Growth hormone releasing hormone or growth hormone treatment in growth hormone insufficiency?

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SUMMARY Sixteen prepubertal children who were insufficient for growth hormone were treated with growth hormone releasing hormone (GHRH) 1-40 and GHRH 1-29 for a mean time of nine months (range 6-12 months) with each peptide. Eleven children received GHRH 1-40 in four subcutaneous nocturnal pulses (dose 4-8 µg/kg/day) and eight (three of whom were also treated with GHRH 1-40) received GHRH 1-29 twice daily (dose 8-16 µg/kg/day). Altogether 73% of the children receiving GHRH 1-40 and 63% receiving GHRH 1-29 showed a growth response. Double the daily dose of GHRH 1-29 was required to obtain equivalent growth response to pulsatile GHRH 1-40. A significant linear correlation was shown between growth hormone secretion and height velocity on GHRH 1-40 but not on GHRH 1-29 and there was a significant correlation between plasma GHRH and serum growth hormone concentrations during GHRH 1-40 administration.

Response to conventional growth hormone treatment in a matched group of children was significantly better than the response after GHRH. A significant improvement in height velocity was observed in the children transferred to growth hormone replacement. Growth hormone remains the treatment of choice in growth hormone insufficiency. GHRH treatment may be of benefit in children with less severe growth hormone insufficiency in the presence of pulsatile endogenous growth hormone secretion.

Growth hormone secretion is controlled by inhibitory somatostatin and stimulatory growth hormone releasing hormone (GHRH).¹⁻³ In subjects with growth hormone sufficiency or insufficiency intravenous GHRH results in growth hormone release from the pituitary.⁴⁻⁶ Preliminary reports indicate that GHRH may have a therapeutic role in the treatment of children who are growth hormone insufficient.⁷⁻⁹ Evaluation of different treatment regimens, however, has been limited. The aims of this study were to investigate the relation between GHRH administration and growth hormone secretion and the therapeutic effects of alternative modes of administration and dose regimens of GHRH in children with growth hormone insufficiency and to compare the linear growth response with conventional growth hormone replacement.

Patients and methods

Sixteen prepubertal children (11 boys, five girls)

with bone ages of less than 10 years were recruited to the study between January 1985 and July 1986. The pretreatment clinical details are summarised in table 1. Auxological observation over a period of one year had shown a low growth velocity, and

Table 1 Pretreatment clinical data (n=16)

	Mean (SD)	Range
Age (years)	8.5 (2.2)	4.9 to 11.9
Bone age (TW2)	5.9 (2.1)	2.5 to 8.8
Height SDS	-2.4 (1.0)	-4.8 to -0.6
Height velocity (cm/year)	3.8 (0.9)	1.7 to 5.7
Height velocity SDS	-2.2 (0.8)	-4.1 to -0.9
Maximum growth hormone concentration (on insulin tolerance test) (mU/l)	6.5 (5.3)	1.9 to 17.0
Maximum growth hormone concentration (during sleep) (mU/l)	9.3 (5.7)	4.0 to 26.0
Sum % of 24 hour growth hormone pulse amplitude (mU/l)	27.0 (24.0)	4.0 to 93.0

hypothalamo pituitary assessment, using insulin induced hypoglycaemia with thyrotrophin releasing hormone and luteinising hormone releasing hormone stimulation, confirmed isolated growth hormone insufficiency in 10 children and multiple pituitary hormone deficiencies in six children. The children with multiple pituitary hormone deficiencies received appropriate hormonal replacement before GHRH, and pituitary hypoplasia was shown in all cases by high resolution computed tomography (GE 8800). One child had been treated with human growth hormone for four years but stopped treatment three months before starting GHRH. The remaining children had received no treatment for growth hormone insufficiency.

The studies were approved by the Middlesex Hospital Clinical Investigation Committee, the Department of Health and Social Security, and USA Food and Drugs Administration. Written parental consent was obtained.

Before starting GHRH a 24 hour endogenous growth hormone profile was obtained from each child by withdrawing blood samples (1 ml) every 20 minutes. Serum growth hormone profiles were also obtained throughout the study during each treatment regimen. An intravenous GHRH study (1 µg/kg) was performed before GHRH treatment and in 11 children at intervals of three months throughout the study (blood samples taken at -20, -10, 0, 2, 5, 10, 20, 30, and at 15 minute intervals up to 120 minutes).

During the GHRH 1-40 regimen profiles for plasma immunoreactive GHRH concentrations were obtained on each dose schedule in six children in conjunction with serum growth hormone estimations.

Anthropometric measurements were made every three months by the same observer using standard techniques, and bone age assessment was performed at the beginning of the study and after any change in treatment.^{10 11} The measurements obtained were described as raw height velocity (cm/year) when assessing the short term (three month) results or as the standard deviation score (SDS) when assessing the overall results.¹²

GHRH TREATMENT

The first 11 children received GHRH 1-40 (supplied by Professor MO Thorner) by nocturnal pulsatile infusion every three hours for four pulses per night. A minipump (Zyklomat, Ferring) delivered each pulse over one minute via a 27 gauge infusion needle inserted subcutaneously into the anterior abdominal wall. A dose of 1 µg/kg/pulse (4 µg/kg/day) was given for the first three months in six children followed by 2 µg/kg/pulse (8 µg/kg/day) for the rest

of the study period. The remaining five children received the higher dose regimen throughout the GHRH 1-40 study period.

Eight children received GHRH 1-29 (KabiVitrum) subcutaneously into anterior abdominal wall in an initial dose of 4 µg/kg/dose (8 µg/kg/day) twice daily for three months followed by a dose of 8 µg/kg/dose (16 µg/kg/day) for the remaining treatment period. Three GHRH 1-40 'responders' were transferred directly to GHRH 1-29. The mean treatment duration on both GHRH peptides was nine months (range 6-12 months).

On completion of the GHRH study each child was transferred to somatrem (Somatonorm, KabiVitrum) in a dose of 2 IU growth hormone subcutaneously six times a week. The growth results (age, bone age, height SDS, height bone age SDS, and height velocity SDS) on GHRH were compared with a matched group of naive idiopathic insufficient children who received somatrem from January 1986 in an identical regimen.

GROWTH HORMONE ASSAY

Serum growth hormone concentration was assayed using a Tandem R human growth hormone immunoradiometric assay kit (Hybritech) with the standards calibrated against the National Institute of Health human growth hormone reference preparation HS 2243E. The mean intra-assay coefficient of variation was 6.3% at 4.9 mU/l with an interassay coefficient of variation of 9.6%. All samples from the same sampling period were stored at -20°C and assayed together.

GHRH 1-40 ASSAY

Plasma immunoreactive GHRH was assayed by radioimmunoassay using a specific rabbit GHRH 1-40 antiserum (NIBSC code No 84/591) as previously described.¹³ Sensitivity of the assay was 10-20 pg/ml and at a dilution of 1 in 300 000 the antiserum bound 30-40% of freshly labelled GHRH. Intra-assay and interassay coefficients of variation were 6.5% and 9% respectively at a concentration of 400 pg/ml.

STATISTICS

Analysis of variance was used to compare the pretreatment characteristics of children receiving GHRH and growth hormone. Analysis of variance, multiple regression analysis, and Mann-Whitney U test were used to analyse the response to treatment.

Results

Both regimens were well tolerated with only one child experiencing slight stinging at the injection site

Fig 1 95% Confidence limits after 1 $\mu\text{g/kg}$ GHRH 1-40 given intravenously in growth hormone insufficient children ▨ and normal (adult) subjects ▩

(GHRH 1-29). Most of the children in each group undertook their own injections under parental supervision (GHRH 1-40, nine children; GHRH 1-29, six children).

All children responded to intravenous GHRH (mean 31 mU/l; range 5-7-107) (fig 1). The response to intravenous GHRH was highly variable both between and within subjects throughout the study and there was no correlation with the growth response observed.⁶ The 95% confidence limits of the growth hormone response to GHRH in normal adults and the study children is shown in fig 1. Within these limits there is no overlap in the two groups between GHRH administration and the first observed growth hormone response within 10 minutes of the study.

GHRH 1-40

All 11 children showed pulsatile GH secretion of growth hormone in response to nocturnal pulsatile GHRH administration from the first night of treatment. Eight of the 11 children had a significant growth response and maintained pulsatile growth hormone secretion throughout the study (fig 2). The growth velocity achieved reflected the dose regimens (table 2) with no significant improvement during the lower schedule. A positive correlation was present between the stimulated growth hormone secretion and growth velocity ($r=0.78$; $p<0.001$; fig 3) and between the peak plasma immunoreactive GHRH concentrations obtained and the peak growth hormone secretion with each GHRH pulse ($r=0.75$; $p<0.001$; fig 4).

Three children failed to show a linear growth

response to GHRH 1-40. In one child a diminishing growth hormone response with time was observed in association with a falling growth rate. In another child GHRH administration resulted in a high frequency growth hormone response which was

Fig 2 Growth hormone response to subcutaneous GHRH 1-40 during treatment period 2 $\mu\text{g/kg/pulse}$ in one child. Responses to intravenous 1 $\mu\text{g/kg}$ GHRH 1-40 are shown in the left panels.

Fig 3 Relation between growth hormone secretion and growth velocity on GHRH 1-40 ($r=0.78$, $p<0.001$).

Fig 4 Relation between peak GHRH and growth hormone concentrations during nocturnal pulsatile GHRH 1-40 ($r=0.75$, $p<0.001$).

associated with no change in height velocity SDS and in a third inadequate growth hormone pulse amplitude (>15 mU/l) was achieved.

GHRH 1-29

The five naive patients and the three transfer patients showed pulsatile (two pulses) growth hormone secretion to twice daily GHRH 1-29 (fig 5). The higher growth hormone pulse amplitude observed after GHRH 1-29 in comparison with that after pulsatile GHRH 1-40 was not reflected in a more significant height velocity (table 2). Twice the daily dose of GHRH 1-29 was required to achieve equivalent growth responses. The relation between growth hormone secretion and velocity was only maintained by the transfer patients (transfer patients $r=0.78$, $p<0.02$; all GHRH 1-29 patients $r=0.41$, NS; 'naive' patients $r=0.18$, NS). Five children responded to GHRH 1-29 and the non-responders included two naive (one psychosocial; one noncompliance) and one transfer child (inadequate growth hormone secretion).

There was a highly significant increase in height velocity after GHRH 1-40 8 $\mu\text{g/kg/day}$ and both GHRH 1-29 regimens ($p<0.001$), when compared with the pre-treatment height velocity, with no significant difference between the groups (table 2). When analysing the results of the 'responders' only there was a significant difference ($p<0.01$) between the height velocity on GHRH 1-40 8 $\mu\text{g/kg/day}$ and GHRH 1-29 8 $\mu\text{g/kg/day}$ (table 2).

GROWTH HORMONE TREATMENT

When comparing the change in height velocity SDS after GHRH with the change in height velocity SDS in the matched group of children receiving growth hormone there was a significant difference in the growth response observed (mean (SD) change in height velocity SDS; children on growth hormone +4.2 (1.9); children on GHRH +2.8 (2.3); $p<0.04$). The 10 GHRH treated children who have completed one year of somatrem treatment have shown a

Table 2 Growth response to GHRH 1-40 and GHRH 1-29

	Pretreatment	GHRH 1-40		GHRH 1-29	
		4 $\mu\text{g/kg/day}$	8 $\mu\text{g/kg/day}$	8 $\mu\text{g/kg/day}$	16 $\mu\text{g/kg/day}$
All patients	(n=16)	(n=6)	(n=11)	(n=8)	(n=8)
Mean (SEM) height velocity (cm/year)	3.8 (0.2)	4.5 (0.6)	6.4 (0.7)	5.1 (0.3)	5.9 (0.4)
Responders only		(n=3)	(n=8)	(n=5)	(n=5)
Mean (SEM) height velocity (cm/year)		5.4 (0.8)	7.6 (0.6)	5.5 (0.5)	6.5 (0.5)

Fig 5 Growth hormone response to GHRH 1-29 in two dose regimens in one child.

significantly greater growth response to pharmacological growth hormone replacement (mean (SD) height velocity: children on growth hormone 8.7 (2.5); children on GHRH 5.8 (2.0); $p < 0.01$). In one child there was no difference in height velocity on either treatment and in another the height velocity was greater on GHRH.

Discussion

Intravenous administration of GHRH is capable of stimulating growth hormone release from the pituitary of subjects with growth hormone insufficiency.

To determine the therapeutic effect of GHRH in the treatment of growth hormone insufficiency different routes of administration, dose and frequency of GHRH administration, appropriate patient selection, and cost effectiveness of the regimen used require evaluation. The aim of this study was to investigate the relation of GHRH administration and growth hormone secretion and growth effect in growth hormone insufficient children.

The study was started at a time when growth hormone for therapeutic purposes was not available. Initially, an attempt was made to mimic (augment) physiological growth hormone secretion, an aim in which it was successful.⁸ During the initial phase of the study a 1-29 analogue of the naturally occurring GHRH 1-40 became available and was used in some of the children to see if the growth they had achieved on GHRH 1-40 could be maintained on a twice daily regimen. Contemporaneously, another study reported some success using this analogue and regimen⁹ and we enrolled a further cohort of patients to evaluate its success in 'naive' patients who had not previously been treated in this manner. All patients subsequently were treated with growth hormone when the biosynthetic product became available.

All the children showed a growth hormone response to intravenous GHRH administration and the test was of no predictive value in determining the growth response to therapeutic GHRH administration. We used 95% confidence limits to compare the responses with those of normal subjects because of the highly variable responses observed in both normal and growth hormone insufficient subjects. The growth hormone peaks overlap between the groups but the lag between GHRH administration and the first observed growth hormone response was different within the first 10 minutes of the study. This difference would not have been apparent if the first blood sample after GHRH administration had been taken at 15 minutes as with previous studies.⁴⁻⁵ More frequent sampling and further evaluation are required before the role of the dynamic GHRH study is determined in growth hormone insufficiency.

Altogether 75% of the children receiving nocturnal pulsatile GHRH 1-40 and 63% receiving GHRH 1-29 showed a significant change in height velocity SDS. A dose response relation was observed in the pulsatile study and was reflected in the relation between plasma GHRH and growth hormone concentrations as well as between the growth velocity and stimulated growth hormone secretion. A maximum dose response effect was observed in the GHRH 1-29 study with no difference in the growth

response when the daily dose of GHRH 1–29 was doubled. The relation between stimulated growth hormone secretion and growth velocity was only maintained by the children who were transferred from the pulsatile regimen.

Methods of subcutaneous administration include intermittent injection, pulsatile or continuous infusion. Pulsatile GHRH administration was used initially to mimic endogenous pulsatile growth hormone secretion.¹⁴ Thorner *et al* reported the successful treatment of two growth hormone insufficient children receiving pulsatile GHRH 1–40 every three hours throughout the 24 hours.⁷ Smith *et al* reported their initial findings in five children receiving nocturnal pulsatile GHRH 1–40.⁸ This study was to compare the growth response between different GHRH regimens and conventional growth hormone treatment.

Intermittent injection of GHRH has also been investigated by several groups. Hummelink *et al* concluded that a single daily injection of 10 µg/kg was inadequate to achieve a significant growth response.¹⁵ Ross *et al* reported a 44% success rate after the administration of GHRH 1–29 twice daily in a dose of about 25 µg/kg/day.⁹ The growth response of their 'responders' was comparable with the results we found when we used one third of the daily dose of GHRH 1–40 administered in a pulsatile manner.

We recognise that the lack of prospective design strictly diminishes the scientific value of the therapeutic comparison we have made. Nevertheless the results do show that both GHRH regimens were capable of stimulating growth hormone secretion and an increased height velocity in growth hormone insufficient children. Pulsatile GHRH administration, stimulating a more normal pattern of growth hormone secretion, was by far the more cost effective both physiologically and pharmacologically.

Neither GHRH regimen, however, achieved comparable results with growth hormone treatment in either naive growth hormone insufficient children or in children transferred to growth hormone after study completion. Growth hormone treatment is the most appropriate treatment of choice in conventional growth hormone insufficiency. Selection of children for GHRH treatment may require more careful evaluation and GHRH treatment may have a greater role in children who have less severe growth hormone insufficiency in the presence of suboptimal endogenous growth hormone secretion. The results suggest that a more practical long term approach may be the development of a GHRH depot release preparation, which would combine the advantages

of several growth hormone pulses per day and the convenience of infrequent administration.

We are grateful to Ms PJ Pringle and Mr B Rafferty for undertaking the growth hormone and GHRH assays respectively and to Professor M Thorner and KabiVitrum for supplying us with GHRH 1–40 and GHRH 1–29. We are also grateful to Children Nationwide Medical Research Fund for financial support.

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Accepted 15 December 1987