b) Report of the XXI Annual Meeting of the Latinamerican Society for Pediatric Endocrinology

By Horacio Domené, MS, Marco A. Rivarola, MD and A. Belgorosky, MD, PhD, with the collaboration of Henry Marcano, MD, Tatiane Sousa e Silva, MD, Héctor G. Jasper MD, Roberto Lanes MD, and Carlos A. Longui, MD.

Sociedade Latinoamericana de Endocrinologia Pediátrica (SLEP)
XXI Reunião Anual
Costa do Sauípe (BA) – Brasil
27 a 30 de outubro de 2010
Presidente SLEP 2010: Gil Guerra-Junior
Secretária Geral SLEP: Alicia Belgorosky

Plenary Session Speakers.
Alicia Belgorosky – Hospital de Pediatria Prof. Dr. J. P. Garrahan – Buenos Aires (Argentina)
Annette Grüters-Kieslich – Humboldt University e Charité-Universitätsmedizin – Berlim (Alemanha)
Carlos Alberto Longui – Faculdade de Ciências Médicas da Santa Casa – São Paulo (Brasil)
Irene Néchine – Hôpital d’Enfants Armand Trousseau e Faculté de Médecine Pierre et Marie Curie – Paris (França)
Rodolfo Rey – Hospital de Niños Ricardo Gutiérrez – Buenos Aires (Argentina)
Stephanie Seminara – Harvard Medical School e Reproductive Endocrinology Unit of Massachusetts General Hospital – Boston (EUA)
Veronica Mericq – Institute of Maternal and Child Research, Faculty of Medicine, University of Chile e Pediatric Department at Clinica Las Condes – Santiago (Chile)
Wieland Kiess – Hospital for Children and Adolescents, Medical Faculty, University of Leipzig – Leipzig (Alemanha)

As expressed by President Gil Guerra-Junior “This meeting was attended by distinguished professors and international researchers (Europe, USA and Latin America), that offered excellent lectures and updates in several areas of the Pediatric Endocrinology. It also had more than 200 scientific works - from basic to translational research - that promoted discussions about the paths of the Pediatric Endocrinology in Latin America.” The Meeting was attended by over 800 professionals mainly from Brazil, Argentina and Chile, but also from almost every other Latin American country: Barbados, Bolivia, Chile, Colombia, Costa Rica, Cuba, Ecuador, El Salvador, Guatemala, Honduras, Mexico, Nicaragua, Panama, Peru, Dominican Republican, Uruguay and Venezuela. There were
also were present professionals from Japan, Portugal, Spain and USA. The program included 8 Plenary Conferences, 2 Symposium, 48 oral communications, and 157 posters presented in 2 sessions.

The inaugural conference: Aromatase Deficiency was presented by Dr. Alicia Belgorosky, MD, PhD (Endocrine Unit, Garrahan Pediatric Hospital, Buenos Aires, Argentina). She stated that aromatase is the enzyme complex that catalyzes the synthesis of estrogens from androgens and that the biological importance of the aromatase complex activity is related not only to its role in the synthesis of estrogens but also to its potential influence in the balance of the androgen–estrogen ratio in several tissues. In humans cP450arom appears to be the product of a single gene (CYP19) that is located in the chromosome 15q21.1. The enzyme complex, which is expressed in the ER of cells, consists of cP450arom coupled with a ubiquitous flavoprotein, NADPH-cP450 reductase. In humans, several molecular CYP19 gene alterations associated with complete cP450arom deficiency have been described.

Since 1992 well documented cases of aromatase deficiency have been reported in 46,XX females with ambiguous genitalia due to cP450arom deficiency. They presented absence of breast development and menarche, ovarian cysts, high serum androgen levels and abnormal serum gonadotropins. However, partial aromatase deficiency has been also described. Affected 46,XY males with cP450arom deficiency show normal or slightly elevated basal serum androgens and gonadotropins, mainly FSH. However, a marked increase of LH and FSH is found after GnRH. A decreased motility and number of spermatozoa are also observed, but the pubertal development is reported to take place at normal age. In females during infancy as well as childhood and puberty, minimal amounts of estrogens are required for the feedback mechanism of FSH. Estrogens are not involved in the regulation of FSH secretion in infant boys, but they appear to be one of the components of gonadal feedback of LH and FSH in adult men. Estrogens are required for fertility in men. Dr Belgorosky described a patient with partial aromatase deficiency. Molecular studies revealed that the mutation c655>A was associated to exon 5 skipping. In vitro studies showed that this splicing variant was associated to an inactive protein. On the other hand, this splicing variant might be involved in the complex regulation of aromatase expression in normal steroidogenic tissues. The metabolic syndrome has been described in males with this deficiency. During fetal life, the increment of androgens in males or the lack of androgens might be involved in a mechanism of fetal programming of insulin sensitivity and of the HPG axis. Patient with aromatase deficiency represent another “experiment of nature” that has proven to be a fertile source of knowledge of estrogen physiology and pathology, in particular the role of estrogens on bone maturation, gonadotropin modulation in both sexes, ovarian cyst formation, testicular function and glucose and lipid metabolism.

Dr. Wieland Kiess MD, PhD (Hospital for Children and Adolescents, Medical Faculty, University of Leipzig, Leipzig, Germany) presented his talk: Genome-wide strategies and genetic factors associated with obesity and/or diabetes. He began his lecture with a summary about obesity as a public health problem, and then elaborated on the topic of obesity and its multiple causes ending his presentation with the relationship between obesity and type-2 diabetes.
Reviewing the issue of candidate genes, he first talked about “Inflammation” of visceral adipose tissue relating to high-risk obesity phenotypes. In human obesity, the stroma vascular fraction (SVF) of white adipose tissue (WAT) is enriched in macrophages. These cells may contribute to low-grade inflammation and to its metabolic complications. The first review was on the FTO (fat mass and obesity associated) gene on chromosome 16q12.2, that is consistently strongly associated with early-onset and severe obesity in both adults and children of European ancestry. FTO contributes to human obesity and hence may be a target for subsequent functional analysis. However, the cause of obesity might be multifactorial and involved in the regulation of insulin sensitivity and glucose homeostasis. Indeed, genes and environment interact in the pathogenesis of obesity.

Dr. Stephanie Seminara, MD (Reproductive Endocrine Unit, Massachusetts General Hospital, USA) spoke on Genes controlling human reproduction. She presented a review on the genetic spectrum abnormal GnRH secretion, such as constitutional delay of puberty, hypothalamic amenorrhea, congenital and adult-onset idiopathic hypogonadotropic hypogonadism (IIH). She pointed out that mutations in the genes involved in GnRH deficiency are able to induce abnormalities on olfactory development (WDR11), GnRH neuronal fate specification and migration path-finding (KAL1, NELF, PROK2, PROKR2, FGFR8, FGFR1), synthesis of GnRH (GNRH1), release of GnRH and pulse generation (KISS1R, TAC3, TACR3) and signaling (GNRHR). Some genes for GnRH deficient states affect olfactory development, some affect GnRH release, and several others have other undefined functions. She also discussed the physiology of the kisspeptin pathway, and the use of kisspeptin as a probe of GnRH neuronal function. Because kisspeptin is a powerful stimulus for GnRH release, it can be used as a probe of GnRH neuronal function. In normal men, kisspeptin resets the GnRH pulse generator. In the last part of the conference Dr. Seminara made comments on the physiology of other neuropeptides, such as neurokinin B, that also cause GnRH deficiency and are co-expressed with kisspeptin. These neuropeptides may play different roles in regulating GnRH secretion in the mini-puberty of infancy and in the puberty of adolescence.

Dr. Irene Netchine MD, PhD (Hôpital d’Enfants Armand Trousseau, Paris, France) disserted on Genetic and epigenetic abnormalities of IGF system leading to fetal growth disorders. Dr Netchine said that by definition ~2.3% of the newborns present height or weight bellow normal limits. About 10% of them have a persistent short stature during infancy. Considering the crucial role of IGF-I in intrauterine growth, only a small number of children have been reported to present homozygous IGF1 gene defects or heterozygous inactivating mutations in the IGF1R gene. She also proposed that aberrant genomic imprinting of a given region of the gene might be involved in intrauterine growth restriction. The human chromosome 11p15.5 region contains a cluster of imprinted genes. This cluster is organized in two neighboring imprinted domains, the IGF2/H19 and the KCNQ1 domains, each of them under the control of its own imprinting center, ICR1 and ICR2, respectively. Aberrant genomic imprinting of the 11p15.5 region has a pivotal role in both Beckwith-Wiedeman syndrome (BWS – a syndrome characterized by fetal overgrowth associated with an enhanced tumor risk) and Silver-Russell syndrome (SRS –
characterized by intrauterine and postnatal growth restriction). Epigenetic alterations account for ~ 60-70% of BWS and SRS cases. Dr Netchine presented data on 129 small for gestational (SGA) children classified as SRS or non SRS by using a clinical scoring system (relative macrocephaly at birth, postnatal growth retardation, prominent forehead, body asymmetry and severe feeding difficulties). Fifty eight SRS children were diagnosed by using the clinical score system. In this cohort, 63% presented 11p15.5 ICR1 LOM and only 5.2% maternal uniparental disomy at chromosome 7 (mUPD7).

Dr. Annette Grüters, MD (Charity University Children’s Hospital, Research Institute for Pediatric Endocrinology, Humboldt University Berlin, Germany), presented two conferences. In the first conference: Gene mutations, endocrine disruptors and primary hypothyroidism she described the molecular genetic defects resulting in some cases of congenital hypothyroidism (CH). The most common etiology of primary CH is a spectrum of defective thyroid gland development (thyroid dysgenesis) including athyreosis (no visible thyroid tissue in imaging studies), thyroid ectopy, hypoplasia and hemithyroidea. These forms represent 75-85% of all cases of permanent CH, while defects of thyroid hormone biosynthesis characterized by a normal sized or enlarged thyroid gland in the normal position are present in only 15-20% of the patients.

Congenital hypothyroidism is usually sporadic but organification defects are often recessively inherited. Candidate genes associated with CH can be divided in two different groups: those causing thyroid gland dysgenesis and those causing dyshormonogenesis. Molecular defects leading to thyroid dysgenesis in non-syndromic CH include the TSHR gene and the genes PAX-8, FOXE-1 (TTF-2) and NKX2-1 (TTF-1) in CH associated with different complex syndromes.

Dyshormonogenesis may result from genetic defects involving several different steps in the synthesis of thyroid hormones: sodium iodide symporter (NIS), dual oxidase 2 (DUOX2), thyroid peroxidase (TPO), thyroglobulin (TG) and pendrin (PDS).

TPO gene mutations represent the most common defect in hormonogenesis (30-40% of dyshormonogenesis). These patients have an increased risk for nodules and there is a report of metastatic thyroid carcinoma associated with some mutations located in exon 14. Dr. Gruters presented a recent study where, by using array comparative genomic hybridization (CGH) to map copy number variation (CNV), her group was able to demonstrate an 8.75% of novel CNV affecting exclusively patients with athyreosis or thyroid hypoplasia. It remains to be determined the affected pathways involved in the cause of the disease.

Dr. Annette Grüters, MD also disserted on Update on the management of congenital hypothyroidism (CH). a) Screening strategies. There are three different approaches: 1) primary TSH and backup T4 (this may miss TBG deficiencies, hypothalamic-pituitary hypothyroidism, and hypothyroxinemia with delayed TSH elevation); 2) primary T4 and backup TSH (it will miss delayed TSH elevation with initial normal T4) and 3) primary T4 and TSH: it would be the ideal screening approach. In all cases blood samples should be taken at 2-4 days of age or at time of discharge.
b) Confirmational diagnosis. Primary hypothyroidism may result from thyroid agenesis, ectopic gland, or even normal or augmented non-functional gland. By combining thyroid ultrasound plus T4 and TBG measurements with perchlorate discharge the following three scenarios can be established: 1) negative ultrasound with low levels of T4 and thyroglobulin (TG) are indicative of athyreosis; 2) negative ultrasound associated to measurable levels of both T4 and TG are suggestive of an ectopic gland; 3) while a positive ultrasound with low T4 levels and measurable TG would suggest an hypoplastic gland or, when goiter is present, an organification defect.

c) Outcome. Treatment should be started no later than the first two weeks of life using a “high” dosage regime of L-thyroxine (10-15 µg/kg.day). Adequate treatment results in normal IQ at long term in almost all the detected patients with CH, although they may have reduced IQ relative to siblings.

d) Persistent or transient hyperthyrotropinemia are observed in a low percent of children (relatively frequent in Down syndrome). Among transient causes, iodine contamination, maternal immunoglobulins (TSH receptor Ab) and maternal antithyroid drug therapy must be investigated. Genetic causes of neonatal hyperthyrotropinemia are rare and could be related to congenital syndromes (such as Williams-Beuren-Syndrome, trisomy 21), or TSH-receptor mutations, defects of signal transduction (PHP 1a) or thyroid hormone resistance (defects of TRβ). Indications for treatment with L-T4 of hyperthyrotropinemia are a progressive decrease in T4 levels, increasing TSH (> 10 mIU/l after one month) or when goiter or severe hypoplasia is present.

e) Proposed guidelines for treatment of CH include: initial dose: 10–15 µg/kg.day, full term: 50 µg/day, preterm: 25–37.5 µg/day, adjustments according to TSH, T4 (FT4) and T3 concentrations. At the end of the first year: 5 µg/kg.day. Maintain TSH levels < 1.4 mU/L. No thyroxine before blood is drawn.

Dr. Rodolfo Rey, MD, PhD (Endocrine Unit, R. Gutiérrez Children’s Hospital, Buenos Aires, Argentina) presented The César Bergadá Lecture, in memoriam of the founder of SLEP. He was introduced by Marco A. Rivarola. It was entitled: Fetal hypogonadism and disorders of sex development: what we learned from Anti-Mullerian Hormone. Anti-Mullerian hormone (AMH) initially known as Mullerian Inhibiting Substance (MIS) has been studied since 1947 in the Jost’s pioneering experiments in sex differentiation. AMH is a member of TGF super family and has an essential role in sex differentiation. The Persistent Mullerian Duct Syndrome (PMDS) is associated with normal male external genitalia, cryptorchidism, persistence of uterus, abnormal AMH production (AMH gene mutation) or insensitivity to AMH (AMHR gene mutation). While prepubertal, patients with PMDS present higher serum AMH concentration in defective AMHR type II gene than that observed in defective AMH gene. Mutations span through the whole AMH and AMHR-II genes, without hot spot regions. In the male fetus, the biological effect of AMH correlates with regression of Mullerian Duct (MD). MD formation starts at week-5 after fertilization. MD regression starts at week-8 and finishes at week-10 by apoptosis types I and II. Other biological effects of AMH are described in Leydig and Granulosa cells and extragonadal tissues. AMH expression is a useful marker of Sertoli cell function to identify the fetal, neonatal and infantile hypogonadism. In the fetal testis, the regulation of AMH expression is
gonadotropin independent during the 8-12 weeks of fetal life. During postnatal life AMH is elevated in the prepubertal male; in puberty testosterone down-regulates AMH in a gonadotropin dependent manner. In patients with defects of androgen synthesis or action higher levels of serum AMH are observed. These findings are also described in Sertoli cell-specific Androgen Receptor knockout mice (S-ARKO). Sertoli cells are physiologically androgen-insensitive from fetal life until 6-12 months after birth, becoming increasingly sensitive by the age of puberty, when AMH inhibition by androgens occurs. During the FSH regulation of AMH expression, SOX9 and SF1 sites are essential for cAMP-mediated AMH up-regulation. PKA mediates the activation of SOX9 and SF1 expression in the SMAT1 cell line. cAMP induces SF1 nuclear translocation in SMAT1 cells. AMH has been measured in infants with congenital multiple pituitary deficiencies, in congenital hypogonadotropic hypogonadism, and in patients with disorders of sex development (DSD). Fetal sex development consists of morphogenesis of gonadal and genital primordial, gonadal differentiation, and androgen- and AMH-driven genital differentiation. Early fetal-onset hypogonadism in Y-chromosome bearing patients is usually related to a whole gonadal dysfunction determining a DSD state with uterus persistence. Leydig cell-specific dysfunction is typically represented by a DSD state without the presence of uterus. Sertoli cell-specific dysfunctions include males with uterus. In whole gonadal dysfunction, AMH is undetectable / low; in Leydig cell-specific dysfunction AMH is normal / high and in Sertoli cell-specific dysfunction AMH is undetectable. AMH represents not only an excellent marker of gonadal function in boys with either primary or central hypogonadism, but also of follicular reserve in girls.

Dr. Wieland Kiess’s second conference was entitled: Metabolic syndrome in children and adolescent. There is an increased prevalence of obesity worldwide, due in part to an increase in the intake of calories and to sedentary activities (time spent watching television and playing video games), with a decrease in physical activity. Although there is a high cell turnover in adipose tissue, the number of fat cells is determined in childhood and this in turn determines the fat mass. The number of fat cells does not change during adulthood (whether the individual is obese or not), but an increased storage of lipids per fat cell in adults may occur. Obesity in childhood may lead to an increased risk of cardiovascular disease in adulthood. An increase in prevalence of the metabolic syndrome which is characterized by obesity, high blood pressure, increased levels of LDL and fasting glucose and a decrease in HDL concentrations has been noted both in children and in adults. Increased levels of insulin, proinflammatory markers (IL-6 and TnF alpha), leptin, and of adhesion molecules (ICAM, VCAM, selectins) has been noted in obese children. Endothelial progenitor cells, which play a role in vascular healing, are decreased in peripheral blood in obese subjects as has been demonstrated by Dr Kiess. Intervention may include lifestyle changes, nutritional therapy, increased physical activity, the use of pharmacological agents (sibutramine, fenfluramin, phentermin, orlistat, recombinant leptin, metformin) and surgery. Short and long term goals need to be established while clear agreements, constant evaluation and stepwise implementation of behavioral changes need to take place. Questions such as why should I lose weight? How much do I want to lose? How much effort do I want to invest? self image (wish/fact), need
to be addressed by the patient. Vulnerable groups include children, socio-economically disadvantaged populations, and aboriginal populations, (ethnic groups). Decreased weight gain and a reduction in fat mass following a healthier lifestyle and school and family based intervention can be achieved. However positive results have been limited to a relatively small percentage of the targeted subjects and long term goals are difficult to maintain. In conclusion, the prevalence of obesity in children and adolescents is increasing in most countries. Co-morbidities of obesity such as type 2 diabetes are already present in childhood in some populations. Metabolic and cardiovascular risks are interrelated and BMI and the degree of obesity are directly related to the prevalence (and the extent) of these co-morbidities. Prevention of obesity and type-2 diabetes in children and adolescents can be achieved and should be a primary goal for public intervention. Prevalence of these problems need to address social, cultural and economic issues, as obesity is a disease with severe individual and societal consequences.

**Two symposia were included in the Meeting:**

**S1: Symposia 1**

1) **Stephanie Seminara**  
**Current treatment of hypogonadism**

2) **Veronica Mericq**  
**Intrauterine and perinatal influences in development of gonadal function**

**Chairpersons:** Romolo Sandrini & Raquel Burrows

1) **Current treatment of hypogonadism** by Stephanie Seminara, MD (Reproductive Endocrine Unit, Massachusetts General Hospital, USA). Dr. Seminara stated that androgen replacement is the clinical mainstay of boys with hypogonadism from adolescence onwards. Androgen is able to induce and maintain secondary sexual characteristics, to optimize accrual of bone mineral content and muscle mass, and to promote physical and social well being. There is a spectrum of GnRH Deficiency, ranging from absence of sexual maturation to partial puberty to near-normal testicular size (“fertile eunuch” variant) to an acquired form of IHH that occurs after completion of puberty. It can be related to neuroendocrine pattern (absence of GnRH induced LH pulses to disorders of LH pulse amplitude, frequency, and bioactivity) or to phenotypic anomalies (midline facial defects, renal agenesis, synkinesia and cryptorchidism). Pubertal development can be complete, partial or absent; cryptorchidism can be unilateral or bilateral; microphallus is usually present; reduced testicular volume is also found. Dividing patients according to presence or absence of prior pubertal development allows discrimination between groups. Clinical and biochemical markers of testicular growth and differentiation can discriminate between GnRH deficient men with and without puberty. Mean gonadotropin levels and current pattern of LH secretion are not reliable surrogate markers. Kallmann syndrome (KS) represents the most complete form of GnRH deficiency, and the vast majority of patients have: complete lack of sexual development, lack of pulsatile GnRH induced LH secretion, high incidence of cryptorchidism and microphallus, low inhibin B levels and histologically immature testes. In contrast, non-Kallmann IHH patients display the widest clinical spectrum of GnRH deficiency. Traditional treatments include sex steroids, exogenous gonadotropins or pulsatile GnRH. Dr. Seminara emphasize some of the major findings during the
treatment of hypogonadism: i) the efficacy of hCG monotherapy: even in the absence of FSH, hCG can complete spermiogenesis in partial IHH, and the response is predicted by the initial testicular volume; ii) pulsatile GnRH vs. exogenous gonadotropins: more rapid spermatogenesis and higher testicular volume with GnRH when compared with the treatment with gonadotropins. Patients who did not respond to gonadotropins usually achieve adequate testicular stimulation in response to GnRH; iii) comparison of testicular responses to pulsatile GnRH vs. exogenous gonadotropins: in severe GnRH deficiency (TV< 4 ml) the pulsatile GnRH therapy for 2 yr does not enhance testicular growth, hasten the onset of sperm, or increase sperm output during the concomitant use of hCG/hMG.

Outcome of GnRH deficient men during GnRH treatment is generally successful in inducing virilization and spermatogenesis. However, a subset of men fails to reach normal TV and produce sperm. The prognosis of this population can be stratified according to degree of prior pubertal development: Group I: absent puberty; Group II: partial puberty; Group III: adult onset HH.

Dr. Seminara described the favorable predictors for achieving adult testicular size and optimizing spermatogenesis: normalization of LH/Leydig cell/Testosterone axis is achieved more uniformly than the FSH/Sertoli cell axis. Favorable predictors include: prior history of sexual maturation; baseline inhibin B > 60 pg/mL and absence of cryptorchidism. She also pointed out some important aspects during the follow-up: once spermatogenesis is induced, it can be qualitatively maintained by hCG alone. However, declining sperm counts indicate that FSH is required for normal spermatogenesis. Low sperm count does not preclude fertility: men with IHH can initiate conception even when their sperm concentration is well below 20 millions/ml.

Dr. Seminara also talked about the reversal of IHH/KS in 10% of hypogonadotropic hypogonadal population, and its biological implication: GnRH neuronal network is in the hypothalamus; following sex steroid therapy, the GnRH neurons become able to fire in a pulsatile fashion and to induce/sustain a normal reproductive HPG axis activity. She hypothesized that could be an analogy between ‘reversal’ of GnRH deficiency in men and hypothalamic amenorrhea in women. She suggests that the main clinical implication of these findings is the need of periodic discontinuation of therapy to identify the reversal and if persistent therapy is still needed. She finished her presentation showing the preferential activation of GnRH secretion by neurokinin B during the neonatal period (mini-puberty period). Mutation in TAC3-neurokinin B or TACR3 abrogate HPG axis activation at this period, but can be reversed in adulthood after androgen therapy.

2) Intrauterine and perinatal influences in development of gonadal function by Dr. Verónica Mericq, MD (Institute of Maternal and Child Research, Faculty of Medicine, University of Chile and Pediatric Department at Clínica Las Condes, Santiago, Chile).

1. Evidence exists for a secular trend towards an earlier onset of puberty in several countries. Variations in pubertal timing and progression of puberty may be related to ethnicity, genetic background, geographic localization, nutrition, environmental factors, endocrine disrupting compounds such as “xenoestrogens” (estrogen mimics), antiandrogens and other perinatal factors.

2. The number of ovarian germ cells reaches a peak during the sixth month of prenatal life and then tends to decrease gradually during childhood and
adolescence. 3. Overnutrition may accelerate puberty. The increased incidence of obesity in girls may contribute to an increase in the secretion of androgens and in LH pulse frequency. An increased incidence of the metabolic syndrome has also been noted in these females. 4. In children born small for gestational age (SGA), particularly in those with a rapid weight gain following birth, an increase in insulin secretion (basal and postprandial) has been reported. Some studies have reported small ovaries and uterus, FSH hypersecretion in infancy and adolescence, reduced ovulation rates, hyperinsulinemia, low grade inflammation and increased incidence of PCOS in females born SGA. However, the reported association of low birth weight and hyperandrogenism, manifested as an increased incidence of precocious pubarche and of polycystic ovarian syndrome remains controversial. One can conclude that intrauterine growth retardation predisposes towards insulin resistance, but not to hyperandrogenism in young women. 5. In females born SGA controversy exists regarding the progression of puberty and the timing of menarche. Some studies report normal pubertal timing, while others suggest slightly earlier age of menarche and a rapid pubertal progression. 6. Reports of reduced testicular size, decreased testosterone and inhibin B and increased LH levels in males born SGA can be found. However other studies report normal pituitary-gonadal function in adolescent males.

7. In a recent Chilean study, appropriate for gestational age (AGA) and SGA children were studied at the beginning of puberty and followed for 3 years. Both groups were found to have similar waist circumference and central adiposity, final height for target height and age of menarche. However, the tempo of puberty in females, particularly in regard to breast development, advanced more rapidly in children born SGA and estradiol, leptin and stimulated LH concentrations were increased in these subjects. While HOMA was increased at 1 year of follow up and DHEAS and 17OH-P were increased at 2 years in children born SGA, these values were similar to those of children born AGA at 3 years of follow up.

S2: Symposia 2
1) Irène Netchine
Recent advances in genetic anomalies of GH deficiency: isolated and Hypopituitarism
2) Carlos Longui
Clinical impact of the identification of glucocorticoid sensitivity
Chairpersons: Ivo Arnhold & Hector Jasper

1) Recent advances in genetic anomalies of GH deficiency: isolated and hypopituitarism by Dr. Irene Netchine, MD, PhD (France)
Dr. Netchine revised the genetic defects described in the somatotrophic axis, pointing out that at least 18 different genes have been identified in which molecular defects result in alteration of GH secretion or action. She mentioned: HESX1, GLI2, OTX2, SOX2, SOX3, LHX3, LHX4, PROP1, POU1F1, GHRHR, GHSR, GH1, GHR, STAT5B, IGFALS, IGF1, IGF2 and IGF1R. In the particular case of GH deficiency, it is useful to follow a candidate gene approach that include: 1) the clinical phenotype, 2) an endocrine evaluation to determine whether GH deficiency is isolated or associated to other pituitary hormones (combined pituitary hormone deficiency – CPHD), 3) family history, 4) associated malformations and 5) pituitary morphology.
CPHD may result from defects in genes involved in pituitary determination (e.g. \textit{HESX1} or \textit{SIX3}), pituitary organogenesis (\textit{LHX3} or \textit{LHX4}) or specification (\textit{PROP1} or \textit{POU1F1}). Gene defects affecting early pituitary determination or organogenesis are usually associated with syndromic forms of CPHD, while more distal genetic defects result in non syndromic CPHD.

\textit{HESX1} (also known as Rathke Pouch Homeobox, \textit{RPX}) is the earliest known marker for pituitary primordium, suggesting that it has a role in the early determination of the pituitary. The first molecular defect in this gene was described by Dattani et al in 1998 in two sibs with septo-optic dysplasia (SOD). Thirteen different \textit{HESX1} gene mutations have been described in patients with SOD and hypopituitarism. They display an autosomal recessive or dominant mode of inheritance. Although they may present as isolated GH deficiency, most frequently they show CPHD. Hormone deficiency include GH associated to TSH, LH and FSH deficiencies, and in some cases ACTH deficiency. Neuroradiological findings include anterior pituitary (AP) hypoplasia or aplasia and ectopic posterior pituitary (PP). \textit{LHX3} gene is early expressed in the anterior pituitary and the motoneurons. Patients carrying \textit{LHX3} gene defects present CPHD associated to impaired neck rotation. \textit{LHX4} gene defects result in GH deficiency associated in some patients to TSH or ACTH deficiencies. Neuroradiological findings include hypoplasia of AP, ectopic PP and malformations of sella turcica. Six different \textit{LHX4} gene mutations have been described an in all of them inheritance is autosomal dominant.

Originally described by Wu et al in 1998, at least 24 different mutations have been reported in \textit{PROP1} gene, resulting the most frequent gene defects associated to CPHD. \textit{POU1F1} gene defects were simultaneously described by Radovick et al and Pfaeffle et al in 1992. In both cases CPHD is non syndromic and GH deficiency is associated to TSH and PRL. LH and FSH deficiencies may also be present in \textit{POU1F1} deficient patients, and ACTH deficiency may appear later in life.

2) Clinical impact of the identification of glucocorticoid sensitivity by Dr. Carlos Longui, MD (Pediatric Endocrinology Unit, Santa Casa SP, School of Medical Sciences, São Paulo, Brazil). He conducted his presentation in terms of molecular mechanisms of action, glucocorticoid sensitivity (in vitro and in vivo) and clinical implications of tissue variability in GC sensitivity.

Glucocorticoids exert a wide variety of physiological and pathological responses, most of which are mediated by the ubiquitously expressed glucocorticoid receptor (GR). The glucocorticoid response varies among individuals, as well as within tissues from the same individual, and this phenomenon can be partially explained through understanding the process of generating bioavailable ligand and the molecular heterogeneity of GR. Levels of circulating cortisol are controlled systemically by the HPA axis and locally by the action of 11\textbeta-HSD enzymes. This dual regulation ensures the maintenance of glucocorticoid homeostasis. Perturbations of either of these systems contribute to the development of diseases of metabolic origin. Heterogeneity within the GR is emerging as an additional mechanism for modulating endogenous and exogenous glucocorticoid responses. Alternative processing of the GR gene has the potential to explain tissue-specific glucocorticoid responses. Individual-specific heterogeneity manifests as
polymorphisms within the GR and might predispose individuals to disease in addition to influencing their responses to glucocorticoid therapy. A more in-depth understanding of the molecular diversity of GR will aid in the development of tailored, individualized glucocorticoid therapies. Chemical changes in the cortisol molecule can enhance glucocorticoid or mineralocorticoid activities, determining improved therapeutic properties and decreased side effects. Anti-inflammatory potencies have been defined in studies based on *in vivo* and *in vitro* methods. In vivo, other tests to determine GC sensitivity have been described, but only employing pharmacological doses. A very low dose of dexamethasone was able to create a gradient of cortisol suppression that could be useful in identifying an individual's sensitivity to glucocorticoids. The wide variability of responses to corticotherapy suggests a role for individual recognition of steroid sensitivity in order to customize treatment.

The activity of the hypothalamic-pituitary-adrenal axis is usually modulated by several stress factors, including exercise. Different responses are induced by physical training according to the duration and intensity of exercise. During prolonged training, cortisol remains normal or decreased as a consequence of altered cortisol secretion, metabolism and excretion, and possibly by changes in glucocorticoid sensitivity. Prolonged physical training is able to reduce glucocorticoid sensitivity, which can have a beneficial impact in chronic stress conditions. Prolonged physical training decreases the glucocorticoid sensitivity and the mRNA levels of the *GR* gene combined with decreased mRNA of genes related to the *NFkB* pathway.

**SLEP AWARDS.**

The Hormone Research Prize was awarded to the following oral presentation:

**Intracellular trafficking effects of Insulin-Like Growth Factor Acid Labile Subunit gene (IGFALS) mutations identified in idiopathic short stature (ISS) children.** Guida MC<sup>1</sup>; Nitschke R<sup>2</sup>; Domené HM<sup>1</sup>; Karabatas L<sup>1</sup>; Scaglia P<sup>1</sup>; Martinez A<sup>1</sup>; Keselman A<sup>1</sup>; Bergada I<sup>1</sup>; Cassinelli H<sup>1</sup>; Bengolea SV<sup>2</sup>; Heinrich JJ<sup>1</sup>; Pipman V<sup>4</sup>; Ropelato MG<sup>1</sup>; Ballerini MG<sup>1</sup>; Rey RA<sup>1</sup>; Jasper HG<sup>1</sup>-<sup>1</sup>CEDIE/CONICYT, División Endocrinología – Hospital de Niños Ricardo Gutiérrez, Buenos Aires, Argentina; <sup>2</sup>Life Imaging Center, ZBSA - Albert-Ludwigs-Universität, Freiburg, Germany; <sup>3</sup>Endocrinología - Hospital Fernandez, Buenos Aires, Argentina; <sup>4</sup>Pediatría - Hospital Tornú, Buenos Aires, Argentina. Heterozygous IGFALS gene mutations are present in 11.2% of ISS children. Three of these mutations (P287L, A330D and R548W) were selected to determine their *in vitro* effects on intracellular ALS trafficking. ALS, IGFBP-3 and IGF-I serum levels were < -2 SDS in the patients carrying the P287L or R548W mutations but normal in the patient with A330D. The effects of the mutations on intracellular trafficking were studied by generating expression vectors of ALS, wild type (WT) or mutated, tagged to a fluorescent protein (pECFPN1-ALS). CHO cells were cotransfected with pECFP-N1-ALS WT or mutated and a fluorescent trans-Golgi marker vector, TGN38. YFP. We found a significant increase in localization of P287L and R548W ALS mutants, but not of A330D, in trans-Golgi as compared to ALS-WT (Pearson's colocalization coefficient between ALS and TGN38, mean±SE: WT 0.32±0.03; P287L
Two abstracts were selected to receive the SLEP-2010 Awards:

1) Glucocorticoid-remediable aldosteronism (Familial primary hyperaldosteronism type 1) in children: Prevalence, clinical and biochemical characteristics. Bancalari R1; Garcia H2; Aglony M3; Martinez-Aguayo A4; C Carvajal5; Campino C6; Mericq V7; Fardella C6 - 1Pontificia universidad catolica - pediatric; 2Universidad Catolica – Endocrinologia Infantil; 3Universidad Catolica - Nefrologia Infantil; 4Pontificia Universidad Catolica de Chile - Pediatria; 5Universidad Catolica de Chile - Endocrinologia; 6Universidad Catolica - Endocrinologia; 7Institute of Maternal and Child Research (IDIMI) - School of Medicine, University of Chile, Santiago, Chile.

Familial hyperaldosteronism type I (FH-1) is caused by a chimeric gene (CG).

Aim: To report the prevalence of FH-1 in hypertensive children and to describe their clinical and biochemical characteristics. Patients and methods: We studied 120 untreated hypertensive children in which we determined plasma potassium (K), Plasma Renin activity (PRA), serum aldosterone (SA) and the aldosterone to renin ratio (ARR).

Results: We found 4/120 (3.3%) children with ARR>25. A CG was confirmed in all of them. The same study was conducted in 20 first degree relatives, 8 of whom were affected, making a total of 12 patients. The PRA was suppressed (< 0.3 ng/mL/h) in 6/12 and hypokalemia (K <3.5 mEq/L) was present only in 3/12 patients. A high SA levels (>16 ng/dL) was present in 10/12 patients. The ARR >25 was observed in 11/12 affected subjects, the lower value corresponds to a young woman which had pre-hypertension.

Conclusion: The prevalence of FH-1 in pediatric hypertensive population was high when a ARR >25 was used as screening, we suggest that a lower ARR cut off should be determined in them.

2) WNT/Beta -Catenin Pathway y Dysregulation in Childhood Adrenocortical Tumors (ACT). Bancalari R1; Garcia H2; Aglony M3; Martinez-Aguayo A4; C Carvajal5; Campino C6; Mericq V7; Fardella C6 - 1Pontificia universidad catolica - pediatric; 2Universidad Catolica – Endocrinologia Infantil; 3Universidad Catolica - Nefrologia Infantil; 4Pontificia Universidad Catolica de Chile - Pediatria; 5Universidad Catolica de Chile - Endocrinologia; 6Universidad Catolica - Endocrinologia; 7Institute of Maternal and Child Research (IDIMI) - School of Medicine, University of Chile, Santiago, Chile.

Introduction: In Brazil the high incidence of Childhood ACT is associated with the R337H p53 mutation. Abnormalities of the Wnt pathway have been described in ACTs. In vitro, p53 regulates the Wnt pathway but this action has not been demonstrated in vivo in adrenal disease.

Objective: To investigate abnormal expression of Wnt pathway genes in childhood ACT and its relationship with p53 mutation and outcome.

Methods: 53 patients with ACT from two reference centers in Brazil were evaluated (12M/41F; age: 3.4±3.7
All but one patients present hormone excess. Tumor stage: I (n=28), II (n=8), III (n=8), IV (n=9). Expression of CTNNB1, Axin, DKK3, MYC, WISP2, and p53 was measured by RT-PCR and calculated by 2-ΔΔCt. Controls: 8 normal adrenals. **Results:** R337H p53 mutation was present in 87% of the patients. ACTs overexpressed CTNNB1 (p=0.01) and underexpressed DKK3 (p<0.0001), Axin (p=0.05) and MYC (p=0.01). ACTs III/IV presented lower expression of p53 than ACTs I/II and controls (p=0.06). Underexpression of p53 was observed in patients with R337H p53 mutation (p=0.04). There was a correlation of p53 and DKK3 (p=0.04), Axin (p<0.0001), MYC (p=0.0003) and WISP2 (p=0.0017). **Conclusion:** Wnt pathway is dysregulated in childhood ACTs and correlates with the R337H P53 mutation.