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The growth hormone receptor exon 3-deleted/full-length polymorphism and response to growth hormone therapy in prepubertal idiopathic short children

G. Hellgren [a,*], C.A. Glad [b], B. Jonsson [c], G. Johannsson [b], K. Albertsson-Wikland [d]

[a] Department of Pediatrics, Institute of Clinical Sciences, The Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden
[b] Department of Endocrinology, The Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden
[c] Department of Women and Child Health, Uppsala University, Uppsala, Sweden
[d] Department of Physiology/Endocrinology, Institute of Neuroscience and Physiology, The Sahlgrenska Academy, University of Gothenburg, Gothenburg, Sweden

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ABSTRACT

Objective: The primary aim of the study was to evaluate d3-GHR as a possible cause of increased GH sensitivity in children with delayed infancy-childhood transition (DICT). The secondary aim was to investigate the impact of the GHR exon 3 deleted/full-length (d3/fl) polymorphism on GH treatment response in prepubertal children classified as having idiopathic short stature (ISS).
Design: Study subjects included 167 prepubescent longitudinally followed children classified as having ISS. Children were randomized to standard-dose GH treatment (33 μg kg^{-1} day^{-1}), to double-dose treatment (67 μg kg^{-1} day^{-1}), or to an untreated control group. Growth and metabolic outcome were evaluated at birth (n = 166), after one year of treatment (n = 59) and at adult height (n = 145). Genotyping of the GHR d3/fl polymorphism was performed using TaqMan SNP genotyping of tagSNP rs6873545.
Results: Birth and early growth data did not reach the predetermined level of statistical significance for difference between genotypes. Growth and IGF-1 response after one year of GH treatment did not differ between genotypes. IGFBP-3_{SDS} was higher in untreated d3-GHR carriers than in untreated fl/fl individuals, whereas there was insufficient evidence for higher IGFBP-3_{SDS} in treated d3-GHR carriers. Genotype did not explain the growth response to treatment, and no differences in $height_{SDS}$, height gain, or difference in height to midparental $height_{SDS}$ between genotype groups were found at adult height.
Conclusion: The common GHR d3/fl polymorphism is probably not a cause of DICT in children with ISS, and our results do not suggest that the d3-GHR genotype is associated with increased sensitivity to GH in children with ISS.

1. Introduction

Children who are short for unknown reasons are classified as having idiopathic short stature (ISS) [1]. To identify a patient as ISS, systemic diseases, hormone deficiencies, psychosocial deprivation, genetic diseases, and syndromes known to cause short stature should be excluded [1]. Growth hormone (GH) treatment of children classified as ISS has been shown to increase height velocity and adult height [2–6], and was approved for this indication by the United States Food and Drug Administration in 2003.

Diverse mechanisms likely underlie ISS. Among children classified as having ISS, there is great variability in parameters such as height, body mass index (BMI), insulin-like growth factor 1 (IGF-1), IGF-binding protein 3 (IGFBP-3) levels, degree of bone maturation, and timing of puberty, all of which are in some way influenced by GH. As expected in such a heterogeneous patient group, the growth response to GH treatment is highly variable, ranging from no gain to approximately +3 standard deviation score (SDS) [2].

The age at which GH begins to significantly regulate growth through the GH-IGF-1 axis is represented by the transition period from infancy to childhood growth (ICT) [7,8]. In Western countries this period normally occurs at an age of 6-12 months [9,10]. In a Swedish population

Abbreviations: GHR, growth hormone receptor; d3, exon 3 deletion; DICT, delayed infancy-childhood transition; ISS, idiopathic short stature; GH, growth hormone; BMI, body mass index; IGF-1, insulin-like growth factor 1; IGFBP-3, IGF-binding protein 3; SDS, standard deviation score; ICT, infancy-childhood transition; fl, full length; ITT, intention to treat; PP, per protocol; AH, adult height; ICP, infancy-childhood-pubertal; AITT, arginine-insulin- tolerance test; SGA, small for gestational age; GH_{max}AITT, GH max peak after AITT; GH_{max}24h, GH max peak in 24 h profile; GP-GRC, Gothenburg Pediatric Growth Research Center; SWEDAC, Swedish Board for Accreditation and Conformity Assessment; DNA, deoxyribonucleic acid; PCR, polymerase chain reaction.

* Corresponding author at: Department of Pediatrics, Institute of Clinical Sciences, Sahlgrenska Academy, University of Gothenburg, The Queen Silvia Children's Hospital, SE-416 85 Gothenburg, Sweden. Tel.: +46 708636412; fax: +46 31 848952.
E-mail address: gunnel.hellgren@gu.se (G. Hellgren).

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of 2 432 children, the timing of the ICT significantly impacted adult height, suggesting that stunting in early life is not only a problem in developing countries [11]. Delayed ICT (DICT) was suggested to be one of the factors underlying ISS. To further elucidate this issue, a group of ISS children included in a randomized controlled GH treatment study [2] were re-analyzed for DICT; as many as 44% were identified as having DICT. Not one case of catch-up growth, which normally occurs after short periods of stunting at other ages, was identified among children with DICT [12], supporting the hypothesis that many children with ISS is caused by DICT. Children with DICT had lower IGF-1_{SDS} and lower BMI at the start of GH treatment and were more sensitive to GH treatment (in terms of growth) than children without DICT [13]. However, the IGF-1 response did not differ between DICT and non-DICT groups [13].

Homo- or heterozygous carriers of the common exon 3 deletion of the GH receptor (d3-GHR), the exon 3 deleted/full-length (d3/fl) polymorphism, are other groups reported to be more sensitive to GH treatment, although the data are inconsistent. Some studies showed no difference in growth response to GH treatment [14–19], whereas an increased growth response was observed in others [20–24]. However, a meta-analysis indicated that the d3-GHR polymorphism increased growth velocity by an additional 0.5 cm during the first year of treatment [25]. Furthermore, the presence of d3-GHR was reported to be negatively associated with birth $weight_{SDS}$ and fetal growth [26] but positively associated with first year postnatal growth in healthy infants [27] and catch-up growth in preterm infants [28]. Most published data support no or minor impact of the d3-GHR genotype on adult height. Similarly, there are contradictory results regarding the impact of the d3-GHR and fl-GHR isoforms on metabolic factors influenced by GH. Associations [29] as well as no associations with IGF-1 levels, insulin secretion, and sensitivity in children and adolescents have been reported [30–32].

Since GH treatment response is highly variable in children classified as ISS it is important to identify the factors that influence this variation in response. Increased growth response to GH treatment has been identified in both children with DICT and in children who are hetero- or homozygous for d3-GHR. Less is known about the impact of d3-GHR and/or DICT on the metabolic response to GH treatment. Thus, the primary aim of this study was to evaluate d3-GHR as a possible cause of increased GH sensitivity in children with DICT. Our secondary aim was to investigate the impact of the GHR d3/fl polymorphism on GH treatment response in prepubertal children classified as having ISS.

2. Materials and methods

2.1. Ethical considerations

A randomized, controlled study (TNR 80-080) was conducted in accordance with the Declaration of Helsinki and good clinical practice guidelines. The study protocol was approved by the Ethical Committees at the Universities of Gothenburg, Lund, Linköping, Uppsala, and Umeå, Sweden, and at Karolinska Institute in Stockholm, Sweden. Informed consent was obtained from all children and their parents.

2.2. Study design and subjects

A flow diagram, from inclusion of patients to one year on treatment is shown in Fig. 1. In total, 177 short ($height_{SDS} < -2$) non-GH-deficient children, defined as GH_{max} at two independent GH stimulation tests above 10 μg/L, from six pediatric units in Sweden were enrolled in the study between 1988 and 1999. Study design and subjects were previously described in detail [2]. Briefly, children who still remained prepubertal after one pre-study year were randomized into three groups: treatment with a standard dose of GH (33 μg kg^{-1} day^{-1}, 0.1 U kg^{-1} day^{-1}), treatment with a double dose of GH (67 μg kg^{-1} day^{-1}, 0.2 U kg^{-1} day^{-1}), or no treatment (control group). The children were followed every 3 months until their adult height was reached. DNA samples were

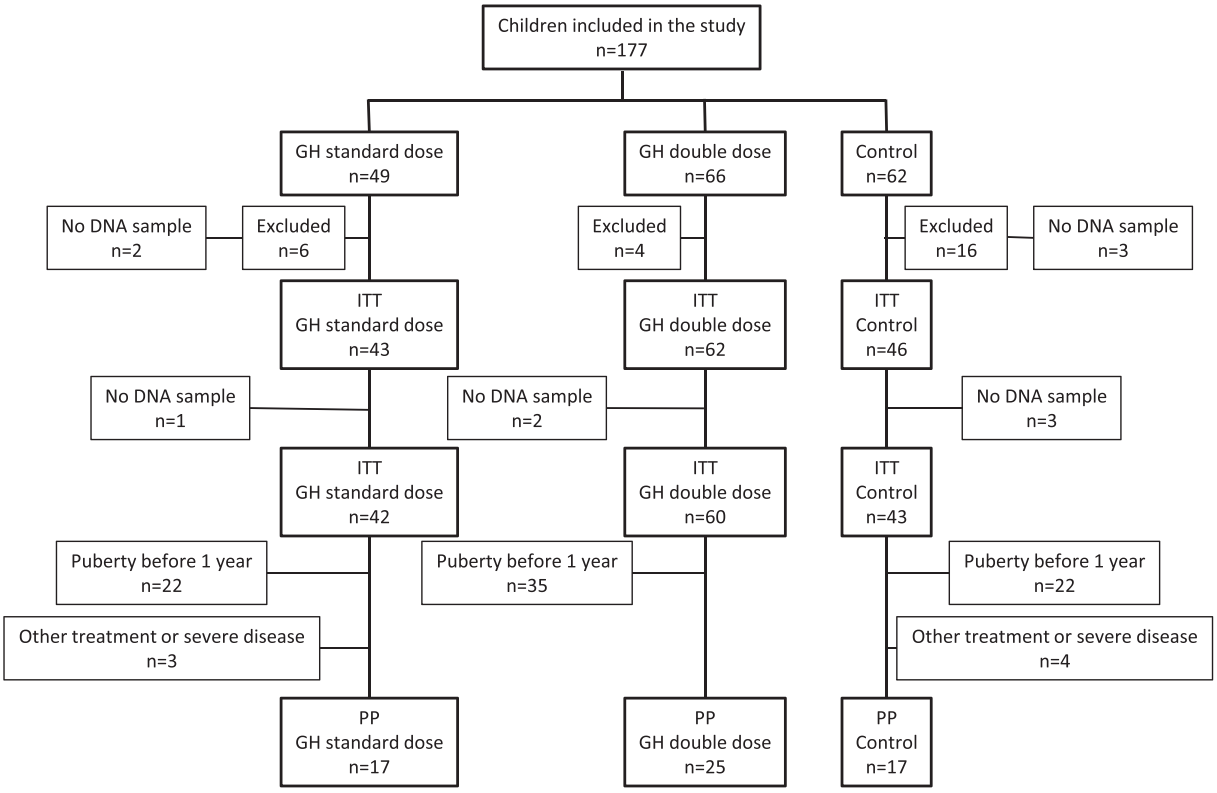

Fig. 1. Flow diagram of the study design according to treatment groups.