● 308

HUMAN GROWTH HORMONE INCREASES THE INTRACELLULAR FREE CALCIUM IN CULTURED HUMAN IM-9 LYMPHOCYTES. M. Mapoko Ilondo*, P. Bouchelouche# and P. De Meyts*. Hagedorn Research Institute*, Gentofte, Denmark and University Hospital#, Herlev, Denmark.

We studied the effects of human GH (hGH) on intracellular free calcium concentration ($[Ca^{2+}]_i$) in cultured IM-9 lymphocytes, an established human cell line possessing typical somatogenic receptors. $[Ca^{2+}]_i$ was measured on cell suspensions by fluorescence spectrometry using the fluorescent probe fura-2. Resting $[Ca^{2+}]_i$ levels were 93 ± 15 nM. After the addition of 5 nM hGH, $[Ca^{2+}]_i$ levels remained unchanged for about 20 min, then increased steadily and continuously, up to 186 ± 20 nM 40 min after GH stimulation . No change in $[Ca^{2+}]_i$ was observed for up to 1 h in unstimulated cells. The effect of hGH was both specific (not observed after either insulin or IGF-I) and dose-dependent (maximal at 5 nM), and depended on the presence of calcium (1 nM) in the extracellular medium, suggesting a calcium influx. Somatostatin-14 (SMS) in the range 10^{-10} to 10^{-8} M had no effect on $[Ca^{2+}]_i$ but, when added together with hGH, completely prevented the GH-induced rise in $[Ca^{2+}]_i$. Preincubation of the cells with pertussis toxin (PTX) did not have any significant effects on the GH-stimulated $[Ca^{2+}]_i$ increase, but it completely suppressed the inhibition of the GH effect by SMS. In conclusion: 1) GH promoted a slow, progressive and sustained increase in $[Ca^{2+}]_i$ in cultured human IM-9 lymphocytes. 2) This effect probably involves a cation channel negatively regulated by a PTX-sensitive G-protein. 3) Measurements of $[Ca^{2+}]_i$ provide an additional tool for investigating the mechanisms involved in the transduction of the GH signal.

309

TURNER'S SYNDROME: EFFECT OF GROWTH HORMONE AND/OR OXANDROLONE ON HEIGHT VELOCITY AND TURNER SPECIFIC BONE AGE TAKING THE AGE OF ONSET OF TREATMENT INTO ACCOUNT. C.J. Partsch*, E.E. Joss°°, P.E. Mullis°°, W.G. Sippell*, Departments of Paediatrics, Universities of Kiel* Germany and Bern°° Switzerland.

Girls with Turner's Syndrome (TS) were treated with three different regimens: a) Oxandrolone (Oxa) 0.1 mg/kg/day, b) recombinant human growth hormone (rhGH) 28 U/m²/week and c) rhGH 24 U/m²/week with Oxa 0.06 mg/kg/day (rhGH+Oxa). Growth data are presented for the first two years of treatment. None of the girls were given any estrogens. Bone age (BA) was assessed by the TW2-RUS method and expressed in Turner specific bone age (BA'TS) based on the mean BA data for untreated TS (1). Therefore, in contrast to using BA deriving from non affected girls, the increase in BA'TS in untreated TS is by definition 1 year/ year at any age.

	Oxa (n=19)	rhGH (n=26)	rhGH+Oxa (n=12)
Height velocity SDS [mean (SD)]			
- before treatment	-0.8 (0.6)	-0.3 (0.8)	-0.4 (0.7)
- 1st yr of treatment	2.4 (1.1)	3.9 (1.4)	6.2 (1.3)
- 2nd yr of treatment		2.4 (1.3)	4.8 (0.9)
Increase in BA'TS [mean (SD)]			
- in 1st yr (yr/ 1yr)	1.4 (0.8)	1.2 (0.5)	1.8 (0.7)
- in 2 yrs (yr/ 2yrs)		2.7 (0.7)	3.3 (1.0)

In all three treatment regimens, the increase in BA'TS is more pronounced the younger the girls are at onset of treatment. Δ Height SDS for BA'TS, as an index of the gain in final height, is less in girls younger than 8 yrs at the beginning of the treatment.
Conclusion: 1) There is no benefit of any growth promoting treatment in girls with TS before the age of 8 yrs. 2) Adding a small dose of Oxa to rhGH the same increase in predicted final height (Δ Height SDS for BA'TS) is achieved in a shorter time.
1) Ranke M. et al. *Eur.J Pediatr* 1983;141:81-88.

310

COMPARISON OF TWO ALGORITHMS FOR ANALYSIS OF GROWTH HORMONE (HGH) PULSATILE SECRETION BY RECEIVER OPERATED CHARACTERISTIC CURVE ANALYSIS. B.P.Hauffa, P.Beyer, H.Stolecke, Department of Pediatric Endocrinology, University of Essen, F.R.Germany

Parameters of pulsatile hGH secretion are currently evaluated for diagnostic use in children with growth disorders. However, biophysical validation of hGH pulses is lacking. Computer-based peak identification algorithms therefore rely on arbitrary user-defined criteria. Set at increasing stringency, they result in different proportions of true-positive (TPF) and false-positive fractions (FPF)(=receiver operating characteristics,ROC). **Objectives:** To compare two commonly used algorithms (CLUSTER, PULSAR) at different sampling intervals and assay systems (RIA,IRMA), but similar stringency levels. **Methods:** The PULSAR program was used with 5 different sets of G-criteria that decreased the false-positive rate from 0.46% to 0% in noise series. CLUSTER t-statistics were then adjusted to give the same false-positive rate. 4 sets of hGH data obtained during 24 h-sampling (20 min- or 1 h-intervals) from children with growth disturbances were analysed for pulses by 4 observers: A:1 h/RIA (n=50) B:20 min/RIA (n=11) C:1 h/IRMA (n=13) D:20 min/IRMA (n=20). The results were used as a standard, to which PULSAR and CLUSTER results were compared by ROC curve analysis. **Results:** PULSAR: A-SUB-Z values were high (0.93-0.96). TPF ranged from 0.93 to 0.99 without differences between sampling intervals and assay systems, indicating good recognition of real peaks. CLUSTER: TPF ranged from 0.87 to 0.92 at 20 min-intervals, but were lower at 1 h-intervals (0.76-0.84) because peaks at the margin of a profile were sometimes not detected. **Conclusions:** Under the chosen conditions, lowering the sampling frequency decreased recognition of real hGH peaks by the CLUSTER but not the PULSAR algorithm, indicating that both algorithms handle noise series and real peak series differently.

311

CLINICAL EXPERIENCE OF PSYCHO-SOCIAL DWARFISM. R. Stanhope, G. Hamill, D. Skuse, B.C. Thomas, The Medical Unit and The Department of Psychiatric Medicine, The Institute of Child Health, 30 Guilford Street, London WC1, United Kingdom

We describe 65 children (32 girls, 33 boys) from 51 families with an average age of 6.6 years (range, 0.9 to 16.5 years, all but 5 prepubertal) diagnosed by a multidisciplinary team as having environmental growth failure. In 67% the patients lived in families with 3 or more children, but in 73% it was the first or the second born child. 45% of the parents were divorced, in 31% the father was unemployed.
In 56 children the birth weight was known and only in 29% it had been above 3000 g; 21% were premature, 29% had features of a low-birth-weight-syndrome (including four with Russell-Silver-Syndrome); the average birth weight was 2786 g.
In all patients the predominant reason for referral was short stature or growth failure. In 28% psychosocial dwarfism had been suspected, and in a further 29% social or emotional problems were known to the referring physician but not suspected as the aetiology of the growth failure. Social services were already involved in 60% at presentation.
At the first presentation and during the attendance of our clinic additional signs leading to the suspicion of psycho-social dwarfism were found; 42% behavior problems, 54% eating problems, 26% soiling, 18% nocturnal enuresis, and 12% deliberately wetting. During the observation period assessment revealed that 27 of our patients had occult sexual or physical abuse (group A), in 15 patients (group B) such abuse was highly suspected, but in 23 patients (group C) only emotional deprivation could be related to growth failure. In group A, behavior problems, bizarre eating habits, soiling and nocturnal enuresis was more common than in group C.

312

LONG-TERM GROWTH DATA AND GROWTH HORMONE SECRETION IN 65 PATIENTS WITH PSYCHO-SOCIAL DWARFISM. B.C. Thomas, G. Hamill, D. Skuse, R. Stanhope, The Medical Unit and The Department of Psychiatric Medicine, The Institute of Child Health, 30 Guilford Street, London WC1, United Kingdom

We describe the growth of 65 children (32 girls, 33 boys) from 51 families with an average age of 6.6 years (range, 0.9 to 16.5 years, all but 5 prepubertal) diagnosed by a multidisciplinary team as having environmental growth failure.
27 of our patients had occult sexual or physical abuse (group A), in 15 such abuse was highly suspected (group B), but in 23 only emotional deprivation could be related to growth failure (group C).
51 (78%) patients had an endocrine assessment, of whom 34 (67%) had repeated testing (25 (49%) during one uninterrupted hospital admission). 14 (41%) showed reversible growth hormone deficiency, 9 (26%) increased their growth hormone peak level during one admission, 6 (18%) had growth hormone deficiency in repeated testing. In all patients from group A and B the repeated testing demonstrated a noticeable result. Following an insulin stimulation test, only 4 patients demonstrated a cortisol peak concentration of less than 450 nmol/l. The average weight gain during hospital admission was 274 g/week.
27 patients had a change in their environment (10 were separated from their family), all had an increase in their height velocity SDS (from -0.76 (SD 1.6) to +2.39 (SD 2.5)), and 19 (70%) showed catch-up growth (change in height velocity >2).
15 patients were treated with growth hormone before the diagnosis of psycho-social dwarfism was appreciated, and 4 (26%) achieved catch-up growth. All 7 patients who had growth hormone treatment coincident with environmental change achieved catch-up growth.

313

THE GROWTH OF GROWTH HORMONE TREATED AND NON TREATED CHILDREN WITH MYELOMENINGOCELE BEFORE AND AFTER TETHERED CORD RELEASE. D. Rotenstein, D. Reigel, R. Yuretich, J. Lucke, Medical College of Pennsylvania, Allegheny Campus, Pittsburgh, PA 15212, USA.

Children with myelomeningocele (MM) are very short and respond to treatment (Rx) with recombinant human growth hormone (RHGH) with acceleration of growth rate (GR). This complex patient group frequently has tethering of the spinal cord (TSC) which untreated, may cause scoliosis and loss of neurofunction. The effect of tethered cord release (TCR) on the GR of children with MM alone or with RHGH Rx has not been assessed. We compared short term GR and standard deviation score for stature (SDS) of 13 pre-pubertal children (7 males, 6 females of mean age 6.1 ± 2.5 yrs) with MM, pre and post symptomatic TCR to a MM control group (MC) matched for age, sex and level of lesion who were followed over the same time period, and did not have TCR. We also compared these two groups to 7 pre-pubertal children (4 males, 3 females of mean age 6.5 ± 2.0 yrs) with MM and GH inadequacy who had TCR and were then treated with RHGH. We evaluated GR from recumbent lengths measured on a horizontal stadiometer. GR was based on annualization of data points approximately 6 months pre and post TCR. For MC, we annualized lengths pre and post 6 months of the age of TCR to match the TCR group. For the RHGH Rx group, annualization of lengths, 10 months pre and post TCR, were used.

	n	Mean Pre GR cm/yr (SD)	Mean Post GR cm/yr (SD)	Gain GR	Pre SDS	Post SDS	Gain SDS
MC	13	5.4 (2.5)	5.5 (2.6)	0.1	-2.13 (1.2)	-2.17 (1.2)	-.05
TCR	13	5.2 (2.0)	7.0 (2.3)	1.8	-0.98 (1.2)	-0.73 (1.1)	.24
TCR&RHGH	7	4.7 (1.6)	10.7 (3.7)	6.0*	-2.82 (1.3)	-2.26 (1.3)	.56**

* Compared with MC and TCR p<.01 ** Compared with MC p<.03

TSC influences the GR of children with MM. Within the TCR group, GR increased significantly (p<.01). For TCR and TCR & RHGH groups, RHGH provides an increased gain of GR over TCR alone (p<.01). Among the three groups, TCR & RHGH in combination provides the largest gain in GR and SDS. Further study of TCR and RHGH will be necessary to assess their influence on adult stature of this extremely short population.