertà (*e la necessità di eventuali terapie aggiuntive in tale periodo*);
  - atti a valutare la necessità di un diverso management tra maschi e femmine (*dosi, trattamento nel periodo puberale)*.

G.I. Baroncelli, F. Massart
UO Pediatria Universitaria – AOUP -Pisa

A. Michelucci, F. Baldinotti
A. Fogli
Molecular Biology – AOUP - Pisa

L. Iughetti
Modena



Thanks for your attention

SHOX Gene & Short Stature: Conclusions

- SHOX gene haploinsufficiency likely represents a main cause of short stature in humans.
- In gene mutations, clinical findings may be uninformative in infancy and in young childhood.
- Deterioration of growth pattern has been reported from puberty onset onward (mainly in females).
- GH treatment improve height outcome both in short- and long-term follow-up studies.
- Association of GH and GnRH analogs may be useful in late treated children (but must be confirmed).
- Additional data in larger series of SHOX deficient patients are needed.