58 J.P. BOURGUIGNON, A. GERARD* and P. FRANCHIMONT*
Pediatric Clinic and Radioimmunoassay Laboratory, University of Liège, Belgium.

Priming effect of estradiol upon LHRH induced gonadotrophin release by cultured pituicytes of prepubertal and pubertal rats.

Monolayer cultures were prepared from pituitaries of prepubertal and pubertal male rats aged 21 and 50 - 60 days respectively. By comparison with controls in steroid-free medium, pituicytes were incubated for 45 h with 1.10^{-8} M estradiol (E_2), various LHRH concentrations (1.10^{-10} to 1.10^{-5} M) being added for the last 5 h period.

The release of immunoreactive rat gonadotrophins showed a dose related response to LHRH. E_2 did not significantly change basal gonadotrophin release. In pubertal cultures, E_2 induced a significant ($p < .001$) increase of LH response to low LHRH concentrations (1.10^{-10} to 3.10^{-9} M), no significant change being observed with higher LHRH dosages. In contrast, in prepubertal cultures, high LHRH concentrations (3.10^{-9} to 1.10^{-6} M) were required to observe a significant ($p < .02$) priming effect of E_2 upon LH release. Similar findings were obtained for FSH.

It is concluded that E_2 can stimulate gonadotrophin responsiveness to LHRH in prepubertal and pubertal pituicytes "in vitro", suggesting that inhibitory effects of E_2 upon prepubertal gonadotrophs "in vivo" do not result from a pituitary mechanism. In addition, priming effect of E_2 requires the presence of less LHRH in pubertal than prepubertal cultures, suggesting a possible role of increasing pituitary sensitivity to LHRH at puberty.

59 F.COMITE*, T.H.SHAWKER*, W.F.CROWLEY*, D.L.LORIAUX* AND G.B.CUTLER* (Intr. by J.Sidbury). DEB/NICHD/NIH, Bethesda, MD 20205 and M.G.H., Boston, MA 02114, USA

Mechanism of precocious puberty in McCune-Albright syndrome (MAS).

The hypotheses to explain precocious puberty in the MAS include premature pubertal gonadotropin secretion, and autonomous ovarian estradiol (E_2) secretion without gonadotropin (LH and FSH) activation. We have used the observation that luteinizing hormone releasing hormone (LHRH) analogs suppress LH and FSH to prepubertal levels (Clin Res 29:504A,1981) to test the hypothesis of autonomous ovarian E_2 secretion in MAS. A 5 y/o female presented with classic MAS (café-au-lait spots, polyostotic fibrous dysplasia, and precocious puberty). She was treated with D-Trp^6-Pro^9-NEt-LHRH ($LHRH_a$), 4µg/kg/s.c. daily, a regimen which decreases LH, FSH and E_2 to prepubertal levels in all females with idiopathic precocious puberty (IPP). Results (mean±SE) are shown below.

Rx Week	0	3	10	13	18	25	38
LH (mIU/ml)	2±.1	2±.2	3±.1	3±.2	3±.1	3±.2	3±.1
FSH(mIU/ml)	.7±.1	.8±.1	.9±.1	1±.1	1±.3	2±.1	1±.1
E_2 (pg/ml)	64± 4	893±58	31±3	220±33	28±3	814±141	55±10
Ovarian cyst (cm)	2.0		0.3		0.4	3.4	1.8

LHRH-stimulated gonadotropins remained <3mIU/ml throughout therapy. In contrast to IPP, this patient with MAS had marked cyclic E_2 elevations during $LHRH_a$ therapy with no change of gonadotropins. E_2 appeared to correlate with ovarian cyst size by ultrasound. We conclude that in MAS changes in ovarian estradiol secretion appear to be independent of changes in plasma gonadotropin concentration.

60 A. Parra, R. Ramos*, C. Cervantes* and M. Sánchez* Unidad de Investigación and Hospital de Pediatría, CMN, IMSS, Mexico, City.

Changes in plasma gonadotropins in relation to body composition in normal and underprivileged boys.

Plasma FSH and LH were measured in 250 underprivileged boys -- (group 1)aged 9.0 to 15.9 years living on a State's orphanage and in 357 healthy boys(group 2)of similar age and analyzed by chronologic age(CA),weight(Wt),heigth(Ht) and calculated lean body mass (LBM)and total body fat(TBF).Permission was granted by the orphanage authorities as well as by parent(s)and children.Group 1 had a two years delay in the clinical onset of puberty. In both groups, mean Wt at time of initiation of the adolescent spurt in Wt and a time of peak velocity in Wt gain were similar and agreed with values reported for Caucasian boys;the same was true for their Ht. Mean plasma FSH in group 1 was lower than in group 2 from 11.0 to 13.9 years CA($p<0.01$)and the subsequent steep rise occurred in -- group 1 two years later than in group 2;however, when plasma FSH was examined based on Wt,Ht,LBM or TBF, no such quantitative or - qualitative differences emerged. Mean plasma LH in group 1 was -- higher than in group 2($p<0.05$)before 12.9 yrs CA, 42kg Wt, 147cm Ht, 34kg LBM and 9kg TBF;however, the steep rise seen afterwards in group 2 was absent in group 1.Thus, the delayed puberty may be due to a retarded release of FSH or LH or both. Although the steep rise in FSH and LH appeared in group 1 at a later CA than in group 2, it occurred in both groups associated with a similar Wt, Ht or LBM. It is suggested the existence of an association between a -- "critical" level of body composition and the major changes in plasma FSH and LH,both in healthy and multiple deprived boys.

61 H. KULIN, N. BWIBO*, D. MUTIE* and S. SANTNER*. Dept. of Peds., The PA State U. School of Med., Hershey, PA, USA, and U. Nairobi School of Med., Nairobi, KENYA.

The effect of chronic childhood malnutrition on growth and gonadotropin excretion during puberty.

There is little information available in man regarding gonadal vs. hypothalamic-pituitary contributions to malnutrition-related delayed puberty. To investigate this question, a cross-sectional study was made of 342 rural (R) and 358 urban (U) Kenyan (Bantu) chilren, ages 10-17. Comparative data in early pubertal girls revealed the following mean differences ($p<.01$) in anthropometry:

	Age(yr)	Ht(cm)	Wt(k)	Skinfolds(mm)	Arm Circ(mm)	% Body Fat
R	12.6	143.1	31.8	7.6	17.6	13.5
U	11.1	146.7	36.9	15.2	19.9	18.0

FSH and LH excretion (mIU/hr) was determined by radioimmunoassay of timed urine specimens. Mean gonadotropin values for individuals in middle and late pubertal stages were similar between the 2 nutritional groups. In early pubertal childhood, however, LH secretion (but not FSH) was lower in malnourished boys (50±7.0 vs. 76±5.5 mIU/hr, $p<.05$) and girls (55±8.8 vs. 99±12 mIU/hr, $p<.01$), equivalent to levels in the prepubertal, urban group. On the basis of age urinary gonadotropins were consistently lower in rural children 10-13 yr old. Conclusions: 1) The onset of puberty in man is not determined by size. 2) Gonadotropin relationships to age and pubertal stage in the 2 nutritional categories suggest pituitary rather than gonadal factors mediate the delayed onset of puberty associated with malnutrition.

62 E.M. RITZÉN, G.V. SHAH*, and E. DICZFALUSY*
Pediatric Endocrinology Unit and Swedish Medical Research Council, Reproductive Endocrinology Research Unit, Karolinska sjukhuset, Stockholm, Sweden

An improved sensitive and specific in vitro bioassay for FSH

In recent years, it has become increasingly clear that immunological reactivity does not always reflect biological activity of gonadotropins. The present in vitro bioassay for human FSH is based on the FSH dependent aromatase activity of immature rat Sertoli cells. Testes from 10 day old rats are minced and dispersed by enzymatic digestion, and plated in tissue culture micro-wells. After an initial 24 h culture, the FSH containing sample, a phosphodiesterase inhibitor and the substrate (19-hydroxyandrostendione) is added to the cells in a defined medium, and the accumulation of 17β-estradiol during the following 24 h is measured. Recent modifications have increased the sensitivity of the method to 0.1 mIU per sample (0.4 mIU/ml), and the specificity is such that hLH, hCG, hTSH showed <0.6% of the activity of highly purified hFSH, while ACTH, hGH, hPRL, and LH-RH preparations showed no detectable FSH activity. All FSH preparations studied showed a dose-response curve that was parallel to the 1st IRP of human gonadotropins for bioassay (69/104). Serum and urine normally contain inhibitory proteins, that must be removed before the assay.

63 Ch. SULTAN*, J.M. EMBERGER*, B. DESCOMPS* and A. CRASTES de PAULET* (intr. by J. Bertrand) INSERM U58 MONTPELLIER - France.

ANDROGEN METABOLISM AND SPECIFIC 5α-DIHYDROTESTOSTERONE (DHT) RECEPTORS IN HUMAN AMNIOTIC FLUID FIBROBLASTS.

This study was designed to find out whether human amniotic fluid cells have steroid 5α-reductase activity and specific intracellular androgen receptors, and whether they could be used for antenatal diagnosis of male pseudohermaphroditism due to androgen resistance. "Fibroblast-like" cells cultures were raised from 12 amniotic fluids collected at mid-gestation. Specific receptor bound-DHT in the cell lysate was measured by a charcoal adsorption method. The binding specificity of the DHT-receptor was investigated. Physico-chemical properties of the binding component were studied by Sephadex G25 column chromatography, by density gradient ultra-centrifugation and by polyacrylamide gel electrophoresis. The dissociation constant of the 3H-DHT receptor complex varied between .38 and 2.10 x 10^{-9}M and the number of binding sites ranged from 110 to 644 fmoles.mg DNA. Moreover, amniotic fluid fibroblasts were capable of metabolizing androgens : when the cell were incubated with testosterone, DHT and Androstane-diols were measured by gas liquid chromatography. 5α-reductase activity and high-affinity specific DHT binding protein are expressed in amniocentesis cells.

From these data, it is thus possible to identify, prior to birth, individuals with 5α-reductase deficiency or with no detectable androgen receptor.