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DECREASED SERUM SOMATOMEDIN-C (Sm-C) RESPONSE TO GROWTH HORMONE (GH) IN HYPOPHYSECTOMIZED RATS FED A LOW PROTEIN DIET : EVIDENCE FOR A POSTRECEPTOR DEFECT

Reduced number of liver GH receptors may participate to the fall of Sm-C in fasted and protein restricted rats. However, a possible role for a postreceptor defect has not been evaluated. To address this question, the liver GH binding sites and the serum Sm-C responses to single injections of saline or bovine GH (0.25; 0.5; 1 and 5 mg/rat; 5 rats/dose) were determined in hypophysectomized female rats, submitted for 7 days to a normal (15 %; n = 25) or a low protein diet (5 %; n = 25). Binding studies were performed with ^{125}I-bovine GH. The liver GH binding capacities (pmol/mg DNA; mean $\pm$ SE) of the low protein-fed rats (0.85 $\pm$ 0.07) were not significantly different (P > 0.05) from those of normal protein-fed animals (1.09 $\pm$ 0.16). Also, the affinity constants were not affected by protein intake. Basal serum Sm-C levels, determined by RIA, were similar in both groups and increased in a dose-dependent manner in response to GH. However for each GH dose, the Sm-C response in the low protein-fed rats was lower. The maximal Sm-C response to GH (5 mg) was in the malnourished rats (0.13 $\pm$ 0.04 U/ml) significantly reduced when compared to the controls (0.36 $\pm$ 0.03 U/ml; P < 0.001).

In conclusion, the blunted maximal Sm-C response to GH together with unaltered liver GH binding suggest that protein malnutrition induces a GH postreceptor defect.

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LOCALISATION OF THE X-LINKED HYPOPHOSPHATAEMIC RICKETS GENE.

Cloned human X chromosome fragments identifying DNA sequence polymorphisms have been used as genetic markers in linkage studies to localise the abnormal gene that causes X-linked hypophosphataemic rickets. These linkage studies were performed in 11 families in which the disease had occured in 3 or more generations. Leucocyte DNA was extracted and digested with restriction endonucleases (Pst I, Pvu II, EcoRI or Taq I), electrophoresed on 0.8% agarose gels, transferred to Hybond-N membranes and hybridised with an appropriate X chromosome radioactive probe: 782, 754, D2, 99.6 and L128 for the short arm and 52A, DXYS1 and S9 for the long arm. Lod scores for linkage between the restriction fragment length polymorphism (RFLP) and the disease were calculated using the MLINK computer program and the order of gene loci determined with LINKMAP. Linkage was established between hypophosphataemic rickets and an RFLP for probe 99.6 (peak lod score = 4.8, Θ = 0.10) and a distal position within 10 centimorgans of the 99.6 locus is favoured. This localises the hypophosphataemic rickets gene to the region Xp22.31 - Xp21.3.

Thus the gene regulating phosphate excretion is mapped to the short arm of the X chromosome. Defining the gene locus may provide genetic markers for earlier diagnosis and further elucidate the biochemical defect.

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CELL CYCLE DEPENDENT MODULATION OF INSULIN AND IGF I RECEPTOR BINDING TO HUMAN BURKITT TYPE ALL CELLS

Insulin stimulates intracellular carbohydrate protein and lipid metabolism, whereas insulin-like growth factor I (IGF I) promotes cell division. We recently demonstrated that human Burkitt type ALL cells in longterm culture displayed high affinity binding for insulin and IGF I. This rapidly proliferating cell line expresses large numbers of receptor binding sites both for insulin and IGF I.

Cell cycle specific separation was performed by counterflow centrifugation. Cells were analyzed by flowcytometry. Receptor binding was determined by Scatchard analysis. Cells could be enriched to 60-80% purity for G1-S-G2 phase. The insulin receptor displayed 10-15000 receptor sites/cell in G1-, 1000-5000 in S- and 40-50000 in G2-phase. The affinity of insulin binding decreased continously during cell cycle. The IGF I receptor displayed 2000 receptor sites/cell in G1-, 5000 in S- and 15000 in G2-phase The affinity of the IGF I receptor was high in G1-phase showing a sharp decrease towards S-phase followed by a slight increase towards G2-phase.

IGF I shows high affinity binding during G1 in Burkitt type ALL cells. This suggests that IGF I is important for initiation of proliferation. The outstanding reduction in insulin receptor binding sites during S-phase indicates refractoriness of the cell to the metabolic action of insulin during DNA replication.

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TREATMENT OF PRECOCIOUS PUBERTY IN GIRLS WITH INTRANASAL LHRH AGONIST (BUSERELIN)

Ten girls with idiopathic precocious puberty were treated for 1 - 3 years with Buserelin (B) intranasally at a daily dose of 200 - 900 ug. The data are compared to those obtained previously in 21 untreated and in 13 girls with precocious puberty treated with cyproterone acetate (Pediat Res 8,248-256,1974). B was clinically effective in suppressing menses in all but one girl who had low gonadotropins at onset. Breast size did not increase in 8, but pubic and axillary hair development progressed in 8 patients. Peak LH (170.3 $\pm$ 39.5 vs 99.1 $\pm$ 24.9 ug/l) and FSH (451 $\pm$ 73.4 vs 164.8 $\pm$ 29.1 ug/l) after LHRH were markedly suppressed during the first months of treatment and remained low thereafter. Basal LH increased intially (18.6 $\pm$ 3.8 vs 71.1 $\pm$ 17.0 ug/l) and persistently while basal FSH remain unchanged. Prl. E1. E2 levels were unaffected. DHEA gradually increased as seen in normal girls. Differences in height SDS at the beginning (1.72 $\pm$ 0.44). at 1 yr (2.01 $\pm$ 0.43) and at 2 years of treatment (1.95 $\pm$ 0.53) were minimal as were corresponding SDS for bone age (3.27 $\pm$ 0.53; 4.15 $\pm$ 0.54; 4.19 $\pm$ 0.53) and for height prediction related to target height (-1.20 $\pm$ 0.61; -1.40 $\pm$ 0.48; -173 $\pm$ 0.39). The data of statural and skeletal development of B-treated girls are not significantly different from those of untreated girls nor from those treated with cyproterone acetate.

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LONG TERM FOLLOW-UP OF LHRH-A TREATMENT OF PRECOCIOUS PUBERTY BY URINARY GONADOTROPINS (UG) : ESCAPE OF FSH DESENSITIZATION IN GIRLS

The amounts of gonadotrophins in 24 h urines (U-LH, U-FSH) reflect spontaneous gonadotrophin secretion and should be taken as a good marker of the onset of puberty. Thirteen cases of precocious puberty (10 girls and 3 boys) have been treated with monthly IM injections of a long acting D-TRP-6 LHRH (DECAPEPTYL). 24 h urines are collected on days 15 and 30 following each injection. In every case, the efficiency of treatment was certified by the decrease of clinical manifestations, growth velocity, uterine size, plasma levels of testosterone and with no response to LHRH test (J. Clin. Endocrinol. Metab., 1986, 4, 670). In both sexes, after a short rise, U-LH rapidly fall to prepubertal levels and remained unchanged thereafter. In girls, U-FSH decreased dramatically during the first month (p < 0.001), and then rose significantly (p < 0.002) during 2nd-4th month remaining in a prepubertal range. After 5 months of treatment, an important increase of U-FSH was observed in 3 out of the 10 girls with secretion ranging stade II of puberty (> 2.14 IU/24 h) and a bone age > 12 yrs. In boys, this escape of U-FSH has not been observed. Our data show the usefulness of UG excretion in LHRH treatment of precocious puberty. But in bone age > 12 yrs an increase of U-FSH without any clinical relapse suggests a selective escape of desensitization of FSH which could be taken as an index of hypothalamo-pituitary maturation and therapeutic interruption.

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DESENSITIZATION OF PITUITARY VERSUS GONADAL SECRETORY FUNCTIONS OF PERIPUBERTAL MALE RATS BY CHRONIC TREATMENT WITH A GnRH AGONIST

Daily treatment with a GnRH agonist (GnRH-A) rapidly inhibits reproductive functions. In order to evaluate the relative importance of desensitization at the pituitary and the gonadal levels, immature male rats were treated daily with the GnRH-A Buserelin^R (5 ug/d in oil) starting at 40 days of life. After 1 day treatment, testicular content of LH receptors and responsiveness to LH stimulation were already markedly depressed. After 4 days of treatment, inhibition of testicular functions was complete, whereas each pituitary parameter tested was affected but not completely inhibited: plasma FSH was significantly decreased and GnRH receptor content reduced by 50%. LH response to GnRH was blunted but not abolished. After 8 days of treatment, all endocrine functions were extensively impaired. Pituitary LH content dropped to values below 5% of those of control rats. In conclusion, it appears that chronic GnRH-A treatment is very effective to arrest sexual maturation in the male rat and that Leydig cell functions are first affected. Pituitary desensitization was characterized by a post GnRH receptor defect of the LH release process and impairment of LH biosynthesis. Thus, inhibition of sexual functions by GnRH-A includes two sites of action in the male rat, a situation which may not be analogous to that of man for which desensitization of testicular functions appears to be less important.