P1-d2-375 Sex Dif ferentiation 1

The European DSD register – a platform for international collaborative research

Martina Rodie1; Jipu Jiang1; Richard Sinnott2; S. Faisal Ahmed2
1University of Glasgow, Department of Child Health, Glasgow, United Kingdom; 2University of Glasgow, National e-Science Centre, Glasgow, United Kingdom

Background: Effective research into understanding the aetiology of Disorders of Sex Development (DSD), as well as long-term outcome of these rare conditions, requires multicentre collaboration often across national boundaries. The EU-funded EuroDSD programme (www.eurodsd.eu) is one such collaboration involving clinical centres and clinical and genetic experts. At the heart of the EuroDSD collaboration is a DSD Register that supports the sharing of DSD data.

Methods: Over the past 12 months the DSD Register has remained central to the EuroDSD project. In addition, the number of individual users has increased by approximately 30% with 23 new users from 14 new centres in 7 new countries and in 1 new continent. Users are provided role based secure access.

Results: At last review (February, 2011) there were 847 cases on the Register from 12 countries. The United Kingdom has the largest number of cases on the Register currently (n=248), followed by the Netherlands (n=179). The age of first presentation ranges from <1 month to 53 years and the median year of birth was 1995 (range 1927-2010). 58% (n=489) cases were assigned female sex and 42% (n=358) were assigned male sex. There was a history of infertility or parental consanguinity in 8% (n=68) and 11% (n=89) cases respectively, and 29% (n=227) cases had associated malformations.

Conclusions: The Register has attracted much international interest over the last year and is changing from a European initiative to an international activity. It provides a virtual research environment within which clinicians and investigators can interact and develop new DSD related studies.

P1-d2-376 Sex Dif ferentiation 1

Exploring the utility of urine steroid metabolite ratios in evaluating 544 suspected cases of disorders of sex hormone synthesis

Martina Rodie1; David Shapiro2; Rosanne Howarth9; Norrice Liu1; Malcolm Donaldson1; M Gutter Shaike1; S. Faisal Ahmed2
1University of Glasgow, Department of Child Health, Glasgow, United Kingdom; 2University of Glasgow, National e-Science Centre, Glasgow, United Kingdom; 9Glasgow Royal Infirmary, Department of Clinical Biochemistry, Glasgow, United Kingdom

Background: Calculation of a urinary steroid metabolite ratio (uSMR) may be a useful method of improving diagnostic yield when investigating disorders of steroid hormone synthesis.

Objective and hypotheses: To determine the range of uSMR and the relationship between the uSMR and the diagnostic outcome in abnormal cases.

Population/methods: Ten ratios were calculated on steroid metabolite data previously analysed by GC-MS in urine samples from 544 patients under 18 years collected between 2008-2010.

Results: Of 544 patients, 337 (62%) were female, 189 (35%) were male and in 18% (3) the sex was not documented. Indicators for performing the test included adrenal or XY virilisation in 238 (44%) cases and XY DSD in 68 (13%) cases. Median age at test was 7.4 years (1 day, 18 years) with 11%, 4%, 3%, 3%, 7%, 16%, 40%, 6%, 6% and 4% of tests performed in the following age bands, <1wk, 2wks-2mths, 2-5mths, 6-11mths, 1-3yrs, 4-6yrs, 7-9yrs, 10-12yrs, 13-15yrs and 16-18yrs, respectively. In 97 (18%) patients, one or more abnormal uSMR was associated with an identified carrier of the NR5A1 gene. In the remaining 447 cases, no abnormal uSMR was detected.

Conclusions: Applying simplified diagnostic ratios to urinary steroid profile data results in a yield of about 20% cases being abnormal. There is a need to improve the sensitivity and specificity of the uSMR and the reference ranges for these ratios.

P1-d2-377 Sex Dif ferentiation 1

Study of the NR5A1 gene in a cohort of Italian patients with 46,XY Disorders of Sex Development (DSD) without adrenal insufficiency: identification of 7 novel mutations

Lilia Balsazzzi1; Antonio Balsamo2; Annalisa Nicoletti7; Soara Menabò3; Diego Rinaldi5; Giuseppe Cangemi5; Claudia Balsamo6; Piero Pirazzoli5; Alessandro Cicognani5
1S. Orsola-Malpighi University Hospital, Gynecology, Obstetric and Pediatric, Bologna, Italy; 2University of Bologna, Gynecology, Obstetric and Pediatric, Bologna, Italy

Background: The NR5A1 gene encodes the Steroidogenic factor 1 (SF1), a nuclear receptor involved in adrenal and gonadal development. NR5A1 heterozygous mutations were associated with 46,XY DSD in humans. Patients may present with severe underandrogenization and hormone profiles suggestive of PAIS.

Objective and hypotheses: Analysis of the NR5A1 gene in the 16 46,XY DSD Italian patients without AR/SDX52A mutations.

Methods: All patients present with severe underandrogenization at birth: female external genitalia with enlarged clitoris and inguinal retained testis. With the exception of the 2 most recently observed, all were submitted to gonadectomy and female correction of external genitalia in the first years of life. To date, adrenal insufficiency has not occurred in any of them. The NR5A1 gene analysis was performed by PCR and direct sequencing.

Results: Heterozygous SF1 mutations were found in 7 cases. Two mutations are predicted to disrupt RNA stability: p.E51X in the DBD and the frameshift mutation c.1016_1025delCCGGGCT in the LBD; five are missense mutations affecting conserved residues: two in the DBD (p.T29K, p.Y47C), three in the LBD (p. R313H, p.A351E, p.Q460R). As far as we know, all the identified mutations are novel. DNA analysis of the parents and clinical evaluation of the identified carriers are running. The degree of conservation of the affected residues, as well as the type of substitution, suggest an implication in the phenotypes observed also for the missense mutations.

Conclusions: This study, that enlarges the spectrum of the NR5A1 gene mutations, confirmed the high frequency of these mutations among 46,XY DSD patients without adrenal insufficiency, thus underlining the role of SF1 haploinsufficiency in humans. It also confirms the importance of performing NR5A1 gene analysis in this group of patients in order to optimize the genetic counselling and the treatment/follow up of both the patients and the 46,XX carriers.

P1-d2-378 Sex Dif ferentiation 1

DNA methyl transferase 3A -448A>G variant in foreskin tissue from children with hypospadias

Roberta Minz1; Isabella Viani1; Francesca Savina1; Chiara Sartrori1; Sergio Bernasconi1; Lucia Ghizzoni2; Alessandra Vottero2
1University of Parma, Department of Pediatrics, Parma, Italy; 2University of Turin, Department of Internal Medicine, Turin, Italy

Background: A hypermethylation of the AR gene associated with an increased expression of DNA methyl transferase 3A (DNMT3A) was detected by our group in foreskin tissue from hypospadiac patients compared to controls, suggesting a possible role of DNMT3A in the etiopathogenesis of this disease. Hong et al. reported that the DNMT3A promoter variant -448A (rs150017) was associated with a higher promoter activity (> two fold) compared to the wild-type -448G allele.

Objective and hypotheses: The aim of this study was to analyze whether in hypospadiac patients the DNMT3A promoter genetic variant -448A>G is associated with the increased DNMT3A transcript.

Methods: Genomic DNA was isolated from foreskin tissue obtained from 20 patients with isolated hypospadias and 20 age-matched (3.5±0.6 vs 5.0±1.0 yr, mean±SE) control children undergoing surgical procedure for circumcision.
In some 46,XY DSD cases, it was observed the occurrence of mutations in the development (DSD) caused by null or decreased synthesis of dihydrotestosterone.

**Background:**

the SRD5A2 protein. This mutation was predicted to affect protein function with a highly deleterious tolerance index score using the sequence homology based SIFT tool, and by the structure-based approach PolyPhen.

**Clinical cases:**

We studied another extended kindred with multiple affected members presenting gonadal dysgenesis in 4 generations. Three 46, XX patients presented premature ovarian failure (one of them was not available for study), a 3.8 year-old 46,XX girl presented high FSH levels. Four 46,XY individuals presented severe hypospadias at birth, one of them associated with microopenis and cryptorchidism. The other three developed spontaneous male puberty, one has fathered five children. Mutational analysis of the NR5A1 gene revealed a novel heterozygous mutation in 46,XY and 46,XX individuals. We have previously reported extreme within-family variability in 46,XY affected patients of two unrelated families. Phenotypes range from severe fetal undervirilization, prompting female sex of rearing, to male spontaneous and progressive puberty. Even though low ovarian reserve with preserved fertility has been reported in females harboring NR5A1 gene mutations, up to date there is no report of spontaneous fertility in the 46,XY affected individuals raised as males.

**Preserved fertility in a patient with a 46,XY disorder of sexual development due to a new heterozygous mutation in NR5A1 gene**

**Methods:**

The cDNA sequence showed absence of exon 3.

**Results:**

The cDNA sequence showed absence of exon 3.

**Conclusions:**

We described for the first time fully preserved fertility in a patient with a 46,XY DSD due to a novel heterozygous mutation in NR5A1 gene.

**References:**

- For further details, please refer to the original research paper published in *Horm Res*.

**Poster Presentations**

1. **P1-d2-379 Sex Dif ferentiation 1**

   **Preserved fertility in a patient with a 46,XY disorder of sexual development due to a new heterozygous mutation in NR5A1 gene**

   **Background:**

   Steroidogenic factor 1 (NR5A1 / SF-1) is a nuclear receptor that regulates the transcription of an array of genes involved in reproduction, steroidogenesis and male sexual differentiation. In humans, mutations in NR5A1 have been reported to cause gonadal dysgenesis with or without adrenal failure in both 46,XY and 46,XX individuals. We have previously reported extreme within-family variability in 46,XY affected patients of two unrelated families. Phenotypes range from severe fetal undervirilization, prompting female sex of rearing, to male spontaneous and progressive puberty. Even though low ovarian reserve with preserved fertility has been reported in females harboring NR5A1 gene mutations, up to date there is no report of spontaneous fertility in the 46,XY affected individuals raised as males.

   **Clinical cases:**

   We studied another extended kindred with multiple affected members presenting gonadal dysgenesis in 4 generations. Three 46, XX patients presented premature ovarian failure (one of them was not available for the study), a 3.8 year-old 46,XX girl presented high FSH levels. Four 46,XY individuals presented severe hypospadias at birth, one of them associated with microopenis and cryptorchidism. The other three developed spontaneous male puberty, one has fathered five children. Mutational analysis of the NR5A1 gene revealed a novel heterozygous mutation c.938G>A, predicted to cause a p.Arg313Hys amino acid change. The aminoacid substitution affects a highly conserved aminoacid of the putative ligand-binding domain of the mature protein. This mutation was predicted to affect protein function with a highly deleterious tolerance index score using the sequence homology based SIFT tool, and by the structure-based approach PolyPhen.

   **Conclusion:**

   We described for the first time fully preserved fertility in a patient with a 46,XY DSD due to a novel heterozygous mutation in NR5A1 gene.

2. **P1-d3-381 Sex Dif ferentiation 2**

   **“Idiopathic” partial androgen insensitivity syndrome in newborn and infant males: prenatal exposure to environmental endocrine-disruptor chemicals?**

   **Background:**

   Over the past few years, we have investigated 47 46,XY disorders of sexual development (DSD) in newborn/infant males with undervirilization. Basal/post-stimulation iHCG testing revealed deficient testosterone (T) in 16 patients (mutation of the key genes involved in testis determination was identified in 6 cases). The 31 remaining patients with normal/high plasma T were considered as presenting partial androgen insensitivity syndrome (PAIS). Sequences of the androgen receptor (AR) and the steroid-5αR-2 (SRD5A2) genes identified AR gene mutations in only 3 patients. We focused on parents’ occupational/environmental exposure to endocrine-disruptor chemicals (EDCs) before/during gestation since most EDCs present both estrogenic and anti-androgenic activity capable of interfering with fetal male sexual differentiation.

   **Methods:**

   1) All parents were interviewed about occupational/environmental exposure to EDCs before/during patients’ gestation.
   2) Estrogen bioactivity of patient serum was analyzed with an ultrasensitive bioassay (JCEM,2002).
   3) To definitively exclude other genetic defects, sequences of SF1 and MAML1, recently associated with PAIS, were performed.

   **Results:**

   1) Among the families of the 28 “idiopathic” PAIS patients, 11 (39%) reported fetal exposure to EDCs (cf. tab.1).
   2) The mean estrogen bioactivity found in these 11 patients was elevated (6.7±1.5pg/ml) vs. that of controls (1.1±0.4pg/ml; p<0.05).
   3) The sequence of SF1 was normal; one double polymorphism of MAML1 was identified (case 6).

   **Conclusions:**

   The high estrogenic bioactivity found in 9/11 newborns/infants in combination with the occupational/environmental exposure to EDCs before/during gestation reported in 11 patients (39%), strongly suggests that “idiopathic” PAIS may be related in some cases to EDC contamination during fetal life.

3. **P1-d2-380 Sex Dif ferentiation 1**

   **The g.49529G>A mutation in SRD5A2 gene causes exon 3 skipping that leads to 5α-reductase type 2 deficiency**

   **Background:**

   The 5α-reductase type 2 is the main enzyme in the male urogenital tract that is produced by the steroid 5-alpha reductase type 2 gene (SRD5A2). Mutations in SRD5A2 gene result in 46,XY disorder of sex development (DSD) caused by null or decreased synthesis of dihydrotestosterone. In some 46,XY DSD cases, it was observed the occurrence of mutations in SRD5A2 gene splicing sites. The splicing of the pre-mRNA is a crucial mechanism for the gene expression in eukaryotes. When an imprecise recognition of splice junctions occurs (exon/intron), an abnormal and unstable mRNA is formed. The mutation p.G183S in SRD5A2 gene was primarily described as a missense mutation. Site-directed mutagenesis creating the mutation in vitro showed the mutated enzyme as responsible for the NADPH affinity reduction. The mutation results from the guanine to adenine nucleotide change (g.49529G>A). However, nucleotide is located in the last position of exon 3, which is within the conserved splice donor region.

   **Objective and hypotheses:**

   We report here the experimental data demonstrating that the g.49529G>A mutation actually leads to abnormal splicing before producing the enzyme with p.G183S mutation. To achieve this conclusion we used the construction of mini-gene that consisted of a fragment from exon 2 to exon 4 of SRD5A2 gene.

   **Methods:**

   The mini-gene cloned in a mammal expression vector was transfected in COS-7 cells. After the mini-gene transcription occurred, mRNA was extracted and reverse transcribed into cDNA for sequencing.

   **Results:**

   The cDNA sequence showed absence of exon 3.

   **Conclusions:**

   The g.49529G>A mutation results in the exon 3 skipping and promotes the junction of exon 2 - exon 4 in the processed mRNA. Therefore, it was evidenced that primary biological effect of this mutation is to produce an altered mRNA by an anomalous SRD5A2 splicing process, instead of the reduction in the enzymatic activity caused the p.G183S amino acid change in the SRD5A2 protein.
Precocious prepubertal activation of genes involved in spermiogenesis as a result of activation of mini-puberty
Faruk Hadziselimovic1; Nils Hadziselimovic1; Philip Demougin2; Eduard Oakeley3
1Childrens Day Care Hospital, Pediatrics, Liestal, Switzerland; 2Biocentrum, Molecular Genetics, Basel, Switzerland; 3Novartis Institutes for Biomedical Research, Molecular Biology, Basel, Switzerland

Background: During mini-puberty, increased gonadotropin and testosterone secretion stimulate transformation of gonocytes into Ad spermatogonia. Cryptorchid boys lacking Ad spermatogonia will develop infertility despite successful orchidopexy at early age (<2 years) Objectives: This study analyzed the data of whole genome expression signatures in boys with undescended testes with or without impaired mini-puberty.

Methods: Whole genome expression analysis was examined in 19 testicular biopsies from 18 boys with cryptorchidism. Results: In the prepubertal testes we found strong expression of 10 Leydig cell genes [ CYP17A1, DHC7R, HSD17B3, IGFI, INHA, INSL3, NES, PTGDS, STAR, VCAM-1] and 11 Sertoli cell genes [AMH, CLU, CTSI, HSF4, JAG1, SFRP1, SF1, SF3B1, SF3B2, SF3B3, SF3B4] Furthermore, 6 genes purported to be specific biomarkers of adult human spermatogonia were expressed or under-expressed in the group of cryptorchid boys with impaired mini-puberty. A reliable indicator of this maturation process is the presence of Ad (dark) spermatogonia in prepubertal testis. Annotated loci were associated with spermatogenesis.

Conclusion: Molecular events initiating the testicular expression program at the onset of puberty and maintaining it during adulthood occur very early in prepubertal testes and are of crucial importance for male fertility potential.
P1-d3-384 Sex Dif ferentiation 2

Molecular diagnosis and endocrine evaluation in aromatase deficiency, three novel mutations and a possible founder effect

Roxana Maring1; Natalia Perez Garrido1; Nora Saraco1; Carlos Roccó2; Gabriela Guercio1; Mariana Costanzo1; Diana Warman1; Marta Ciaccio1; Gladys Pena1; José García1; Miria Miras1; Luis De Lacerda1; Marco A Rivalola1; Alicia Belgorosky1; Hospital de Pediatría Garrahan, Endocrine Service, Buenos Aires, Argentina; Hospital de Pediatria Garrahan, Laboratory of Cellular Biology and Retrovirus, Buenos Aires, Argentina; Hospital Infantil Municipal de Córdoba, Endocrine Service, Córdoba, Argentina; Hospital Regional de Concepción, Endocrine Service, Tucumán, Argentina; Hospital de Niños de la Santísima Trinidad de Córdoba, Endocrine Service, Córdoba, Argentina; Federal University of Paraná, Pediatric Endocrinology Unit, Department of Pediatrics, Curitiba, Brazil

Background: Several molecular CYP19A1 gene alterations, associated with cP450 aromatase deficiency, have been described in both sexes. Since 1991, 19 cases of aromatase deficiency, 11 females and 8 males, have been reported. Phenotype is dependent on sex, age and might be variable according the enzyme activity level.

Objective: To detect CYP19A1 mutations in aromatase deficiency patients. To characterize clinical and endocrinological features. To construct haplotypes linked to a recurrent mutation.

Methods: Sequencing of the coding and flanking intronic regions of the CYP19A1 gene in all patients and parents.

Clinical cases: Seven female patients were studied. They were born with ambiguous genitalia. Signs of maternal virilization during pregnancy were absent in 4/7. During follow-up ovarian enlargement/cysts was detected in 5/7 and 3/7 presented spontaneous breast development.

Results: 4/7 nonrelated patients presented the previously described c655G>A splice mutation, two in homozygosis and two in compound heterozygosis with other mutation. c655G>A was previously described in one aromatase deficiency man from Argentina. Two novel missense mutations were found in three patients (p.Y81C, p.R192C) in homozygous or compound heterozygous state and a novel splice mutation was found in one patient in homozygous state (IVS9+5 G>A). To investigate a possible common ancestry linked to the recurrent mutation c655G>A in our population, the haplotypes of the four patients who were carriers of c655G>A were characterized by data taken from 22 polymorphic markers within the gene and compared with those published from the caucasian american population. The four patients shared a common haplotype in the c655G>A allele. This mutation was found to be linked to the same haplotype with a significantly higher frequency than it was expected (p<0.019).

Conclusion: These results suggest that c655G>A appeared just once in an ancestral haplotype and its high frequency in our population is a consequence of a founder effect.

Poster Presentations
Results: Clinical data: Case 1: born at term with normal birth weight and a PAIS phenotype, External Masculinisation Score (EMS) = 5. Karyotype was XY, with a post-HCG testosterone of 9 nmol/L. There was some response to androgens. Case 2: had adult gynecomastia, genitalia were normal with EMS at birth =12. He was fertile with two children. Serum gonadotrophins and testosterone were increased. Functional studies: 870Val showed normal binding (0.35nM; WT 0.37nM) and expression. Functional activity at 0.25uM mibolerone exposure was similar to WT at 1.0 nM. N/C interaction was elevated. The 870Gly mutant had reduced binding affinity (2.3nM) and N/C interaction while the 870Glu mutant was inactive.

Conclusions: A La870 is key to ligand binding and a conformational change to active AR. 870Val was similar to WT but N/C interaction was enhanced, suggesting a detrimental gain of function. The other mutants displayed variable dysfunction. We suggest that genetic background, epigenetics, somatic mosaicism and the environment may interplay to formulate the variable phenotype in PAIS.

P1-d3-388 Sex Dif ferentiation 2

Posttraumatic stress in parents of children diagnosed with a disorder of sex development (DSD)
Kiki Mastroyannopoulou; Ieuan Hughes; Vickie Pasterk
University of Cambridge, Paediatrics, Cambridge, United Kingdom

Background: Disclosure in the case of DSD is a matter which should be handled in particularly sensitive manner. Difficulties associated with the experience of learning of DSD for parents can have lasting effects on their mental health. Studies have suggested increases in negative emotions long after the event. No study, however, has measured posttraumatic stress in this context.

Objective and hypotheses: The objective of the current study was to assess psychological well-being and posttraumatic stress in parents whose children have been diagnosed with a disorder of sex development and to relate these findings to disclosure experiences.

Methods: Semi-structure interviews designed to assess participants’ experiences regarding the learning of their child’s diagnosis were conducted in person or over the phone. Participants also completed standardised questionnaires assessing symptoms of psychological distress (Brief Symptom Inventory; BSI) and posttraumatic stress (Impact of Events Scale; IES).

Results: 33 mothers and 13 fathers completed the interview and assessment measures. Simple effects analysis suggested that mothers, but not fathers, experienced higher than expected levels of psychological symptoms such as depression and anxiety. Mothers, but not fathers, also reported higher levels of posttraumatic stress compared to a community sample. Thematic analysis of the interviews suggested that confusion and lack of information may have contributed to the negative experiences of these parents.

Conclusions: Indeed, our findings suggest that symptoms of anxiety, depression and posttraumatic stress can be long lasting. Recommendations for case management aimed at reducing such outcomes include: early disclosure in a sensitive manner. Difficulties associated with the experience of learning of their child’s diagnosis were conducted in person or over the phone. Participants also completed standardised questionnaires assessing symptoms of psychological distress (Brief Symptom Inventory; BSI) and posttraumatic stress (Impact of Events Scale; IES).

P1-d2-389 Thyroid 1
Severe pulmonary hypertension and Graves’ disease: an uncommon association
Carla Bizzarro1; Andrea De Zorzì2; Anwar Baban3; Francesca Creà1; Annalisa Deodati1; Marco Cappa1; 1Bambino Gesù Children’s Hospital, Endocrinology, Rome, Italy; 2Bambino Gesù Children’s Hospital, Cardiology, Rome, Italy; 3Bambino Gesù Children’s Hospital - Tor Vergata University, Endocrinology, Rome, Italy

Background: The association of pulmonary arterial hypertension (PH) and thyroid hyperfunction has been reported. The hemodynamic recovery of PH usually follows the achievement of euthyroidism.

Objective and hypotheses: We describe a 8.5 year old girl with severe PH associated with recent-onset hyperthyroidism.

Methods: At admission she was on methimazole therapy (0.5 mg/kg/day); TSH was 0.006 uIU/mL, FT4: 2.86 ng/dL, FT3: 14.3 pg/ml, with positive anti-thyrotropin and anti-TSH-receptor antibodies. Thyroid scan showed diffuse increased uptake suggestive of Graves’ disease.

Conclusions: Severe pulmonary hypertension and Graves’ disease were excluded. Chest CT scan revealed a diffuse alteration of parenchymal density suggesting interstitial fibrosis. Anti-nuclear antibodies (ANA) with homogeneous pattern) were 1/160, anti-centromere antibodies (CENP) were 72 U/ml (nonreactive: 0-7).

Results: Methimazole dose was gradually increased up to 1.5 mg/kg/day, the girl was biochemically euthyroid, but no PH reduction was evident at echocardiography and a rapidly progressive right heart failure appeared. She underwent I-131 treatment (5 millicuries), but 2.5 months after I-131 administration hyperthyroidism relapsed. Total thyroidectomy was performed and a complete resolution of PH, tricuspid regurgitation and right heart failure was achieved after 3 months of surgery.

Conclusions: PH in hyperthyroidism is usually related to the endothelial injury induced by the high cardiac output and to the increased vascular resistances. In contrast with previous data, in our patient biochemical euthyroidism induced by methimazole and then by I-131 did not exert any effects on PH, while surgery normalized cardiac manifestations. We hypothesize that pulmonary artery system and thyroid gland share common antigens and they represent different targets of a multi-organ autoimmune process. Probably, total thyroidectomy removed the main stimulus for antibody production and so determined the improvement of pulmonary hemodynamic function.
Thyroid 1

Increased incidence of congenital hypothyroidism in Asian infants in Washington State and high prevalence of transient congenital hypothyroidism within this population at long-term follow

Background: Recent published data document a 73% increased incidence of congenital hypothyroidism (CH) in the United States since 1987 but no definitive causes could be identified. The Washington State Newborn Screening Program and Seattle Children’s Hospital (SCH) report a disproportionate increase of CH among Asian/Native Hawaiian/Pacific Islander (ANHPI) infants compared to other ethnicities since the transition from primary T4 to primary TSH screening in 2004 with incidence rates increasing from 1:1200 to 1:550. We conducted a retrospective cohort study to assess the racial distribution and longterm clinical course of patients with CH at SCH.

Methods: Patients born in Washington State with confirmed CH and followed at SCH between 2000-2010 (N=258) were included for analysis. Demographic, diagnostic and therapeutic data were retrieved from medical records. We used Chi-square or Fischer’s exact tests to determine racial distribution, gender ratio, and clinical differences between groups.

Results: The percentage of ANHPI patients with CH increased from 20% (2000-2004) to 30% (2004-2010). A greater proportion of males were diagnosed in the ANHPI population (65%) as compared to Caucasians (41%) and Hispanics (43%), p=0.003. Of 210 patients who underwent thyroid imaging, 72% Hispanics and 61% Caucasians had abnormal thyroid imaging, compared to only 20% ANHPI (p=0.001). By age 4 years, 27 of 34 (79%) ANHPI patients could be treated off Levothyroxine compared to 28 of 74 (38%) Caucasians, and 3 of 21 (14%) Hispanics (p=0.001). Ultimately 53% ANHPI, 31% Caucasians and 14.3 % Hispanics had transient CH.

Conclusions: ANHPI patients were more likely to be male, have normal thyroid anatomy, and have transient CH at longterm follow up, all implying an underlying genetic susceptibility to milder forms of dyshormonogenesis. Although the vast majority of ANHPI were treated off Levothyroxine, only 53% were able to permanently stay off. Longterm studies are needed to determine causative factors in ANHPI CH patients, their transient CH prevalence and longterm outcomes.

Prevalence, presentation and clinical evolution of Graves’ disease in children and adolescents with type 1 diabetes mellitus

Fortunato Lombardo1; Maria Francesca Messina1; Giuseppina Salzano1; Ivana Rabbone2; Donatella Lo Presti1; Valentina Calcaterra1; Tommaso Aversa1; Filippo De Luca1; Malgorzata Wasiewska1

1University of Messina, Department of Pediatric Sciences, Messina, Italy; 2University Hospital of Turin, Department of Pediatrics, Turin, Italy; 3University Hospital of Catania, Department of Pediatrics, Catania, Italy; 4University of Pavia and Foundation IRCCS Policlinico S. Matteo, Department of Pediatrics, Pavia, Italy

Background: The epidemiological reports on the association between Type 1 diabetes mellitus (T1DM) and Graves’ disease (GD) are few and based on limited study populations.

Aims: a) to ascertain the prevalence of Graves’ disease (GD) in 1323 Caucasian children with Type 1 diabetes mellitus (T1DM); b) to compare the course of GD in T1DM patients with the one observed in 109 Caucasian peer patients with GD but without T1DM (Group B).

Results: Only 7 patients (0.53%) of T1DM series presented also GD (Group A), that was diagnosed many years after diabetes presentation. At GD diagnosis the prevalence of preclinical hyperthyroidism was higher in Group A (p=0.0001), whereas serum TSH receptor antibodies (TRAb) were higher in Group B (p=0.04). The subsequent course under methimazole therapy and after its withdrawal was very similar in both groups.

Conclusions: a) GD prevalence in T1DM patients was 0.53%, i.e. superimposable to the one reported in the general population; b) GD was diagnosed many years after T1DM presentation; c) at GD diagnosis clinical picture was milder and TRAB serum levels were lower in diabetic patients; d) preclinical diagnosis and early treatment of GD were not associated with better responsiveness to therapy; e) screening programs based on periodical TRAB assessments are not useful in T1DM.

Possible link between thyroid dysfunction, insulin resistance and oxidative stress in obese pre-pubertal children

Valentina Chiavaroli1; Chiara De Leonibus1; Valentina Corazzini1; Cosimo Giannini1; Alessandra De Remigi2; Francesco Chiarelli1; Angelika Mohr1

1University of Chieti, Department of Paediatrics, Chieti, Italy; 2University of Chieti, Department of Endocrinology, Chieti, Italy

Background: During the last years, there has been an increasing focus on the relationship between a moderate elevation of thyrotropin (TSH) levels and childhood obesity. Although the pathogenesis is not completely clear, insulin resistance (IR) and oxidative stress seem to play an important role in the development of thyroid impairment in obese patients.

Objective and hypotheses: The aim of this study was to evaluate thyroid function in obese pre-pubertal children and to explore the possible association between IR, oxidative stress and thyroid dysfunction.

Methods: We studied 186 obese (95 boys and 91 girls; 8.0 ± 1.9 yrs) and 70 normal weight pre-pubertal children (30 boys and 40 girls; 7.5 ± 2.4 yrs). Anthropometric parameters were assessed and blood samples were collected for evaluating free thyroxine (FT4), free triiodothyronine (FT3), TSH, fasting insulin and glucose. Homeostatic model assessment (HOMA-IR) was used as index of IR. In a subgroup of obese and control children urinary PFG-2α levels were obtained as a marker of oxidative stress. All children underwent a thyroid ultrasound scan.

Results: Obese children showed significantly higher levels of TSH than controls (p<0.001); while FT3 and FT4 levels were comparable. As expected, fasting glucose (p=0.005), insulin levels (p=0.001) and HOMA-IR (p=0.001) were significantly higher in obese group compared to controls. Similarly, significantly increased PFG-2α (p=0.001) levels were documented in obese children. A significant correlation was found between serum TSH and BMI-SDS (r=0.187; p=0.003), insulin levels (r=0.283; p=0.001), HOMA-IR (r=0.274; p=0.001) and PFG-2α (r=0.243; p=0.04). The multiple stepwise linear regression documented that HOMA-IR was significantly related to TSH, independently of age, sex, SDS-BMI and PFG-2α.

Conclusions: In conclusion, hyperthyrotopinemia is an emerging problem in obese pre-pubertal children and it appears to be an association between IR and increased TSH levels in this age group.

Primary congenital hypothyroidism (CH) due to 2q12.1-2q14.2 deletion: a new complex syndrome

Iva Stoeva1; Ilana Boneva1; Ivanka Dimova1; Savina Hadjidjekova1; Radka Tincheva1; Radoslava Emilova1; Daniela Avdileva1; Liubomir Spassov2; Blaga Rukova3; Draga Toncheva3

1University Pediatric Hospital Sofia, Screening and Functional Endocrine Diagnostics, Sofia, Bulgaria; 2University Pediatric Hospital Sofia, Cytogenetic Laboratory, Sofia, Bulgaria; 3Medical University Sofia, Medical Genetics, Sofia, Bulgaria; 4University Pediatric Hospital Sofia, Endocrinology, Diabetes and Genetics, Sofia, Bulgaria; 5University Pediatric Hospital Sofia, Cardiology, Sofia, Bulgaria

Background: CH could be associated with other diseases or congenital malformations, incl. chromosomal aberrations.

Objective and hypotheses: Description of the complex phenotype in a patient with CH and undergoing new aberration in chromosom 2.

Methods: Hormonal investigations by Delfta®, Cytogenetics: G banding, ge-

Poster Presentations
nomic hybridization on microarrays; Aulotopical and complex clinical follow up until 8yrs of age.

Results: Primary CH (TSH screening and serum confirmation), associated with SGAs status, eutopic thyroid gland, congenital heart malformation (ASD, VSD spontaneously closed, stenotic left A. pulmonalis), dysmorphic features, muscle hypotonia, profound skeletal, developmental and growth delay despite early and sufficient L-Thyroxine therapy was evident in a boy from the Bulgarian CH cohort picked up by neonatal thyroid screening. This complex, syndromic phenotype was accompanied by a deletion in chromosome 2 (G banding): 46,XX,del(2)(q13-q21). Comparative genomic hybridization on microarrays, covering the whole genome at a mean density of 1 BAC clone/0.8 Mb, (for fine mapping of the aberration in the patient and direct linking to gene sequence data base), refined the manifest deletion to the region 2q12.1-2q14.2 consisting of 16.7 Mbp. The region encompasses several known pathological syndromes and conditions. Among the involved genes, the deletion of PAX8 (2q12-q14.2) is the strongest candidate for the thyroid dysfunction in the patient.

Conclusions: CH patients have substantial growth and developmental delay (despite early diagnosis and sufficient treatment) should be characterized thoroughly in respect of possible underlying chromosomal aberrations; Our index patient represents a natural model for the phenotype of PAX 8 haploinsufficiency. Deletion of 2q12.1-2q14.2 could represent a syndrome with thyroid dysfunction and multiple organs’ anomalies.

P1-d2-395 Thyroid 1

Reduced false positive newborn screening for congenital adrenal hyperplasia using cutoff based on both gestational age and birth weight

Joseph Sack

Ministry of Health, Community Genetics, Tel-Aviv, Israel

Background: The overload clinics with false positive newborn screening cases are a reflection of low Positive Predicted Value (PPV) for congenital adrenal hyperplasia (CAH).

Objective: To evaluate whether cutoff based on both gestational age and birth weight improves PPV for CAH while maintaining overall adequate levels of sensitivity and specificity.

Design: In a two year pilot (2008 - 2009), neonatal screening was performed using 17-hydroxy-progesterone (17OHP) using AutoDELFIA Neonatal 17OHP B24 kits (PerkinElmer). Statistical analysis for 17OHP levels for the combination of gestational age and birth weight was obtained. The cutoff was defined to flag 0.1-3.0 percent as positive levels (full term and over 2500 gram to less than 32 weeks and below 2500 gram respectively).

Results: Previous data obtained from clinically diagnosed patients revealed that nationwide incidence of 21-hydroxylase deficiency (21OHD) was 1:19,000 live births (1:30,000 for Jews 1:8,000 for Arabs). The M:F ratio was 1:2.5 suggesting that 21OHD male patients in the general population might have been missed or died early due to a salt-losing crisis. In the 2008-2009 period 319,394 newborns were screened and 15 CAH patients were detected, 8 males and 7 females. The 17OHP levels were between 202 and 609 nmol/l. Overall prevalence was 1:21,300; among them 8 were Jews 1:28,000 and 7 females. The cutoff was defined to flag 0.1-3.0 percent as positive levels (full term and over 2500 gram).

Conclusions: Previous norms included data from hospitalized patients, some of whom would have been missed due to a salt-losing crisis. In the 2008-2009 period 319,394 newborns were screened and 15 CAH patients were detected, 8 males and 7 females. The 17OHP levels were between 202 and 609 nmol/l. Overall prevalence was 1:21,300; among them 8 were Jews 1:28,000 and 7 females. The cutoff was defined to flag 0.1-3.0 percent as positive levels (full term and over 2500 gram respectively).

P1-d2-396 Thyroid 1

Sex differences in general metabolism and thyroid function in mice with deleted Wolframin (Wfs1) gene

Klaini Normets1; Sulev Kõrak; Marina Aunapuu; Andreas Arendt; Marlin Ilves1; Eero Vasar; Vallo Tillmann1

1University of Tartu, Faculty of Medicine, Department of Pediatrics, Tartu University Hospital, Children’s Clinic, Tartu, Estonia; 2University of Tartu, Faculty of Medicine, Department of Physiology, University of Tartu, Centre of Translational Research, Tartu, Estonia; 3University of Tartu, Faculty of Medicine, Department of Anatomy, Chair of Histology and Embryology, Tartu, Estonia; 4University of Tartu, Faculty of Medicine, Department of Physiology, Tartu, Estonia

Background: Wolfram syndrome is a rare autosomal recessive neurodegenerative disorder caused by mutation in the gene encoding wolframin (Wfs1) characterized by juvenile-onset diabetes mellitus, progressive optic atrophy, diabetes insipidus and deafness. Patients may also have different neurological and endocrine disorders.

Objective and hypotheses: Our aims were to describe general metabolism and thyroid function in Wfs1 deficient (KO) and wild-type (wt) mice.

Methods: 16 males (8KO, 8wt) and 16 females (8KO, 8wt) were studied alone in a metabolic cage for 48 hrs Food, water and O2 consumption, activity, CO2 and heat production were recorded. Serum levels of TSH and FT4 were measured, thyroid glands histologically examined.

Results: Oxygen consumption, activity, CO2 and energy production was not different. Food intake was significantly lower in KO males (1.95+/-.23g vs. 3.74+/-.042g; p<0.05) and they tend to drink more (p>0.06) than wt males. KO males were smaller (20.98+/-.051g vs. 23.74+/-.096g; p=0.05) and lost more weight in cage than wt males (20.43% vs. 16.42%; p<0.05). Female KO were lighter (17.21+/-.067g vs. 20.28+/-.074g; p<0.05) and also had lower food intake than wt females (3.29+/-.063g vs. 3.52+/-.062g; p<0.05). Serum TSH level was not different between the groups and gender, but FT4 in KO males was higher than in wt males (85.98+/-.15.73 ng/ml vs. 40.05+/-.2.67ng/ml; p<0.05). Opposite was seen in females: 68.15+/-.13.14mg/ml vs. 143.27+/-.14.6 ng/ml; p<0.05. Preliminary histology showed the thyroid gland of a KO male had smaller and KO female bigger follicles and both had more connective tissue than wt ones. Further histology studies are in process.

Conclusions: The role of wolframin in general metabolism and thyroid function differs between the sexes. Further investigations are needed to clarify the mechanism for these sex differences.

P1-d2-397 Thyroid 1

Normal age-related thyroid hormone levels based on samples from over 11,000 children and adolescents

David Stritch1; Shalom Edri2; David Gillis3

1Pediatric Specialists ClinicClalit Health Services, endocrinology and diabetes, Jerusalem, Israel; 2Health Information Center, Clalit Health Services, Jerusalem, Israel; 3Hadassah-Hebrew University Medical Center, Department of Pediatrics and Pediatric Endocrinology Unit, Jerusalem, Israel

Background: Current normal values for thyroid function tests in childhood and adolescence are based on data from relatively small numbers of mostly hospitalized patients or patients recruited from hospital-associated clinics. The purpose of this study was to develop new normal pediatric values based on a large sample of ambulatory children.

Design: Data were collected from a computerized data base of the Clalit health services in Jerusalem, Israel. Exclusion criteria were positive anti-thyroid peroxidase or anti-thyroglobulin antibodies, treatment with levothyroxine, methimazole, propylthiouracil and recombinant thyrotropin. Overall after exclusion over 11,000 samples were included. Samples were tested for TSH, FT3 and FT4 with the ADVIA® CentaurTM system.

Results: The upper normal limits for thyroid stimulating hormone (TSH) increased by about 1 mIU/l and the free triiodothyronine (FT3) lower normal limit increased by 0.5-1.5 pmol/l in different age groups. There was no significant change for upper or lower normal limits of FT4 values. The new normal values bring about a change in interpretation in 5.5% of FT3 samples, 6.8% of TSH and only 1.5% of free thyroxine (FT4) samples.

Conclusions: Previous norms included data from hospitalized patients, some...
of whom probably had a mild form of sick euthyroid syndrome. The changes in normal FT3 and TSH values reflect exclusion of such patients. In view of the findings, the reference data developed in this study should replace current normal values.

P1-d3-398 Thyroid 2

**Recurrent goitre in an adolescent caused by a compound heterozygous mutation in the thyroxine peroxidase gene**

**Stuartie Straelenmaier**; Sara Van Aken; W Vinck

1University Hospital Ghent, Pediatric Endocrinology, Berchem, Belgium; 2University Hospital Ghent, Pediatric Endocrinology, Ghent, Belgium; 3St Augustinus Hospital, Endocrinology, Antwerp, Belgium; 4University Hospital Ghent, Medical Genetics, Ghent, Belgium

**Background:** We documented a compound heterozygous TPO gene mutation in a 13-year-old boy who showed a recurrence of a non-malignant goitre despite regular thyroxine supplementation after thyroidec- 

tomy during childhood.

**Clinical description:** A 13.5-year-old boy who was treated with thyroxine after total thyroidec- 

tomy for a dysshormonogenic goitre at the age of 6.5 years was seen for increasing respiratory distress. A mediastinal mass compressing the trachea was seen at CT. An urgent surgical resection of a massive retrosternal goitre was performed. Personal history revealed an unremarkable birth and pregnancy. Neonatal TSH screening was positive. A thyroid scintigraphy showed a normal position and shape of the thyroid. TSH was slightly elevated and FT4 was normal, but thyroglobulin was elevated (207 ng/ml). A subtle thyroid dysshormogenesis was suspected but could not be explained at that time. No thyroxine treatment was started. At the age of 5 years, a visible goitre had developed. TSH was elevated (15.9 mU/L, normal 0.7-5.7), FT4 was low (0.61 ng/dl, normal 1.0-2.1) at that moment. Ultrasound showed a lobulated enlarged thyroid with small hypoechoic nodules. A 1123 scintigraphy showed a highly captating asymmetrically en- 

larged thyroid and positive perchlorate washout test. Thyroxine supplementation was started. TSH and FT4 normalised. At the age of 6.5 years a total thyroidec- 

tomy was performed because of increase in goitre size despite regular thyroxine supplementation and fear for malignancy.

**Genetic analysis:** Sequence analysis of the TPO-gene demonstrated a novel heterozygous mutation c.650A>G and a known heterozygous mutation c.1184_1187dup. The mother is heterozygous for c.650A>G and the father is heterozygous for c.1184_1187dup. At ultrasound examination the proband showed a normal position and shape of the thyroid. TSH was slightly elevated and FT4 was normal, but thyroglobulin was elevated (207 ng/ml). A subtle thyroid dysshormogenesis was suspected but could not be explained at that time. No thyroxine treatment was started. At the age of 5 years a visible goitre had developed. TSH was elevated (15.9 mU/L, normal 0.7-5.7), FT4 was low (0.61 ng/dl, normal 1.0-2.1) at that moment. Ultrasound showed a lobulated enlarged thyroid with small hypoechoic nodules. A 1123 scintigraphy showed a highly captating asymmetrically en- 

larged thyroid and positive perchlorate washout test. Thyroxine supplementation was started. TSH and FT4 normalised. At the age of 6.5 years a total thyroidec- 

tomy was performed because of increase in goitre size despite regular thyroxine supplementation and fear for malignancy.

**Conclusion:** Thyroid remnant growth can develop in patients with dysshor- 

mogenesis due to TPO gene mutations, even after regular thyroxine supple- 

mentation and total thyroidec- 

tomy. Abnormal expression of thyroid growth factors and/or of their receptors might be involved in this process of recurrent goitre development.

P1-d3-400 Thyroid 2

**Novel compound heterozygous mutations in the SBP2 gene: clinical manifestations and the experience with GH replacement therapy**

**Takashi Hamajima**; Yuichi Mushimoto; Kazumichi Onigata

1Aichi Children’s Health and Medical Center, Department of Pediatric Endocrinology and Metabolism, Aichi, Japan; 2Shimane University Faculty of Medicine, Department of Pediatrics, Shimane, Japan

**Background:** Selenocyteinse insertion sequence binding protein 2 (SBP2) has a crucial role in selenoprotein synthesis. Mutations in the SBP2 gene have been reported to lead to a multisystem selenoprotein deficiency disorder, which includes abnormal thyroid hormone metabolism, short stature, delayed bone age, and developmental delay. To our knowledge, there have been no reports of GH replacement therapy for the patient with SBP2 gene mutations.

**Case:** We describe herein a 10-year-old Japanese boy, the second child of unrelated parents, born after uncomplicated pregnancy and delivery. The prob- 

and visited to our hospital due to short stature (-3.7 SD) at 2 years of age. His bone age was markedly delayed (6 months of age) and he had psychomotor retardation, strabismus, and mild conductive hearing loss. Thyroid function tests revealed elevation of FT4 of 2.90 ng/dl (normal range: 0.90-1.70), low FT3 of 2.28 pg/ml (2.30-4.30), and normal TSH of 4.98 µU/ml (0.50-5.00). These abnormal thyroid function tests consisting of high FT4 and low FT3 have continued since then. His full scale IQ evaluated by WISC-III was 66 at 6 years of age. Serum selenium was 41 µg/L (107-171). These findings prompted us to investigate SBP2 gene, and we identified novel compound heterozygous mutations in the SBP2 gene (M151Q/G63X/97Q/G97X). M151Q/G63X was de novo mutation, and Q79X was transmitted from his mother. No oth- 

er family members, except his father with Graves' disease, showed thyroid hormone abnormalities. He has received growth hormone (GH) replacement therapy due to partial GH deficiency since 4 years of age. At 10.8 years of age, his height SD score improved from -3.4 SD to -1.6 SD, however, the difference between chronological age and bone age was little changed: from 3.8 years to 3.1 years.

**Conclusions:** The proband with novel compound heterozygous mutations in the SBP2 gene showed typical thyroid function abnormalities and other clinical symptoms. GH improved height SD score without accelerating bone age.

P1-d3-399 Thyroid 2

**Impact of total body irradiation on the prevalence of primary hyperparathyroidism in patients undergoing bone marrow transplantation (BMT) during childhood for malignant hematologic disorders**

**Maritza Vivanco**; Jean-Hugues Dalle; Meriem Benmerad

1Robert Debré Hospital, Paediatric Endocrinology Department, Paris, France; 2Robert Debré Hospital, Paediatric Haematology Department, Paris, France; 3Robert Debré Hospital, Paediatric Haematology Department, Paris, France; 4Robert Debré Hospital, Clinical Epidemiology Unit, Paris, France

**Background:** The risk of radiation-induced benign and malignant thyroid nodules in patients undergoing bone marrow transplantation (BMT) during childhood for malignant hematologic disorders (CMHD).

**Objective:** The aim of this study was to determine, retrospectively, the preva- 

lence of thyroid nodules after total body irradiation (TBI) preceding BMT for CMHD.

**Methods:** We studied all survivors who had undergone TBI and BMT for CMHD between 1989 and 2009 and had been followed in our department. Thyroid ultrasound scans were performed during follow-up and/or at the time of the study. There were 95 survivors, 19 of whom were lost to follow-up or not evaluated.

**Results:** The study population (n = 76, 80% of the initial cohort) was representative of the entire cohort and was followed for a median interval of 5.1 years after BMT. A nodule (>5 mm) was found in 21 (28%) patients and was either benign (n = 15, 71%) with a median diameter of 10 (7-28) mm, or ma- 

lignant (n = 6, 29%) with a median diameter of 25 (20-30) mm. Results are shown as median (25-75th percentile).

**Age (years) Benign Malignant B+M Without nodules nodules nodules nodules nodules**

| At CMHD diagnosis | 4.4 (3-6) | 2.1 (1.5-2.8) | 3.4 (2.5-5.3) | 3.5 (3.5-9.8) | a |
| At TBI+BMT | 7 (4.1-8.4) | 6.1 (5.6-8.6) | 6.7 (4.6-8.5) | 9 (5.9-12.3) | b |
| At ultrasound examination | 17.1 (13.9-19.1) | 16.4 (14.3-21.7) | 17.1 (14.19-1) | 13 (10.3-16.8) | c |
| Interval since the age of BMT (years) | 10.1 (9.4-11) | 9.3 (3.6-15.3) | 9.9 (8.7-11) | 3.6 (2.1-6.5) | d |

*p = 0.02 benign vs malignant; p = 0.005 p<0.04 p =<0.005 p =0.001 with vs without nodules

**Conclusions:** In conclusion, the risk of thyroid nodules increased with follow-up duration and was greater for children exposed to TBI at younger ages. After 10 years of follow-up, 28% of long-term survivors are diagnosed with thyroid nodules, about 30% of which are malignant, indicating a need for life-long surveillance of this population.
P1-d3-401 Thyroid 2

Triiodothyronine (T3) treatment in children after cardiac surgery: long-term follow-up of a double-blind, randomised, placebo controlled study
Janna Mittnacht1; Carolin Knopp1; Romuald Brunner2; Peter Parzer2; Matthias Gorenflo2; Markus Bettendorf2
1Children’s Hospital Heidelberg, Endocrinology, Heidelberg, Germany; 2University Heidelberg, Department of Child and Adolescent Psychiatry, Heidelberg, Germany; 3Children’s Hospital Heidelberg, Cardiology, Heidelberg, Germany

Objective: Transient secondary hypothyroidism occurs in children with congenital cardiac malformations after cardiopulmonary bypass (CPB) operations. We previously reported significant benefits of acute postoperative T3 treatment on their recovery (1). Now we evaluated the long-term outcome on cognitive and motor development in these children.

Methods: 40 children (median age 0.6 years) with congenital cardiac malformations were initially randomized in a double blind protocol and treated either with one daily infusion of 1 μg/kg T3 or saline for a maximum time of 12 days after surgery. 28 of these 40 children (placebo n=14, T3 n=14) could now be recruited for a structured follow-up examination (median age 11.8 years) including tests for neuropsychological functioning (HAWIK III, Trail Making Test Part B (TMT B), d2 Test of Attention (d2), Lincoln-Osetezky Motor Development Scale (LOS KF 18), Child Behaviour checklist) and evaluations of thyroid (TSH, FT4, FT3, fT3) and cardiac functions (echocardiography). Results are given as mean and confidence interval. The study was approved by the local ethics committee.

Results: The full scale IQ (HAWIK III: 100;94-106) of all children were within the reference range (91-109) and similar in the placebo and the T3 group (101;93 –108 and 99;87 –111; p=ns). Tests for motor and cognitive functions (LOS KF 18, TMT B and d2) as thyroid functions and indices of cardiac function were also not significantly different in the two study groups.

Conclusions: Overall intellectual development is preserved in adolescents with congenital cardiac malformations previously treated with CPB operations in infancy irrespective of low postoperative thyroid hormone concentrations. While the acute postoperative T3 treatment in children after CPB improves recovery, no significant long-term effects on cognitive and motor developments could be detected. (1) Bettendorf M et al. Lancet. 2000;356(9229):529-34.

P1-d3-402 Thyroid 2

Complete thyroxine-binding globulin deficiency in a girl caused by a novel mutation in the TGB-gene from maternal origin
Patricia Papendieck1; Cecilia M. Oclos2; Fiorella S. Bellforte2; Cinta E. Cittero2; Hector M. Targonki2; Carina M. Rivolta2; Laura Gruñeiro-Papendieck1; Ana Chiesa1
1Ricardo Gutiérrez Children’s Hospital, Division of Endocrinology, Buenos Aires, Argentina; 2Laboratorio de Biología Molecular, Cátedra Genética y Biología Molecular, Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires, Buenos Aires, Argentina

Background: Thyroxine-binding globulin (TBG) is the main thyroid hormone transport protein in serum. Inherited TBG defects caused by mutations in the TGB-gene (locus: Xq22.2), are X-linked transmitted and lead to complete (TBG-CD) or partial deficiency (TBG-PD).

Objective and hypotheses: Describe clinical, biochemical and molecular characterization of a patient with TBG-CD inherited from her mother.

Method: DNA was extracted from the patient and her parents. TBG exons and flanking intronic regions were amplified by PCR and sequenced. Polymorphisms were ruled out by SSCP analysis in normal population.

Case report: A 10 year old insulin dependent diabetic girl thyroid function was studied during follow up finding total thyroxine (T4) of 3.7 ug/dl (NR: 6-14), free T4 (FT4) of 1.33 ng/dl (NR 0.8-2.2), thyrotropin (TSH) of 4.67 mU/L (NR 0.3-6.5) and negative thyroid antibodies. Serum TBG levels were <3,5ug/ml (NR:15-40). She is the only child of a non-consanguineous couple. The mother’s thyroid profile showed also low serum levels of thyroid hormones with undetectable TBG and positive thyroid antibodies. The father had normal thyroid profile and TBG values.

Results: A novel and heterozygous mutation in the TBG gene, a T deletion at position 2 of intron 1 (g.IVS1+2delT), was identified in the patient and her mother. This change was not present in normal population.

Conclusions: We report a novel mutation (g.IVS1+2delT) in the TGB gene responsible for TBG-CD. Proposed mechanisms for the expression of the disease in two heterozygous females may be selective X-chromosome inactivation or dominant X-linked inheritance, which will be studied further. Identification of the molecular basis of this disorder will lead to an accurate diagnosis and will be useful to understand the pathophysiology of abnormalities in the thyroid hormone transport.

P1-d3-403 Thyroid 2

Pediatric primary thyroid carcinoma: an institutional experience
Leena Priyambada1; Suttipa Sen1; Aravindan Nair2; Jacob Chacko2; Anna Simon1
1Christian Medical College, Pediatricians, Vellore, India; 2Christian Medical College, Pediatric Surgery, Vellore, India

Background: Thyroid cancer is rare in children and adolescents. The incidence of this malignancy is increasing over the years.

Objective: To describe the clinical presentation, course and factors with disease outcome in pediatric primary thyroid carcinoma.

Methods: Records of patients younger than 18 years presenting with primary thyroid malignancy between 2003–2010, were analysed retrospectively.

Results: Thirty four patients with a mean age of diagnosis of 13.5 ± 3.1 years were included in the study. A female predominance (65%) was seen in patients older than 10 years. None of the patients (except for medullary carcinoma) had any known risk factors. All were euthyroid. 90% presented with painless goiter and 2/3 had nodal metastases. Metastases were seen in regional lymph nodes (67%) and lungs (12%). FNAC was inconclusive or negative in 50% of cases. Histopathologically, the majority were papillary (62%), 25% were follicular variant of papillary, 10% follicular and 3% were medullary carcinomas. Total thyroidectomy was done for all patients, with additional modified radical neck dissection in 38% cases. Residual malignancy on post-operative iodine scan was seen in >90%. 80% underwent radioiodine ablation. External beam radiotherapy was given for extensive regional metastasis (13%). The follow-up duration ranged from 6 to 84 months. Recurrence or nodal, was seen in 16% patients. More than 50% of the patients were in remission. Patients with follicular variant of papillary carcinoma had a favourable outcome. Half of the patients with lung metastases showed remission.

Conclusions: Pediatric primary thyroid carcinoma presents as asymptomatic neck swelling. Post-operative radioiodine ablation is important for disease control. Male gender, metastases, multicentricity, and extrathyroidal extension on biopsy are associated with disease persistancy.

P1-d3-404 Thyroid 2

Identification of orphan GPR39 receptor and ghrelin receptor in thyroid tissues in patients with immune and non-immune thyroid diseases
Artur Bossowski1; Barbara Czarnocka2; Krzysztof Bardadin2; Anna Lyczkowska1; Jolanta Czerwinska3
1Medical University in Bialystok, Department of Pediatrics, Endocrinology, Diabetology with Cardiology Division, Bialystok, Poland; 2Medical Center of Postgraduate Education, Department of Molecular Biology and Clinical Biochemistry, Warsaw, Poland; 3Medical Center of Postgraduate Education, Department of Pathomorphology, Warsaw, Poland

Background: Body weight depends on the balance between energy intake and consumption. The preproghrelin is a gene responsible for generates ghrelin and obestatin. These are two gastric peptides with opposite effects on food intake and gastrointestinal tract function. Obestatin suppress food intake and digestive motility and antagonize ghrelin’s stimulatory effect through interaction with the orphan G protein –coupled receptor GPR-39 (GPCR). In addition, Ghrelin is supposed to be a link connecting metabolism and energy homeostasis with growth as results of activation of growth hormone secretagogue receptor (GHSR).

Objective and hypotheses: The aim of this study was to estimate expression of GPCR and ghrelin receptor (GHSR) in thyroid tissues from 12 pa-
tients with Graves' disease (GD), 6 with non-toxic nodular goiter (NTNG) and 6 with toxic nodular goiter (TNG). Criteria for qualification of Graves’ patients: goiter large, ophthalmopathy, TRAb > 5, positive titre of anti-TPO and anti-TG antibodies and concentration of TSH <0.3 more the 2-3 months from onset of disease.

Methods: Detection of GPCR and GHSR in thyroid tissues was performed by Western Blot. These analysis was confirmed by immunohistochemistry using monoclonal antibodies in DAB chromogene visuality and marked by Mayer’s haematoxylin.

Results: Identification of orphan GPR-39 receptor and GHR in the thyroid follicular cells revealed higher expression of both proteins in patients with Graves’ disease (+/+; +/) in comparison to patients with NTNG (+/-; +) and TNG (++; +). The detection of orphan GPR-39 receptor was presented in thyroid autoimmune disease in bands p51, p30 (kDa), in NTNG and TNG in band p51. Identification of ghrelin receptor was found in all examined groups in band 70p.

Conclusions: We conclude that expression of ghrelin receptor family in thyroid tissues may suggest role of gastric peptides in thyroid functions.

P1-d3-405 Thyroid 2

Congenital transient hypothyroidism: comparison with permanent cases

Zeynep Atay; Tulay Guran; Serap Turan; Haliloglu Belma; Fatih Gunay; Abdullah Bereket

Marmara University, Pediatric Endocrinology and Diabetes, Istanbul, Turkey

Background: Congenital hypothyroidism (CH) can be transient in neonates with eutopic thyroid gland due to several reasons including maternal antibody passage, iodine deficiency or excess.

Objective: We aimed to compare characteristics of patients with transient and permanent congenital hypothyroidism.

Methods: Diagnosis of transient CH was made in patients with eutopic-normal sized thyroid gland in whom thyroid replacement therapy can be gradually tapered and stopped without causing hyperthyrotoxicemia and hyperthyroxinemia in the course. Thyroid function tests (TFTs) were checked at least twice up to 4 months after discontinuation of L-T4 therapy and those who maintain normal TFTs were considered as transient hypothyroidism.

Results:

<table>
<thead>
<tr>
<th></th>
<th>Transient</th>
<th>Permanent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median age of diagnosis (day)</td>
<td>22</td>
<td>25</td>
</tr>
<tr>
<td>Median TSH (mIU/ml)</td>
<td>30</td>
<td>37</td>
</tr>
<tr>
<td>T4 (mcg/dl)</td>
<td>6.4 ± 3.5</td>
<td>5.9 ± 4.5</td>
</tr>
<tr>
<td>Free T4 (ng/ml)</td>
<td>0.93 ± 0.4</td>
<td>0.83 ± 0.4</td>
</tr>
<tr>
<td>Dose of L-T4 (mcg/kg/day)</td>
<td>8.3 ± 3.2</td>
<td>9.4 ± 3.8</td>
</tr>
<tr>
<td>Age at discontinuation of L-T4 (year)</td>
<td>2.5</td>
<td>NA</td>
</tr>
<tr>
<td>TSH at the time of discontinuation</td>
<td>3.8</td>
<td>NA</td>
</tr>
<tr>
<td>Free T4 at the time of discontinuation</td>
<td>1.4</td>
<td>NA</td>
</tr>
<tr>
<td>N</td>
<td>58</td>
<td>117</td>
</tr>
</tbody>
</table>

Although hyperthyrotoxicemia and hyperthyroxinemia was a bit milder in transient cases, TSH, T4 and freeT4 levels were not statistically significant between those with transient and permanent patients with CH. These results suggest that in our center approximately 1 in every neonate (33%) referred for congenital hypothyroidism turns out to have a transient form of the disease. These transient cases are essentially indistinguishable from those with permanent cases at presentation in respect to thyroid function tests. In fact 10 of 58 transient cases had initial TSH ≥ 100 mU/ml with severe hyperthyroxinemia demonstrating that severe CH can be transient as well.

Conclusions: Gradual and careful tapering of L-T4 replacement in CH cases with normal thyroid imaging allows discontinuation of treatment before 3 years of age in considerable number of cases.

P2-d1-406 Adrenal and HPA Axis 1

Intramuscular glucagon stimulation test for assessing adrenal function in short children

Liat de Vries; Ariel Tenenbaum; Moshe Phillip
Schneider Children’s Medical Center of Israel, The Jesse Z and Sara Lea Shafer Institute for Endocrinology and Diabetes, National Center for Childhood Diabetes, Petah Tikva, Israel

Background: The glucagon stimulation test (GST) has been shown to be effective in evaluating growth hormone (GH) secretion in children but there are few data on its use in evaluating the hypothalamic-pituitary axis (HPA).

Objective: To investigate the diagnostic value of the GST in evaluating the adrenocortical response in short children.

Methods: Intramuscular glucagon was used to assess the HPA axis in addition to GH in children evaluated for short stature. A total of 194 children aged 7.7±4.4 years were evaluated (158 healthy children; 36 with various disorders). Adrenal function was considered normal if peak cortisol was >550 nmol/l and/or absolute increase of cortisol was >250nmol/l. A 250µg ACTH test was performed in 31 children with inadequate response to GST.

Results: Abnormal adrenal response to GST was found in 25.7% of the cohort. Inadequate cortisol response was significantly more common among males than among females (28.7% vs. 16.4%, p<0.04) and among children >6 years than among younger children (32.7% vs. 18.4%, p<0.02). Both mean basal and mean peak cortisol levels were significantly higher in the females than in the males: 381±165 vs. 319±151 nmol/l (p=0.003) and 741±102 vs. 595±208 nmol/l (p=0.001), respectively. By 180 minutes peak cortisol was achieved in 98% of the patients, with the highest proportion (44%) of patients showing peak cortisol response at 180 minutes. In only 4 of the 31 patients undergoing an ACTH stimulation test was peak cortisol <550 but higher than 500 nmol/l. There were no significant differences in proportions of patients with abnormal cortisol response based on GH secretory status. Analyses including only healthy children yielded the same results.

Conclusions: GST may serve as a useful screening tool for adrenal function in both healthy and “abnormal” children with suspected hypopituitarism, especially in children <6 years old and in female girls. The adrenal response to GST is age and gender related. Larger studies are needed for establishing the best cut-off level for adequate cortisol response to the GST.

P2-d1-407 Adrenal and HPA Axis 1

Cortisol responses to a simplified low dose short synacthen test in children with asthma

Joanne Blair1; Jon Couriel2; Matthew Peak3; David Lacy4; Paul Newland5; Daniel Hawcutt6; Chris Gardner2; Teresa Moorcroft6; Naomi Wallin1; Mohammed Didi1

1Alder Hey Children’s NHS Foundation Trust, Endocrinology, Liverpool, United Kingdom; 2Alder Hey Children’s NHS Foundation Trust, Respiratory Medicine, Liverpool, United Kingdom; 3Alder Hey Children’s NHS Foundation Trust, Research, Liverpool, United Kingdom; 4*Wirral University Hospital NHS Foundation Trust, Paediatrics, Arrowe Park, United Kingdom; 5*Alder Hey Children’s NHS Foundation Trust, Biochemistry, Liverpool, United Kingdom; 6*Liverpool University, Division of Developmental and Reproductive Medicine, Liverpool, United Kingdom; 7Department of Endocrinology, Alder Hey Children’s NHS Foundation Trust, Liverpool, United Kingdom; 8Medicines for Children Research Network, Cheshire, Merseyside and North Wales Medicines for Children Local Research Network, Liverpool, United Kingdom; 9Medicines for Children Local Research Network, Cheshire, Merseyside and North Wales Medicines for Children Research Network, Liverpool, United Kingdom

Adrenal and HPA Axis 1

Intramuscular glucagon was used to assess the HPA axis in children with asthma.

Background: Adrenal suppression can be difficult to identify clinically. A low dose (500ng/1.73m2) short Synacthen test (LDSST) is effective at producing a cortisol response and correlates well with results of an insulin stress test. We present data from a simplified version of the LDSST in a population of children taking inhaled corticosteroids (ICS) +/- other corticosteroids.

Objective and hypotheses: To describe cortisol response to a simplified LDSST and to compare these data to studies using more intensive sampling.

Methods: The results of LDSSTs from children participating in ongoing observational research studies were examined. A Synacthen dose of 500ng/1.73 m2was administered intravenously and samples were collected at 0, 15, 25, 35 minutes. Data were compared with a previously published cohort studied with more intensive sampling.
Background: Despite several publications have shown aldosterone is secreted diurnally, and at midnight the aldosterone concentration is highest. However, other studies have found aldosterone levels are highest in the morning in young adults. The aldosterone concentration according to sleep stage was determined using the polysomnography method. Our aim was to determine the nocturnal aldosterone secretion pattern in healthy children and adolescents.

Methods: Twenty-three healthy children and adolescents (12 females, 11 males) aged 4–23 years but with and without growth hormone deficiency were examined. The aldosterone concentration was determined in serum samples collected over the 12-h night in pulsatile manner (Krauth et al. 1990; Charloux et al. 1999). We analyzed the 12-hour nocturnal secretion of ADS, GH, DHEA, and MLT in children and adolescents with short stature aged 4 to 23 years with and without growth hormone deficiency.

Results: The ADS concentration between the two groups. The evaluation of the polysomnographies showed that ADS peaks occur more frequently during wake periods and sleep stage 2 rather than during REM sleep and delta sleep. Conclusion: In children the nocturnal secretion of ADS is pulsatile and ADS peaks occur during the second half of the night, similar to young adults. The reason for the earlier peak ADS level in pre-pubertal patients remains unclear. We found no association between ADS secretion and REM-nonREM sleep.

Background: Mutations in the DAX1/NROB1 gene cause X-linked adrenal hypoplasia congenita (AHC), a rare developmental disorder of the adrenal cortex that is classically associated with hypogonadotropic hypogonadism. Interestingly, a rare occurrence of precocious sexual development in affected boys has been reported recently.

Objective and hypotheses: We hypothesize that precocious puberty is a disorder associated with X-linked AHC.

Methods: We report a new case and present a summary of the n=17 published cases of patients with NROB1 mutations exhibiting precocious puberty.

Results: A boy with a nonsense mutation in the NROB1 gene, who had been treated with hydrocortisone and 9α-fludrocortisone since early infancy developed sexual dimorphism and precocious puberty at the age of 4.5 years. While his height was 115.9 cm (90-97 percentile), he presented a pubic hair ( Tanner stage II), penile enlargement (length 7 cm), advanced bone age (6.83 years) and elevated testosterone levels (0.5 µg/l). To stop the precocious sexual development we increased the dosage of hydrocortisone to 19.5 mg/m2/d as suggested in similar published cases. However, this treatment resulted neither in a convincing clinical effect nor a change in laboratory results. While elevated testosterone levels (testosterone range 0.5-4.78 µg/l) were found in nearly all patients of the n=17 published cases of patients with AHC and precocious puberty, the clinical presentation varied to a great extent (age at onset (range 0.5-6.5 years), pubic hair (range Tanner stage I – III), with/without testicular or penile enlargement).

Conclusions: Precocious puberty expands the spectrum of clinical heterogeneity of congenital adrenal hypoplasia in patients with DAX1 gene mutations – a case report and review of the literature.
Background: Final adult height is often jeopardized in children with CAH, which is explained by the combination of hyperandrogenism and iatrogenic hypercorticism. An assay of the association of an antiandrogen (flutamide) and an aromatase inhibitor (letrozole) was proposed by a US NIH team (Merke et al., JCEM 2000; 85; 1114-1120) to prevent the effects of sex steroids and to reduce hydrocortisone dose at the same time. We propose the first reproduction of this assay.

Methods and population: 10 children with classical CAH with poor height prognosis (9 boys and one girl) were submitted to letrozole (2.5 mg/d) and flutamide (250 mg/d), mean chronological age before initiating treatment was 8.5 years (y) ± 1.4 while bone age was 12.3 y ± 1.4, mean duration of the combined treatment was 3 y ± 1.8 [1-6]. Hydrocortisone dose was gradually lowered on an auxological basis.

Results: Hydrocortisone dose was reduced from 15,7 ± 3,7 mg/m² to 8,8 ± 3 mg/m² two years later (p = 0.001) and predicted height increased from 1,8 ± 0.9 DS at the beginning to +0.5 ± 0.8 DS after 2 years (p = 0.03). All 4 patients achieved a final height of about 0 SDS. Under this treatment, patients had elevated 17 hydroxyprogesterone, ACTH, testosterone and Delta 4-androstenedione levels, as expected. No specific abnormalities in electrolytic, hepatic, renal, or hematological functions were observed during the treatment. Testicular adrenal rests were detected by a systematic ultrasound screening in four boys, in hypothetic relation with the withdrawal of the adrenal suppression.

Conclusion: Combined antiandrogen and aromatase inhibitor treatment is encouraging in terms of growth, according to the period of observation. However, a concern for fertility is induced by intrauterine adrenal rests.

Poster Presentations

P2-d1-412 Adrenal and HPA Axis 1

Treatment with anti-androgens and aromatase inhibitors in addition to low hydrocortisone doses in children with congenital adrenal hyperplasia (CAH)

Hanan El Ouahabi1; Jacques Weill2; Isabelle Guemas3; Maryse Cartigny-Maciejewski1
1Paediatric Endocrine Unit, Paediatics, Lille, France; 2Paediatric Endocrine Unit, Paediatics, Lens, France

Background: Final adult height is often jeopardized in children with CAH, which is explained by the combination of hyperandrogenism and iatrogenic hypercorticism. An assay of the association of an antiandrogen (flutamide) and an aromatase inhibitor (letrozole) was proposed by a US NIH team (Merke et al., JCEM 2000; 85; 1114-1120) to prevent the effects of sex steroids and to reduce hydrocortisone dose at the same time. We propose the first reproduction of this assay.

Methods and population: 10 children with classical CAH with poor height prognosis (9 boys and one girl) were submitted to letrozole (2.5 mg/d) and flutamide (250 mg/d), mean chronological age before initiating treatment was 8.5 years (y) ± 1.4 while bone age was 12.3 y ± 1.4, mean duration of the combined treatment was 3 y ± 1.8 [1-6]. Hydrocortisone dose was gradually lowered on an auxological basis.

Results: Hydrocortisone dose was reduced from 15,7 ± 3,7 mg/m² to 8,8 ± 3 mg/m² two years later (p = 0.001) and predicted height increased from 1,8 ± 0.9 DS at the beginning to +0.5 ± 0.8 DS after 2 years (p = 0.03). All 4 patients achieved a final height of about 0 SDS. Under this treatment, patients had elevated 17 hydroxyprogesterone, ACTH, testosterone and Delta 4-androstenedione levels, as expected. No specific abnormalities in electrolytic, hepatic, renal, or hematological functions were observed during the treatment. Testicular adrenal rests were detected by a systematic ultrasound screening in four boys, in hypothetic relation with the withdrawal of the adrenal suppression.

Conclusion: Combined antiandrogen and aromatase inhibitor treatment is encouraging in terms of growth, according to the period of observation. However, a concern for fertility is induced by intrauterine adrenal rests.

Poster Presentations

P2-d1-411 Adrenal and HPA Axis 1

LC-MS/MS-based determination of basal and ACTH-stimulated serum concentrations of 17-hydroxyprogesterone, 11-deoxycortisol and cortisol can differentiate normal controls from heterozygote carriers of CYP21A2 mutations

Alexandra E. Kulle; Dorothee Roessner; Jessica Schmitz; Lena Niemeyer; Paul-Martin Holterhus; Felix Repp
Children's Medical Center of Israel, Petah Tikva, The Jesse Z. and Lea Shafer Institute for Endocrinology and Diabetes, Kiel, Germany

Background: 21-hydroxylase deficiency (21OHD) has a high prevalence of asymptomatic heterozygote carriers in the general population.

Objective and hypotheses: To determine basal an ACTH-stimulated values of 17-hydroxyprogesterone (17OHP), 11-deoxycortisol (11S) and cortisol (F) in genotypic normal subjects and heterozygote carriers of CYP21A2 mutations, to generate reference ranges for these hormones, and to determine a distinctive basal or ACTH-stimulated parameter for separation of heterozygote carriers from normal controls.

Methods: 17OHP, 11S, and F were measured by LC-MS/MS. The study included 58 heterozygote carriers (35 males, 23 females, age range 6-78 years) and 44 control subjects (25 males, 19 females, age range 8-58 years). For all basal values, ACTH-stimulated values after 30 and after 60 minutes of 17OHP, 11S and F were determined. Ratios for 17OHP/F and 17OHP/11S were calculated.

Results: Basal mean value steroid concentrations in controls were 0.73 ±0.6 ng/mL (17OHP), 0.2 ±0.18 ng/mL (11S) and 137.9 ± 68 ng/mL (F). ACTH-stimulated mean values after 30 min in controls were 1.07 ±0.5 ng/mL (17OHP), 0.69 ng/mL (11S) and 248.5 ng/mL (F). After 60 minutes, 1.14 ng/mL (±0.8 SD) (17OHP), 0.64 ±0.4 ng/mL (11S) and 217.3 ± 70 ng/mL (F), and after 60 min were 1.42 ng/mL (±0.5 SD) (17OHP), 0.69 ng/mL (11S) and 268 ng/mL (F). Basal F was significantly lower in heterozygote carriers (mean value: 104 ng/mL ± 38) (P<0.0003). 17OHP was in both ACTH-stimulated values significantly higher in heterozygote carriers (mean value: 2.76 ng/mL ± 1.4 (30 min) and 2.95 ng/mL ± 1.5 (60 min)) (P<0.0001). The ratio of 17OHP/11S was significantly higher in heterozygote carriers.

Conclusions: We determined reference range for these hormones after ACTH stimulation for normal controls, detected significant differences between heterozygote carriers and normal controls on the basis of the stimulated 17OHP and the quotient of 17OHP/11S. However, we are not able to distinguish on the individual level. The inclusion of additional steroids may improve the specificity of the approach.
Adrenal and HPA Axis 1

Increased levels of adrenal androgens and cortisol in neonates with primary congenital hypothyroidism (CH)

Antonis Voutetakis1; Maria Dracopoulou2; Alexandra-Maria Magiakou2; Christina Kanaka-Gantenbein1; Catherine Dacou-Voutetakis1; Chryssanthi Mangelis1; 1University of Athens, Medical School, First Department of Pediatrics, Division of Endocrinology, Diabetes and Metabolism, Athens, Greece; 2Agia Sophia Children’s Hospital, Institute of Child Health, Biochemical Laboratories, Athens, Greece

Background: In animals with spontaneous or induced hypothyroidism, decreased adrenal weight and corticosteroid levels were detected and were attributed to primary adrenal dysfunction. In hypothyroid men, however, a significant hypercortisolism associated with decreased metabolic clearance and normal production rate of cortisol were reported.

Objective and hypotheses: To prospectively evaluate adrenal steroid levels in neonates with CH and in appropriate controls and to correlate these values with various thyroid function parameters, testing the hypothesis that CH alters adrenal function and/or metabolism of adrenal steroids.

Methods: Nine female neonates with CH, aged 15.2±9.8 days and 10 normal female neonates matched for age, birth weight and pregnancy duration were studied. Thyroid hormones, Thyroid Stimulating Hormone (TSH), Cortisol (C), 17-OHP, progesterone (P), Δ4 androstenedione (Δ4), Testosterone (T), and DHEAS were determined by chemiluminescence.

Results:

<table>
<thead>
<tr>
<th></th>
<th>TSH µIU/ml</th>
<th>T3 ng/dl</th>
<th>T4 ng/dl</th>
<th>C µg/dl</th>
<th>Δ4 ng/ml</th>
<th>T ng/dl</th>
<th>17-OHP ng/ml</th>
<th>DHEAS ng/ml</th>
<th>P ng/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>3.7</td>
<td>171</td>
<td>13.6</td>
<td>2.4</td>
<td>3.1</td>
<td>21.4</td>
<td>3.6</td>
<td>467</td>
<td>1.7</td>
</tr>
<tr>
<td>CH</td>
<td>501</td>
<td>98.6</td>
<td>4.2</td>
<td>8.8</td>
<td>8.5</td>
<td>58.5</td>
<td>5.2</td>
<td>839</td>
<td>3.1</td>
</tr>
<tr>
<td>p</td>
<td>0.001</td>
<td>0.019</td>
<td>0.015</td>
<td>0.027</td>
<td>0.004</td>
<td>0.007</td>
<td>0.026</td>
<td>0.048</td>
<td>0.247</td>
</tr>
</tbody>
</table>

The mean values of the parameters tested in neonates with normal (I) and high TSH values (II) are shown in the Table. In the total group, there was a significant positive correlation between TSH and DHEAS, T, C, and a significant negative correlation between T3, T4 and 17-OHP, Δ4, DHEAS, T (Spearman correlation coefficient).

Conclusions: The data show significantly higher levels of adrenal steroids in the hypothryoid state. The mechanisms involved are not clear. However our findings of increased values of both androgens and corticosteroids rather exclude an enzymatic block in adrenal steroidogenesis and favor a defect in the metabolism and/or excretion of adrenal steroids. Being aware of such data, misinterpretation of relevant findings in neonates with CH can be avoided.

Adrenal and HPA Axis 1

High proportion of structural abnormalities of the CYP21A2 gene in neonatal dried blood spots with moderately elevated 17-OHP levels

Jana Malíková1; Felix Votava1; Ondrej Cinček1; Jan Lebl1; 1University Hospital Motol and Second Faculty of Medicine, Charles University in Prague, Department of Pediatrics, Prague, Czech Republic; 2University Hospital Kralovske Vinohrady and Third Faculty of Medicine, Charles University, Department of Pediatrics, Prague, Czech Republic

Background: The neonatal screening for congenital adrenal hyperplasia (CAH) leads to identification of a certain proportion of newborns with moderately increased 17-hydroxyprogesterone (17-OHP) who require a follow-up until normalization of 17-OHP. Among these individuals, we aimed to assess the number of copies of the CYP21A2 gene and its pseudogene and the prevalence of point mutations.

Methods: We analyzed 527 randomly chosen neonatal dried blood spots from subjects within the 17-OHP “grey zone” (41-230 nmol/l), who decreased to normal 17-OHP levels thereafter. DNA was extracted from dried blood spots. The CYP21A2 gene was amplified and sequenced in all subjects. The copy number ratio between the CYP21A2 gene and its CYP21P pseudogene was tested by two methods utilizing the difference in the sequence of exon 3 (8bp deletion): two-coloured real-time quantification, and fragment analysis. The samples clustered into four major clusters according to the gene to pseudo-gene ratio, representative samples from each group were analyzed by the multiplex ligation-dependent probe amplification (MLPA).

Results: In 160 samples (30%), we detected an abnormal CYP21A2 to CYP21P ratio, suggestive of heterozygous structural abnormalities. Furthermore, eight subjects (1.5%) carried a heterozygous point mutation (seven Q318X, one I172N). Over 50 samples (10%) could not be classified due to failure of one or more methods, because of the low quality of DNA obtained from the dried blood spots.

Conclusions: As we found an unexpectedly high proportion of presumed structural abnormalities within the CYP21A2/CYP21P region, the use of simple genetic methods as a second tier in the CAH screening seems unlikely, at least in our population. The genetic efforts towards final confirmation of no less than one functional allele of the CYP21A2 gene would be far too costly and laborious as compared to the current strategy of recall measurements of 17-OHP.

Adrenal and HPA Axis 1

Five novel mutations in the SCN1A gene causing autosomal recessive pseudohypoaldosteronism type I

Maik Welzel1; Leyla Akirı2; Anja Büscher3; Tulay Guran4; Berthold Hautfle; Wolfgang Högler5; Julia Leonardo6; Beate Karges6; Heiner Kentrup7; Birgul Kirei8; Emine Yalinbas Senses9; Neşilhan Tekin10; Paul-Martin Holterhaus11; Felix Riepe11; 1University of Kiel, Pediatrics, Kiel, Germany; 2Erciyes University, Pediatrics, Kayseri, Turkey; 3University of Essen, Pediatrics, Essen, Germany; 4Birmingham Children’s Hospital, Pediatrics, Birmingham, United Kingdom; 5St Marien-Hospital, Pediatrics, Bonn, Germany; 6RWTH University Aachen, Pediatrics, Aachen, Germany; 7University of Essen, Pediatrics, Essen, Germany; 8Bethlehem Krankenhaus, RWTH Aachen, Pediatrics, Aachen, Germany; 9Osmangazi University Faculty of Medicine, Eskişehir, Turkey

Background: Autosomal recessive pseudohypoaldosteronism type I (arPHA I) is caused by mutations in the genes encoding for the α (SCN1A), β (SCN1B) or γ (SCN1G) subunit of the epithelial Na+ channel (ENaC). Affected children suffer from neonatal onset of multi-organ salt loss and often exhibit cystic fibrosis-like pulmonary symptoms.

Objective: We searched for underlying mutations in 7 unrelated children with arPHA I, all offspring of healthy consanguineous parents.

Methods and results: Amplification of the SCN1A gene and sequencing of all 13 coding exons unravelled 5 novel homozygous mutations (c.587+588insC, c.1342_1343insTACA, c.742delG, c.189C>A, IVS8-2A>G) and one known mutation (c.1474C>T) leading to truncation of the eNaC protein. All parents were asymptomatic heterozygous carriers of the respec-
tive mutations, confirming the autosomal recessive mode of inheritance. Four out of seven patients exhibited pulmonary symptoms in the neonatal period.

**Conclusion:** The alpha subunit is essential for ENaC function and mutations inactivating the pore-forming part of the protein lead to systemic arHPA-1. Based on our current knowledge, we cannot satisfactorily predict the pulmonary phenotype.

**P2-d1-417 Adrenal and HPA Axis 1**

**Long term outcome in adult patients with congenital adrenal hyperplasia due to 21 hydroxylase deficiency**

Mahdi Kamoun1; Mouna Minfi; Nadia Charfi; Nozha Kallel; Mohamed Tahar Star; Mohamed Habib Star; Mongia Hachicha; Mohamed Abid

1Hedi Chaker Hospital, Endocrinology Department, Sfax, Tunisia; 2Tahr Star Hospital, Pediatric Department, Mahdia, Tunisia; 3Tahr Star Hospital, Endocrinology Department, Mahdia, Tunisia; 4Hedi Chaker Hospital, Pediatric Department, Sfax, Tunisia

**Background:** Congenital adrenal hyperplasia (CAH) is an autosomal recessive disorder affecting adrenal steroid synthesis. More than 95% of cases are caused by 21-hydroxylase (OH) deficiency. Data concerning adults with CAH are scanty and studies have reported conflicting results.

**Objective and hypotheses:** To evaluate the impact of CAH due to 21-OH deficiency and its treatment on final height (FH), bone health, cardiometabolic risk, fertility, neurocognitive outcome and quality of life.

**Methods:** Twenty-six patients (11H, 15F, salt wasting: n=10, simple virilizing: n=8, non classical forms: n=8), aged over 16 years, were recruited.

**Results:** Mean age at presentation was 27.4 ± 8.2 years. Mean FH was 159.5 ± 9.77 cm (-1.35 ± 1.67 DS). Serum osteocalcin and 25-hydroxyvitamin D were below the normal range in 15 (57.7%) and 22 (84.6%) patients respectively. Ten patients (38.4%) exhibited osteopenia and 1 patient (3.8%) osteoporosis. Metabolic abnormalities included obesity in 8 patients (30.7%), insulin resistance in 7 patients (27%), carbohdrate metabolism disorders in 5 patients (19.2%), hyperleptinemia in 3 patients (11.5%) and hypercholesterolemia in 2 patients (7.7%). 24-hour ambulatory blood pressure monitoring discovered hypertension in 1 patient (3.8%) and “Non dipper” status in 5 patients (19.2%). Carotid ultrasound showed increased intima-media thickness in 11 patients (42.3%). Inhibine B was decreased in 4/11 males patients and AMH in 4/15 female patients. Semen analysis showed abnormalities in 4 patients. Testicular tumors were detected in 6 patients. Pelvic sonograms showed evidence of polycystic ovaries in 5 patients. Brain MRI revealed white matter changes and hippocampal dysgenesis in 6 (23%) and 2 (7.7%) patients respectively. Quality of life, assessed by the SF-36 scale was altered in 11 patients (42.3%).

**Conclusions:** Our study emphasizes the importance of regular follow-up, lifestyle interventions, bone health assessment, testicular ultrasound and psychological management in patients with CAH.

**P2-d1-418 Adrenal and HPA Axis 2**

**A novel mutation of the CYP11B2 gene in an infant with congenital isolated hyperreninemic hypoaldosteronism**

Carla Bizzarri1; Annalisa Decodati2; Giuseppe Scirè2; Paola Bencivenega; Romana Marinì; Valentina Pampanini2; Anna Taranta2; Marco Cappa1

1Bambino Gesù Children’s Hospital, Endocrinology, Rome, Italy; 2Bambino Gesù Children’s Hospital – Tor Vergata University, Endocrinology, Rome, Italy; 3Bambino Gesù Children’s Hospital, Nephrology and Dialysis, Rome, Italy

**Background:** Isolated hyperreninemic hypoaldosteronism is due to mutations in the CYP11B2 gene encoding aldosterone synthase. It is characterized by failure to thrive, vomiting and dehydration which can be life-threatening in newborn infants.

**Objective and hypotheses:** We describe a 2 month old female infant with clinical and biochemical diagnosis of hypoaldosteronism and an unusual genotype. She was referred with failure-to-thrive, recurrent vomiting and dehydration. She was born at term from healthy unrelated Caucasian parents.

At birth, weight was 3.75 Kg (0.1 SDS) and length: 50 cm (0.1 SDS). The reported daily food intake was appropriate for weight.

**Methods:** At admission weight was 3.65 Kg (-1.9 SDS) and length was 54 cm (-1.8 SDS). Plasma exams showed Na: 123 mEq/L, K: 5.99 mEq/L, Cl: 94 mEq/L; osmolality: 267 mOsm/Kg. Urinary osmolality was 177 mOsm/Kg with urinary Na concentration: 16 mEq/L. Blood gas analysis showed pH: 7.23, ABG: –5.1 mmol/L, HCO3: 21.5 mmol/L. Plasma aldosterone was 98 pg/ml (normal values 320-1300 pg/ml) with renin > 310 pg/ml (normal values: 5-270 pg/ml). ACTH was 19.1 pg/ml with cortisol: 14.91 mcg/dl.

**Results:** The simultaneous presence of these laboratory data strongly suggested the diagnosis of primary hypoaldosteronism. Sequence analyses of the CYP11B2 gene showed 3 heterozygous mutations: a novel nonsense (p.Q170X) and 2 already known missense (p.E1980 and p.V386A) mutations. During hospitalization the infant underwent iv rehydration and oral salt supplementation (NaCl 10 mEq 3 times a day). Therapy with fludrocortisone was started at the initial dose of 0.025 mg/die, progressively increased up to 0.1 mg/die; with prompt resolution of the symptoms and normalization of plasma electrolytes and blood gas analysis. Renin levels normalized within 10 days. The child manifested a remarkable catch-up growth.

**Conclusions:** The genotype of our patient has never being described up to now. The presence of 3 different mutations, could indicate that 2 aminoacid substitutions are insufficient to abolish aldosterone synthase activity.

**P2-d1-419 Adrenal and HPA Axis 2**

**541 ACTH-tests in children and adolescents lead to 321 patients to molecular genetic testing of the 21-hydroxylase gene and reveal 71 mutations**

Jennifer Krueger1; Andreas Schuster1; Judith Große-Sudheus1; Ulrich Glaubtl; Heinrich Maria Schulte1; Annette Richter-Unruh2

1Endokrinologikum Ruhr, Pediatric Endocrinology and Diabetology, Bochum, Germany; 2Endokrinologikum Hamburg, Molecular Genetics, Hamburg, Germany; 3Endokrinologikum Hamburg, Endocrinology, Hamburg, Germany

**Background:** From 2006 until early 2011 we performed an ACTH-test in 541 children and adolescents (aged 7 months to 19 years; 475 females, 66 males). Indications for ACTH-test were suspected congenital adrenal hyperplasia (CAH), accelerated bone age, premature pubarche, hyperandrogenemia or signs of androgenization like hirsutism, acne or menstrual irregularities.

**Objectives:** Aim of this study was the evaluation of the cut-off value of Δ17OHP for initiating molecular genetic testing.

**Methods:** For ACTH-test we administered 250 µg recombinant ACTH i.v. (125 µg for children < 1 year) and measured basal adrenocorticotropic hormone (ACTH) and 17-hydroxy-progesterone (17OHP) and stimulated 17OHP after 60 minutes. If the increase in 17OHP (Δ17OHP) was 2.5 µg/l or more, we initiated molecular genetic testing for 21-hydroxylase gene (CYP21A2) by DNA sequencing or Multiplex ligation-dependent probe amplification.

**Results:** 321 patients underwent molecular genetic testing. 71 of them (22.1%) had a positive result. We found e.g. several partial or complete gene deletions, gene conversions or duplications, nucleotide exchanges, 46 single heterozygous point mutations, 6 compound heterozygous mutations, one homozygous mutation and two cases with multiple mutations. 18 of these patients received treatment with hydrocortisone, two of them in combination with a GnRH-agonist due to true central precocious puberty. 11 patients received an oral hormonal contraceptive (OHC). The lowest Δ17OHP in patients treated with hydrocortisone was 3.5 µg/dl. The established cut-off for Δ17OHP of 2.5 µg/l has in our cohort a sensitivity of 98.6% and a specificity of 17.3%. A higher cut-off of 3.5 µg/l leads to a higher specificity of 56.6% and a higher positive predictive value of 35.7%.

**Conclusions:** Mutations in the 21-hydroxylase gene are very common in patients with signs of androgen excess. Therefore the ACTH-test is an appropriate mean of identifying patients with adrenal disorders. However we suggest a higher cut-off point for Δ17OHP of 3.5 µg/l for initiating molecular genetic testing.
Corticosterone/18-OH-Corticosterone ratio (30.9) and normal 18-OH-Corticoesterone/18-OH-Corticosterone ratio (5.8) confirming mild ASD type1, despite no mutations detected in the CYP11B2 gene. Patient 2 was a boy born in another Hospital presenting at 19 days of life with weight loss, poor feeding, hyperactivity, salt loss (NaK= 116±8.44 mEq/L). Normal 170HP on neonatal screening ruled out 21OHD, while both aldosterone (40 µg/ml) and renin (>500 microU/ml) levels were judged high and oriented the diagnosis to pseudo-hypoadosteronism. Treatment was begun with 0.05 mg/day of fludrocortison, and NaCl administration restored electrolyte equilibrium. Multistother analysis, after fludrocortison withdrawal, showed an elevated 18-OH-Corticosterone/Aldosterone ratio (5.8) confirming mild ASD type1, despite normal Aldosterone/Aldosterone ratio (30.9).

Results: Patient 1 was a girl born at term with normal female genitalia presenting at 28 days of life with vomiting, failure to thrive and salt loss (NaK=125±6.2 mEq/L). Normal cortisol production, low aldosterone levels and high plasma renin activity induced suspected ASD. Substitutive treatment with fludrocortison and NaCl administration restored electrolyte equilibrium. Multistoider analysis, after fludrocortison withdrawal, showed an elevated Corticosterone/18-OH-Corticosterone ratio (30.9) and normal 18-OH-Corticosterone/18-OH-Corticosterone ratio (5.8) confirming mild ASD type1, despite no mutations detected in the CYP11B2 gene. Patient 2 was a boy born in another Hospital presenting at 19 days of life with weight loss, poor feeding, hyperactivity, salt loss (NaK= 116±8.44 mEq/L). Normal 170HP on neonatal screening ruled out 21OHD, while both aldosterone (40 µg/ml) and renin (>500 microU/ml) levels were judged high and oriented the diagnosis to pseudo-hypoadosteronism. Treatment was begun with 0.05 mg/day of fludrocortison, and NaCl administration, however with limited electrolyte control.

The baby was transferred to our Center where the re-evaluation of the endocrine and electrolyte pattern (aldosterone 40 µg/ml, NaK=133±5.8 mEq/L) redirected us to suspect ASD and the rise of mineralocorticoid therapy (0.1 mg/day) normalized the clinical picture. Multistoider analysis was not possible for this patient, but molecular analysis of CYP11B2 gene identified two novel mutations in compound heterozygosis (p.R141X and p.E198G) confirmed in the parents. Functional evaluation of theE198G mutation is ongoing.

Conclusions: Although CAH due to salt losing 21-hydroxylase deficiency is the most frequent cause of adrenal mineralocorticoid deficiency, ASD has to be taken into consideration as a possible alternative cause of neonatal endocrine salt loss.

P2-d1-422 Adrenal and HPA Axis 2
Prevalence of testicular adrenal rest tumors in boys with congenital adrenal hyperplasia due to 21 hydroxylase deficiency
Esera Deniz Papanya Cakir1; Erdal Eren1; Fatma Sentürk Mutlu2; Aiyile Oztemel Pasar2; Halil Saglam1; Omer Faruk Tarim1
1Uludag University Faculty of Medicine, Pediatric Endocrinology, Bursa, Turkey; 2Uludag University Faculty of Medicine, Radiology, Bursa, Turkey

Background: Testicular adrenal rest tumors (TART) consists of ectopic adrenal tissue localized in testes. Early diagnosis and treatment is important for gonadal functions and fertility protection in boys with CAH.

Objectives and hypotheses: In this study we investigated prevalence of TART in our 21 hydroxylase deficient CAH boys.

Methods: Two different radiologist performed scrotal ultrasonography in thirteen boys with CAH due to 21 hydroxylase deficiency aged between 0 to 18 years. Chronological ages, bone ages, anthropometric measures, and mean serum ACTH and 17-α-OHP levels of the patients were evaluated during the follow up period.

Results: Study group consisted of 13 patients. The mean age of the patients was 9.79±5.23 (0.76-18.25) years. Forty percent of the percent were diagnosed as salt wasting and 45 percent as simple virilizing form of the 21 hydroxylase deficiency. The mean age at diagnosis was 3.1±2.66 (0.03-6.33) years. The mean serum ACTH and 17 alpha hydroxyprogesterone levels during the follow up period were 84±53.21 pg/ml (21.7-178.4) and 5.66±3.33 ng/ml (0.77-11.42), respectively. Two patients (patients 3 and 9) (15.4%) had images representing TART and three patients (patients 10,12 and 13) (23.1%) had testicular microtheliasis in scrotal ultrasonography. There were bilateral large testicular tumors in both of the patients. The mean ages of the patient 10,12 and 13 diagnosed as testicular microtheliasis were 14.25, 8.67 ve 18.24 years, and their ages at the time of the diagnosis of CAH were 6.32, 6.25 ve 6.33 years, respectively. Mean ACTH levels were 74.95±5.311 (27-161.8) pg/ml and renin levels were 7.43±2.58 ng/ml (4.7-10.30) in patients with TART or testicular microtheliasis.

Conclusions: Microthelias or TART may be frequently seen during the follow up of the patients with CAH. In order to prevent late complications including infertility we suggest yearly ultrasonographic evaluations to be performed in all of these patients.

P2-d1-423 Adrenal and HPA Axis 2
Genotype and phenotype correlation of patients with congenital adrenal hyperplasia
Eunice Marumudi1; B Kulshreshtha1; A Sharma2; Rajesh Khadgawat1; M.L. Khurana3; A.C. Ammin2
1All India Institute of Medical Sciences, Department of Endocrinology & Metabolism, New Delhi, India; 2All India Institute of Medical Sciences, Department of Anatomy, New Delhi, India

Background: Congenital adrenal hyperplasia (CAH) is an autosomal recessive disorder caused by steroid 21-hydroxylase deficiency.

Objectives and hypotheses: In the present study we evaluated the CAH patients referred to the Endocrine Clinic at AIIMS Hospital, New Delhi for underlying mutations in the CYP21A2 gene for genotype phenotype correlation.

Methods: Detailed family history including genital appearance, sex of rearing, gender identity, sexual development and hormonal studies were done for fifty five patients. Standard Radioimmunoassay (RIA) kit based methods were used for hormonal analysis. Informed consent to carry out molecular genetic
analysis was obtained from these patients/parents. DNA was isolated from blood samples of CAH patients by using Qiagen DNA isolation kit. PCR amplification was done as described previously (Öriola et al., 1997).

**Results:** Fifty patients were females and five were males. Their mean age at the time of study was 14.9 years. Their mean hormone levels were: 17 OHP- 21.5 ng/ml; ACTH-52.6 ng/ml; Cortisol-8.8 μg/dl; DHEAS-125.9 μg/dl; FSH-6.0 mIU/L; L-H-5.5 mIU/L & T-1.63 ng/ml.Forty one (76.4%) had simple virilizing (SV) type, twelve (22%) had salt wasting type (SW) and two (3.6%) had non classic type (NC) of CAH based on their phenotype and hormonal profile. Molecular genetic analysis revealed that thirty seven (70.0%) was found to have abnormal genotype. Twenty eight were simple virilizers, eight were salt wasters and one was non classic type. The genotype analysis according to the type of CAH showed a significant difference. In SV type, Intron 2 and Q318X mutations were found to be high (50% & 50%) followed by P30L (25%), 8-bp del (12.5%) and I172N (12.5%).Whereas in SW type, P30L mutations were found to be high (64%) followed by Intron 2 (36%), I172N (32%), Q318X(29%), 8-bp del (25%), R356W (18%).In one NC patient, only P30L & Intron 2 mutations were identified.

**Conclusions:** This is the first study to evaluate the largest group of CAH patients with good genotype and phenotype correlations from India.

**P2-d1-424 Adrenal and HPA Axis 2 A novel mutation of two boys affected by congenital adrenal hypoplasia Aleksandra Rojek1; Maciej Flader2; Elzbieta Malecka2; Marek Niedziela1 1Poznan University of Medical Sciences, Department of Pediatric Endocrinology and Rheumatology, Molecular Endocrinology Laboratory, Poznan, Poland; 2Korol Jonscher’s Clinical Hospital, Department of Pediatric Endocrinology and Rheumatology, Poznan, Poland

**Introduction:** X-linked Adrenal Hypoplasia Congenita (AHC, OMIM® 300200) is a rare disorder that can be inherited in an X-linked or autosomal recessive pattern. Primary adrenocortical failure due to the lack of permanent adult cortical zone of the adrenals occurs in X-linked AHC and is caused by mutations in DAX1 (NR0B1) gene (OMIM®300473) located on the short arm of the X-chromosome (Xp21.3) and encoding for a nuclear hormone receptor.

**Patients and methods:** Here we present two boys with AHC aged 7 years 10 months and 1 year 3 months, respectively, who came to our attention at the age of 15 days and 10 days, respectively, in a life-threatening state. Laboratory studies showed hyponatremia (124mEq/l and 112mEq/l) and hyperkaliemia (6,06mEq/l and 7,8mEq/l), respectively. All laboratory results are shown in the Table 1. Primary adrenocortical failure due to the lack of permanent adult cortical zone of the adrenals occurs in X-linked AHC and is caused by mutations in DAX1 (NR0B1) gene (OMIM®300473) located on the short arm of the X-chromosome (Xp21.3) and encoding for a nuclear hormone receptor.

**Results and conclusions:** Molecular analysis of the DAX1 gene coding region and the adjacent splicing sites revealed a novel mutation c.315G>A (W105X) in exon 1 that resulted in a premature stop codon generation and destroying the ligand binding domain of DAX1 protein. Similar, but not identical c.314G>A (W105X) mutation was only found in one patient described by Choi et al (Horm Res Paediatr, 2005). c.315G>A mutation was also present in both heterozygous healthy mothers, paternal grandmother and her two out of six sisters, while was absent in the maternal grandmother indicating the existence of gonadal mosaicism. We conclude the novel DAX1 mutation in this family and show that this analysis is important for the confirmation of the diagnosis and highlight the critical role of genetic counseling in families with AHC patients.

**P2-d1-425 Adrenal and HPA Axis 2 Mutational spectrum and genotype-phenotype association in patients with congenital adrenal hyperplasia due to 21-Hydroxylase deficiency Hayva Nur Paltek Kendirci; Zehra Ayca; Semra Cetinkaya; Tulay Tos; Sebahat Yılmaz Agadiloglu; Asan Önder Dr. Sami Ulus Women Health, Children’s Education and Research Hospital, Clinics of Pediatric Endocrinology, Ankara, Turkey

**Objective:** The aims of this study were to determine the mutational spectrum of CYP21A2 and to evaluate the genotype-phenotype association in patients with CAH due to 21-OH deficiency(21OHDD).

**Methods:** We investigated 80 CAH patients from Turkey for the large gene deletions, P30L, IVS2-13A/C, I172N, V281L, Q318X, and R356W mutations using PCR-RFLP. The clinical subtypes of the patients were determined based on the clinical manifestations and the levels of the relevant steroid metabolites and electrolytes in plasma. Genotypes were categorized in 5 mutation groups (null, A, B, C, D) according to their predicted functional consequences and compared to the clinical phenotype.

**Results:** Forty two patients (52.5%) had salt-wasting type, 19 patients (23.7%) had simple virilising and 19 patients (23.7%) had nonclassic form. Mutations were detected in 57 patients: 36 patients had homoygous for one mutation; 9 patients had heterozygotes, 5 patients had compound heterozygotes with different mutations on each chromosomes; 7 patients had complex mutations, and 23 patients harbored none of the tested mutations. The most frequent mutations were IVS2-13A/C (18.1%), followed by Q318X (17.5%), V281L (13.8%), large gene deletion (7.5%), R356W (7.5%), I172N (7.5%) and P30L (5.6%). In mutation group null/Large deletion, Q318X, R356W homoygous/heteryozygous, 35 of 19 patients (94.7%) had the classic salt-wasting form. In group A (IVS2-13A/C homoygous/compound heterozygous with a null mutation), 10 of the 15 patients (66.7%) had the salt-wasting form, 4 patients (26.7%) had simple virilizing CAH. In group B (I172N homoygous/compound heterozygous with a null mutation), all of 3 patients (100%) had simple virilizing CAH. In group C (P30L and V281L homoygous/compound heterozygous with a null/A/B mutation), 10 of 13 patients (76.9%) had non-classic form. In group D contained 7 patients, who had complex mutations, all of them had saltwasting form.

**Conclusions:** A good genotype-phenotype correlation was observed in patients with 21OHDD.

**P2-d1-426 Adrenal and HPA Axis 2 Insulin sensitivity in children with classic 21-hydroxylase deficiency with impaired adrenomedullary function Belén Huidobro-Fernández1; Maite Echeverría-Fernández2; Elena Dulin-Iríguez; Begona Ezqueta-Zubiaray3; Amparo Rodriguez-Sánchez4 1Hospital General Universitario Gregorio Marañon, Pediatric Endocrinology, Madrid, Spain; 2Hospital General Universitario Gregorio Marañon, Laboratorio de Metabolopatías de la Comunidad de Madrid, Madrid, Spain; 3Hospital General Universitario Gregorio Marañón, Laboratorio de Diagnóstico Molecular, Madrid, Spain

**Background:** Reduced insulin sensitivity has been reported in children and adults with CAH. Many mechanisms have been implied: glucocorticoid replacement, hyperandrogenism and impaired adrenomedullary function. **Aim:** To investigate whether there are differences in insulin resistance in children with classic CAH with normal (NAF) versus impaired adrenomedullary function (IAF).

**Population and methods:** 18 children with CAH were included (4 girls, 14 boys). We analyzed glucose and insulin in an overnight fasting blood sample and during an oral glucose tolerance test (OGTT) and calculated HOMA and QUICKY index. Children were classified as having impaired adrenomedullary function (IAF).

**Results:** 8 children had IAF (9.19±2.67 years, BMI SD 0.51±1.0) and 10 NIF (10.47±3.15 years, BMI SD 0.85±1.9). There were not statistically significant differences in fast glucose (IAF 3.55±0.35 vs. 3.65±0.55 mmol/L), fast insulin (2.88±2.32 vs. 2.89±2.89 mmol/L), HOMA (0.45±0.36 vs 0.51±0.69) and QUICKY index (0.48±0.08 vs 0.48±0.82) between both groups and all children presented values in the reference range for children of our country.
There were also no differences in glucose and insulin values after OGTT:

<table>
<thead>
<tr>
<th>Glucose (mmol/L)</th>
<th>IAF</th>
<th>NAF</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glu 0’</td>
<td>3.55 ± 0.35</td>
<td>3.65 ± 0.55</td>
<td>0.44</td>
</tr>
<tr>
<td>Glu 30’</td>
<td>6.31 ± 1.38</td>
<td>6.28 ± 1.66</td>
<td>0.75</td>
</tr>
<tr>
<td>Glu 60’</td>
<td>5.53 ± 1.14</td>
<td>6.28 ± 1.66</td>
<td>0.13</td>
</tr>
<tr>
<td>Glu 90’</td>
<td>5.68 ± 0.93</td>
<td>5.36 ± 0.89</td>
<td>0.82</td>
</tr>
<tr>
<td>Glu 120’</td>
<td>5.47 ± 1.15</td>
<td>5.63 ± 0.77</td>
<td>0.55</td>
</tr>
</tbody>
</table>

Results: Patient #1 had small adrenal glands by CT scanning and ultrasound examination while her mother had bilateral diffuse lipid hyperplasia. Patient #2 had diffuse enlarged adrenal glands with lipid infiltration as well as her mother.

Conclusions: We observed adrenal lipid hyperplasia in women with proven heterozygote STAR gene mutations and without clinical signs of adrenal insufficiency. In patients with genetically proven congenital adrenal gonadal hyperplasia and their 1st degree relatives present with clinically proven CLAH and their 1st degree relatives with proven heterozygote mutations. The objective was to determine CT scanning picture in patients with genetically proven CLAH and their 1-st degree relatives in patients with congenital adrenal hyperplasia and their relatives with proven heterozygote mutations.

CT findings in patients with genetically proven congenital lipoid adrenal hyperplasia and their relatives with proven heterozygote mutations

Aygul Voronova1; Maria Kareva1; Oleg Remizov2; Valentina Peterkova
1Endocrinology Research Centre, Institute of Pediatric Endocrinology, Moscow, Russian Federation; 2Endocrinology Research Centre, Department of radiology and interventional radiology, Moscow, Russian Federation

Background: Congenital lipoid adrenal hyperplasia (CLAH) is the most severe form of congenital adrenal hyperplasia, caused by mutations of STAR-protein gene. Affected individuals fail to transport cholesterol to the inner mitochondrial membrane. The adrenal cortex becomes engorged with cholesterol and esters. Abdominal computed tomography (CT) scanning demonstrates diffuse enlargement and lipid infiltration of adrenal glands.

Objective and hypotheses: The objective was to determine CT scanning picture in patients with genetically proven CLAH and their 1-st degree relatives with proven heterozygote mutations.

Methods and patients: Two girls with CLAH and their mothers were examined by CT (Toshiba Aquilion One with a 320-row detector). Two genetic girls aged 6 (patient #1) and 8 years (patient #2) had adrenal insufficiency from early infancy. Levels of cortisol and 17-hydroxyprogesterone did not elevate in response to ACTH stimulation. Their mothers had no clinical signs of adrenal insufficiency. Mother of patient #1 had normal basal level of cortisol and ACTH. STAR gene mutations see in Table 1.

<table>
<thead>
<tr>
<th>STAR gene mutations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient #1</td>
</tr>
<tr>
<td>IVS5-1G&gt;C/IVS5-1G&gt;C</td>
</tr>
<tr>
<td>Mother #1</td>
</tr>
<tr>
<td>IVS5-1G&gt;C/IVS5-1G&gt;C</td>
</tr>
<tr>
<td>Patient #2</td>
</tr>
<tr>
<td>129 G/W250X</td>
</tr>
<tr>
<td>Mother #2</td>
</tr>
<tr>
<td>W250X/-</td>
</tr>
</tbody>
</table>

Results: Patient #1 had small adrenal glands by CT scanning and ultrasound examination while her mother had bilateral diffuse lipid hyperplasia. Patient #2 had diffuse enlarged adrenal glands with lipid infiltration as well as her mother.

Conclusions: We observed adrenal lipid hyperplasia in women with proven heterozygote STAR gene mutations and without clinical signs of adrenal insufficiency. In patients with genetically proven congenital lipoid adrenal hyperplasia we saw both adrenal hypoplasia and adrenal lipid hyperplasia.

P2-d1-427 Adrenal and HPA Axis 2

Ultrasound evaluation of intima-media thickness in children with CAH due to 21-hydroxylase deficiency

Thomas MK Volki1; Heiko von Goesself; Jörg Jüngert; Helmut G Dörr2
1Division of Paediatric Endocrinology and Diabetology, Department of Paediatrics and Adolescent Medicine, Friedrich-Alexander University of Erlangen-Nuremberg, Erlangen, Germany; 2Paediatric Ultrasound Division, Department of Paediatrics and Adolescent Medicine, Friedrich-Alexander University of Erlangen-Nuremberg, Erlangen, Germany

Objective: In adults increased intima-media thickness (IMT) has been shown to be a measure of subclinical atherosclerosis and a predictor of myocardial infarction and stroke. A recent study revealed higher IMT values in adult patients with congenital adrenal hyperplasia (CAH) (Sartorato et al., JCEM 2007). Our purpose was to analyse IMT in children and adolescents with CAH as an early parameter vascular disease.

Methods: We studied 36 CAH patients (19 f, 17 m), aged between 7 and 18 years (median 13.6). All patients had genetically proven classic CAH (salt wasting, simple virilizing) and received standard steroid substitution therapy. The patients underwent echo colour Doppler ultrasonography of left and right common carotids (CC), carotid bulbs (CB), and common femoral arteries (CF) using a GE LOGIQ 9 at a 60° angle of incidence. For statistical analyses we used standard non-parametric tests.

Results: BMI of n=14 patients (39%) indicated overweight or obesity (>90th percentile). IMT of CF were significantly higher in obese CAH patients than in lean patients (mean IMT [mm] ±SD; lean, n=22, vs. obese, n=14; right CF: 0.317 ±0.118 vs. 0.417 ±0.103, p=0.027; left CF: 0.314 ±0.091 vs. 0.417 ±0.083, p=0.005). There was no difference between lean and obese patients for IMT of CC and CB. In addition, there was a significant difference between lean and right IMT values of CC (p=0.002), CB (p=0.002), but not for CF (p=0.690). IMT of CC, CB, and CF were not different between boys and girls. BMI SDS showed a significant correlation with IMT of CF (right CF: r =0.481, p=0.005; left CF: r =0.553, p=0.001), but not with IMT of CC and CB. None of the IMT parameters showed a correlation with chronological age, bone age, height SDS, or laboratory parameters of metabolic control.

Conclusions: IMT of the common femoral arteries (CF) but not of the carotids (CC, CB) may be an early marker of vascular disease in obese children and adolescents with CAH.

P2-d1-428 Adrenal and HPA Axis 2

Endocrine manifestation and scorpion envenomation in children

Sana Abourazzaz1; Sanee Achour2; Samir Almani1; Sana Chaouki1; Moustapha Harrandou3; Abdelhak Bouharrou1; Mou斯塔pha Hida4
1Mère-Enfant Hospital, Pediatrics, Fez, Morocco; 2University Hassan II Hospital, Toxicology, Fez, Morocco; 3Mère-Enfant Hospital, Anaesthesiology and Reanimation, Fez, Morocco

Background: Scorpion sting is a public health problem in Morocco. The most severe cases occur in children. Scorpion envenomation can be accompanied by hyperglycemia resulting from an increase in hepatic glycogenolysis inhibition of secretion and insulin action and increased secretion of glucagon. Objective and hypotheses: The aim of the present prospective study is to describe the endocrine manifestations of severe scorpion envenomation in children.

Methods: We report a prospective study including children admitted in the Hassan II University Hospital in Fez for severe scorpion envenomation during two years (2009-2010).

Results: 46 cases required medical attention. Male children constituted 80% of the cases. The mean age of the patients was 69 ± 46 months. The mean time that elapsed between sting and first medical attention was 5 hours. Neurological distress (altered consciousness, coma, restlessness, convulsive crisis) was founded in 56.5% of children with severe envenomation. Cardiac failure was present in 68% and respiratory distress in 40%. The mean of glycemia values was 26.7±19 mmol/l (higher than 8.3 mmol/l in 20%). The glycemia is statistically associated with the neurological failure (p=0.05).
Conclusions: Scorpion envenomation causes an autonomic storm releasing massive amounts of catecholamines confirming the biochemical changes and endocrine manifestations of envenomation produced by scorpion envenomation in animals.

P2-d2-430 Adrenal and HPA Axis 3
Congenital adrenal hyperplasia in a female with normal genitalia due to HSD3B2 mutation
Yael Levy-Shrags; Kinaret Mazor-Aronovitch; Dalit Modan-Moses; Ort Pinhas-Hamiel
Safa Children's Hospital, Sheba Medical Center, Pediatric Endocrine and Diabetes Unit, Ramat Gan, Israel

Background: 3beta-Hydroxysteroid dehydrogenase (3b-HSD) deficiency is a rare cause of congenital adrenal hyperplasia (CAH). It results from mutations in the structure of type II 3b-HSD gene (HSD3B2) and is classified as classical and nonclassical forms. Classical 3b-HSD deficiency is characterized by salt wasting. In males it is associated with incomplete virilization of the external genitalia, whereas females exhibit normal external genitalia or mild virilization.

Methods: The patient is a full term female infant. Her parents are 2nd degree cousins of Jewish ethnicity from the Caucasus. Physical examination was unremarkable with normal external genitalia. Newborn screening for 17-hydroxyprogesterone showed elevated level (153 nmol/l). Repeated venous sample revealed a 17-hydroxyprogesterone level of 181 nmol/l, testosterone >55 nmol/l, androstenedione >34.5 nmol/l, cortisol 292 nmol/l and aldosterone 1000 pmol/l. On the 6th day of life she developed salt wasting (serum K-7.3 meq/l Na-132 meq/l) and a combined therapy with hydrocortisone, fludrocortisone and saline was initiated. Karyotype was 46XX. Abdominal and pelvic US revealed normal uterus and adrenal hyperplasia.

Genetic analysis: Evaluation of the CYP21A2 and CYP11B1 genes for the CYP21A2 and CYP11B1 genes for the

P2-d2-431 Adrenal and HPA Axis 3
Prevalence of non classic 21 hydroxylase deficiency in adolescents with primary amenorrhea
Heshmat Moayeri; Ali Rabbani
Tehran University of Medical Sciences, Pediatric Endocrinology, Tehran, Islamic Republic of Iran

Background: Congenital adrenal hyperplasia (CAH) owing to 21-hydroxylase deficiency (21-OH) can occur in a classic (simple virilizing or salt wasting) or a non classic form (NC-CAH). Patients with NC-CAH present with signs of hyperandrogenism such as cystic acne and hirsutism. Patients with NC-CAH may have delayed or absent male secondary sexual characteristics. The common mutations in the Jewish population was negative. Sequencing of HSD3B2 was performed.

Results: A homozygote missense mutation in exon 4 of the HSD3B2 gene was found. This C>A mutation results in the substitution of proline for threonine and saline was initiated. Karyotype was 46XX; Abdominal and pelvic US revealed normal uterus and adrenal hyperplasia.

Conclusions: Evaluation of the CYP21A2 and CYP11B1 genes for the

P2-d2-432 Adrenal and HPA Axis 3
Novel mutation in glucocorticoid receptor gene causing primary generalized glucocorticoid resistance (Chrousos syndrome)
Nicolas C. Nicolaides¹; Eliza Geer²; Amalia Sertedaki²; Evangelia Charmandari²¹Biomedical Research Foundation of the Academy of Athens, Division of Endocrinology and Metabolism, Athens, Greece; ²Mount Sinai School of Medicine, Division of Endocrinology, Department of Medicine, New York, United States; ³Aghia Sophia Children's Hospital, Division of Endocrinology and Metabolism, First Department of Pediatrics, Athens, Greece

Background: Primary Generalized Glucocorticoid Resistance or Chrousos syndrome is a rare genetic disorder characterized by partial, generalized target-tissue insensitivity to glucocorticoids. The molecular basis of the condition has been ascribed to mutations in the human glucocorticoid receptor (hGR) gene, which impair the molecular mechanisms of hGR action, altering tissue sensitivity to glucocorticoids. We identified a new case of Chrousos syndrome caused by a novel hGR mutation in the ligand-binding domain (LBD) of the receptor.

Patient and methods: A 30 year-old woman presented with a long-standing history of hirsutism, acne, hair-loss, hypertension, anxiety and irregular menstrual cycles. She was otherwise asymptomatic and had no clinical manifestations of Cushing's syndrome. Endocrinologic evaluation revealed elevated 08:00h plasma ACTH [207 pg/mL, normal range (nr)<52], serum cortisol (26 mcg/dL) and androstenedione (252 ng/dL, nr<235), concentrations, and increased urinary free cortisol excretion (97-122 nmol/day; nr<50). There was no resistance of the hypothalamic-pituitary-adrenal axis to high-dose dexamethasone suppression. A pituitary magnetic resonance imaging scan was normal. DNA was extracted from peripheral lymphocytes and the entire coding region of the hGR gene was amplified and sequenced.

Results: A single, heterozygous nucleotide (A → G) substitution was identified at nucleotide position 2177 (exon 8), resulting in histidine (His, CAT) to arginine (Arg, CGT) substitution at amino acid position 726 in the LBD of the receptor. Molecular studies to determine the mechanisms through which the mutant receptor hGRuH726R impairs glucocorticoid signal transduction are currently underway.

Conclusions: We describe a new case of Chrousos syndrome caused by a novel, heterozygous hGR gene mutation. The location of this mutation in the LBD of the receptor may predict impaired affinity for the ligand, while manifestation of the disease at the heterozygote state may indicate a dominant negative effect of the mutant GR upon the wild-type receptor.

P2-d2-433 Adrenal and HPA Axis 3
Longitudinal growth and pubertal development pattern of girls from precocious adrenarche until ovarian hyperandrogenism
Nancy Villareal Perig1; Carlos Pavià2; Carmen Valle3
1Hospital de Nens de Barcelona, Pediatric Endocrinology Unit, Barcelona, Spain; 2Hospital Sant Joan de Déu, Biochemistry, Barcelona, Spain

Background: Hyperandrogenism from ovarian etiology in pubertal girls is commonly seen in outpatient. There are few studies about their longitudinal follow-up, specially on their growth and pubertal development. Objective: To analyse growth and pubertal characteristics of girls with hiperandrogenism from precocious adrenarche.

Methods: We monitored 67 girls for ten years, they were observed for the first time for precocious pubarche and at puberty they developed both classical clinical and hormonal criteria that agreed with hyperandrogenism. Growth progression was determined every 6 months by a Harpenden stadiometer. A hand X ray was obtained at the same intervals. Enzymatic defects of steroidogenesis were eliminated after ACTH test.
**Results:** Age of pubarche range between 4-8 yr and telarche between 9-12 yr. Minimal height velocity (MHV) was observed between 8 -12 yr, whereas puberal growth spurt (PHV) ranged from 9 to 13 yr. Progression of growth velocity was into normal limits, according normal spanish population and maternal ones; as well as the interval MHV-PHV. Menarche appeared between 9 - 15 yr. The final height was 163.2 ±5.1cm, was not different neither for reference values nor the predicted ones. Advanced bone maturation (calculated by a numerical method) was detected respect to reference population. The corresponding peak was attained two years before normal girls.

**Conclusions:** Growth pattern in these patients was into the normal limits. The onset and the duration of puberty was, in spite of the hyperandrogenism, not different from normal population.

---

**P2-d2-434 Adrenal and HPA Axis 3**

**Determining optimum sampling intervals for cortisol day curves**

**Leonor Boto1; Nathan Hill1; Caroline Fall1; Evangelia Charmandari1;**

**David R. Matthews2; Peter Hindmarsh3**

1Hospital Santa Maria, Paediatrics, Lisbon, Portugal; 2Churchill Hospital, Oxford Centre for Diabetes, Endocrinology and Metabolism, Headington, Oxford, United Kingdom; 3Southampton General Hospital, Environmental Epidemiology Unit, Southampton, United Kingdom; 4Biomedical Research Foundation of the Academy of Athens, Division of Endocrinology and Metabolism, Athens, Greece; 5Institute of Child Health, University College London, Developmental Endocrinology Research Group, London, United Kingdom

**Background:** In clinical practice cortisol profiles are used as diagnostic tools and to evaluate replacement therapy with hydrocortisone in conditions such as congenital adrenal hyperplasia (CAH).

**Objective and hypothesis:** To determine the optimum sampling interval for the construction of cortisol profiles.

**Methods:** 24h serum cortisol profiles were conducted with 20 min sampling interval in 36 children (13M aged 6.1 to 18.8 years) with CAH on standard replacement therapy and 66 healthy adults (43M, aged 61-72 years). Profiles at different sampling intervals were generated by removal of increasing number of data points to create new profiles. Fourier Transformation (FT) was used to compare profiles.

**Results:** In adults mean serum cortisol concentration was 9.2 (1.3) ug/dL. When sampling intervals increased beyond 100 mins there was a loss of definition of the frequency component of the profile although mean concentrations were unaffected. Mean serum cortisol concentrations in CAH patients receiving hydrocortisone was 6.5 (1.9) ug/dL. FT analysis showed an oscillatory pattern with a periodicity of 740 min which was lost for sampling intervals greater than 120 minutes. Mean concentrations showed a decline in oscillatory pattern with a periodicity of 740 min which was lost for sampling intervals greater than 120 minutes. Mean concentrations showed a decline in estimated value with sampling intervals greater than 140 min.

**Conclusions:** These findings indicate that day case cortisol profiles for diagnostic purposes can use long sampling intervals of 180 min if estimation of the frequency domain is not required. In CAH patients the minimum sampling interval for definition is 2 hours. Sampling protocols used in clinical practice need to reflect these observations.

---

**P2-d2-435 Adrenal and HPA Axis 3**

**The potential modulating role of inflammation on the hypothalamic pituitary adrenal axis in children with inflammatory bowel disease**

**SC Wong1; A. Smyth2; E. McNeill1; K. Hassan1; P. McGrogan1;***

**Christiaan Mooij1; Silvia Parajes1; Ian Rose1; Angela Taylor1;**

**Wiebke Arlt1; Nils Krone1**

1University of Birmingham, Centre for Endocrinology, Diabetes and Metabolism, Birmingham, United Kingdom; 2Royal Hospital for Sick Children, Department of Internal Medicine, Glasgow, Turkey; 3Oxford Centre for Diabetes, Endocrinology and Metabolism, Churchill Hospital, Department of Endocrinology, Oxford, United Kingdom; 4University of Dundee, School of Medicine, Dundee, United Kingdom; 5University of Manchester, Endocrine Sciences Research Group, Manchester, United Kingdom

**Background:** Steroid 11β-hydroxylase (CYP11B1) deficiency (11OHĐ) is the second most common form of congenital adrenal hyperplasia. Cases of non-classic 11OHĐ are rare compared with the incidence of non-classic 21-hydroxylase deficiency.

**Objective and hypotheses:** To analyse the functional consequences of five CYP11B1 mutations (p.P42L, p.R143W, p.A297V, p.L382R, p.G444D) found in three adult female patients with 11OHĐ within the UK-CaHASE* study. The p.A297V mutation was also found in two Turkish brothers with 11OHĐ. Four mutations are novel mutations; the p.G444D mutation has been previously described but not functionally characterized. Four patients presented with mild 11OHĐ, one patient presented with virilisation of the external genitalia two years after birth.

**Methods:** Functional analyses employing a COS7 cell in vitro expression system comparing wild-type (WT) and mutant CYP11B1 activity. The previously described p.P425 mutation was analysed to compare to activity of the novel p.P42L mutation. Western blot analysis was performed to analyse translation of CYP11B1 WT and mutant proteins.

**Results:** Three missense mutations (p.P42L, p.R143W and p.A297V) had activity between 10 to 27% residual CYP11B1 activity. The previously characterized p.P425 showed 13% of WT activity, within the same range as the previously published 15%. The p.L382R and p.G444D mutations caused complete loss of CYP11B1 enzymatic activity. These results are consistent with the in silico analyses, using our three-dimensional CYP11B1 protein model.

**Conclusions:** Four missense mutations in the CYP11B1 gene are associated with 11OHĐ. The phenotypic presentation is variable, with severe virilisation of the external genitalia caused by the P42L mutation. Our data are important...
to predict phenotypic expression and provide important information for clinical and genetic counselling in 11OHD. Furthermore, mild 11OHD might be more common than previously thought, which is supported by the identification of recent cases.

P2-d2-437 Adrenal and HPA Axis 3

Hypothalamic-pituitary-adrenal axis suppression in school age children at the allergy clinics of Cape Town - prevalence and associated factors

Ekkehard W Zeelenberg1; Carl Lombard2; Ushma Gala1; Stephen F Hough1; Elvis M Insuen3; Eugene Weinberg1

1University of Stellenbosch, Paediatrics, Cape Town, South Africa; 2Medical Research Council, Biostatistics Unit, Cape Town, South Africa; 3University of Stellenbosch, Medicine, Cape Town, South Africa; 4University of Cape Town, Lung Institute, Cape Town, South Africa

Background: Hypothalamic-pituitary-adrenal axis suppression (HPAS) in the setting of an asthma clinic is thought to be rare. It is possible that recovering HPAS function may account for this observation. Risk and/or associated factors of HPAS have not been described.

Objective: To determine the prevalence and associated factors of HPAS in asthmatic children treated with corticosteroids at the allergy clinics of Cape Town.

Methods: 143 asthmatic children, 5-18 years old, on inhaled corticosteroids (ICS) with or without additional steroids for other atopic diseases were recruited. Clinical features compatible with HPAS were documented. Daily and cumulative steroid dose, adherence, asthma score and lung functions were recorded. The overnight metyrapone test was performed if the 08:00 hr cortisol was >83nmol/l. Spearman correlations coefficients (r) were calculated between the post-metyrapone (PMTP) ACTH, 11-deoxycortisol (11DOC), 11DOC + cortisol, and each variable. The cut-offs were 106pg/ml, 208nmol/l and 400nmol/l respectively. Hypocortisolaemia was diagnosed if 08:00 hr serum cortisol <83nmol/l.

Results: Prevalences: All HPAS 58.0 (50.0-66.1) %; low PMTP 11DOC, 11DOC+cortisol 36.2 (28.2-44.1) %; low PMTP ACTH, 11DOC, 11DOC+cortisol 17.0 (10.8-23.2) %; hypocortisolaemia 5.6 (2.45-10.7) %. Gastro-intestinal symptoms in hypocortisolaemic patients were the only clinical features associated with HPAS (Odds ratio 20.55 [1.3-329.2], p 0.016).

Variable r (ACTH) r (11DOC) r (11DOC+cortisol)
Age 0.084 -0.009 -0.224*
BSA1 0.145 0.026 -0.227*
Daily ICS2/m2 -0.204* -0.171* -0.034
Daily N3S2/m2 -0.245* -0.243* -0.194*
Cumulative NS2/m2 (previous yr) -0.289* -0.0324* -0.269*
Daily TS4/m2 -0.110 -0.111 0.006
Adherence % ICS -0.234 -0.111 -0.148
Adherence % NS -0.362* -0.197* -0.145
Asthma score 0.020 0.106 0.212*
FEV1 % -0.026 0.039 0.148

*p<0.05 1body surface area 2inhaled corticosteroids 3nasal steroids 4topical steroids

Conclusions: More than half the number of asthmatic children in the clinic setting may have HPAS. In a third of the patients the adrenals may still be suppressed while hypothalamic and pituitary function may already have recovered. This may be associated with poor adherence. Symptoms may only be expected if the patients are hypocortisolaemic. Younger age, smaller size, the daily dose of ICS and NS, and the cumulative dose of NS for the previous year may contribute to the development of HPAS.

P2-d2-438 Adrenal and HPA Axis 3

A rare cause of delayed puberty: 17-alpha-hydroxylase deficiency

Ayla Govan; Fatma Dursun; Heves Kirmizibekmez

Goztepe Educational and Research Hospital, Pediatric Endocrinology, Istanbul, Turkey

Introduction: 17-alpha-hydroxylase deficiency is a rare form of congenital adrenal hyperplasia. The CYP17 gene encodes an enzyme that has both 17-hydroxylase and 17,20-lyase activities. Consequence of 17-alpha-hydroxylase deficiency is mineralocorticoid excess. Reduction of cortisol production by the 17-hydroxylase defect results in increased ACTH secretion and, therefore, increased production of corticosterone, deoxycorticosterone and 18-hydroxy-deoxycorticosterone.

Case report: 12 10/12 years old female patient was consulted for short stature, delayed puberty and malaise. Parents were second degree related; any familial disease was not described. In physical examination, weight: 37.4 kg (3-10 p), height: 146.5 cm (3-10 p) and blood pressure was 145/120 mmHg. Breast stage 1, axillary and pubic hair was not present. Baseline cortisol: 2.34 µg/ml, ACTH: 140 pg/ml, FSH: 31.8 µIU/ml, LH: 11.8 µIU/ml, E2: < 5 pg/ml and carotopolypeptide was 46 XX. GnRH stimulation test showed peak LH: 94.2 µIU/ml, peak FSH: 64.4 µIU/ml and E2: < 5 pg/ml. Cortisol increased to 4.05 µIU/ml with 250 mcg ACTH stimulation, progestosterone was slightly high (12.6 – 13.1 ng/ml), androgen precursors did not increase, renin and aldosterone were normal. Normal level of deoxycorticosterone high (11.9 pmol/ml; N: 0.5-0.6 pmol/ml) supported the diagnosis of 17-alpha-hydroxylase deficiency. There was not any evidence of end-organ injury due to hyper tension. Osteopenia was present in bone mineral densitometry performed for delayed puberty. Hydrocortisone, estradiol, amlopidine and enalapril treatments were prescribed.

Result: In case of adrenal and gonadal steroidogenesis defects, presence of salt and water retention, metabolic alkalosis and hypertension should remind 17-alpha-hydroxylase deficiency. 46 XX individuals may present with delayed puberty, while XY ones generally indicates different degrees of sexual development disorders. This rare disorder demonstrates the importance of proper blood pressure measurement.

P2-d2-439 Adrenal and HPA Axis 3

Severe aldosterone synthase deficiency – a novel mutation in the Cyp 11B2 gene

Erwin Lankes1; Heiko Krude2; Ritta Bernhard2; Dirk Schnabel2

1Charite Universitätsmedizin, SPZ Endokrinologie, Berlin, Germany; 2Universität des Saarlandes, Biochemie, Saarbrücken, Germany

Background: Aldosterone synthase deficiency is a rare disease which is characterised by failure to thrive, hyponatraemia, hyperkalaemia and hypotonia. The aldosterone synthase is an enzyme of the cytochrome P450 family and catalyses the 11 beta hydroxylation, the 18-hydroxylation and the 18-oxidation in the aldosterone biosynthesis.

Methods: We present a family of Turkish origin with 4 children of related parents. Our index patient is the third child, a now 7 year old girl, who was presented with failure to thrive. Hyperkalaemia (6.4 mmol/l) and hyponatraemia (133 mmol/l) was documented. The further work up showed an elevated level for renin 1080 ng/l (normal 5.9-132 ng/l) and a low aldosterone 64 ng/l (normal 70-83ng/l) while ACTH, cortisol and 17-OH-Progesterone was normal. With the tentative diagnosis of Aldosterone synthase deficiency we performed the molecular sequencing of the CYP11B2 Gene and found a homozygous mutation in the second exon (c.393C>G p.Phe131Lys). Her brother was born five years later. He showed the same changes in electrolytes, renin and aldosterone. The molecular analysis showed the same alteration in the CYP11B2 Gene.

Introduction: Aldosterone synthase deficiency is a rare disease which is characterised by failure to thrive, hyponatraemia, hyperkalaemia and hypotonia. The aldosterone synthase is an enzyme of the cytochrome P450 family and catalyses the 11 beta hydroxylation, the 18-hydroxylation and the 18-oxidation in the aldosterone biosynthesis.

Methods: We present a family of Turkish origin with 4 children of related parents. Our index patient is the third child, a now 7 year old girl, who was presented with failure to thrive. Hyperkalaemia (6.4 mmol/l) and hyponatraemia (133 mmol/l) was documented. The further work up showed an elevated level for renin 1080 ng/l (normal 5.9-132 ng/l) and a low aldosterone 64 ng/l (normal 70-83ng/l) while ACTH, cortisol and 17-OH-Progesterone was normal. With the tentative diagnosis of Aldosterone synthase deficiency we performed the molecular sequencing of the CYP11B2 Gene and found a homozygous mutation in the second exon (c.393C>G p.Phe131Lys). Her brother was born five years later. He showed the same changes in electrolytes, renin and aldosterone. The molecular analysis showed the same alteration in the CYP11B2 Gene.
P2-d2-440 Adrenal and HPA Axis 3

Adrenal hemorrhage in newborns: a retrospective review

Gülay Karaguzel¹; Mehmet Mutlu¹; Yakup Aşlan¹; Aysegul Cansul¹; Ayşenur Ökten²

¹Karadeniz Technical University, School of Medicine, Pediatric Endocrinology, Trabzon, Turkey; ²Karadeniz Technical University, School of Medicine, Neonatology, Trabzon, Turkey

Background: Adrenal hemorrhage (AH) is a relatively uncommon condition in neonates. Its clinical features are variable and nonspecific in newborns.

Objective: The aim of this study was to review the risk factors and clinical, laboratory and ultrasonographic features of AH in newborns.

Methods: We retrospectively reviewed the medical records of patients with AH admitted to our neonatal intensive care unit.

Results: Of the 13 newborns with AH, eight (62%) were term and 10 (77%) were male. Clinical presentations of the newborns were neonatal jaundice (85%), paleness and/or flank mass (38%), discoloration of the scrotum (15%) and hypotonia/lethargy or hypotension (8%). Five newborns had anemia and four had adrenal insufficiency. Adrenal insufficiency was observed in 80% of preterm newborns with AH. AH was observed on the right side in nine (69%) patients. The median age of the hemorrhage was 6±4.5 (range 4-16) weeks.

Conclusions: AH should be kept in mind in newborns with unexplained jaundice. Adrenal insufficiency is more frequent in preterm than in term newborns. Abdominal USG should also be done in a newborn presenting with scrotal swelling and bluish discoloration in order to detect AH. Serial USG is the best modality for monitoring AH and precludes unnecessary surgery.

P2-d2-441 Adrenal and HPA Axis 3

Gender identity and sexual orientation in adult patients with classical virilizing congenital adrenal hyperplasia (CAH)

Marlene Inacio¹; Tânia A. S. Bacheque¹; Vinicius N. Britó¹; Guiomar Madureira¹; Larissa G. Gomes¹; Ari Oliveira¹; Eliisa Verudegue²; Ivo J. P. Arnhold¹; Berenice B Mendonça³

¹Unidade de Endocrinologia do desenvolvimento, LIM/42 da Disciplina de Endocrinologia da FMUSP, Internal medicine, Sao Paulo, Brazil; ²University of Sao Paulo, Internal Medicine, Sao Paulo, Brazil; ³University of Sao Paulo, Sao Paulo, Brazil

Background: There are few reports on gender identity and sexual orientation in adult patients with virilizing congenital adrenal hyperplasia. Objective: to evaluate the effect of prenatal and postnatal androgen excess in gender identity and sexual orientation in an adult Brazilian female cohort with virilising CAH.

Patients and methods: 46 XX DSD patients, 52 with classical 21OH and 3 with classical 11-hydroxylase deficiency were selected: from 210OH patients 2 patients had the SF form; 7 patients were reared as males and 4 out of them changed to female social sex after medical guidance. From the 48 females registered as females, 5 changed to male social sex. The degree of external genitalia virilization was obtained in 44 cases and was classified according Prader (P): P2 n= 5, P3 n= 25, P4 and P5 n= 14. House Tree and Person Test and Bleger’s questionnaire were used to evaluate gender identity and sexual orientation. The patients were grouped according to the chronological age of treatment (before and after 2 years) and according to hormonal control during growth periods: (good - 3 out of 4 normal androgen levels (n=20), poor - high androgen levels in more than 2 out of 4 annual measurements (n=29).

Results: All patients with good control presented female gender identity irrespective of Prader score and age at diagnosis. Homosexual orientation was found in 2 SW (P 3 and good control) and in 3 SV patients (P 3 and bad control) and bisexual orientation was found in 3 SV patients. Five 210OH patients, all with >P III, late diagnosis and poor control presented male gender identity and changed to male social sex. No predictors of sex change were found at multivariate analysis. All three SV patients reared as males, presented gender and sexual orientation adequate to male social sex.

Conclusions: Females with virilising CAH with good control did not present gender identity problems observed in patients with bad control. Prolonged postnatal androgen excess is associated with male gender identity or homosexual or bisexual orientation in adult female patients with CAH.

P2-d2-442 Adrenal and HPA Axis 4

Relationships of basal level of serum 17—hydroxyprogesterone with that of serum androstenedione and their stimulated responses to a low dose of ACTH in young adult patients with congenital adrenal hyperplasia due to 21—hydroxylase deficiency

Min Jae Kang; Shin Mi Kim; Young Ah Lee; Jieun Lee; Ju Young Yoon; Choong Ho Shin; Sei Won Yang

Seoul National University Children’s Hospital, Pediatrics, Seoul, Republic of Korea

Background: A single measurement of serum 17α—hydroxyprogesterone (17αOHP) level, used in the assessment of treatment adequacy in patients with 21—hydroxylase deficiency, can be unreliable because of its marked diurnal variation. Serum androstenedione (AD) level has a smaller diurnal variation, which is not measured routinely in Korea.

Objective and hypotheses: In the present study, we investigated the relationship between serum 17αOHP and AD levels, and whether the responses of these two hormones to low—dose ACTH stimulation are correlated in patients with 21—hydroxylase deficiency.

Methods: Baseline serum 17αOHP and AD levels were measured in 87 patients (46 males and 41 females). A low—dose (1 μg) ACTH stimulation test was performed in 41 patients to measure the basal and peak levels of serum 17αOHP and AD.

Results: The basal 17αOHP level correlated positively with the basal AD level independently of sex, type of 21-hydroxylase deficiency, and the time of day of blood sampling (n=87, R²=0.75, P<0.001). Peak levels of both hormones were significantly higher than their basal levels. The area under the curve of 17αOHP and AD correlated positively with their respective basal levels. The fold—change increase in 17αOHP after low—dose ACTH injection correlated negatively with the basal 17αOHP level, but that of AD did not correlate with the basal AD level.

Conclusions: The random serum 17αOHP level, as currently measured in the clinic, may be a reliable guide in the management of patients with 21—hydroxylase deficiency. A low—dose ACTH stimulation test provides no extra benefit for assessing the treatment adequacy in these patients.

P2-d2-443 Adrenal and HPA Axis 4

Role of genotype in the diagnosis of children with 21-hydroxylase deficiency

Paolo Cavazza; Monica Vincenzi; Francesca Teatoli; Rossella Gaudino; Elena Monti; Evelina Maines; Francesca Doro; Luciano Tata; Marta Camilot; Franco Antoniazzi

Division of Paediatric, University of Verona, Department of Life and Reproduction Sciences, Verona, Italy

Background: Mutations of CYP21A2 gene are responsible of 21-hydroxylase deficiency (21-OHD), the most common enzymatic defect causing congenital adrenal hyperplasia (CAH).

Objective and hypotheses: The aim of our study was: to confirm the diagnosis of 21-OHD by the analysis of CYP21A2 in infants with clinical features of 21-OHD; to analyze the genotype-phenotype relationship in these infants.

Methods: We studied 21 children with clinical features of 21-OHD: 4 babies presented a salt-wasting form of CAH, 12 a premature pubarche and 5 an elevated 17α-OHP level at newborn screening. All of them and their parents were submitted to genetic analysis of CYP21A2, performed by PCR, MLPA and exons and promoter sequencing. Patients were classified in 3 groups according to predict mutations’ severity: severe (group A), moderate (group B) and mild (group C).

Results: The most frequent mutation in our population was V281L. All children in group A (2) and B (2) presented a salt-wasting form of CAH. Eight children were in group C and had a non classical form of CAH. Four infants were heterozygotes for 21-OHD and other 4 children did not present mutations in CYP21A2. A girl clinically presenting a non classical form of CAH...
was a compound heterozygote for Q318X and R255S, a new mutation whose residual enzymatic activity is not known, moreover she shown 4 genes at MLPA analysis. Analyzing the correlation between the 17-OHP levels after ACTH stimulation test and the genotype, we found an optimal relationship in patients of all three groups. All affected children presented a 17-OHP level after ACTH stimulation greater than 100 nmol/L.

**Conclusions:** CYP21A2 analysis permitted to confirm the diagnosis of 21-OHD in 61.9% of our children. To improve this percentage we suggest to perform the analysis of CYP21A2 only when 17-OHP after ACTH stimulation is greater than 100 nmol/L. We confirm an optimal genotype-phenotype relationship in the 21-OHD patients.

**P2-d2-444 Adrenal and HPA Axis 4**

**Vitamin D status and bone mineral density in Turkish children with congenital adrenal hyperplasia**

Fatma Demirel; Ozlem Kara; Derya Tepe; Iscan Esen
Ankara Child Diseases Hematology and Oncology Education and Research Hospital, Pediatric Endocrinology Department, Ankara, Turkey

**Background:** Congenital adrenal hyperplasia (CAH) is the most common adrenal disorder in children. Life-long glucocorticoid (GC) replacement is necessary to reduce overproduction of adrenal androgens. Careful dose adjustment is essential for optimal growth and bone health. There were some reports about relationship between supraphysiologic GC doses and decreased bone mineral density (BMD) among adult patients with CAH, but data were less informative in growing children.

**Objective and hypotheses:** Aim of the study was to determine vitamin D status and BMD in children with CAH.

**Methods:** Thirty females and 21 males; 32 salt wasting and 19 simple virilizing cases with average of 12.9 years (ranges 5-24 years) were studied. Areal BMD values of lumbar vertebrae (L1-L4) which determined by dual X-ray absorptiometry (DXA) were used to calculate z scores according to sex and chronological age. Z scores below -2.0 were accepted as decreased BMD. A serum 25(OH)D3 level > 20 ng/mL was considered as indicative of vitamin D sufficiency.

**Results:** The mean BMD z score was -0.2±1.5 (-2.8 to 3.6). Decreased BMD was determined in two adolescent patients (3.9%). The mean 25(OH)D3 level was 14.8 ng/mL (4 to 45 ng/mL). Forty (74.5%) children had vitamin D deficiency and it was more prevalent in pubertal age (P<0.05). BMD z score was not found different according to sex, age and pubertal level. There were no correlation between BMD z scores and average daily or cumulative steroid doses and 25(OH)D3 levels.

**Conclusions:** Harmful effect of long term steroid therapy on bone health of CAH patient has not been emerged yet in childhood. But high rate of vitamin D deficiency was remarkable finding in this group. For maintaining bone health during long-term GC therapy, serum 25(OH)D3 level > 20 ng/mL was considered as indicative of vitamin D sufficiency.

**P2-d2-445 Adrenal and HPA Axis 4**

**Incidence of 21-hydroxylase deficiency in Republic Bashkortostan, Russia**

Oleg Malievsky1; Galina Pechenina2; Dilara Nurmukhametova3
1Bashkir State Medical University, Department of Pediatrics, Ufa, Russian Federation; 2Republican Perinatal Center, Neonatal Screening Laboratory, Ufa, Russian Federation; 3Republik Children Hospital, Department of Endocrinology, Ufa, Russian Federation

**Background:** In most world countries, for early diagnostics of 21-hydroxylase deficiency (21-OHD), the neonatal screening is performed. In Republic Bashkortostan, examination of newborns for this disease has begun as late as in 2006.

**Objective and hypotheses:** To determine incidence of 21-hydroxylase deficiency in Republic Bashkortostan, Russia.

**Population and/or methods:** The level of 17-hydroxyprogesterone in the newborn blood spot was determined by the immunofluorescent method. In the newborns with the 17-hydroxyprogesterone level in the whole blood exceeding cut-off, the content of 17-hydroxyprogesterone, potassium, and sodium was determined in the blood serum.

**Results:** During the whole period of screening (2006-2010) we examined 134 618 newborns, which accounted for 99.5% of all newborns. The 17-hydroxyprogesterone level in the whole blood above the cut-off was revealed in 806 (0.6%) newborns included in the program of screening. In studying the 17-hydroxyprogesterone level in the blood serum of these children, its increased concentrations were revealed in 113 (8.4%) cases. In 98 newborns, at dynamic observation, the 17-hydroxyprogesterone level was decreasing to the normal level, which has allowed us to rule out their 21-hydroxylase deficiency. In 15 newborns the 17-hydroxyprogesterone level remained elevated; in this connection, analysis of the gene CYP21 was performed, which allowed verification of this diagnosis. Incidence of 21-OHD in Republic Bashkortostan amounted to 1 case per 8974 newborns. This parameter does not differ from that in the majority of other regions of Russia, in which the 21-hydroxy- lase deficiency incidence is present from 1 : 7617 (Krasnodar Krai) to 1 : 10014 (Moscow). The 21-OHD incidence in Bashkortostan also does not differ essentially from that in the majority of the European countries.

**Precise conclusions:** Incidence of 21-hydroxylase deficiency in the Republic of Bashkortostan amounts to 1 case per 8974 newborns and does not differ essentially from that in the majority of other regions of Russia and European countries.

**P2-d2-446 Adrenal and HPA Axis 4**

**Plasma synacthen levels are highly variable following administration of 1mcg intravenous synacthen and bear no relationship to peak cortisol levels**

Charlotte Elder1; Pooja Sachdev2; Martin Loxley3; Trevor Johnson4; Jerry Wales5; Neil Wright6
1University of Sheffield, Human Metabolism, Sheffield, United Kingdom; 2Sheffield Children’s Hospital, Endocrinology, Sheffield, United Kingdom; 3Sheffield Teaching Hospitals NHS Trust, Clinical Chemistry, Sheffield, United Kingdom; 4Sheffield Children's Hospital, Pharmacy, Sheffield, United Kingdom

**Background:** Since 2003, three meta-analyses have supported the use of the 1mcg Short Synacthen test (SST) over the supra-physiological 250mcg standard SST. Our survey of British paediatric endocrinologists in 2009 reported 90% were using a low-dose SST (LDST) but that there was considerable variation with regards to dose, timings of peak cortisol, diagnostic cut-offs and methods of making up low-dose Synacthen (14 different methods in use).

**Objective and hypotheses:** To look at the inter-individual variability of Synacthen levels in the LDST and the relationship of peak Synacthen and peak cortisol.

**Method:** We performed a LDST on 12 healthy adult males who had been dexamethasone suppressed. Volunteers had paired samples of Synacthen and cortisol taken at 0, 5, 10, 15, 20, 30, 40, 50, 60, 75, 90, 120, 150 and 180 minutes. The Synacthen was measured on a competitive radioimmunoassay quoting 100% cross-reactivity with endogenous ACTH.

**Results:** Despite all 12 volunteers receiving the same intravenous dose there was considerable variation in peak cortisol, diagnostic cut-offs and methods of making up low-dose Synacthen (14 different methods in use). There were no correlation between BMD z scores and average daily or cumulative steroid doses and 25(OH)D3 levels.

**Conclusions:** Harmful effect of long term steroid therapy on bone health of CAH patient has not been emerged yet in childhood. But high rate of vitamin D deficiency was remarkable finding in this group. For maintaining bone health and prevention of osteoporosis in adult age, vitamin D replacement should be recommended to these children.

**Poster Presentations**
Methods: A retrospective case-note review of 294 consecutive insulin tolerance tests over a five year period [January 2004—December 2009].

Results: Hypoglycaemia was achieved in 285 tests (97%), this was optimal in 267 (94%). Ninety-five patients (33%) had a suboptimal cortisol response. Factors associated included, higher nadir glucose (median 1.9 vs 1.7 mmol/L, P = 0.007), lower basal cortisol (median 198 vs 328 mmol/L, P < 0.0001), older age (mean 13.2 vs 12.1 years, P = 0.009) and earlier cortisol peak (mean 37.5 vs. 48.4 minutes, P = 0.012). Those who were growth hormone deficient were not more likely to be cortisol deficient (p = 0.66). Following application of a multiple linear regression model a significant positive correlation remained (r = 0.62, p < 0.0001) between basal and peak cortisol. Nadir glucose and age were negatively correlated with peak cortisol (p = 0.036 and <0.0001 respectively). Forty-two patients with peak cortisol < 500 mmol/L on ITT had a low-dose (1µg) ACTH stimulation test. Thirty-one (74 %) had an adequate cortisol response to low-dose Synacthen (peak cortisol > 500 mmol/L).

Conclusions: Despite being the gold-standard test, cortisol deficiency appears to be relatively common on ITT even in children with a low pre-test probability of same. As an alternative test of cortisol status, the low-dose (1 µg) ACTH stimulation test would appear to lack sensitivity when a 500 nmol/L cut-off is used.

---

**P2-d2-450 Adrenal and HPA Axis 4**

**Twin male sibs with xp21 contiguous gene syndrome**

**Ahmet Uçakturk1; Cengiz Karpı; Serdar Ceylaner2; Figen Gunindı3; Murat Aydin1**

1Ondokumayis University Faculty of Medicine, Department of Pediatric Endocrinology, Samsun, Turkey; 2Intergen Genetics Centre, Medical Genetics, Ankara, Turkey

**Introduction:** X-linked adrenal hypoplasia congenital (AHC) is characterized by primary adrenal insufficiency caused by deletion or mutation of the DAX-1 gene and frequent association with hypogonadotropic hypogonadism. It can occur as a part of Xp21 contiguous gene syndrome together with glycerol kinase deficiency and Duchenne muscular dystrophy. We report a new case of this rare disease.

**Case report:** Twin male sibs at the ages of 30 day-old were hospitalized because of feeding difficulties, vomiting and weight loss. Parents had no consanguinity and there was no family history of endocrine or renal diseases. One of the twins died in a few hours. Other patient’s physical examination revealed that weight was 2100 g (his twin’s was 1800 g), body length 49 cm. Blood pressure: 70/40 mm-Hg; heart rate: 140 beats/minute, respiratory rate: 50 breaths/minute, body temperature: 36 °C. He was dehydrated and lethargic. External genitalia were well developed with intrascrotal testes and a normal vaginal opening. Chromosome analysis showed normal male karyotype. Laboratory tests were consistent with primary adrenal insufficiency. The twins were referred, respectively at 6 and 5 y of age for precocious puberty, accelerated growth, bone age advance. According to the molecular genetics of 21-hydroxylase gene, HA revealed to bear a non classical form (Triple mutation in exon 6 and I121T mutation in exon 5 of CYP 21), and DA a classical one (Large lesion of CYP 21 and genic conversion if this gene), missed at neonatal screening.

**Conclusions:** HSG is essential to detect in the treatment of CAH -and of all diseases treated by glucocorticoids-, although its molecular basis remains unexplained in the present case.
associated with salt-losing congenital adrenal hyperplasia. On ultrasound examination, adrenal gland could not be visualized. Therefore, a diagnosis of AHC was established. Further, we investigated for contiguous gene syndrome. Serum creatinine phosphokinase (CK) [9974 U/L, (N:35-195)] and triglyceride (TG) [439 mg/dl, (N:0-200)] levels were markedly elevated. On follow-up period of 9-month, his motor and mental development were retarded. Molecular studies confirmed a contiguous Xp21 deletion.

**Conclusions:** Serum CK and TG levels should be measured in all male patients who present with an adrenal hypoplasia. These simple tests may help early diagnosis and appropriate genetic counseling for next pregnancy.

<table>
<thead>
<tr>
<th>Initial Laboratory Tests</th>
<th>Standard dose ACTH stimulation test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Na (mEq/L)</td>
<td>Normal (N:135-145)</td>
</tr>
<tr>
<td>K (mEq/L)</td>
<td>5.1</td>
</tr>
<tr>
<td>BUN (mg/dl)</td>
<td>N:5-24</td>
</tr>
<tr>
<td>ACTH (pg/ml)</td>
<td>N:0-50</td>
</tr>
<tr>
<td>Cortisol (µg/dl)</td>
<td>&lt;1</td>
</tr>
<tr>
<td>17-hydroxyprogesterone</td>
<td>(ng/ml) 2,21</td>
</tr>
<tr>
<td>Renin activity (mU/ml)</td>
<td>0-2-5</td>
</tr>
<tr>
<td>Dehydroepiandrosterone</td>
<td>24</td>
</tr>
<tr>
<td>17-hydroxyprogesterone</td>
<td>18</td>
</tr>
<tr>
<td>11-deoxycortisol (ng/ml)</td>
<td>6 ACTH</td>
</tr>
<tr>
<td>Luteinizing hormone</td>
<td>(mU/ml) 0.7-7</td>
</tr>
<tr>
<td>Dehydroepiandrosterone</td>
<td>0.43</td>
</tr>
<tr>
<td>Follicle stimulating hormone</td>
<td>N:0.16-4.1</td>
</tr>
<tr>
<td>Total testosterone</td>
<td>N:0.7-4</td>
</tr>
</tbody>
</table>

**Objective and hypotheses:** Endogenous Cushing Syndrome in children is a rare disorder that is most frequently caused by pituitary or adrenocortical tumors. Cushing Disease associated with leukemia in children is very rare.

**Case:** A 3,8 year-old girl was admitted to our center with the complaints of excessive weight gain and depression during the last month. Her medical history revealed that she had diagnosed with B cell ALL 1.5 years ago. After induction protocol (BFM95), she has been treated with maintenance therapy (purinethol and methotrexate) for the last 10 months. CNS invasion was not detected during the follow-up period of 9-month. She became hypothyroid and hypocortisolemic during hospitalization. Baseline and stimulated cortisol levels were 16.7 mcg/dl and after overnight single dose (0.5 mg) dexamethasone suppression test, morning cortisol was 1.23 mcg/dl. During follow-up, her weight gain and depression were controlled. During hospitalization, she was started on appropriate steroid treatment only, without any antihypertensive medication.

**Conclusion:** We suggest bilateral adrenalectomy might be a good choice for treatment of refractory hypertension in patients with congenital adrenal hyperplasia due to 11-beta-hydroxylation deficiency.

**Background:** Adrenalectomy has been used recently as an alternative treatment in patients with congenital adrenal hyperplasia. We describe our experience with bilateral adrenalectomy for severe hypertension in a 20 year-old female with 11-beta-hydroxylation deficiency (21beta HD).

**Case:** The patient was diagnosed with congenital adrenal hyperplasia due to 21beta HD at three years of age and did not attend to regular follow up visits. The compliance with medication was not proper until age 13 years. The patient was severely virelized and hypertensive when she was presented to our clinic at age 13. Laboratory investigations showed 17 OH P: 73.2 ng/ml (N:0.4-1.02), ACTH: 2708 pg/ml (N:10-50), Androstenedione: 40 ng/ml (N: 0.1-2.9), Total testosterone: 591 ng/ml (11-80), 11 deoxycortisol: 45.7 ng/ml (<8). She was restarted appropriate steroid replacement and antihypertensive treatment. Although androgen levels were suppressed to normal levels, hypertension remained uncontrolled and led to complications. Bilateral adrenalectomy was performed successfully. Blood pressure normalization was achieved immediately after intervention. On her recent visit at 6 months after adrenalectomy, she was clinically well and normotensive while taking appropriate steroid treatment only, without any antihypertensive medication.

**Conclusion:** We suggest bilateral adrenalectomy might be a good choice for treatment of refractory hypertension in patients with congenital adrenal hyperplasia due to 11-beta-hydroxylation deficiency.
Conclusions: AI is common in children with AC1 and early post-surgery phase. Very high levels of cortisol are likely to be associated with high risk of mortality. Being the highest frequency of hyperglycemia in Group 3 shows the importance of glucose monitoring besides cortisol at the early post-surgery phase. The frequency of other endocrine dysfunctions are also high in children with AC1.

P2-d-345 Autoimmune Endocrine Disease 1
Atrophic body gastritis in children with familial autoimmune thyroid disease
Cecilia Volta1; Silvia Cerasi1; Giuseppe Lippi1; Francesco Di Mario1; Sergio Bernasconi1
1University of Parma, Dpt. Pediatrics, Parma, Italy; 2AUSL Parma, Dpt. Laboratory Medicine, Parma, Italy; 3University of Parma, Dpt. Gastroenterology, Parma, Italy

Background: A frequent association between autoimmune thyroid disease (AITD) and atrophic body gastritis (ABG), with a significant family clustering, has been evidenced in adults. Few studies have analyzed this association in children. Gastropanels represents a non-invasive instrument that indicates the morphological status of the gastric mucosa through the measurement of serum levels of pepsinogens (PGI, PGII), gastrin -17 (G-17) and Helicobacter pylori antibodies.

Objective and hypotheses: Our aim was to determine the existence of an association between AITD and ABG in children, the possibility of an early detection of children at risk through Gastropanel, and predictiveness of its markers.

Methods: We evaluated gastropanels markers, parietal cell antibodies (PCA), thyroid function and autoantibodies (TPO, TG) in 20 children with AITD (group 1), in affected family members (group 2) and in a control group (n=19; group 3).

Results: TSH, T4, TPO and TG levels were not correlated with the gastropanel markers in the group as a whole. A higher percentage of pathological levels of PGI (10% vs 6.2%), PGII (30% vs 6.2%), G-17 (25% vs 12.5%) and PCA (5% vs 0%) were demonstrated in group 1 compared to controls, and the comparison between mean values evidenced that PGI, PGII and PCA levels in group 1 were situated between those in groups 2 and 3.

Conclusions: Correlation between age and PCA and PGI levels confirmed the importance of age in onset of autoimmune gastritis. Discrepancy between PGI and G -17 levels was evidenced in children with AITD, with a higher percentage of hypergastrinemia compared to lower levels of PGI, allowing us to hypothesize that gastric autoimmunity tends to appear at a later age and that the first manifestation may be modest hypergastrinemia. Our results indicate that children with AITD, in particular those with family clustering for autoimmune diseases, should be evaluated for gastric autoimmunity through gastropanel.

P2-d-345 Autoimmune Endocrine Disease 1
Hyperthyroidism and vasculitis associated with antineutrophil cytoplasmic antibodies (ANCA) in two girls receiving benzylthiouracil
Mongia Hachicha1; Imen Chaaboub1; Lamia Ben Mansour1; Najer Aloulou1; Nabilia Rekk1; Khawla Kamoun1; Mohamed Abid1; Jamil Hachicha1; Thuraya Kamoun1
1Chu Hedi Chaker, Pediatrie, Sfax, Tunisia; 2Chu Hedi Chaker, Nephrologie, Sfax, Tunisia

Background: Vasculitis associated to antineutrophil cytoplasmic antibodies (ANCA) is a rare complication of therapy with antithyroid medication. It was mainly described in patients treated by propylthiouracil (PTU), carbimazole, methimazole and rarely by benzylthiouracil (BASDEN).

Methods: We describe two cases of antineutrophil cytoplasmic antibodies (ANCA) vasculitis induced by Benzylthiouracil (BUTU).

Results: * Observation 1: F…, a 12-years-old girl, developed after two years benzylthiouracil medication for Grave’s disease, a vasculitis associated with cutaneous involvement (generalized ulcer necrotic purpura) and glomerulonephritis with proteinuria of 24 hours at 26 mg/kg/day, microscopic hematuria and renal failure with high creatinemia level (135 μmol/l). The ANCA type anti-MPO (myeloperoxidase) were positive. The histological study of the renal needle biopsy showed a focal necrotizing glomerulonephritis and crescents with different evolutive stages. The discontinuation of benzylthiouracil and the treatment by the corticoids induced a disappearance of cutaneous lesions, a negative result of proteinuria, a normalization of the renal function and a disappearance of hematuria and ANCA. Clinical and biological remission was obtained during six years. * Observation 2: A…, a 12-years-old girl, was treated by benzylthiouracil for Grave’s disease, since the age of five years. Three years later, she suffered from arthritis and generalised purpura. The ANCA anti-TPO and anti-MPO were positive. The withdrawal of benzylthiouracil caused disappearance of skin lesions and arthritis. Although, at the age of ten, Grave’s disease relapsed. ANCA remained positive without renal dysfunction.

Conclusions: Since benzylthiouracil may induce severe forms of vasculitis, ANCA must be measured when confronted to systemic manifestations during treatment.

P2-d-346 Autoimmune Endocrine Disease 1
Sardinian family with APS1: unusual presentation and elevated IFNω and IFN α autoantibodies in early infancy
Antonella Meloni1; Michela Atzeni1; Maria Furcas1; Maria Teresa Scaslas2; Teresa Tuveri1; Anthony Meager2
1Pediatric Clinic II, Ospedale Microcitemico, University of Cagliari, Dipartimento di Scienze Biomediche e Biotecnologie, University of Cagliari, Cagliari (Sardinia), Italy; 2Laboratorio di Genetica Molecolare, II Clinica Pediatria, Ospedale Microcitemico, Dipartimento di Scienze Biomediche e Biotecnologie, University of Cagliari, Cagliari (Sardinia), Italy; 3National Institute for Biological Standards and Control, Division of Immunology, Blanche Lane, South Mimms EN6 3QG, United Kingdom

Background: APS1 is a rare AR disorder (OMIM 240300) caused by mutation in the AIRE gene. The major manifestations are chronic mucocutaneous candidiasis (CMC), hypoparathyroidism and Addison disease. Multiple additional autoimmune components occur variably.

Objective: Because of the rarity and the variability at presentation and clinical features, APS1 diagnosis may be extremely difficult. We emphasized the role of the mutational analyses of the AIRE gene and specific IFNω and IFNα autoantibodies (aAbs) in diagnosis of APS1.

Methods: Mutational analysis was performed by DNA sequency of AIRE gene: Anti-type I IFN aAbs were titrated using ELISA and anti-viral interferon neutralization assays.

Patients and results: The index case, a 3-year-old girl, was the first of two children born to healthy, consanguineous Sardinian parents. She was examined for acute hepatitis (ALT 312 UI/L, AST 226 UI/L). Autoimmune hepatitis was eventually diagnosed by elevated serum aAbs (LKM-1:640) and liver biopsy. APS1 was first suspected due to history of recurrent oral candidiasis. Genetic analysis revealed homozygous R139X mutation; the family screening allowed preclinical diagnosis of APS1 in the younger brother (6 months of age). High titer of IFNω and IFNα aAbs were found in the patients (tab); parents and young controls were all negative.

Conclusions: APS1 has a high prevalence in Sardinian population (1:14.400). Autoimmune hepatitis is a serious, relatively common symptom in Sardinian patients (28.6%); it may appear at presentation, beside CMC. The single Sardinian major mutation, R139X, should help us diagnose earlier APS1 with unusual presentation and provide treatments to prevent serious/fatal complications. IFNω and IFNα aAbs may be elevated very early in life, even before the occurrence of any clinical manifestation. Our data suggests IFNω and IFNα aAbs may be effective markers of preclinical APS1; they may allow precocious diagnosis in heterogeneous populations in which genetic analyses of the AIRE gene may be complex.

The table below shows the antibody titers and binding activity in the index case (6 years) and the younger brother (3 years).

<table>
<thead>
<tr>
<th>AIRE mutation</th>
<th>Clinical manifestation</th>
<th>Anti-IFNω NAb titre</th>
<th>Anti-IFNω binding OD</th>
<th>Anti-IFNα NAb titre</th>
<th>Anti-IFNα binding OD</th>
<th>Other aAbs tested</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Index Case</td>
<td>R139X</td>
<td>LKM-1 (+++)</td>
<td>ANA (+)</td>
<td>ASA (+)</td>
<td>APCA (-)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R139X</td>
<td>ANA (+)</td>
<td>ASA (+)</td>
<td>APCA (-)</td>
<td>GAD (-)</td>
<td></td>
</tr>
<tr>
<td>Brother</td>
<td>R139X</td>
<td>Preclinical APS1</td>
<td>5.000</td>
<td>3.9</td>
<td>12.000</td>
<td>2.89</td>
</tr>
<tr>
<td></td>
<td>R139X</td>
<td>LKM (+)</td>
<td>ANA (+)</td>
<td>ASA (+)</td>
<td>APCA (-)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R139X</td>
<td>ANA (+)</td>
<td>ASA (+)</td>
<td>APCA (-)</td>
<td>GAD (-)</td>
<td></td>
</tr>
</tbody>
</table>

50th Annual Meeting of the ESPE
Horm Res 2011;76(suppl 2) 143
**Background:** Hashimoto's encephalopathy is a rare condition of presumed autoimmune origin characterized by increased antithyroid antibody titers and responsive to glucocorticoid therapy. The pathogenesis of the disease is unclear; some studies suggested localized cerebral edema, autoimmune vasculitis or toxic effect of antibodies as possible causes.

**Objective and hypotheses:** We present a case of acute encephalopathy associated with positive antithyroid antibody titers.

**Case report:** A 15yr old boy was admitted to our Pediatric Center because of an acute onset of attention deficit, somnolence, cognitive decline and temporary aphasia. Some days later he presented sleep disturbances and neuropsychiatric symptoms (paranoia, agitation, and aggressiveness). Toxic, metabolic and infectious causes of encephalopathy were excluded with routine blood tests, serological screening, neuroimaging and CSF examination. The EEG showed a widespread slowing of the background activity, in particular in the left cerebral hemisphere. Positron emission tomography demonstrated diffuse hypometabolism in parietal, occipital and temporal zone. Thyroid hormone levels were in normal range and antibody titers were increased (anti-Tg Ab 121 UI/L;anti-TPO Ab 69 UI/L; reference values respectively<115 and <34). The thyroid US was normal while the ECG showed sinus bradycardia.

**Results:** Glucocorticoid therapy (Prednisone 50 mg orally) was started with an improvement of neuropsychiatric symptoms after 72 hours. The symptoms receded completely with normalization of EEG, ECG and serum level of antithyroid antibodies within 3 weeks. months after the stop therapy, positive antithyroid antibody titers reappeared, however thyroid function and clinical conditions remained normal during follow-up.

**Conclusions:** Hashimoto's encephalopathy appears to be a rare disorder in adolescent patients, but it must be considered in case of “investigation negative encephalopathy”.

---

**Introduction:** Chronic autoimmune disease of thyroid gland (Hashimoto Thyroiditis) and permanent intolerance in gluten proteins (Celiac disease) are documented to be associated with type 1 Diabetes in children and adults.

**Purpose:** To determine the prevalence of Hashimoto Thyroiditis and Celiac disease in children with type 1 Diabetes.

**Methods:** 45 children with type 1 Diabetes were included in the study. Serological laboratory testing of Gliadin antibody (IgG, IgA), anti-tissue Transglutaminase, T4, TSH, TPO and TG autoantibodies were determined in " Fati Im " laboratory in Pristina based in two methods: Time resolved flurometry (DELFIA Walac) and ELISA (ELISA) DRG. Jejunal biopsy was done in five patients with positive serological testing of antibodies in celiac disease.

**Results:** The prevalence of Celiac Disease was in 7 of 45 children. In five patients diagnose of Celiac disease was made in the same time when it was confirmed type 1 Diabetes and in two patients one to four years later. The prevalence of Hashimoto thyroiditis was in 9 of 45 children. In four patients diagnosis of Hashimoto thyroiditis was in the same time when Diabetes was confirmed and in five another after one to five years later.

**Conclusions:** Children diagnosed with Diabetes type 1 must be tested for celiac disease and Hashimoto thyroiditis in time of diagnose of Diabetes and testing must be repeated every year in those that results negative.
P2-d1-461 Bone, Growth Plate and Mineral Metabolism 1

Novel ENPP1 loss-of function mutation causing generalized arterial calcification of infancy and autosomal recessive hypophosphatemic rickets

Cécile Brachet1; Nicole Van Regemorter2; Anne Clerckx1; Denise Blum3; Tim Storm3; Claudine Heinrichs1

1Hôpital Universitaire des Enfants Reine Fabiola-ULB, Pediatrics, Brussels, Belgium; 2Hôpital Erasme-ULB, Genetics, Brussels, Belgium; 3Institut für Humangenetik des Klinikums rechts der Isar der Technischen Universität München, Genetics, Munich, Germany

Background: Ecto/nucleotide pyrophosphatase/phosphodiesterase 1 (ENPP 1) is a major generator of extracellular pyrophosphate, which inhibits hydroxyapatite crystal deposition. ENPP1 loss of function mutations have been described in patients with autosomal recessive hypophosphatemic rickets (HR), in patients with generalized arterial calcification of infancy (GACI) and in patients with both conditions.

Objectives: We describe a patient homozygous for ENPP1 loss of function mutation.

Method: Case report.

Results: She was the first child of consanguineous Tunisian parents. She was born at 36 weeks of gestational age after a pregnancy characterized by hydramnios and preeclampsia. Birth weight was 2.3kg, birth length 44 cm and head circumference 33 cm. A large placental infarct was observed. She had fetal distress necessitating respiratory and hemodynamic support. Central deafness was diagnosed neonatally. On plain Xrays, generalized arterial and peri-articular calcifications were noted. At one week of age, she developed reno-vascular hypertension and congestive heart failure. This was treated by fluid restriction, aldactone, captopril, salicylic acid and enalapril. Arterial calcifications disappeared radiologically after the 5th month of life, hypertension subsided at 5 years of age. At 5 months of age, she developed clinical and radiological rickets initially attributed to biphosphonates. Hypophosphatemic rickets was diagnosed and treated with 1,25OHD and P at 3.8 years of age. She is now 22 years old, her height is 155 cm, without major bone deformities (after 2 osteotomies), her intelligence is near normal with profound deafness. ENPP1 sequencing showed a homozygous 1 bp deletion within intron 3, NM_006208.2:c.[430+1delG]+[430+1delG].

Conclusion: We report on a patient with a novel homozygous ENPP1 loss of function mutation that presented with GACI that disappeared after infancy and subsequently developed hypophosphatemic rickets. Deafness could result from calcification of the supplying arteries and has been previously reported.

P2-d1-463 Bone, Growth Plate and Mineral Metabolism 1

Surgical treatment of children with hyperparathyroidism: a single centre experience

Swethan Alagaratnam1; Caroline Brain1; Helen Spoudeas2; Mehul Dattani3; Jeremy Allgrove3; Peter Hindmarsh3; Tom Kurzawinski4; William Van’t Hoff4

1University College London Hospital, Department of Endocrine Surgery, London, United Kingdom; 2UCL Institute of Child Health, Department of Paediatric Endocrinology, London, United Kingdom; 3Royal London Hospital, Barts and the London NHS trust, Department of Paediatric Endocrinology, London, United Kingdom; 4UCL Institute of Child Health, Great Ormond Street Hospital for Children, Department of Nephrology, London, United Kingdom

Background: Hyperparathyroidism (HPT) in children is a rare disease, with delayed diagnosis and limited evidence with regards to outcomes following surgery.

Objective and hypotheses: We report our experience of surgical management of hyperparathyroidism, over a 25 year period, for children under the age of 18 years including neonates.

Methods: Retrospective case review of all children under the age of 18 years from 1978 to 2010 who underwent parathyroid surgery at our institution.

Results: Twenty-four children met our inclusion criteria. This group included 6 neonates with severe neonatal hyperparathyroidism (SNHPT), 11 children with primary HPT and 7 children with familial HPT. Imaging studies used included ultrasound (sensitivity 88.9%, specificity 100% in identifying single gland disease), Sestamibi scan (sensitivity 87.5%, specificity 100% in single gland disease) and parathyroid venous sampling in 2 children prior to 1980. All children with SNHPT and 5 of 7 children with familial causes of HPT underwent total parathyroidectomies. One child with MEN 2a and one child with MEN1 underwent removal of 3 glands, due to remaining intrathyroidal glands. In children with primary HPT, two underwent subtotal parathyroidectomies prior to 1980, four children underwent neck explorations and removal of enlarged glands between 1980 to 2002, and minimally invasive parathyroidectomies were carried out since 2002 in 5 children, with the last 3 cases involving intraoperative parathyroid hormone measurements. No post operative complications were noted in our patient group. None of our neonate group and children with primary HPT required reoperations. The child with a subtotal parathyroidectomy for MEN1 related HPT, developed a recurrence of hypercalcemia and required a reoperation for removal of the remaining gland.

Conclusions: Our experience demonstrates good outcomes without complications for surgery in our patient group. Minimally invasive parathyroidectomy should be regarded as the operation of choice in primary HPT.

P2-d1-462 Bone, Growth Plate and Mineral Metabolism 1

Reduced volumetric trabecular bone mineral density of the lumbar spine in adolescents with anorexia nervosa

Lily M Wheeler1; M Zulf Mughal2; Judith E Adams3; Jane Whittaker4; Swethan Alagaratnam1; Caroline Brain1; Helen Spoudeas2; Sarah Ehlichman1

1University of Manchester, Medical School, Manchester, United Kingdom; 2Royal Manchester Children’s Hospital and The University of Manchester, Endocrinology, Manchester, United Kingdom; 3Manchester Royal Infirmary and The University of Manchester, Radiology, Manchester, United Kingdom; 4Royal Manchester Children’s Hospital, Child Psychiatry, Manchester, United Kingdom; 5Royal Manchester Children’s Hospital, Endocrinology, Manchester, United Kingdom

Introduction: It is well known that AN in adolescents is associated with reduced bone mineral density (BMD). Its aetiology is multi-factorial.

Objectives: A retrospective evaluation of patients with AN referred for endocrine and metabolic bone health assessment between February and September 2010, in whom lumbar spine (LS) BMD was measured by dual-energy X-ray absorptiometry (DXA) and quantitative computed tomography (QCT). Their distal radial total and trabecular BMD was measured by peripheral QCT (pQCT).

Methods: BMD of L1-L4 was measured by DXA. This data was expressed as bone mineral apparent density (BMD, g/cm^3) and values transformed to Z scores using local normative data (ARC,2007;9(2):1:53-9). Volumetric trabecular BMD (TBMD, mg/cm^3) of L1-L3 was measured by QCT and values transformed to Z scores using Mindways sofwareTM (Austin,Texas). Distal radial (4%) total and TBMD was measured by peripheral QCT (pQCT) and values transformed to Z scores using local normative data (Osteoporos Int. 2009;20(8):1337-46).

Results: 12 females and one male aged 13.6-16.9 years with a diagnosis of AN were evaluated. The mean duration of AN was 3.5 years (0.7 to 8). 11 of the females had amenorrhoea; 2 primary, 9 secondary (mean duration 1.2 years). Mean BMI SDS was -1.74 (-4.2 to 0.19). A table of median Z scores of LS BMD, LS TBMD, distal radial total BMD and distal trabecular BMD is shown below.

<table>
<thead>
<tr>
<th>BMD Test</th>
<th>Median Z score</th>
<th>Range Z scores</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>LS BMD</td>
<td>-0.45</td>
<td>-2.36 to 0.79</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>LS TBMD</td>
<td>-0.91</td>
<td>-3.42 to -0.42</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Distal radius total BMD</td>
<td>0.06</td>
<td>-1.83 to 1.08</td>
<td>0.656</td>
</tr>
<tr>
<td>Distal radius trabecular BMD</td>
<td>-0.05</td>
<td>-1.91 to 0.81</td>
<td>0.416</td>
</tr>
</tbody>
</table>

Conclusions: We report our experience of surgical management of hyperparathyroidism, over a 25 year period, for children under the age of 18 years including neonates.

Objective and hypotheses: We report our experience of surgical management of hyperparathyroidism, over a 25 year period, for children under the age of 18 years including neonates.

Methods: Retrospective case review of all children under the age of 18 years from 1978 to 2010 who underwent parathyroid surgery at our institution.

Results: Twenty-four children met our inclusion criteria. This group included 6 neonates with severe neonatal hyperparathyroidism (SNHPT), 11 children with primary HPT and 7 children with familial HPT. Imaging studies used included ultrasound (sensitivity 88.9%, specificity 100% in identifying single gland disease), Sestamibi scan (sensitivity 87.5%, specificity 100% in single gland disease) and parathyroid venous sampling in 2 children prior to 1980. All children with SNHPT and 5 of 7 children with familial causes of HPT underwent total parathyroidectomies. One child with MEN 2a and one child with MEN1 underwent removal of 3 glands, due to remaining intrathyroidal glands. In children with primary HPT, two underwent subtotal parathyroidectomies prior to 1980, four children underwent neck explorations and removal of enlarged glands between 1980 to 2002, and minimally invasive parathyroidectomies were carried out since 2002 in 5 children, with the last 3 cases involving intraoperative parathyroid hormone measurements. No post operative complications were noted in our patient group. None of our neonate group and children with primary HPT required reoperations. The child with a subtotal parathyroidectomy for MEN1 related HPT, developed a recurrence of hypercalcemia and required a reoperation for removal of the remaining gland.

Conclusions: Our experience demonstrates good outcomes without complications for surgery in our patient group. Minimally invasive parathyroidectomy should be regarded as the operation of choice in primary HPT.
Bone density analysis in children born small for gestational age (SGA)  

Danilo Fintini1; Claudia Bruftan2; Annalisia Deodati1; Graziaria Ubiergi1; Stefano Cianfarani1; Francesca Creata1; Marco Cappa1  
1Bambino Gesù Children Hospital, Cardiorespiratory and Sport Medicine Unit, Rome, Italy; 2Bambino Gesù Children Hospital, Scientific Directorate, Rome, Italy; 3Tor Vergata University, Department of Public Health and Cell Biology, Rome, Italy; 4Bambino Gesù Children's Hospital and Tor Vergata University, Molecular Endocrinology Unit, Rome, Italy; 5Bambino Gesù Children Hospital, Endocrinology Unit, Rome, Italy  

Background: Conflicting results about influence of birth size (BW) on bone mineral density (BMD) and bone mineral content (BMC) are present in the literature. Fetal growth pattern seems to influence bone accrual not only in small for gestational age (SGA) but also when birth weight is maintained in the normal range.  

Aim: We analyzed BMD and body composition in children born SGA compared with age and sex matched children born appropriate for gestational age (AGA).  

Patients: 53 SGA (26 f; mean age 10.9±2.5 yrs; BW 2.4±0.4 kg, percentile 2.9±0;4; BMI SDS 1.2±1.9; pubertal stage 2.2±1.4) vs 40 AGA (27 f; mean age 11.6±2.9 yrs; BW 3.4±0.4 kg, percentile 4.8±1.6; BMI SDS 1.3±1.6; pubertal stage 2.6±1.4) were studied using DXA scan (Hologic) in order to obtain lumbar and whole body BMC, normalized for height BMD (BMDAD and nWBMD), lumbar Z score and fat and lean mass. None of the patients were taking medication influencing growth or bone content.  

Results: No differences were found in bone parameters between SGA and AGA children, both as a whole group and divided for gender. SGA children showed higher fat mass expressed as g but no difference in lean and fat mass was found when expressed as percentage of body weight. The multivariate analysis showed independent positive correlation between BW and whole body BMC (beta 0.479; p<0.001) in SGA and in lumbar areal BMD (beta 0.343; p 0.035) in AGA.  

Conclusion: Our data suggest that although a correlation between birth weight and bone density seems to exist, no statistical difference in bone parameters and body composition is present in peripubertal SGA and AGA children.

A novel mutation of the parathyroid hormone gene (PTH) in a girl with isolated hypoparathyroidism  

Diana Alexandra Ertti1; Susanne Stary2; Berthold Streubel3; Adalbert Raimann1; Gabriele Haeusler1  
1Medical University of Vienna, University Clinic of Paediatrics and Adolescent Medicine, Paediatric Pulmology, Allergology and Endocrinology, Vienna, Austria; 2Medical University of Vienna, Institute of Pathology, Genetics, Vienna, Austria; 3Medical University of Vienna, Clinical Institute for Pathology, Genetics, Vienna, Austria  

Case report: A female patient (parents first-degree cousins) presented with severe symptomatic hypocalcaemia (1.44 mmol/l) at the age of 4 months. Treatment with 1,25-(OH)2-cholecalciferol and calcium was immediately started and serum calcium concentrations were stabilized at the lower normal range. During the 6 years of observation, the serum levels of PTH were always low but detectable, with values between 5.3 and 2.5 pg/ml (normal 15 - 65 pg/ml). Disturbances in the vitamin-D metabolism, autoimmune polyendocrine syndrome (APS), chromosomal anomalies or mutations in the calcium-sensing receptor gene (CaSR) were excluded. Microarray analysis revealed loss of heterozygosity for PTH. Sequencing analysis of PTH showed the presence of c.68C>A homozygous point mutation that causes a premature stop-codon (p.Ser23X) resulting in a non-functional PTH-precursor. The parents and the 2 siblings are heterozygous for the mutation. A complementary measurement conducted at a specialized institution showed undetectable PTH levels.  

Discussion: Isolated hypoparathyroidism is a diagnostic challenge. Besides chromosomal defects (CATCH22, Di George Syndrome), differential diagnosis includes APS and mutations in the CaSR. The detection of measurable PTH serum concentrations in our patient made a defect in the PTH gene rather unlikely - only three PTH mutations have been published so far.

A case of Hypoparathyroidism-Retardation-Dysmorphism syndrome complicated by gastrointestinal and respiratory insufficiency  

Corinna Grasemann1; Katja Schaaf1; Wei-Shin Liu1; Uwe Millies2; Patrik Gerner3; Simone Kathemann1; Florian Stehling3; Stephanie Gross1; Beate Albrecht1; Bernhard Horsthemke1; Berthold Hauffe1  
1Kinderklinik II, UK-Essen, Pediatric Endocrinology, Essen, Germany; 2Kinderklinik III, UK-Essen, Pediatric Respiratory Medicine, Essen, Germany; 3Kinderklinik II, UK-Essen, Pediatric Gastroenterology, Essen, Germany; 4UK-Essen, Department of Human Genetics, Essen, Germany; 5Kinderklinik II, UK-Essen, University of Essen-Duisburg, Department of Pediatric Endocrinology, Essen, Germany  

Background: Hypoparathyroidism-Retardation-Dysmorphism Syndrome (HRD) is a rare disorder characterized by congenital hypoparathyroidism, mental retardation, severe growth retardation and typical dysmorphic features. An MRI series in 4 children also revealed pituitary hypoplasia with possible growth hormone deficiency and adrenal insufficiency in these patients.  

Case report: We report a 3-year-old girl with typical features of HRD from a family of Moroccan origin. The patient presented SGA with congenital hypoparathyroidism and cortical thickening. Serum IGF-I levels were low with normal serum IGFBP-3 and cortisol levels. Serum calcium levels remained stable at the low normal range with an oral calcium (500 mg/d) and calcitriol (60 ng/kg BW/d) supplementation. HRD in this patient was complicated by severe gastrointestinal motility disorder with delayed gastric passage and retroplulsion from the small bowel into the stomach. Despite feeding via jejunal tube and prokinetic treatment enteral feeding alone did not cover the caloric needs. The patient developed respiratory insufficiency due to thoracic dystrophy with pulmonary hypoplasia and recurrent aspiration-pneumonia, requiring long term mechanical ventilation. Intercurrent H1N1 viral infection resulted in further deterioration of gastric and pulmonal symptoms, requiring TPN and iv calcium substitution. Genetic analysis revealed homozygosity for the previously described 12 bp deletion in the Tubulin Chaperone E (TBCE) gene, which encodes a chaperone necessary for the folding of tubulin α.  

Conclusion: We report a case of HRD complicated by gastric and respiratory insufficiency not previously described. In HRD families, some patients with suspected HRD die at a very young age. Cortisol deficiency has been discussed as a possible cause. However, facultative respiratory and gastrointestinal involvement with respiratory insufficiency, might limit survival in newborns. Severely affected children may require temporary iv calcium supplementation during intercurrent illness.

Mutational spectrum of SHOX gene in 25 Italian pediatric patients with Léri-Weill dyschondrosteosis (LWD)  

Annalisa Nicciti1; Laura Mazzanti1; Piero Pirazzoli1; Soara Menabò1; Loredana Boccone1; Emanuela Scaroni1; Alessandro Cicogna1; Lilà Baldazzi1  
1University of Bologna, Department of Gynecological Obstetric and Pediatric Sciences, Bologna, Italy; 2Micrornatomy Hospital, Clinical genetics and rare diseases unit, Cagliari, Italy  

Background: SHOX gene maps in the troubled pseudoautosomal region 1 (Xp22.33, Yp11.3) and is the candidate gene in LWD. The mutation mechanisms involving SHOX gene represent a modern model of pathogenetics including: point mutations, gene deletions, complex chromosomal rearrangement.
ments with variable penetrance due to unknown mechanisms.

Objective and hypotheses: In the last six years, the finding of deletions hundreds of megabases from the gene point to the enhancers regulating the SHOX expression, that produce, if disabled, the same LWD phenotype as SHOX gene mutations. In this study each type of mutation in SHOX locus was considered in order to complete the diagnostic procedure in LWD patients and determine their frequency.

Methods: 25 patients with clinical diagnosis of LWD were studied for mutation in SHOX gene and its 3' regulatory region. Genetic analysis was performed on genomic DNA from peripheral blood by SHOX gene sequencing and MLPA analysis of SHOX locus.

Results: Seven point mutations were identified: five new (M1T, R160X, A234Y, IVS4+1G->C, IVS5+1G->A); two known (E144K, R160P). Height deletions were identified, one was in the 3' regulatory region. In this case, the break point was mapped in order to determine the number of CNEs involved. One substitution (IVS4-57C>T) of undetermined significance was found in two unrelated patients.

Conclusions: A condition of SHOX haploinsufficiency was confirmed in 12 patients; in 3 patients the missense mutations require functional studies to determine their causativity. In one case with a LWD phenotype the mutation (R160P) was homozygous, indicating a weak effect of the mutation or a low penetrance due to other factors. The deletion of 3' regulatory region involves CN7 and CN8 and seems to be caused by a NHR1 (non homologous end joining) mechanism. In total an alteration in SHOX gene or its 3' regulatory region was found in 15/25 (60%) patients confirming the SHOX mutation joining (R160P) was homozygous, indicating a weak effect of the mutation or a low penetrance due to other factors. The deletion of 3' regulatory region involves CN7 and CN8 and seems to be caused by a NHR1 (non homologous end joining) mechanism. In total an alteration in SHOX gene or its 3' regulatory region was found in 15/25 (60%) patients confirming the SHOX mutation detection in LWD, with an unexpected equal ratio between point mutation and deletion.

P2-d1-469 Bone, Growth Plate and Mineral Metabolism 1

Neonatal hypocalcaemia with seizures - diagnosis of infantile osteopetrosis

Anke Pyper1; Ansgar Schultz2; Gabriele Hahn3; Angela Huebner1

1Children’s Hospital, Technical University Dresden, Division of Endocrinology and Diabetes, Dresden, Germany; 2University Medical Center Ulm, Department of Pediatrics and Adolescent Medicine, Ulm, Germany; 3Children’s Hospital, Technical University Dresden, Division of Radiology, Dresden, Germany

Background: Infantile malignant osteopetrosis is a rare, severe disease due to autosomal recessive mutations in TCIRG1, CLCN7, OSTM1 or RANK genes. Impaired osteoclast function leads to insufficient bone resorption which results in bone marrow failure and impaired calcium metabolism. Hematopoietic stem cell transplantation was developed as the only curative strategy.

Case reports: 1: A male newborn (38 weeks) presented with seizures 7 days after birth due to hypocalcaemia (calcium 0.6 mmol/l). Despite calcium supplementation hypocalcaemia and seizures persisted and the child was given phenobarbital. During the following weeks the child developed anemia (Hb 6.0 mmol/l), thrombocytopenia and elevation of liver enzymes. The child presented critically ill with near blindness, intermitted nystagnus, bone marrow failure, persistent hypocalcaemia and hyperparathyroidism (PTH 400 pg/ml, NR 7-53). X-ray showed extremely dense sclerotic bones, leading to diagnosis of infantile osteopetrosis. Molecular analysis revealed compound heterozygote mutation in the TCIRG1 gene. The child underwent hematopoietic cell transplantation from his HLA identical sister. Due to bone sclerosis, he is blind. 2: One year later same mother delivered a girl in whom prenatal diagnosis was declined. The child developed hypocalcaemia and signs of beginning bone marrow failure at day 5 of life. X-ray confirmed infantile osteopetrosis, leading to immediate hematopoietic cell transplantation from his HLA identical healthy other brother. The child died due to pneumocystis jirovecii pneumonia at the age of 3.5 months.

Conclusions: Impairment of calcium homeostasis is common in newborns, and in the majority of cases is due to changes in blood pH or to insufficient PTH secretion after birth. Osteopetrosis is very rare, but early diagnosis is required in order to initiate immediate hematopoietic cell transplantation. Hypocalcaemia, hyperparathyroidism and signs of impaired bone marrow function should therefore immediately urge physicians to perform X-ray and genetic analyses.

P2-d2-470 Bone, Growth Plate and Mineral Metabolism 2

A novel CYP27B1 gene mutation in a Turkish family with pseudovitamin D deficiency rickets

Erdem Durmaz1; Minjing Zou2; Iffet Bircan1; Sema Akcurin1;

1Akdeniz University, Pediatric Endocrinology, Antalya, Turkey; 2King Faisal Specialist Hospital and Research Center, Genetics, Riyadh, Saudi Arabia

Background: Pseudovitamin D deficiency rickets (PDDR) is an autosomal recessive disorder caused by 1α-hydroxylase deficiency.

Objective and hypotheses: The index patient is a 14 month-old girl with growth retardation and muscle weakness. Growth and development had been normal until the age of 9 months when she showed difficulties with standing up and poor appetite. Physical examination revealed classical features
of rickets such as large fontanel, rachitic rosary, Harrison’s groove, palpable widening of the distal radius. Systemic examination was unremarkable. Laboratory data showed hypocalcemia (7.5 mg/dL, Normal: 8.6-10.2 mg/dL), hypophosphatemia (2.4 mg/dL, Normal: 2.7-4.5), high serum alkaline phosphatase (5546 U/L, Normal: 0-270 U/L), high serum 25-OH vitamin D (108 ng/mL, Normal: 11.1-42.9 ng/mL), low serum 1, 25 (OH)2 Vitamin D (14 pg/mL16-65 pg/mL) and high serum parathyroid hormone (396 pg/mL, Normal: 15-65 pg/mL). Her mother and father had normal biochemical profile except low level of serum 25-hydroxyvitamin D. Her X-ray showed classical signs of widening and irregular mineralization of the distal radial metaphyses. The patient was treated with calcitriol (1, 25 (OH)2 D3) 1 mcg daily and calcium carbonate 750 mg daily. At the third month of the treatment she showed remarkable symptomatic, biochemical and radiological improvement with normalization of serum calcium and phosphorus.

Method: To identify molecular defect leading to 1α-hydroxylase deficiency, we screened CYP27B1 for mutation. A novel monoallelic c.1079 C>A point mutation at codon 360 (p.S360X) was found in the patient and her father, with normal parental origin. We then screened CYP27B1 in her mother. A novel monoallelic c.1079 C>A point mutation at codon 360 (p.S360X) from father and p.R389H from mother.

Conclusion: The 1α-hydroxylase deficiency was caused by these two mutations in the CYP27B1 gene.

P2-d2-471 Bone, Growth Plate and Mineral Metabolism 2

Important reduction in bone mass acquisition in 43 adolescents with recent anorexia nervosa
Laurent Maimoun1; Giacomo Gastaldi2; Patrick Lefebvre2; Sébastien Guillaume3; Isabelle Rainear1; Anne Wojtusciszyn1; Anne Chalençon2; Denis Mariano-Goulart4; Eric Thomas5; Olivier Coste6; Eric Renard7; Franois Paris8; Charles Sultan9; Jacques Bringer2

43 adolescents who exhibited AN during the peripubertal period were included in our study. We evaluated the effect of AN on bone mass acquisition, measured by total proximal femur, total body and lumbar spine bone mineral density (aBMD) during the peripubertal period. To evaluate the effect of AN on areal bone mineral density, we performed a study in 43 adolescents with recent AN points to the need for early bone investigation in these patients and swift therapeutic response.

Background: Puberty is a crucial period for bone mass acquisition. However, nutritional deprivation as observed in patients with anorexia nervosa (AN) may have a deleterious effect on peak bone mass.

Objective and hypotheses: To evaluate the effect of AN on areal bone mineral density (aBMD) during the peripubertal period.

Methods: Forty-three patients who exhibited AN during the peripubertal period and 25 age-matched controls were recruited for this study. The aBMD at whole body, total proximal femur, and lumbar spine was determined using dual-energy X-ray absorptiometry.

Results: A good response was established in all the cases. We suggest an explanation for the acquisition; rather than resorptive effect of vitamin D over the bone metabolism, limitation of bowel absorption effect of vitamin D over the bone metabolism may be considered a priority in infantile vitamin D intoxication by increasing the bone resorption. Therefore, bisphosphonates are presented as a main modality in vitamin D intoxication due to low calcium resorption.

Conclusion: The classic treatment of vitamin D intoxication consists of intravenous hydration, furosemide, prednisolone and bisphosphonate therapy. It is mostly accepted that 25-OH vitamin D is responsible for hypercalcemia in vitamin D intoxication by increasing the bone resorption. Therefore, bisphosphonates are presented as a main modality in vitamin D intoxication due to low calcium resorption.

Objective: To report our clinical experience with the effectiveness of hPTH in pediatric patients with syndromic conditions including hypoparathyroidism.

Background: Subcutaneous human 1-34 parathormone (hPTH) has been introduced for treatment of hypoparathyroidism, allowing avoidance of vitamin D plus calcium side effects. It is mostly accepted that 25-OH vitamin D is responsible for hypercalcemia in vitamin D intoxication by increasing the bone resorption. Therefore, bisphosphonates are presented as a main modality in vitamin D intoxication due to low calcium resorption.

Conclusion: A good response was established in all the cases. We suggest an explanation for the acquisition; rather than resorptive effect of vitamin D over the bone metabolism, limitation of bowel absorption effect of vitamin D on calcium metabolism may be considered a priority in infantile vitamin D intoxication treatment.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>AN (n=43)</th>
<th>Controls (n=25)</th>
<th>Difference (%)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>17.4 (1.6)</td>
<td>17.3 (1.9)</td>
<td>0.8 0.75</td>
<td>0.43</td>
</tr>
<tr>
<td>Anthropometric data</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>41.8 (6.3)</td>
<td>59.1 (9.7)</td>
<td>-29.2 &lt;0.0001</td>
<td></td>
</tr>
<tr>
<td>Height (cm)</td>
<td>163.3 (6.9)</td>
<td>165.7 (6.9)</td>
<td>-1.4 0.17</td>
<td></td>
</tr>
<tr>
<td>BM (kg/m2)</td>
<td>15.7 (1.8)</td>
<td>21.5 (2.8)</td>
<td>-27.0 &lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Body fat mass (%)</td>
<td>52.3 (3.3)</td>
<td>28.3 (8.5)</td>
<td>-59.9 &lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Body fat-free soft tissue (kg)</td>
<td>16.0 (8.1)</td>
<td>10.0 (8.1)</td>
<td>-42.4 &lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Body fat mass (%)</td>
<td>33.7 (4.7)</td>
<td>21.0 (4.5)</td>
<td>-18.9 &lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Clinical Data</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age of menarche (yr)</td>
<td>12.5 (1.3)</td>
<td>12.2 (1.3)</td>
<td>-2.31 0.436</td>
<td></td>
</tr>
<tr>
<td>Duration of amenorrhea (months)</td>
<td>11.4 (9.1)</td>
<td>-</td>
<td>0.81</td>
<td></td>
</tr>
<tr>
<td>Patients with amenorrhea (nb)</td>
<td>34/43</td>
<td>-</td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>Patients treated with OC (nb)</td>
<td>5/9</td>
<td>-</td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>Patients with amenorrhea (nb)</td>
<td>34/43</td>
<td>-</td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>Age of AN onset (yr)</td>
<td>15.5 (2.0)</td>
<td>-</td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>Duration of AN (yr)</td>
<td>1.8 (2.0)</td>
<td>-</td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>Bone Mineral Density (g/cm2)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole body</td>
<td>1.027 (0.073)</td>
<td>1.066 (0.077)</td>
<td>-3.9 0.04</td>
<td></td>
</tr>
<tr>
<td>Z-score</td>
<td>0.902 (2.7)</td>
<td>1.022 (2.8)</td>
<td>-10.9 0.03</td>
<td></td>
</tr>
<tr>
<td>Lumbar spine (L2-L4) (g/cm2)</td>
<td>0.986 (0.143)</td>
<td>0.990 (0.125)</td>
<td>-12.7 &lt;0.0001</td>
<td></td>
</tr>
<tr>
<td>Z-score</td>
<td>8.6 (10.1)</td>
<td>9.77 (12.7)</td>
<td>-11.4 &lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Femoral neck (g/cm-2)</td>
<td>0.843 (0.107)</td>
<td>0.956 (0.115)</td>
<td>-2.6 &lt;0.001</td>
<td></td>
</tr>
<tr>
<td>T-score</td>
<td>85.8 (11.2)</td>
<td>97.8 (12.2)</td>
<td>-2.9 &lt;0.001</td>
<td></td>
</tr>
</tbody>
</table>

*Two patients exhibited a primary amenorrhea. OC, oral contraceptive
Methods: We conducted a self-controlled study on 6 pediatric patients (4 males, age 9.8±5.1 years) with syndromic hypophosphatasia including 3 with Autoimmune Polynodocinopathy Candidiasis Ectodermal Dysplasia (APC/ED) syndrome, 1 with Di George syndrome, 1 with CHARGE association, 1 with Hypophosphatia-Deafness-Renal Dysplasia (HDR) syndrome. The 3 latter patients had renal dysplasia with borderline high blood creatinine levels in 2 cases and mild renal failure in 1. We compared the clinical and biochemical outcome of the patients under conventional treatment based on oral administration of calcium (1-1.5 g/day in 3 doses) plus oral calcitriol (20-50 mg/kg/day in 2-3 doses) with that implemented with hPTH (Teriparatide, 0.35 µg/kg bid). Therapy shift was introduced following hPTH paralleled by calcium-vitamin D withdrawal until sufficient to maintain clinical control. Blood calcium, phosphorus, alkaline phosphatase, urinary calcium to creatinine ratio before and after hPTH introduction were compared.

Results: rPTH treatment allowed complete withdrawal of calcium and vitamin D treatment in 2 patients, calcium in 2, and a reduction of vitamin D dosage in 2. Mean calcium, phosphorus, and alkaline phosphatase were unmodified, whereas there was a significant reduction of the calcium to creatinine ratio (0.55±0.31 vs 0.07±0.07, p=0.004). There was a consistent reduction in urinary calcium to creatinine ratio before and after hPTH introduction were compared.

Conclusion: hPTH allows a prompt normalization of urinary calcium excretion maintaining adequate calcium levels in spite of a conventional therapy dose reduction in syndromic hypophosphatasia.

Bone density and geometry in adolescents with inflammatory bowel disease

Avril Mason1; Sheila Khanna1; Richard K. Russell2; J. Bishop2; P. McGregor2; S. Faissal Ahmed2
1Royal Hospital for Sick Children, Bone and Endocrine Research Group, Glasgow, United Kingdom; 2Royal Hospital for Sick Children, Department of Paediatric Gastroenterology, Glasgow, United Kingdom

Background: Abnormal bone health is an ongoing concern in children, particularly adolescents, with Inflammatory Bowel Disease (IBD).
Aims: To assess bone health using dual energy x-ray absorptiometry (DXA) and peripheral quantitative computed tomography (pQCT) in children with IBD.

Patients: 43 (m:21) with Crohn disease (CD) and 18 (m:12) with ulcerative colitis (UC) with a median age of 13.8 (range, 10.0-16.6) and 13.3 (10.6-15.1) yrs, respectively.

Methods: DXA of lumbar spine (LS2-4) and total body (TB) for assessing bone mineral content (BMC) and corrected for bone area (BA) and sex and presented as BA SDS (BMBCSDS). pQCT of the forearm was performed with results expressed as sex- and age-specific z-scores for trabecular (Trb) and cortical (Crt) BMD and height corrected z-scores for total bone cross-sectional area (bCSA) and muscle (mCSA).

Results: Median height SDS was -0.16 (-2.62, 2.12) and 0.25 (-1.82, 2.73) and BMI SDS was 0.02 (-1.75, 2.84) and 0.61 (1.94, 3.23) for CD and UC respectively. Median BMC SDS for TB was -0.2 (-0.7, 0.8) and -0.05 (-0.7, 0.8) for CD and UC respectively. mCSA was -1.8 (-4.3, 0.2) and -1.8 (-3.2, 0.4) and bCSA was -1.65 (-5.7, 1.1) and -1.6 (-4.3, 0.4) in CD and UC, respectively. bCSA: mCSA ratio was -0.1 (-4.7, 3.9) and 0.1 (-3.2, 1.2) in CD and UC, respectively and not statistically significantly different. 0 age in the whole group was associated negatively with BMD(Br-c, 0.3, p=0.01) and Trb(Br-c, 0.3, p=0.01) and positively with bCSA: mCSA ratio (0.5, p=0.0001). Delayed bone age in the whole group was associated negatively with BMD(Br-c, 0.4, p=0.002) and BMDLS(r-0.5, p=0.001).

Conclusion: Bone health is adversely affected in CD and the effect may be more pronounced in the adolescent who is older and has delayed skeletal maturation.

Bone health in children and adolescents with inflammatory bowel disease

Saija Leasko; Helena Vaata; Matti Verkasalo; Heli Viljakkainen; Outi Makitie
University of Helsinki, Hospital for Children and Adolescents, Helsinki, Finland

Background: Children with chronic illness are at risk of developing secondary osteoporosis. Low bone mineral density (BMD) has been reported in children with inflammatory bowel disease (IBD).

Objective and hypotheses: The aim of this study was to evaluate prevalence of impaired bone health in children and adolescents with IBD.

Methods: We performed a cross-sectional study of bone health in a cohort of 80 children and adolescents with IBD. Control subjects were 80 age- and gender-matched healthy school children and adolescents. Dual-energy X-ray absorptiometry was performed to assess BMD and vertebral body morphology. BMD values were correlated with disease history, biochemistry, and medications.

Results: Of the 80 subjects (37 males) with IBD 51 (64%) had ulcerative colitis, 26 (32%) had Crohn’s disease, and 3 (4%) had unspecified colitis. Median age of the patients at study assessment was 14.9 yrs (range 5.1 to 20.1 yrs) and median disease duration was 4.3 yrs (range 0.3 to 14.5 yrs). The median bone age adjusted lumbar spine, hip and whole body BMD Z-scores were -0.6, -0.2 and -0.3, and the values were < -2.0 in 8%, 4%, and 6% of the patients, respectively. Healthy control subjects had higher lumbar spine and whole body BMD Z-scores (median for both < 0.1, p < 0.001), while the difference in total hip BMD Z-scores was not significant (median 0.01, p = 0.07). Abnormal vertebral morphology was observed in 7 patients with ulcerative colitis (14%), 4 patients with Crohn’s disease (15%), and in 2 patients with unspecified colitis. Thus altogether 17% of the patients had compression fractures, whereas abnormal vertebral morphology was found only in 4% of the control subjects. Compression fractures did not associate with BMD values.
Conclusions: The high prevalence of abnormal vertebral morphology emphasizes the importance of careful evaluation of bone health in children and adolescents with IBD.

P2-d2-477 Bone, Growth Plate and Mineral Metabolism 2
McCune-Albright syndrome: investigation of negative effects of bisphosphonates on jaws
Daniele Tarsissi1; Patrizia Matarazzo1; Alessandro Musa1; Gerd Tull1; Francesca Verna1; Massimiliano Tordella1; Alessandra Cimr1; Patrizia Detabian1; Roberto Lala1
1University of Turin, Pediatric Endocrinology and Diabetology, Torino, Italy; 2University of Turin, Dental School, Torino, Italy

Background: Osteonecrosis of jaws (ONJ) is a recently described adverse side effect of bisphosphonate therapy, but it has not been studied in McCune-Albright syndrome (MAS).

Objective and hypotheses: The aim of study is to search clinical and radiological ONJ signs, as a possible consequence of intravenous pamidronate treatment, in MAS bone fibrous dysplasia (BFD).

Population and MAS patients (4 males and 6 females) with craniofacial and/or long bone BFD, mean age at the evaluation 20 years and 4 months (range 7-27 years). Patients during pediatric age have been treated with pamidronate at dosage of 1mg/kg/day for 3 consecutive days at 4-6 months interval, over a period superior to 30 months. All patients are analyzed by medical anamnesis, clinical and oral evaluation. During the examination set of teeth, pain, soft-tissue swelling and infection, loosening of teeth, caries, fistula and exposed bone were evaluated. Panoramic radiograph and TC are estimated.

Results: In all patients there were no cases of osteonecrosis of the jaw. Furthermore no patients show alteration of dental eruption or only two patients show caries. No one show loosening of teeth, dental mobility or periodontal diseases. No one show fistula and exposed bone and nobody has radiological signs different from BFD of jaws (these are radioolucent cystic areas, endosteal scalloping and thinning of the cortex). The major phenotype expression of the syndrome is malocclusion (4 patients).

Conclusions: Our experience, although the number of patients is low, rule out the ONJ, as a chronic effect of bisphosphonates treatment, in this MAS population. Good oral hygiene, absence of corticosteroid-therapy or chemotherapy, pediatric age and patient’s compliance to follow-up could represent protective factors: so a multidisciplinary approach between pediatric endocrinologists and dentists is desirable.

P2-d2-478 Bone, Growth Plate and Mineral Metabolism 2
Bone mineralization in children with motor disabilities: determination of bone mineral density in the legs can be a useful tool
Hamiton Cassinelli1; Marina Troiano1; Rodolfo Rey; Ignacio Bergadá1
Centro de Investigaciones Endocrinologicas, Division de Endocrinologia, Hospital de Niños Ricardo Gutierrez, Buenos Aires, Argentina

Background: Children with motor disabilities often suffer from serious locomotor disability, osteoporosis and pathologic fractures. However little is known about their bone mineral density (BMD) and its relationship to fracture risk. The current definition for osteoporosis in children includes a BMD Z-score less than –2.0 adjusted for age, gender, and body size plus a clinically significant history of fractures. Peripheral quantitative computed tomography (pQCT) provides true volumetric BMD. However, the use of pQCT is not yet validated in children with motor disabilities. Dual-emission X-ray absorptiometry (DXA) is the most widely used method for assessment of BMD.

Objective and hypotheses: To assess leg and total BMD in children with motor disabilities by measuring BMD by DXA, in order to detect potential risk zones (legs) for osteoporosis and fractures.

Methods: We evaluated 11 children (8 females) with motor disabilities (7 myelomeningocele, 3 cerebral palsy, 1 Duchenne disease –Wheelchair bearing), with median age 9.3 years old. Total body and leg BMD was measured by DXA using a Lunar DPX-L machine. BMD values from patients were compared with those obtained in 91 normal individuals (45 females) that were classified according to sex, age (5-6, 7-9, 10-12, 13-15, 16-18 years) and Tanner stage. Results are expressed as mean ± SD. Statistical analysis was performed using one sample T test compared to a theoretical mean of 0.

Results: Patients showed a normal Total Body BMD (-0.2±1.4), and only two of them had BMD less than -2 SD. On the other hand, when legs mineralization was compared to normal values, they showed a significant reduction: -2.8±2.7 SD, p 0.006. Seven out of 12 patients (58%) had BMD less than -2.0 SD.

Conclusion: Children with motor disabilities from lower extremities may have less mineralization in their legs. The determination of bone mineral density in the legs can be a useful tool to identify those patients at greatest risk of suffering multiple fractures in their legs. A larger study is necessary to corroborate these results.

P2-d2-479 Bone, Growth Plate and Mineral Metabolism 2
Idiopathic juvenile osteoporosis and vitamin D insufficiency
Evelina Maines1; Grazia Morandi; Elena Monti; Francesco Doro; Paolo Cavarzere; Rossella Gaudino; Silvana Laurioli; Franco Antoniazzi
University of Verona, Department of Life and Reproduction Sciences - Paediatric Section, Verona, Italy

Background: Idiopathic juvenile osteoporosis (JIO) is a transient, primary, nonhereditary form of osteoporosis of unknown aetiology, characterized by spontaneous remission with progression of puberty. Good levels of vitamin D are essential for bone growth and mineralization since vitamin D insufficiency can lead to bone loss, secondary hyperparathyroidism and increased risk of fractures. In the literature few studies have considered the prevalence of vitamin D insufficiency in osteopaenic-osteoporotic children or adolescents and none in patients affected by JIO.

Objective and hypotheses: The purpose of this work is to evaluate the prevalence of vitamin D insufficiency in the patients affected by JIO followed in our Clinic and the relationship between serum 25-hydroxyvitamin D (25OHD) levels and bone parameters.

Methods: We selected 12 Caucasian paediatric patients (9 males and 3 females; mean age 12.63 ± 4.3 years and 12.78 ± 3.9 years respectively) affected by JIO. All had a clinically significant fracture history and a lumbar BMD Z-score adjusted for age, gender and body size, lower than -2.0 (mean Z-score -2.45 ± 0.9 SD). Pubertal stage, serum 25OH D levels, parathyroid hormone and other bone markers, as well as bone mineral density, were obtained.

Results: Vitamin D insufficiency was observed in the majority of patients (autumn: mean 27.68 ng/ml ± 11.8 SD; spring: mean 29.73 ng/ml ± 16.7 SD; summer mean 37.7 ng/ml ± 0 SD). CTX levels are increased (mean 1.43 ng/ml ± 0.5 SD) compared to normal values (0.15-0.45 ng/ml). Total lumbar BMDs are significantly correlated with 25OHD levels (p<0.02) as well as Lumbar BMD Z scores correlate with 25OHD levels.

Conclusions: We reckon that it is important to dose serum 25OHD, especially in patients with JIO; in fact, since lumbar BMDs seem to be directly correlated with vitamin D levels, and that there is evidence of vitamin D insufficiency, we believe that calcium and vitamin D suppletiments are potential preventive measures that must be used before any further treatment in case of osteopaenia or osteoporosis.

P2-d3-480 Bone, Growth Plate and Mineral Metabolism 3
The role of growth hormone in bone maturation evaluation by hand x-ray
Lee Even1; Ze’ev Hochberg2
1Western Galilee Hospital, Pediatrics, Naharia, Israel; 2Rambam Medical Center, Endopediatrics, Haifa, Israel

Background: The process of growth and maturation of the long bones (Radius Ulna, RU) and short bones (phalanges and metacarpals, S) of the hand (enchondroplasia and ossification) differs from the carpal bones (chondral ossification, C).

Objective and hypotheses: We aimed to determine role of GH in the two processes.

Methods: Bone age x-ray (BA) was performed in 12 children with severe GHD, 3 children with Laron syndrome and in 19 ISS children during 3 y of hGH treatment and 12 untreated ISS children, who were followed in parallel. The ISS children age 5.3 ±0.8 (m±sd). Individual bones were evaluated by a single blinded observer according to Greulich and Pyle, and are expressed as
Methods: In GHD, maturation was delayed by 3.1±0.9, 4.5±1.2 and 2.9±0.9 ‘y’ for RU, C and S bones, resp. In Laron syndrome, maturation was delayed by 0.75±0.1, 4.7±0.2 and 0.9±0.1 ‘y’ for RU, C and S bones, resp. In ISS over 3 ‘y’ of GH treatment, RU advanced by a mean 3.5±0.4 ‘y’, as compared with untreated 3.3±0.7 ‘y’ (p<0.10), C advanced by a mean 4.2±0.7 ‘y’ on hGH and 3.3±0.6 ‘y’, in control (p<0.001), and S bones by a mean 3.5±0.9 ‘y’, on hGH and 3.3±0.7 ‘y’ in control (p<0.058). The advanced in the C bones is statistical significant.

Conclusions: These results suggest that GH strongly regulates and GHD interferes with bone maturation by inhibiting chondral osteogenesis (seen in the carpal bones), and less so through delayed enchondroplasia, observed by RU and S maturation. These profiles help in the diagnosis of GHD.

Background: Kearns-Sayre syndrome (KSS) is a mitochondrial disease with a classic triad of signs: onset before 20 yrs of life, progressive extracocular muscle ophthalmoplegia, retinal pigment degeneration concomitant with endocrine, cardiologic, muscular and neurological abnormalities.

Objective: A case report of a girl with KSS.

Methods: Hormonal tests, ophthalmological and audiological examinations, imaging pictures, mitochondrial DNA analysis.

Results: A 16 year old girl was admitted to Department of Endocrinology when 11 years old due to growth deficit (˜2.5 SD). Upon physical examination, she presented with dusky skin, left eyelid ptosis, micrognathia, bilateral hearing loss and was prepubertal. The diagnosis was primary hypoparathyroidism, primary subclinical hypothyroidism, GH deficiency and diabetes type was normal (female). Ophthalmology showed retinal pigment degeneration. An audiogram confirmed moderate reception-type hearing loss. Her karyotype was normal (female). Ophthalmology showed retinal pigment degeneration. An audiogram confirmed moderate reception-type hearing loss. Head CT showed areas of high density in the hypothalamus; there was no contrast uptake of pituitary in MRI. She was administered oral calcium, active vitamin D3, levothyroxine, diabetes diet, then insulin therapy and she was fitted with a hearing aid. When 15 years old, she was admitted to the Department with diabetic ketoacidosis. Following normalization of water-electrolyte and metabolic parameters and bone geometry with the disease severity, fracture history or number of bleedings.

Results: Boys with haemophilia had a decreased trabecular volumetric BMD (mean Z-score -0.5, p<0.01), while their cortical volumetric BMD was normal (mean Z-score 0.4, p<0.05). The volumetric bone mineral content and the bone geometry at the radial diaphysis were normal when adjusted for patients’ shorter body height. Muscle area was decreased (mean Z-score -1.0, p<0.001), irrespective of age. No association was observed of bone quality parameters and bone geometry with the disease severity, fracture history or number of bleedings.

Conclusion: Bone strength measured at the diaphysis of the radius is not impaired in boys with haemophilia. The finding of the decreased trabecular bone density can be most likely attributed to their sarcopenia.

Background: Osteonecrosis (ON) is a severe complication of Acute Lymphoblastic Leukemia (ALL) therapy. Risk factors have been identified: age ≥ 11 yrs, female gender, high BMI and increased dose of corticoids. ON diagnosis leads to discontinuation of corticoid therapy, support care, in some cases surgery and more recently the use of bisphosphonates.

Objective and hypotheses: The aim of our study was to describe the beneficial effects of bisphosphonates on pain and motor function in children who developed ON as complication of ALL therapy.

Methods: Symptomatic ON was diagnosed in 23 of 242 children treated for ALL at our center between April 2000 and September 2008. ON diagnoses were made by MRI. 11/23 patients were treated with IV pamidronate. Clinical
P2-d3-484 Bone, Growth Plate and Mineral Metabolism 3

Pseudohypoparathyroidism and mild AHO features in two siblings with methylation defects in exon a/b of the GNAS locus

Dorothee Schmidt; Bettina Brix; Susanne Thiele; Ulla Doehnert; Ralf Werner; Olaf Hört
University of Luebeck, Department of Pediatrics and Adolescent Medicine, Luebeck, Germany

Background: Pseudohypoparathyroidism (PHP) describes several disease entities with diminished or absent sensitivity to parathyroid hormone. PHP type Ic (PHPic) is defined as the combination of clinical signs of Albright hereditary osteodystrophy (AHO) with normal Gsalpha activity. The pathomechanism in most cases is not known yet. PHPIb patients lack AHO signs and this subtype is caused by methylation changes of the GNAS gene locus. Here we present two siblings with PHP and mild AHO features.

Patients and methods: Patient 1 (P1) was 7 years old at presentation with a height of 129.4 cm (P97.4), weight 34.6 kg (P97.8), BMI 20.6 kg/m² (P97.8). The stature appeared stubby with short hands without brachymetacarpia. Initially, the patient presented with rickets (Ca 1.99 mmol/l, P 2.13 mmol/l, PTH 213.6 pg/ml, 25-OHD 7.4 ng/ml). After therapy of the vitamin D-deficiency PTH stayed elevated. Gsalpha activity was normal in erythrocyte membranes. Patient 2 (P2, little sister) was 5 years old and had a height of 123.5 cm (P99.7) and a weight of 43.1 kg (P100) and BMI 28.62 kg/m² (P100, +5.57 SDS). The same clinical features. Beside she was diagnosed with autism and primary hypothyroidism.

Results: Ca 2.2 mmol/l, P 2.17 mmol/l, PTH 39 pg/ml, 25-OHD 10.7 ng/ml. 3 month later we found massively elevated of PTH (404 pg/ml), Ca and 25-OHD were within the reference range. Molecular genetic analysis of the coding region of both patients in exons 2-13 of GNAS from blood leukocyte derived DNA revealed no sequence variation. Pyrosequencing analysis of bisulfit converted DNA derived from blood leukocytes revealed a loss of methylation in exon A/B of the GNAS locus in both children.

Conclusions: The phenotype of PHP can be highly variable. Methylation defects may explain also hereditary cases. A new classification of the patients based on clinical, biochemical, and molecular data is desperately needed.
in the lower normal range for healthy children. After 3 months of vitamin D supplementation we observed the increase in 25-OHD levels, a drop of PTH levels and an important reduction of pain intensity. Interventions: We subdivided our patients in groups according to their age and their growth velocity for that age; we concluded that since most children had an age, in which growth is low, it could be not appropriate the term “growing pains” used so far. On the other hand, since most children (all with QUS bone density at the lower values of normal range) showed increased bone metabolism markers and pains reduced significantly after the three months of vitamin D supplementation, we reckon that it could be useful to consider vitamin D deficiency in children with musculoskeletal pain.

**P2-d3-487 Bone, Growth Plate and Mineral Metabolism 3**

**Low-dose, single day pamidronat treatment in osteogenesis imperfecta**

**Ahmet Uçakturk; Cengiz Kara; Gunindii Figen Murat Aydin**

Ondokumayis University Faculty of Medicine, Department of Pediatric Endocrinology, Samsun, Turkey

**Background:** Bisphosphonates are potent inhibitors of bone resorption, and they have been reported to have beneficial effects in children with osteogenesis imperfect (OI). Pamidronate has been given in 3-day cycles, every 2 to 4 months depending of the age of the patients. In this schema, the annual dose of pamidronate is 9 mg/kg per year.

**Objectives:** To evaluate the efficacy and safety of low dose, single day infusion of pamidronate in children with OI.

**Patients and methods:** Intravenous pamidronate was administered to eight patients (from 1 month to 8, 6 years old) with severe OI, in a 1,5 mg/kg single dose, at 3-month intervals, one year duration. All patients received 3-day supplementation of calcium after pamidronate infusion. The patients were assessed respecting clinical and laboratory findings at baseline and before each cycle. Bone mineral density (BMD) and fracture rates at pre-treatment were compared to that at end of the first year of therapy.

**Results:** Pain decreased after the first infusion cycle. The median of fracture incidence decreased ($P = 0.027$). Ambulation scores increased all children (Table 1). Alkaline phosphatase decreased by 23%. BMD of the spine increased by a median of 14% ($P = 0.018$). Z-scores increased from a median of -4.4 to -3.1 ($P = 0.028$). Height SD score was not changed. No adverse effects were seen with pamidronate infusions apart from fever during the first infusion cycle.

**Conclusions:** In our country the form of Aredia has only 90 milligrams flacon and it should be used within 24 hours after diluted. Low dose, single day pamidronate therapy with 3-month intervals decreased frequency of fractures and increased bone mineral density without significant adverse events. We think that one-day treatment is far more acceptable than the three consecutive days as used in other protocols. Further investigation for optimal dose of treatment is indicated.

**Clinical and Biochemical Characteristics of the Patients at the Onset and at the End of Pamidronate Therapy**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>At onset</th>
<th>At end of the first year</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Bone pain (n)</strong></td>
<td>8</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td><strong>Fracture rate (n)</strong></td>
<td>2 (1-22)</td>
<td>1 (0-3)</td>
<td>0.027</td>
</tr>
<tr>
<td><strong>Height SDS</strong></td>
<td>-4.9 (-6.78 - -0.8)</td>
<td>-4.65 (-6.7 - -0.5)</td>
<td>0.159</td>
</tr>
<tr>
<td><strong>Ambulation score</strong></td>
<td>0 (0-2)</td>
<td>2 (1-3)</td>
<td>0.016</td>
</tr>
<tr>
<td><strong>Ca (mg/dl)</strong></td>
<td>9.9 (9.4-10.2)</td>
<td>9.5 (9.1-10.1)</td>
<td></td>
</tr>
<tr>
<td><strong>ALP (IU/L)</strong></td>
<td>576 (444-771)</td>
<td>443 (120-737)</td>
<td></td>
</tr>
<tr>
<td><strong>L2-L4 BMD (g/cm2)</strong></td>
<td>0.279 (0.246-0.414)</td>
<td>0.319 (0.286-0.490)</td>
<td>0.018</td>
</tr>
<tr>
<td><strong>BMD Z Score</strong></td>
<td>-4.4 (-5.9 - -3.2)</td>
<td>-3.1 (-4.6 - -2.7)</td>
<td>0.028</td>
</tr>
</tbody>
</table>

**P2-d3-488 Bone, Growth Plate and Mineral Metabolism 3**

**Correlation of bone mineral density measurements with GE Lunar iDXA and Lunar Prodigy in children**

**Joachim Peltz; Oliver Semler; Angelika Stabrey; Eckhard Schoenau**

University children's hospital, Endocrinology/Osteology, Cologne, Germany

**Background:** The new GE Lunar iDXA has a higher resolution and higher precision in bone mineral density measurements compared to the GE Lunar Prodigy. Previous studies showed a good correlation between the results of these devices in adults.

**Objective and hypotheses:** The objective is to investigate if there is a significant correlation between measurements with GE Lunar iDXA and GE Lunar Prodigy in children.

**Methods:** We studied 26 patients (9 girls), with bone relevant diagnosis (e.g. osteogenesis imperfecta, cerebral palsy) median age 12.2 years (5.3 - 15.7) and did whole body and spine ap measurements. Those results were correlated regarding bone mineral density (BMD) and bone mineral content (BMC).

**Results:** For whole body BMD and lumbar spine BMD results see table below. Also the analysis of bone mineral content of whole body ($r = 0.988$, $p < 0.0001$) and spine ap ($r = 0.997$, $p < 0.0001$) showed a highly significant correlation, as well as analysis of whole body area ($r = 0.948$, $p < 0.0001$) and leg BMC ($r = 0.993$, $p < 0.0001$) and leg muscle mass ($r = 0.986$, $p < 0.0001$).

**Conclusions:** GE Lunar iDXA and Lunar Prodigy showed a highly significant correlation in all measurements in children. Therefore measurements of patients and reference data performed with GE Lunar Prodigy, can be used and compared with measurements done with GE Lunar iDXA.

**P2-d1-489 Diabetes and Insulin 1**

**MODY 5 diabetes, study of 4 patients**

**Diana Barreto Campos; Elba Campos Reyes; Diego Yeste Fernandez; Miguel Gussinye; Maria Clemente; Maria Angeles Albisu; Antonio Carrascosa Lezcano**

Hospital Vall de Hebron, Unidad de Endocrinologia Pediatrica, Barcelona, Spain

**Introduction:** MODY5 is a monogenic autosomal dominant type of diabetes, characterised by mutations in the regulating gene of hepatocyte nuclear factor-1B (HNF1B). Insulin secretion deficiency may be accompanied by structural and functional renal (96%), liver (19.4%) and genital (13.3%) anomalies, among others. MODY5 represents 0.6 - 1% of all MODY diabetes.

**Patients:** Retrospective analysis of clinical characteristics at diagnosis and their clinical evolution in patients with genetically-confirmed HNF1B mutation. Mean age: 17.9 ± 4 yrs (range: 13-23 yrs). Three patients were male and 1 female. Diagnosis was established by the presence of hyperglycaemia without ketoadiposis (116-378 mg/dl), (HbA1c %: 5.8-14.5) and renal cystic disease in 3 patients, 2 of whom also presented a moderate rise in liver enzymes. Molecular genetic study revealed common factor-1β (HNF1B). Insulin secretion deficiency may be accompanied by structural and functional renal (96%), liver (19.4%) and genital (13.3%) anomalies, among others. MODY5 represents 0.6 - 1% of all MODY diabetes.

**Patients:** Retrospective analysis of clinical characteristics at diagnosis and their clinical evolution in patients with genetically-confirmed HNF1B mutation. Mean age: 17.9 ± 4 yrs (range: 13-23 yrs). Three patients were male and 1 female. Diagnosis was established by the presence of hyperglycaemia without ketoadiposis (116-378 mg/dl), (HbA1c %: 5.8-14.5) and renal cystic disease in 3 patients, 2 of whom also presented a moderate rise in liver enzymes. Molecular genetic study revealed common factor-1β (HNF1B). Insulin secretion deficiency may be accompanied by structural and functional renal (96%), liver (19.4%) and genital (13.3%) anomalies, among others. MODY5 represents 0.6 - 1% of all MODY diabetes.

**Patients:** Retrospective analysis of clinical characteristics at diagnosis and their clinical evolution in patients with genetically-confirmed HNF1B mutation. Mean age: 17.9 ± 4 yrs (range: 13-23 yrs). Three patients were male and 1 female. Diagnosis was established by the presence of hyperglycaemia without ketoadiposis (116-378 mg/dl), (HbA1c %: 5.8-14.5) and renal cystic disease in 3 patients, 2 of whom also presented a moderate rise in liver enzymes. Molecular genetic study revealed common factor-1β (HNF1B). Insulin secretion deficiency may be accompanied by structural and functional renal (96%), liver (19.4%) and genital (13.3%) anomalies, among others. MODY5 represents 0.6 - 1% of all MODY diabetes.
doses of 1.05 IU/kg/day and 1.45 IU/kg/day, respectively, with a progressive decrease in these requirements in the following weeks. One required insulin treatment 4 years after diagnosis at a dose of 0.3 IU/kg/day. Another still has not required insulin treatment. No other alterations associated with this entity such as facial dysmorphism, genital and thyroid anomalies were present.

**Conclusions:** Study of HNF1B gene should be considered in all patients with hyperglycaemia, family history of type 2 diabetes and renal anomalies (structural or functional). All mutations in the 4 patients studied were the novo.

**P2-d1-490** *Diabetes and Insulin 1*

**Acute onset and complete recovery of retinopathy in a type 1 diabetic adolescent with chronic myeloid leukaemia**

_Silvia Schmidt; Mariarosaria Lang-Muritano; Daniel Konrad; Eugen Schoenle_

University Children's Hospital Zurich, Switzerland, Endocrinology and Diabetology, Zurich, Switzerland

**Background:** The onset of diabetic retinopathy correlates with the long-term quality of glycemic control. Until now there has been no report of retinopathy morphologically completely similar to diabetic retinopathy being induced by chronic myeloid leukaemia (CML).

**Case report:** A 17 years old adolescent with type 1 diabetes presented unexpectedly with acute nonproliferative retinopathy despite good glycemic control. He was suffering from diabetes for 9 years and his average HbA1c value was 7.7% (normal range < 6%, DCA 2000) over the past 5 years. Ophthalmologic examinations were done yearly. Surprisingly, severe nonproliferative diabetic retinopathy was detected at the age of 17 years. The findings were confirmed by fluorescein angiography revealing severe nonproliferative diabetic retinopathy with bilateral macular oedema and haemorrhages. A panretinal photoacoagulation was judged as not yet necessary. Other possible causes of retinopathy like hypertension and thrombophilia could be excluded. Two months later, CML was diagnosed (white blood cell count 1166 G/l (4-16)), haematocrit 39.5% (40-55), haemoglobin 13.2g/l). Chemotherapy was started and after a few weeks the patient was in remission. During this time metabolic control of the diabetes remained good (average HbA1c 7.4%) (Figure 1). Retinopathy resolved completely within 8 months.

**Conclusions:** To our knowledge, this is the first case of CML-triggered retinopathy in a well controlled diabetic adolescent. Haematological disorders have been described to be a cause of rapid progressive retinopathy in patients with diabetes. In case of unexpected retinopathy in patients with type 1 diabetes other potential causes of retinopathy should be considered.

**P2-d1-491** *Diabetes and Insulin 1*

**A novel dominant SUR1 gene mutation causing childhood autosomal diabetes in two siblings**

_Bulent Hacihambaloglu; Merih Berberoglu; Zeynep Siklar; Senay Savas Erdeve; Sian Elwood; Gonul Ocal_

*Ankara University School of Medicine, Pediatric Endocrinology, Ankara, Turkey; 2Peninsula Medical School, Clinical Molecular Genetist, Plymouth, United Kingdom*

**Background:** Increased risk of diabetes later in life in patients with dominant KATP mutations is well defined in the literature. Generally the diagnosis of diabetes mellitus is made in adulthood, although a few patients with heterozygous ABCC8 mutations and diabetes during early adolescent period have been reported.

**Objective and hypotheses:** We describe two siblings with novel heterozygous ABCC8 mutations who developed diabetes at very early ages.

**Methods:** Two siblings were born to non-consanguineous parents. Both were macrosomic and evaluated for hypoglycemia after macrosomic delivery who develop hypoinsulinemic diabetes during adolescence or adulthood, especially if there is a dominant history of diabetes. Endocrine abnormalities in patients with ABCC8 gene heterozygous mutations can be variable, even in the same family, from severe or moderate hypoglycemia to overt hyperglycemia.

**Results:** In patients with heterozygote ABCC8 gene mutations, diabetes can begin as early as 10 years.

**Conclusions:** Dominant KATP mutations should be kept in mind as a differential diagnosis in patients with diazoxide responsive hyperinsulinemic hypoglycemia after macrosomic delivery who develop hyperinsulinemic diabetes during adolescence or adulthood, especially if there is a dominant history of diabetes. Endocrine abnormalities in patients with ABCC8 gene heterozygous mutations can be variable, even in the same family, from severe or moderate hypoglycemia to overt hyperglycemia.

**P2-d1-492** *Diabetes and Insulin 1*

**Lipid profiles 3-5 years after onset of type 1 diabetes mellitus in Danish children correlate with BMI-SDS, HbA1c and gender**

_Jesper S. Sorensen1; Kurt Kristensen1; Gerd Sandgren1; Birgitte Hertz2; Keld Gadé2; Flemming Pocoot3; Niels H. Birkebaek3_

1Aarhus University Hospital, Skejby, Department of Pediatrics, Aarhus N, Denmark; 2Nykobing Falster Hospital, Department of Pediatrics, Nykobing Falster, Denmark; 3Viborg Hospital, Department of Pediatrics, Viborg, Denmark; 4Roskilde Hospital, Department of Pediatrics, Roskilde, Denmark; 5Glostrup Hospital, Research Institute, Dept. of Clinical Experimental Research, Glostrup, Denmark

**Background:** Patients with type 1 diabetes mellitus (T1DM) have a higher risk for developing cardiovascular disease compared to the background population. This may partly be explained by unfavourable lipid profiles.

**Objective:** To determine and describe lipid profiles in T1DM children and healthy controls, and the correlation of lipids with BMI-SDS, HbA1c and gender.

**Methods:** A total of 340 children/adolescents (171 males) with a median age of 13.4 years (range 4.8-18.8) and T1DM for 3-5 years were enrolled; 70 healthy children/adolescents (30 males), with a median age of 12.1 years (range 7.4-16.9), served as controls. The study variables included fasting blood samples for total cholesterol (TC), high-density lipoprotein cholesterol (HDL), low-density lipoprotein cholesterol (LDL), triglyceride (TG), height, weight, body mass index standard deviation score (BMI-SDS), HbA1c and gender. Overweight was defined as BMI-SDS ≥ 1.1 and obesity as BMI-SDS ≥ 2.

**Results:** Significantly more patients than controls had reduced HDL (p=0.05) and elevated TC (p<0.01). 129 patients and 11 controls were overweight (p=0.01), and 30 patients and 2 controls were obese (p=0.06). The group of patients with overweight and obesity had higher TC (p=0.05), higher LDL (p=0.01), higher TG (p=0.02) and lower HDL (p=0.05) compared to normal weight patients. Patients with HbA1c less than 8% had significantly lower TC and TG compared to patients with HbA1c more than 8%, (p<0.05) and (p=0.01), respectively. Boys with T1DM had higher TC (p=0.01) and LDL (p=0.01) compared to girls with T1DM.

**Conclusions:** Overweight and obesity are highly prevalent in Danish children and adolescents with T1DM compared with healthy children. Overweight, increased HbA1c and male gender seem to be risk factors for unfavourable lipid profiles in children and adolescents with T1DM.
Background: In the UK at present, different centres use different insulin regimens for children and young people at diagnosis of diabetes. Some centres start all children on basal bolus or multiple dose injection (MDI) regimens, whereas others still use twice daily (BD) mixed insulins in certain age groups, partly because of difficulties in supporting insulin administration in school. Since 2008, all children newly diagnosed with diabetes attending Nottingham Children’s Hospital have been started on a MDI regimen, regardless of age. Since changing, there has been a perception that we are getting better honey-

Prag 1AICs and the honeymoon period lasts longer when BD regi-
mens were used. This is despite no other changes in diabetes management, education or team occurring.

Methods: A retrospective analysis of HbA1C and duration of honeymoon period in children started on MDI regimens compared with those started on BD regimens, looking at all children and young people registered with the Nottingham Paediatric Diabetes Service in Dec 2010. Honeymoon was de-

Prined using Insulin Dose Adjusted for A1C ie HbA1C + (4 x total daily dose

in units/kg/day) ≤ 9.

Results: 104 complete records were analysed for children and young people

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.

PPPmence with BD regimens, looking at all children and young people.
as its clinical presentation and follow-up, its determinants and its association with thyroid autoimmunity (anti-Tg, anti-TPO) and autoimmunity against pancreatic b-cells (anti-GAD).

Methods: The study included 98 children and adolescents with T1DM, aged (mean±SD) 12.8±4.7yrs with a T1DM duration of 4.6 ±3.8yrs.

Results: The prevalence of APCA was 8.2% (n=8), anti-thyroid antibodies (anti-Tg, anti-TPO) 22.4% (n=22), while the prevalence of anti-GAD was 50.0% (n=49). The majority of APCA (+) patients (5/8) were positive for anti-thyroid antibodies (62.5% vs18.9%, p=0.012) and for anti-GAD (62.5% vs 48.8%, p=0.223). Univariate regression analysis indicated that the presence of gastric and thyroid autoimmunity (thyrogastic autoimmunity) was associated with older patient age and longer T1DM duration. Three years later 82/98 (83.7%) patients, among which 6/8 APCA positive, were re-evaluated in terms of APCA. From these 6 patients, 3/6 became APCA(+) at re-evaluation, while 3/6 developed high titres of APCA. These latter three patients were anti-GAD(+), while 2/3 had thyroid autoimmunity, none had pernicious or iron-deficiency anemia and 2/3 had mild symptoms of gastritis.

One of them underwent gastroscopy, had histological findings typical of H. Pylori gastritis and received treatment for its irradiation.

Conclusions: The prevalence of gastric autoimmunity among T1DM children and adolescents was 8.2% and its occurrence was associated with older age, longer diabetes duration and with the presence of thyroid autoimmunity. Thus, patients with T1DM, especially adolescents with thyroid autoimmunity, should have periodic autoantibody screening for the early diagnosis and follow-up of gastric autoimmunity.

**P2-d1-497 Diabetes and Insulin 1**

**Phenotypical difference of HNF1alpha diabetes from other forms of diabetes in childhood**

Juliana Predescu1; Steven Ghanny2; Lina Nie3; Duanjun Tan3; Sheila Paree3; Sonal Bhandari2; Felicitas Lachbauer1; Amit Bhangoo1; Svetlana Ten2

1Infants and Childrens Hospital at Maimonides and SUNY Downstate, Pediatric Endocrinology, Brooklyn, United States; 2Infants and Childrens Hospital at Maimonides and SUNY Downstate, Pediatric Endocrinology, Brooklyn, United States; 3SUNY Downstate, Molecular Pathology, Brooklyn, United States

**Background:** Monogenic Diabetes (MD) is poorly recognized in childhood. The most common form of MD is caused by mutations in the HNF1A gene. Proper diagnosis is important, since it can change treatment and outcome.

**Objective and hypotheses:** To compare the clinical features of patients with DM caused by mutations in HNF1A to patients with T1DM, T1.5DM and T2DM.

**Methods:** We compared 11 patients with MD caused by mutations in the HNF1A gene to 120 patients with T1DM. We subdivided HNF1A group into 8 patients with the expected phenotype for MD and 3 with a more severe phenotype known as Oji Cree, with severe insulin resistance and morbid obesity.

Results: HNF1A group had earlier age of onset than in T2DM, but later than in T1DM. BMI in the HNF1A group was greater than in patients with T1DM, but less than that of patients in T2DM and T1.5DM. BMI in Oji Cree-like DM patients was greater than BMI in patients with T1DM, T1.5DM and T2DM.

Conclusions: Monogenic diabetes caused by mutations in HNF1A is prevalent in pediatric diabetes clinics. Proper diagnosis is important, since it can change treatment and outcome.
well controlled diabetics and healthy children, which might be risk factors for levels and ophthalmic artery resistivity indexes were increased compared to diabetics \( (p<0,05) \). The duration of diabetes didn’t affect the retinal blood control group, retinal blood flow velocities were similar in diabetics and non-

Conclusions:

Results:

Methods:

Objective and hypotheses:

Background:

Objective and hypotheses:

According to the results of the current study, TGF-β1 plays a role in the development of T1DM by inducing Th17 cells, leading to the destruction of insulin-producing β-cells, causing chronic hyperglycemia.

Finally, patients with severe disease had much higher levels of sLepR at both examinated stages.

Results:

Patients with T1DM were grouped as juvenile-onset T1DM and adult-onset T1DM compared to the controls and healthy controls.

Methods:

Objective and hypotheses:

Background:

Objective and hypotheses:

Comparison between sensor-augmented insulin therapy with either insulin pump (CSI1) or multiple daily injections (MDI) in everyday life: analysis of glucose variability and sensor reliability

Background:

Objective and hypotheses:

Therefore, patients may define the way to develop a characteristic profile of T1D progression.

Conclusions:

Analyze the metabolic profile of patients with T1D through the evaluation of a wide range of metabolic/ inflammatory parameters.

Results:

Then, patients with T1DM had higher sLepR levels than both controls and healthy children. Furthermore, there was a significant negative correlation between sLepR levels and disease duration in patients with T1DM. These findings suggest that high sLepR levels may be a marker of disease severity in patients with T1DM.

Conclusions:

Diabetes and Insulin 2

P2-d1-501 Diabetes and Insulin 2

Analysis of metabolic/inflammatory markers in children with T1D: possible predictive role of metabolic and inflammation of soluble leptin receptor (sLepR)

Rosa Nugnes1; Adriana Franzese1; Enza Mozzillo1; Mariateresa Falco1; Valentina Fattorussou2; Mario Galgani1; Marianna Santopao1; Salvatore De Simone1; Giuseppe Matarese1; Federico II University of Naples, Pediatrics, Naples, Italy; 2Federico II University of Naples, Cellular and Molecular Pathology, Naples, Italy; 3Consiglio Nazionale delle Ricerche (CNR) of Naples, Istituto di Endocrinologia ed Oncologia Sperimentale, Naples, Italy

Background:

Results:

Methods:

Objective and hypotheses:

Comparison between sensor-augmented insulin therapy with either insulin pump (CSI1) or multiple daily injections (MDI) in everyday life: analysis of glucose variability and sensor reliability

Stefano Zucchini; Mirella Scipione; Giulio Maltoni; Alessandra Rollo; Claudia Balsamo; Martina Zanotti; Alessandro Cicognani S.Orsola-Malpighi Hospital, Pediatrics, Bologna, Italy

Background:

Results:

Objective and hypotheses:

Comparison between sensor-augmented insulin therapy with either insulin pump (CSI1) or multiple daily injections (MDI) in everyday life: analysis of glucose variability and sensor reliability

Stefano Zucchini; Mirella Scipione; Giulio Maltoni; Alessandra Rollo; Claudia Balsamo; Martina Zanotti; Alessandro Cicognani S.Orsola-Malpighi Hospital, Pediatrics, Bologna, Italy

Background:

Diabetes and Insulin 2

P2-d1-502 Diabetes and Insulin 2

Comparison between sensor-augmented insulin therapy with either insulin pump (CSI1) or multiple daily injections (MDI) in everyday life: analysis of glucose variability and sensor reliability

Stefano Zucchini; Mirella Scipione; Giulio Maltoni; Alessandra Rollo; Claudia Balsamo; Martina Zanotti; Alessandro Cicognani S.Orsola-Malpighi Hospital, Pediatrics, Bologna, Italy

Background:

The debate on whether CSII is superior to MDI in the treatment of children with type-1 DM is still open. Some studies have demonstrated that sensor-augmented CSI1 therapy resulted in improved metabolic control when compared with MDI therapy.

Objective and hypotheses:

To compare in everyday life mean glucose and inter-intra day glucose variability using RT-CGMS in pts treated with CSI1 or
RT-CGMS specificity ranged from 86% to 98% (higher for 40-80 mg/dl values and 241-400 mg/dl values, lower for 121-240 mg/dl values), sensitivity ranged from 59% to 93% (higher for 240-400 mg/dl values, lower for 40-80 mg/dl values).

Conclusions: During a 4-day period sensor-augmented therapy with CSII seemed more effective than sensor-augmented MDI therapy, both in terms of glucose mean values and intraday variability. Interday variability was not compared.

Results: In the whole group of patients HbA1c correlated positively with mean glucose (r=0.38; p=0.003), AUC>180 (r=0.32; p=0.011), HBGI (r=0.34; p=0.008) and negatively with AUC<70 (r=0.32; p=0.013) and HBGI (r=0.40; p=0.002). The table shows the significant results.

<table>
<thead>
<tr>
<th></th>
<th>CSII (17 pts)</th>
<th>MDI (43 pts)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>mean_glucose</td>
<td>146.5±30.4</td>
<td>164.6±33.9</td>
<td>0.052</td>
</tr>
<tr>
<td>sd_glucose</td>
<td>44.6±10</td>
<td>56.8±13.6</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CV</td>
<td>30.8±5.5</td>
<td>35.3±8.5</td>
<td>0.02</td>
</tr>
<tr>
<td>conga2</td>
<td>49.0±15.2</td>
<td>58.4±14.4</td>
<td>0.036</td>
</tr>
<tr>
<td>conga2_day</td>
<td>53.6±16</td>
<td>64.9±16.4</td>
<td>0.02</td>
</tr>
<tr>
<td>conga2_night</td>
<td>46.7±32</td>
<td>68.4±41.8</td>
<td>0.037</td>
</tr>
<tr>
<td>HBGI</td>
<td>5.2±4.2</td>
<td>8.4±5.5</td>
<td>0.008</td>
</tr>
</tbody>
</table>

---

**P2-d1-504 Diabetes and Insulin 2**

**Growth patterns during puberty in relation to the development of microalbuminuria in young people with type 1 diabetes**

**Loredana M. Marcovecchio1; Neil R. Dalton2; David B. Dunger1**

1University of Cambridge, Department of Paediatrics, Cambridge, United Kingdom; 2King’s College London, Evelina Children’s Hospital, WellChild Laboratory, London, United Kingdom

**Background:** In adults with type 1 diabetes (T1D) short stature has been associated with cardiovascular disease and diabetic nephropathy. However, there are no data on the potential relationship between growth patterns during puberty and the development of diabetic vascular complications.

**Objective and hypotheses:** We tested the hypothesis as to whether pubertal growth is impaired in young people with T1D who develop microalbuminuria (MA).

**Methods:** Repeated height measurements performed during puberty were available for 201 young people (103 boys with childhood onset T1D followed in the Oxford Regional Prospective Study. Longitudinal data on albumin/creatinine ratios and HbA1c were also collected from the study participants. Height standard deviation scores (SDS) were compared between subjects who developed MA (MA+; n=66 (56% males); age at diagnosis: 7.4±3.2y) and normoalbuminuric subjects (MA-; n=135 (51% males); age at diagnosis: 7.4±2.9y).

**Results:** In the whole study population height SDS progressively declined during puberty, from 0.22±0.97 to 0.07±1.04, p=0.001. The decline in height SDS was significantly different between the MA+ and MA- groups (p=0.04) (Fig 1). In particular, height SDS was lower in the MA+ group at the age of 15-16yr: -0.11±1.02 vs 0.22±0.96, p=0.02, and at the age of 17-18yr: -0.17±1.06 vs 0.19±1.01, p=0.02. Within the MA+ group, mean height SDS after MA onset was significantly lower than before MA onset (-0.24±0.96 vs -0.03±0.94, p=0.001). Final height was inversely associated with MA (HR[95%CI]: 0.968[0.943-0.994] p=0.01), although this association was no longer significant after adjusting for HbA1c, which was higher in the MA+ group (10.5±1.7 vs 9.8±1.4%, p=0.003).

**Conclusions:** In this study we detected a significant impairment in growth during puberty in young people with T1D, particularly in those developing MA. A potential explanation for these findings could be the involvement of common factors in the pathogenesis of impaired growth and MA, such as genetic/environmental factors and/or poor glycaemic control.

---

**Background:** Monogenic diabetes is a type of diabetes with inheritance autosomal dominant.

**Objective and hypotheses:** Determine a mutation in a family with suspicion of monogenic diabetes.

**Methods:** A boy aged 12 years and 6 months and a girl aged 11 years and 2 months studied due to hyperglycemia under fasted conditions (103 mg/dl, 109 mg/dl respectively). Family history: Maternal grandparents, paternal grandfather, two paternal uncles, two maternal uncles, father and brother with Diabetes Mellitus type II. Personal history of both individuals: not relevant. BOY aged 12 6 years old. Weight 43.400 Kg. (P50), height 157 cm (P75), BMI: 17.6. Pubertal stage Tanner 2. GIRL 112 years old, Weight 35.7 Kg. (P10-25), size 143 cm (P10-25), BMI:17.5.

**Results:** Complementary examinations: Oral Glucose Tolerance Test: glycemia after 120 minutes, 7.2 mmol/L (131 mg/dl) and 5.9 mmol/L (109 mg/dl) and baseline figures of 4.56 and 4.51 mmol/L (83 and 82 mg/dl). Prediabetes Study (ketonuria and glycosuria negative, peptide C 1 ng/ml, Hb A1c 5.5 and baseline figures of 4.56 and 4.51 mmol/L (83 and 82 mg/dl). Genetic Study:Both siblings: mutation c.-137_-131dupTGGGGGT in heterozygosis in the promoter region of gene HNF 1 alpha. MODY 3. The father and elder brother present 2 mutations: in exon 2 of the GCK Y61X gene (Mody 2) and c.-154_-160dupTGGGGGT in heterozygosis in the promoter region of the gene HNF1a (Mody 3). Genetic study of the mother was normal. The 2nd brother had the family’s Mody 2 mutation, while the 3rd sister had the Mody 3 mutation.

**Conclusions:** It is very rare to share a mutation for Mody 2 and 3 in the same individual and family. The presence of one mutation does not rule out the other.
Results: In our center, 126 pediatric liver transplants were performed between 2001-2009, of which 4% (n=6) developed NODM. 4 of 6 patients were male. Their ages were between 12.6-15.6 years. The etiologies of the liver diseases in cases who developed NODM were Wilson disease (three cases), cryptogenic cirrhosis (one patient), autoimmune hepatitis (one patient), tyrosinemia type 1 (one patient). Two of 6 underwent primary cadaveric related liver transplantation. In 4 cases, hyperglycemia was detected in the first week of operation. In the others, hyperglycemia was detected during methylprednisolone bolus therapy for acute rejection at postoperative 25th and 44th days, respectively. HgA1c levels were normal except two. The doses of insulin infusion were varied between 0.1-0.5U/kg/h and SC insulin doses were between 0.4-3U/kg/day. The duration time of insulin therapy was changed between 1-10 months. Two patients died after one month of discontinued insulin therapy. Three patients were normoglycemic during 3-5 years of follow-up after discontinuation of the insulin therapy. One patient needed to receive antidiabetic drug after 3 years of insulin therapy cessation. Except one, all the patients were received methylprednisolone bolus at least one time at the NODM period. Conclusions: In our center, NODM patients needed high dose insulin at the beginning, but the doses were decreased in the short time. It seems that insulin therapy necessity was related to pulse methylprednisolone therapy and high tacrolimus levels. A less diabetogenic immunosuppressive regimen, individualized dosage of immunosuppressive agent, and close monitoring of plasma levels should be routinely applied to liver transplant recipient.

HbA1c in obese and overweight children: is there any role in diagnosing impaired glucose metabolism

Konstantinos Kitsios1; Maria Papadopoulou1; Nikolaos Kadoglou2; Konstantina Kosta3; Alikisita Kapelouzou2; Lemonia Skoura1; Dimitrios Tserepis2; Michael Chatzidimitriou1; Maria Papagianni1; 1Aristotle University of Thessaloniki, Hippokration General Hospital, Thessaloniki, Greece; 2Third Department of Pediatrics, Thessaloniki, Greece; 3Foundation of Biomedical Research, Academy of Athens, Athens, Greece; 4Aristotle University of Thessaloniki, Medical School, Laboratory of Microbiology, Thessaloniki, Greece; 5Alexandron Technological Educational Institute, School of Medical Laboratories, Thessaloniki, Greece

Background: HbA1c has been recently proposed as diagnostic test for diabetes mellitus or impaired glucose metabolism at cut-off points: 6.5% and 5.7% respectively. Nevertheless, little is known about the validity of this diagnostic approach in obese children at risk for impaired fasting glucose (IFG), impaired glucose tolerance (IGT) or type 2 diabetes (T2D).

Objective and hypotheses: To assess the prevalence of IFG and/or IGT in obese children and to evaluate the validity of HbA1c in the diagnosis of the aforementioned metabolic abnormalities in youth compared to Oral Glucose Tolerance Test (OGTT).

Methods: 54 consecutive obese children (BMI>95%) and 50 overweight (85<BMI<95%) children and adolescents (6-17 years old) attending the obesity clinic were screened for IFG and IGT by means of an OGTT (1.75gr glucose/kg up to 75g) performed according to the International Diabetes Federation (IDF) standards. HbA1c was measured according to the DCCT standards in all the obese and overweight children and in 40 normal-weight age-matched children (control group).

Results: Among obese children 29.6% had IFG, 5.6% IGT and 9.3% both IFG and IGT. Similarly, in overweight children 16% appeared with IFG, 6% with IGT and 2% with both IFG and IGT. HbA1c was significantly higher in obese children (5.1±0.3%) compared to overweight (4.9±0.5%, p<0.001) and normal-weight children (4.5±0.2%, p<0.001). Overweight children had also significantly elevated HbA1c compared to the control group (p=0.027). HbA1c did not differ in obese and overweight children with IFG or/and IGT compared to their counterparts with normal glucose metabolism (4.7±0.3% vs 4.7±0.5%, p=0.476). None of the children with IFG, IGT or both had HbA1c>5.7%.

Conclusions: IFG and IGT are prevalent amongst obese and overweight children identifying a group at risk for T2D. HbA1c, although elevated in obese and overweight children compared to normal weight children, is not sensitive at a cut-off point>5.7% in diagnosing impaired glucose metabolism in youth.

A novel autosomal dominant syndrome of severe insulin resistance and growth retardation in a Turkish kindred

Tulay Gurur1; Eleanor Raffanti2; Abdullah Bereket3; Isabel Huang-Doran4; Keith Porter4; Julie Harris5; Teoman Akcay1; Serap Turan5; Zeynep Atay6; Stephen O’Rahilly2; Robert Semple3; 1Marmara University, Pediatric Endocrinology and Diabetes, Istanbul, Turkey; 2University of Cambridge, Institute of Metabolic Science, Metabolic Research Laboratories, Cambridge, United Kingdom

Background: Obesity-related insulin resistance (IR) is a growing problem worldwide. Wherever some patients with severe IR are lean and have underlying single gene defects. The most common genetic cause of severe IR is heterozygous INSR mutations, which produce severe IR in association with a normal lipid profile and raised adiponectin which is nearly pathognomonic. Growth retardation is only severe in the case of biallelic INSR mutations, though may be severe in patients with mutations in the IGFR gene, encoding the IGFI receptor. The signalling pathways downstream from the highly homologous products of the INSR and IGFR genes are nearly indistinguishable.

Hypothesis: We hypothesized the existence of novel molecular defects causing defective signalling downstream from both insulin and IGFI receptors, which would be expected to lead to a clinical syndrome with features of both insulin and IGFI resistance.

Population and methods: Eight patients from three generations within a non-consanguineous Turkish family were evaluated at clinical and molecular levels.

Results: We identified an extended kindred in which 8 members from 3 generations were affected by severe IR and short stature with facial dysmorphism. All affected family members had normal lipid profiles and normal or elevated adiponectin, consistent with insulin receptoropathy. Surprisingly coding sequence and splice junctions of the INSR and IGFR genes were found to be normal. Protein levels of INSR and IGFR were in the low normal range in dermal fibroblasts from the proband, with mRNA levels 2-3 fold elevated for both receptors. Linkage mapping has implicated a critical interval on chromosome 19, and further studies are currently underway.

Conclusion: We have identified a rare syndrome with features of both insulin and IGFI resistance with normal expression and sequence of both receptors. This is consistent with a novel molecular defect affecting both insulin and IGFI signal transduction.

Rabson Mendehall syndrome confirmed in a six year old boy: a case report

Roberta Diaz Savoldelli1; Michele Gatti Carballo1; Thais Della Mannu1; Maria Angela Zanello Fortes1; Maria Lucia Correa-Gianella1; Danvil Damiani1; 1Instituto da Criança - HCFMUSP, Pediatric Endocrinology Unit, São Paulo, Brazil; 2Faculdade de Medicina – USP, LIM 25, São Paulo, Brazil

Background: Rabson Mendehall Syndrome (RMS) is a rare autosomal recessive disorder caused by mutation in the insulin receptor (INSR) gene. It is characterized by dental anomalies, thick and hyperpigmented skin, hirsutism, macrocogentosomia and severe insulin resistance (IR) that may evolve to diabetes mellitus (DM). The RMS represents an intermediate form among
syndromes related to mutations in the INSR gene as the leprechaunism and type A and B IR syndromes.

Objective: To report the clinical and laboratory characteristics of a patient with RMS.

Patient and methods: A six year old boy born in Bahia, Brazil, from unrelated parents developed hyperpigmentation in areas of skin folds since his first year of life; one first degree cousin with similar physical features was also reported. Examination showed cervical, axillary, inguinal and peri-umbilical acanthosis nigricans; hypertrichosis and dental abnormalities; penis of 5.0 cm in length. Laboratory work-up and molecular analysis were performed.

Results: Glucose and fasting insulin: 70 mg/dL and 178.6 mcU/mL. Oral GTT showed a peak insulin and glucose of 2287.2 mcU/mL and 138 mg/dL, respectively. HBA1c: 5.8%. Molecular investigation demonstrated a homozygous mutation in exon 19 of INSR gene, codon 1135, GGC (alanine) to GTG (valine). Parents were heterozygous. The patient has not developed DM yet, only few episodes of postprandial hyperglycemia (220-234 mg/dL) and prolonged fasting hypoglycemia (minimum 43 mg/dL). There are descriptions of the same mutation changing the alanine codon (GGC) by glutamic acid (GAG) providing insulin resistance. It is known that alanine at codon 1135 is highly conserved in species and valine is one of the amino acids designated as not to be tolerated in this position.

Conclusion: A RMS patient is described with an identified mutation in codon 1135, GGC (alanine) to GTG (valine) in exon 19 of the INSR gene, presenting a typical phenotype, severe IR but not DM until 6 years of age.

P2-d2-510 Diabetes and Insulin 3
Pharmacokinetics of metformin at age 9: a pilot study in girls with early puberty
David Sanchez-Infantes1; Marta Diaz2; Abel Lopez-Bermejo2; Maria Victoria Marcos3; Francis de Zegher4; Lourides Ibáñez2
1Hospital Sant Joan de Déu, University of Barcelona, Endocrinology, Esplugues, Barcelona, Spain; 2Hospital Sant Joan de Déu, Endocrinology, Esplugues, Barcelona, Spain; 3Hospital Dr. Josep Trueta, Pediatrics, Girona, Spain; 4Hospital de Terrassa, Endocrinology, Terrassa, Barcelona, Spain; 5University of Leuven, Woman & Child, Leuven, Belgium

Aim: To study the pharmacokinetics of metformin in young and non-obese children.

Methods: The study population consisted of 6 girls with a combined history of low birthweight and early-normal onset of puberty. At time of study, these girls were aged 9 and had been receiving metformin (850 mg/d at dinner time) for a mean duration of 8 mo. The girls were sampled before metformin intake and for 12 h thereafter. Serum metformin levels were assessed by liquid chromatography–tandem mass spectrometry (LC/MS/MS); the area under the curve (AUC), the maximum concentration (Cmax), the time of maximal concentration (tmax), the elimination half-life, (t1/2), the volume of distribution (Vd) and the total clearance (CL) were calculated.

Results: Metformin concentration-time curves were similar in girls receiving similar metformin doses (range 21-29 mg/Kg); in those girls, mean AUC was 21 mg hr/L, Cmax 3 mg/L, tmax 2.5 h, t1/2 4 hr, Vd 111 L and CL 20 L/hr, these values being comparable to those in adults.

Conclusion: In children aged 9, metformin’s pharmacokinetics compare to those in adults.

P2-d2-509 Diabetes and Insulin 3
Continuous glucose monitoring, glucose tolerance and indices of insulin resistance in children on high dose corticosteroid therapy
Ahmed Elawwa1; Ashraf Soliman2; Bajes Hamad3
1Alexandria University, Egypt and Hamad Medical Center, Pediatrics, Doha, Qatar; 2Hamad Medical Center, Pediatrics, Doha, Qatar

Background: Impaired glucose tolerance (IGT) and steroid-induced diabetes have been reported with systemic use of pharmacological doses of corticosteroids. The effects of glucocorticoids on carbohydrate metabolism may be dose related. The use of continuous glucose monitoring system (CGMS) for early detection of glycemic abnormalities has not been studied in those children on high dose corticosteroid therapy.

Objectives: To assess oral glucose tolerance, 72-h continuous glucose blood concentrations by CGMS, calculate Homeostatic model assessment (HOMA), and the quantitative insulin sensitivity check index (QUICKI) in children and adolescents on pharmacological doses of corticosteroids.

Population: 10 children with nephrotic syndrome receiving pharmacological doses of oral prednisolone for more than 3 weeks.

Results: The mean age of children was 7.3± 2.9 years. The mean cumulative dose of prednisolone given was 1601.3± 984.8 mg, the mean duration of therapy was 84 ± 14 days. The OGTT and the average glucose level recorded by CGMS were normal in all patients. 4 cases had Maximum glucose level > 11.1 mmol/L, 3-hours after meal (Diabetic range), 2 of them had BMI > 2 SD. All others had BG > 7.8 mmol/L (IGT) mean = 10.6 +/- 1.6 mmol/L. HOMA and QUICKI revealed high > 2 (2.9± 2.3) and low levels < 0.35 (0.32±0.05) respectively denoting insulin resistance. Unexpectedly, 4 children had one hypoglycemic episode (RBS < 2.8 mmol/L). The cumulative steroid dose was positively correlated with the insulin levels (r=0.79, P <0.01), c-peptide level (r=0.67, P <0.01), and negatively correlated with the QUICKI (r= -0.66, P <0.01) which suggested that the higher the cumulative dose the higher the insulin secretion and resistance state.

Conclusion: CGMS is superior to OGTT in detection glycemic abnormalities. The cumulative dose of steroid therapy is significantly correlated with insulin secretion and resistance state.
P2-d2-512 Diabetes and Insulin 3

Wolcott-Rallison syndrome in a girl with a novel mutation

Metin Yıldız1; Pinar Isguvvener2; Hyes Kirmizibekmez3; Nurcan Cebecli3; Fatma Dursun4; Ayşe Guven4
1Goztepe Education and Research Hospital, Pediatric Endocrine, Istanbul, Turkey; 2Goztepe Education and Research Hospital, Pediatric Clinic, Istanbul, Turkey; 3Goztepe Education and Research Hospital, Pediatric Endocrine, Istanbul, Turkey

Background: Wolcott-Rallison syndrome (WRS) is a rare, autosomal recessive disorder characterized by early infancy-onset insulin-dependent diabetes mellitus, epiphyseal dysplasia, and growth retardation with frequent additional manifestations, including acute attacks of hepatic and renal dysfunction, developmental delay, exocrine pancreatic deficiency, and neutropenia.

Case: Here we present a Turkish girl with WRS carrying a novel mutation (EIF2AK3:E375X). GU, is a 7.7-year-old girl, presented with diabetic ketoacidosis at 3 months of age. She was born SGA (BWSDS: -3.96), the third child of consanguineous parents. Her complaints were; poor weight gain, vomiting, poliuria and respiratory distress and laboratory results were consistent with diabetic ketoacidosis. HbA1c level was % 15.3, islet-cell and anti-GAD antibodies were negative. With insulin therapy she had normoglycemia but rather high HbA1c levels during follow-up. She had never had a catch-up growth. Thyroid function tests and growth hormone stimulation test were normal. Difficulty in walking and pain in dorsolumbal vertebrae and pelvis became prominent at 4 years of age. X-ray roentgenograms showed spondylo-epiphysial dysplasia. 2 years of age she had transient liver dysfunction which resolved spontaneously. Her renal functions showed mild uremia, hyperkalemia and hyperphosphatemia. Renal ultrasound and scintigraphy revealed no abnormality. At 7.7 years of age her height was 91.3 cm (< 3.5D SDS), weight 11.8 kg (< 3rd percentile), BMISDS: -1.4, showing severe growth retardation. Mutation analysis confirmed WRS in our patient and her parents were heterozygous carrier for the same mutation. In exon 6 of the EIF2AK3 gene sequencing analysis has shown homozygosity for a nonsense mutation, E375X. This mutation as far as we know is a novel mutation published in the literature.

Conclusions: WRS is the most common cause of permanent neonatal diabetes mellitus in consanguineous pedigrees and should be keep in mind in patients with skeletal dysplasia and/or intermittent renal/hepatic dysfunctions.

P2-d2-513 Diabetes and Insulin 3

Do oral glucose tolerance tests (OGTT) alter management in patients at risk of metabolic derangement due to insulin sensitivity?

Karen Loga1; Nirit Brah1; Helen Speoudea; Peter Hindmarsh1; Shalini Santhakumar1; Gary Butler1
1University College Hospital London, Paediatric Endocrinology, London, United Kingdom; 2Imperial College London, Paediatrics, London, United Kingdom

Background: Escalating childhood obesity has prompted early identification of metabolic derangement by oral glucose tolerance tests (OGTT) to avert future cardiovascular risk by treatment intervention. Screening for insulin insensitivity however, is problematic with no clear diagnostic criteria and was not endorsed by the ESP consensus statement (2010).

Objective and hypothesis: We aimed to assess OGTT retrospectively, in a future cardiovascular risk by treatment intervention. Screening for insulin insensitivity, but rather high HbA1c levels during follow-up. She had never had a catch-up growth. Thyroid function tests and growth hormone stimulation test were normal. Difficulty in walking and pain in dorsolumbal vertebrae and pelvis became prominent at 4 years of age. X-ray roentgenograms showed spondylo-epiphysial dysplasia. 2 years of age she had transient liver dysfunction which resolved spontaneously. Her renal functions showed mild uremia, hyperkalemia and hyperphosphatemia. Renal ultrasound and scintigraphy revealed no abnormality. At 7.7 years of age her height was 91.3 cm (< 3.5D SDS), weight 11.8 kg (< 3rd percentile), BMISDS: -1.4, showing severe growth retardation. Mutation analysis confirmed WRS in our patient and her parents were heterozygous carrier for the same mutation. In exon 6 of the EIF2AK3 gene sequencing analysis has shown homozygosity for a nonsense mutation, E375X. This mutation as far as we know is a novel mutation published in the literature.

Conclusions: WRS is the most common cause of permanent neonatal diabetes mellitus in consanguineous pedigrees and should be keep in mind in patients with skeletal dysplasia and/or intermittent renal/hepatic dysfunctions.

P2-d2-514 Diabetes and Insulin 3

The influence of metabolic disorders in children with diabetes mellitus type 1 on changes of QT interval

Guina Megar1; Anzhalkia Soltseva2
1Belarusian State Medical University, First Department of childhood diseases, Minsk, Belarus; 2Minsk City Republican Children’s Medical Centre, Minsk, Belarus

Methods: OGTT and QTC interval were evaluated in 178 children with DM1 (middle age 13.44±0.29 years, duration of disease 5.72±0.28 years) and in 60 sex- and age-matched healthy children. Level of HbA1c in children with DM1 made up 9.8±0.18% (N<6.2%; P<0.001). It is established that values of QTc in girls with DM1 are higher than in boys (428.07±4.51 ms versus 413.6±4.08 ms, P<0.0025). 17.71% of girls and 13.41% of boys with DM1 have autoimmune thyroiditis (the percent of children with values of QTc>440 ms is high in this group). At the same time values of HbA1c are higher in boys than in girls (10.4±0.3 and 9.35±0.2% respectively, P<0.002). Connection of QT with age (r=0.338, P<0.00001) and level of HbA1c (r=0.37, P<0.0001) was established. Feedback connection of QT with pulse rate was revealed (r=0.48, P<0.005). There were no reliable correlations of QTc with Body Mass Index, level of cholesterol, duration of the disease and values of systolic and diastolic blood pressure.

Conclusions: The increase of QT and QTC is noticed in children with DM1. Age, level of glycemic and pulse rate are the factors defining values of QTc. In girls QTc has higher values more often than in boys. Intervals QTc>440 ms are found more often in children with DM1 and autoimmune thyroiditis.

P2-d2-515 Diabetes and Insulin 3

Weight loss and metabolic benefit with the addition of glucagon-like peptide agonists and pioglitazone to type 2 diabetes mellitus adolescents treatment

Nouhad Rassouni1; Sheila Perez2; Fatma Ahmed1; Anmit Bhango3; Swetiana Ten3
1Belgium; 2United States; 3Belgium

Methods: 9 overweight adolescents’ with DMT2 and BMI >24 kg/m2 were

Horm Res 2011;76(suppl 2) 161
studied. GLP-1 agonists 5mg subcutaneous was prescribed twice daily once patients were clinically diagnosed with DMT2 and then increased to 10 mcg twice daily one month later. Lantus averag 30 units daily with Actoplus Met 15mg/850mg once daily was used. Anthropometric parameters, metabolic parame-
ters, Gastrointestinal and neurological complications assessment were obtained at the baseline and 6 months after treatment.

Results: Anthropometric and Biochemical Characteristics of 9 adolescents with DMT2, Base Line and post 6 months treatment.

<table>
<thead>
<tr>
<th>BMI (Kg/m2)</th>
<th>SBP</th>
<th>DBP</th>
<th>HbA1c</th>
<th>LDL</th>
<th>Chol</th>
<th>TG</th>
<th>HDL</th>
<th>AST</th>
<th>ALT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>127.6</td>
<td>72.7</td>
<td>9.1256</td>
<td>182.3</td>
<td>183.4</td>
<td>36.7</td>
<td>7.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>+/15.9</td>
<td>+/36</td>
<td>+/18.5</td>
<td>+/1.7</td>
<td>+/34.4</td>
<td>+/29</td>
<td>+/18.4</td>
<td>+/29</td>
<td>+/9.1</td>
<td></td>
</tr>
<tr>
<td>6 months</td>
<td>120.8</td>
<td>70.6</td>
<td>8.100</td>
<td>100</td>
<td>153</td>
<td>40.6</td>
<td>6.5</td>
<td>39.5</td>
<td></td>
</tr>
<tr>
<td>Follow-up</td>
<td>+/22</td>
<td>+/-2</td>
<td>+/-1.2</td>
<td>+35</td>
<td>+/40</td>
<td>+/6</td>
<td>+/5.8</td>
<td>+/8</td>
<td></td>
</tr>
</tbody>
</table>

Values are means +/- SD

Conclusions: After 6 months of therapy 75% of patients showed a remission of DMT2 based on improved HbA1c. Lantus therapy was discontinued after one month of use only. Short acting insulin use was not necessary. BMI %was decreased in all patients. All patients showed improvement in their liver enzymes. No documented Gastrointestinal or neurological complications were notified.

P2-d2-516 Diabetes and Insulin 3

Metabolic profile in prepubertal type 1 diabetes mellitus and its evolution after antioxidants and fatty acid omega-3 therapy

Isabel González Casado,1 Julio Guerrero-Fernández,1 Elena Ibashez Ezequiel2, Coral Barbabs Arribas;1 Javier Señorans Rodríguez,2 Guillermo Reglero Rada;2 Luis Salamanca Fresno;3 Ana de la Puente Arvantlo;1 Ana Coral Barreda Bonis1, Ricardo Gracia Bouthelier1

Hospital Infantil La Paz, Pediatric Endocrinology Unit, Madrid, Spain; 1Instituto de Investigación en Ciencias de la Alimentación, CIAL-CSIC, Departamento de Bioactividad y Análisis de Alimentos, Madrid, Spain; 2San Pablo-CEU University, Analytical Chemistry, Boadilla del Monte, Spain; 3Universidad Autónoma de Madrid, Ciencia y Tecnología de Alimentos, Madrid, Spain; Universidad Autonoma de Madrid, Institute de Fermentaciones Industriales, Madrid, Spain

Background: The effects of hyperglycemia on oxidative and lipid profiles is well known in patients with Diabetes Mellitus and its influence on the microvascular complications. However advances in metabolomics could help to understand this complex pathophysiological process.

Objective and hypotheses: The aim of this study was to determine new differentiating metabolites using metabolomic techniques in pediatric patients with type 1 Diabetes Mellitus (DM1) compared with healthy controls (CS) and tested the use of meat products that contain antioxidants from oil rose and in there liver enzymes. No documented Gastrointestinal or neurological complications were notified.

Methods: Fifty prepubertal patients aged between 6 and 10 years old has been divided into three groups: 17 took the product DM1 (group 1), 17 DM1 patients (group 2), which was homogeneous at the beginning and 26 healthy patient’s profile. This approach suggests a possible beneficial effect of such products in children with type 1 diabetes mellitus of short evolution.

P2-d2-517 Diabetes and Insulin 3

Glycaemic control in children, adolescents and young adults with type 1 diabetes: the role of physical activity and sociodemographic factors

Angela Gallego1; Andrea Erent1; Maren Lindau1; Peter Friedrich1; Klemens Raile1

1Charité, Universitätsmedizin Berlin, Paediatric Endocrinology and Diabetology, Berlin, Germany; 2Charité, Universitätsmedizin Berlin, Institute for Biometrics and Clinical Epidemiology, Berlin, Germany

Background: Poor glycaemic control is associated with an increased risk of developing microvascular and macrovascular complications in type 1 diabe-
tes. Therefore identifying the factors affecting metabolic control in children and adolescents with type 1 diabetes is important.

Objective and hypotheses: Aim of the study was to examine the relation-
ship between physical activity, psychosocial factors, and glycaemic control in children, adolescents and young adults with type 1 diabetes.

Methods: The cross-sectional study included children, adolescents and young adults with type 1 diabetes up to the age of 22 years. Clinical data and HbA1c levels were collected during outpatient clinic visits. Self-report questionnaires were used to assess physical activity and to collect sociodemographic vari-
ables. Risk factors were analysed by linear regression.

Results: A total of 296 children, adolescents and young adults with type 1 diabetes (age 13.7+/−4.1 years, diabetes duration 6.1+/−3.3 years, HbA1c 8.7+/−1.6%) participated in the study.

HbA1c levels were not different in subjects who were physically active com-
pared to those who were physically inactive (p=0.63). High socioeconomic status was significantly associated with better glycaemic control (HbA1c 8.9+/−1.4%) whereas middle and low socioeconomic status was associated with worse glycaemic control (HbA1c 8.5+/−1.4% and 8.9+/−1.7%; p=0.023 and p=0.001).

HbA1c levels were significantly higher in children and adolescents from im-
migrant families compared to those from non-immigrant families (9.0+/−1.7% vs 8.4+/−1.4%; p<0.001). Glycaemic control was better in two-parent families compared to single-parent families (HbA1c 8.2+/−1.3% vs 9.2+/−1.7%; p<0.001).

Linear regression analysis identified diabetes duration, socioeconomic status, and family status as risk factors for poor glycaemic control.

Conclusions: Longer diabetes duration, lower socioeconomic status, and single-parent status are significant risk factors for poor glycaemic control.

P2-d2-518 Diabetes and Insulin 3

C-peptide secretion in pediatric patients with recent onset of type 1 diabetes

María Martin-Frías; Noelia Alvarez; Mª Angeles Alvarez; Rosa Velmo; Mitagros Alonso; Raquel Barrio

Ramón y Cajal Hospital, Pediatric Endocrinology and Diabetes Unit, Madrid, Spain

Background: In type 1 diabetes (T1DM) a significant secretory function may persist for a long period in some patients, as evidence by the presence of serum C-peptide levels. This phenomenon might reduce the risk of chronic complications, severe hypoglycemias and allow easier metabolic control.

Objective and hypotheses: To investigate the relationship between C-pep-
tide response to glucagon stimulation test (GST) and clinical and biochemical characteristics at diagnosis of childhood T1DM.

Methods: Prospective study of 40 pediatric patients. Mean age at diagnos-
sis 9.3 years (±4.12.5), 51.2% males. All subjects underwent an intravenous GST one or two months after diagnosis (15mg/kg I.V., maximum 1mg). At diagnosis we analyzed: age, weight loss (kg), presence of cardinal symptoms (months), metabolic disturbances: isolated hyperglycaemia, ketosis or ketoacidosis (ISPAD 2010), HbA1c (%) [±5.3+/−0.3] and insulin dose; after one o two months: C-peptide response to GST. Statistical analysis was performed with SPSS program, version 15.0, non parametric tests; data expressed in per-
centage, median and range.

Results: Younger patients had lower C-peptide levels (p=0.01). At diagnosis 23.1% patients had isolated hyperglycaemia, 28.2% ketosis and 48.7% ke-
toacidosis. Patients with ketoacidosis had lower C-peptide levels and were younger (p<0.05). No others significant correlations between C-peptide levels and clinical- biochemical features were found.
**P2-d3-519 Diabetes and Insulin 4**

**Abnormal glucose tolerance in prepubertal patients with cystic fibrosis: the role of oral glucose tolerance test**

María Martín-Frias1; Adelaida Lamas2; Lucrécia Suárez2; Noelia Alvarez3; Milagros Alonso3; Raquel Barrio3

1Ramón y Cajal Hospital. Alcalá University, Paeditric Endocrinology Unit, Madrid, Spain; 2Ramón y Cajal Hospital. Alcalá University, Cystic fibrosis Unit, Madrid, Spain

**Background:** Annual screening for abnormal glucose tolerance (AGT) should begin by age 10 years in Cystic fibrosis (CF) patients (Consensus 2010). Some authors demonstrated AGT in patients younger than 9 and recommend an earlier screening.

**Objective and hypotheses:** To examine the frequency of AGT and its outcome in prepubertal CF patients and the changes in glycemic and nutritional status and lung function over the preceding year.

**Methods:** Retrospective study of 19 prepubertal CF patients (68% males). All subjects underwent an oral glucose tolerance test (OGTT). In follow-up, 49 OGTTs were performed over a mean of 2 years. Results were classified as: normal glucose tolerance (NGT), impaired glucose tolerance (IGT), CF related diabetes (CFRD) or indeterminate glucose (INDET). We related C-peptide basal/ C-peptide 1/2h (ng/ml) to metabolic control and weight, height, BMI. Blood samples were taken between 7:30-8:30 in normoglycemia after the night without episodes of hyper or hypoglycemia. All analysis were made in Department of Radioimmunology Children’s Memorial Health Hospital by ELISA, IRMA and RIA commercial kits.

**Results:** Positive correlation between age and the moment of diagnosis the dose of insulin (r=0.23, p<0.05), concentration of leptin (r=0.40, p=0.001) and negative with the concentration of s-OB1 (r=-0.45, p=0.001) were observed in IDDM children whereas the age correlated positively only with concentration of leptin (r=0.42, p=0.001) and negatively with s-OB1 (r=-0.28, p=0.005) and duration of diabetes only positively with s-OB1 (r=0.28, p<0.05).

**Conclusions:** 1. The age of children at the moment of diagnosis the diabetes better correlate with leptin and s-OB1 than the age and duration of illness.

**P2-d3-520 Diabetes and Insulin 4**

**Does the age of children in the moment of diagnosis the diabetes may influence on adipocytokines levels in children with IDDM**

Mieczyslaw Szalecki1; Roman Janas1; Ewa Pankowska2

1Children’s Memorial Health Hospital, Endocrinology & Diabetology, Warsaw, Poland; 2Mother and Child Institute, Pediatric, Warszawa, Poland

**Background:** Adipocytokines play a great role in IDDM, pathogenesis complications, insulin resistance, dawn phenomenon and fat disorders.

**Objective and hypotheses:** Estimate the influence of age, duration of diabetes, age at the moment of diagnosis on adipocytokines (adiponectin, leptin, resistin, TNF-alpha) and solubles form of their receptors(s-OBR, sTNFR1) levels in children with IDDM.

**Methods:** 67 patients, 15 age-matched healthy children were included into the study. All children were prepubertal, without coexisting diseases, suffering from IDDM from more than two years. All patients were divided into groups according to the kind of therapy: 22 conventional insulin therapy, 21 multiple insulin injections, 24 insulin pumps. There were no statistically significant differences between groups as to the metabolic control, weight, height, BMI. Blood samples were taken between 7:30-8:30 in normoglycemia after the night without episodes of hyper or hypoglycemia. All analysis were made in Department of Radioimmunology Children’s Memorial Health Hospital by ELISA, IRMA and RIA commercial kits.

**Results:**

- Positive correlation between age and the moment of diagnosis the dose of insulin (p=0.23, p<0.05), concentration of leptin (p=0.40, p<0.001) and negative with the concentration of s-OB1 (p=-0.45, p<0.001) were observed in IDDM children whereas the age correlated positively only with concentration of leptin (p=0.42, p<0.001) and negatively with s-OB1 (p=-0.28, p<0.005) and duration of diabetes only positively with s-OB1 (p=0.28, p<0.05).

**Conclusions:** 1. The age of children at the moment of diagnosis the diabetes better correlate with leptin and s-OB1 than the age and duration of illness.

2. The age of children at the moment of diagnosis the diabetes may influence on adipocytokines abnormalities in IDDM children.

3. The kind of therapy appears to have an influence on the levels of adiponectin, leptin, s-OB1 and sTNFR1 but not on resistin and TNF-alpha levels.

**P2-d3-521 Diabetes and Insulin 4**

**Telemedicine follow-up of glycemic variability in children with type 1 diabetes mellitus: an analytical observational cohort study (Pilot)**

Nancy Villarrreal Peña1; Marisa Torres2; Larisa Suarez3; Paula Casano Sancho1; Raquel Iniestra1

1Hospital Sant Joan de Déu, Pediatric Endocrinology Unit, Barcelona, Spain; 2Hospital Sant Joan de Déu; Pediatric Diabetes Unit, Barcelona, Spain; 3Fundación Sant Joan de Déu, Statistics, Barcelona, Spain

**Background:** Glycemic variability consists of acute fluctuations in blood glucose levels and is directly related to microvascular and vascular damage in diabetes mellitus and with acute complications such as hypoglycemia.

**Objective and hypotheses:** To determine the impact of 3 months of telemedicine follow-up on metabolic control and glycemic variability in prepubescent patients with type 1 diabetes mellitus (T1DM).

**Hypothesis:** Metabolic control, measured by glycosylated hemoglobin (HbA1c) and glycemic variability, will improve in children aged between 6 and 10 years with T1DM followed-up by telemedicine for 3 months.

**Methods:** This study was divided into two periods: a 3 month period in which Telemedicine assessment was provided every 2 weeks for 3 months in 13 prepubescent 6-10-year-olds with T1DM compared with a 4 months period without telemedicine assessment. Metabolic control was compared between the two periods. Glycemic variability was evaluated using standard deviation of SD, the average daily risk range (ADR) and the low blood glucose index (LBGI). Blood glucose reports were sent by patients’ families using Accu-Chek Smart Pix software.

**Results:** Follow-up visits was regularly performed during the 7 months of the study. HbA1c values were determined and glycemic variability indexes were calculated at the start of the study, at the end of the telemedicine follow-up and
again 4 months later. Blood glucose was monitored 5.8 times per day. Statistical analyses were performed with non-parametric tests (p < 0.05).

**Results:** HbA1c values significantly decreased (p = 0.012; Figure 1). Non-significant decreases were found in LBOI (p = 0.115), ADRR (p = 0.552) and SD (p = 0.700).

**Conclusions:** Telemedicine follow-up for 3 months improved metabolic control based on HbA1c values and glycemic variability, without increasing acute complications. When telemedicine follow-up was discontinued, then metabolic control worsened; so we suggested that it will be necessary a prolonged period of follow-up with a telemedicine program.

---

**P2-d3-523 Diabetes and Insulin 4**

**First Vietnamese patient with a de novo mutation in the hepatocyte nuclear factor 1B/maturity-onset diabetes of the young type 5 gene**

Chi Dung Vu1; Thi Hoan Nguyen1; Phuong Thao Bui2; Thi Bich Ngoc Can3; Jayne Houghton; Sian Ellard4; Ngoc Khahn Nguyen5

1National Hospital of Pediatrics in Hanoi, Endocrinology, Metabolism and Genetics, Hanoi, Vietnam; 2Pensinsula Medical School, Institute of Biomedical and Clinical Science, Exeter, United Kingdom

**Background:** Maturity-onset diabetes of the young type 5 (MODY5), a type of dominantly inherited diabetes mellitus and nephropathy, has been associated with mutations of the hepatocyte nuclear factor-1B (HNF-1B) gene, mostly generating truncated protein. Various phenotypes are related to HNF-1B mutations.

**Objective:** To describe clinical and genetic findings in the first Vietnamese case identified with HNF-1B mutations.

**Patient:** The proband with diabetes diagnosed at 14.5 years of age and nondiabetic kidney disease who were described.

**Methods:** Case report included information: characteristics of diabetes, renal function and structure, pancreas structure, insulin secretion, and liver test results. Genomic DNA were extracted from WBC of whole blood and HNF-1B mutation was performed using PCR and direct sequencing.

**Results:** The proband is heterozygous for an HNF-1B missense mutation, S148L. This C>T mutation at nucleotide 443 (c.443C>T) results in the substitution of the amino acid leucine for serine at codon 148 (p.Ser148Leu) and has been reported previously. This result confirms a diagnosis of renal cysts and diabetes (RCAD). Pancreas atrophy was observed using abdominal CT. Renal involvement, consisting of structural changes and renal failure, was recognized on admission. Liver enzyme levels were normal. Blood glucose levels were controled using insulin and Metformin (glucophage) 500 mg daily.

**Conclusions:** Maturity-onset diabetes of the young type 5 encompasses a wide clinical spectrum. Analysis for mutations of HNF-1B is warranted, even without a family history of diabetes, in nonobese patients with diabetes and slowly progressive nondiabetic nephropathy, particularly when pancreatic atrophy.

---

**P2-d3-524 Diabetes and Insulin 4**

**The role of leptin, soluble leptin receptor, adiponectin and visfatin in insulin secretory dynamics in preterm born children**

Diana Yann1; Firdevs Bas2; Asuman Coban; Gulay Can3; Feyza Darendeliler2

1Istanbul University Istanbul Faculty of Medicine, Department of Pediatrics, Istanbul, Turkey; 2Istanbul University Istanbul Faculty of Medicine, Department of Pediatric Endocrinology, Istanbul, Turkey; 3Istanbul University Istanbul Faculty of Medicine, Department of Neonatology, Istanbul, Turkey

**Background:** There are still controversies whether insulin resistance develops in preterm born children in early childhood.

**Objective and hypotheses:** To investigate the role of leptin, soluble leptin receptor (sLR), adiponectin, visfatin and insulin secretory dynamics in the pathogenesis of possible insulin resistance in preterm born children in early childhood.

**Methods:** 30 preterm SGA born children (Group 1) (age range 2.5-5 years) and 27 preterm AGA born children (Group 2), matched for sex and age, were included in the study. Gestational age was 33.5±2.2 weeks and not different between the groups. In Group 1 mean birth weight (1275±279g) was significantly lower than in Group 2 (2065±521g) (p=0.000) by definition. Mean age at investigation was 3.3±0.7 years. Fasting blood glucose, lactate, cortisol, insulin, proinsulin, adiponectin, leptin, sLR and visfatin levels were measured.

**Results:** Mean height SDS was -0.8±0.9 and weight SDS was -0.9±1.1 in Group 1 at investigation and were significantly lower than the respective figures in Group 2 (0.0±0.9 and 0.0±1.0, p=0.002). There was no significant difference in BMI between the groups. There was no difference in HOMA-IR, leptin, sLR, adiponectin, proinsulin and visfatin values between the groups. In the total group, there was a positive correlation between adiponectin and
visfatin levels (r=0.292, p=0.034). In group 1, there was negative correlation between gestational age and proinsulin (r=−0.368, p=0.05), between SLR and proinsulin (r=−0.485, p=0.008) and visfatin levels (r=−0.377, p=0.044).

In group 2 there was negative correlation between visfatin and BMI SDS (r=−0.504, p=0.017), positive correlation between visfatin and adiponectin (r=0.475, p=0.019), and negative correlation between adiponectin and SLR (r=−0.407, p=0.043).

Conclusions: Preterm born children whether born AGA or SGA do not show insulin resistance in early childhood if BMI is normal. Significant differences between the preterm SGA and AGA groups regarding the adipocytokine levels were not detected.

**P2-d3-525 Diabetes and Insulin 4**

Outcomes in diabetic children managed on continuous subcutaneous insulin infusion (CSII) therapy

*T Makaya*1; S Chatterjee*2; NP Wright*3

1Sheffield Children’s Hospital, Diabetes and Endocrinology, Sheffield, United Kingdom; 2Sheffield Children’s Hospital, Paediatrics, Sheffield, United Kingdom

Background: Subcutaneous continuous insulin infusion (CSII) therapy use has gradually increased. UK guidelines recommend a trial of CSII in children aged under 12 years where multiple dose injections (MDI) are deemed impractical; or in children over 12 years who fail to achieve an HbA1c less than 8.5% despite multidisciplinary input, or who suffer disabling hypoglycaemia in an attempt to achieve good control.

Objective and hypotheses: We sought to evaluate our CSII service, and to audit it against national guidelines. We also sought to establish whether CSII improved diabetes control.

Methods: We reviewed notes for patients on CSII in our paediatric diabetic clinic. Data was collected for demographic information and pump initiation. We compared HbA1c, BMI SDS, episodes of hypoglycaemia and diabetic ketoacidosis (DKA) pre- and post-CSII.

Results: Of the 54 patients on CSII in our clinic (30.8% of total clinic), 59% were female. Ages ranged from 1.9 to 18.4 years (mean: 12.9 years). Fifty-seven percent of patients were between 11 and 16 years. Two years were less than five years old. Mean age at pump start was 11.7 years (range: 20 days to 16.61 years). Most patients commenced on CSII at age >12 years (66%). Duration of diabetes at start of CSII ranged from 20 days to 12.2 years (mean: 4.4 years). Carbohydrate counting sessions were documented in 90.5% of patients at the time of commencing CSII.

### Results

<table>
<thead>
<tr>
<th></th>
<th>12 months pre-CSII</th>
<th>Just before CSII</th>
<th>6 months into CSII</th>
<th>12 months into CSII</th>
</tr>
</thead>
<tbody>
<tr>
<td>HbA1c %</td>
<td>8.6 ±1.6</td>
<td>9.2 ±1.6</td>
<td>8.8 ±1.4</td>
<td>8.3 ±0.9</td>
</tr>
<tr>
<td>Mean ±SD</td>
<td>(n=49)</td>
<td>(n=52)</td>
<td>(n=48)</td>
<td>(n=28)</td>
</tr>
<tr>
<td>BMI SDS</td>
<td>0.53 ±0.92</td>
<td>0.43 ±0.96</td>
<td>0.60 ±0.76</td>
<td>0.82 ±0.65</td>
</tr>
<tr>
<td>Mean ±SD</td>
<td>(n=48)</td>
<td>(n=52)</td>
<td>(n=39)</td>
<td>(n=25)</td>
</tr>
</tbody>
</table>

*pValue calculated for values just before and 12 months into CSII therapy. (Significant level considered as p=0.007).

Conclusions: CSII resulted in an improved HbA1c in the short-term.

**P2-d3-527 Diabetes and Insulin 4**

Impaired glucose intolerance screening by urinary visfatin in Japanese children

Eiichiro Satake*; Yuichi Nakagawa; Rie Matsushita; Eiko Nagata; Rie Yamaguchi; Kazuki Kitsuta; Shinichiro Sano; Yasuiko Fujisawa; Toshihiko Nakashima; Takehiko Ohteki

Hammatsu University School of Medicine, Pediatrics, Hamamatsu, Japan

Background: Type 2 diabetes mellitus is increasingly being observed among children and adolescents. Early detection is important for preventing diabetes-related morbidity and mortality. Myoinositol (MI) is one form of inositol and is a cyclic D-glucose isomer. It is widely available in organisms and is supplied by food or renal biosynthesis. MI is reabsorbed in the renal tubules, maintaining its blood concentration. It can be measured in urine and has been found to be elevated in patients with diabetes and glucose intolerance in adults. However little is known about in children or adolescents.

Objective and hypotheses: To assess the possibility of using MI as a marker of glucose intolerance in Japanese children.

Methods: Urinary visfatin (UMI) was enzymatically measured before and 2 h after school meal in 148 Japanese children (78 boys and 70 girls: 10-12 years old) without known diabetes. UMI (myoinositol/creatinine ratio: 2-h after meal-before meal) was measured and an oral glucose tolerance test (OGTT) was performed in each subject with over 10 mg/gCr UMI.

Results: The OGTT was performed on five children. They had no glycosuria and were not obese. Among them, two children represented low insulinogenic index (the ratio of increment of plasma insulin to that of plasma glucose at 30 min of OGTT) and one girl represented 1h plasma glucose higher than 10 mmol/l (180mg/dl). That girl exhibited impaired glucose tolerance after 5months.

Conclusions: UMI can be use of a non-invasive and sensitive marker for glucose intolerance in children.

**P2-d3-528 Endocrine Oncology 1**

Intra-operative analysis of autonomous ovarian testosterone production unveils the virilizing tumour site in a young girl

Regina Braun1; Steven Warmann2; Jörg Fuchs2; Gerhard Binder2

1University Children’s Hospital, Pediatric Endocrinology, Tuebingen, Germany; 2University Children’s Hospital, Department for Pediatric Surgery, Tuebingen, Germany

Background: It is a challenge to diagnose ovarian, testosterone producing tumors in childhood and adolescence when the first clinical features appear. Considering the rareness of these tumors imaging techniques may hint to their localization, but can still be afflicted with diagnostic uncertainty.

Aim: Verifying the site of excessive testosterone production by immediate intra-operative separate assessment of testosterone levels in the right and left ovarian vein.

Case: 11.6-year-old girl with a history of deepening of voice, mild hirsutism, minor acne, increased growth velocity, weight gain and cliteromegaly for about six to twelve months. PH3, B2 according to Tanner, no menarche. Laboratory investigation revealed an excessively elevated serum testosterone (343 ng/dl) without any additional endocrine abnormalities (normal DHEAS, androstendione, 17-hydroxyprogesterone). Abdominal ultrasound and MRI scan showed a sparsely noticeable solid mass with a maximum diameter of 13 mm in the center of the right ovary. At laparotomy both ovaries appeared unsuspicious. Blood was separately drawn from the right and left ovarian vein.

Immediate intra-operative measurement of serum testosterone with ADVIA

Conclusions: Preterm born children whether born AGA or SGA do not show insulin resistance in early childhood if BMI is normal. Significant differences between the preterm SGA and AGA groups regarding the adipocytokine levels were not detected.
Centaur testosterone test (28380 ng/dl right ovarian vein, 403 ng/dl left ovarian vein) guided to the site of the tumor within 45 minutes. Both dilutions were diluted (1:4). The sample of the right ovarian vein required a second dilution (1:16). Consent for right salpingo-oophorectomy was carried out during the same surgical procedure. Postoperative testosterone level was in normal range within 24 hours. The histopathological diagnosis was Leydig cell tumor.

Conclusion: Intra-operative assessment of ovarian testosterone production combined the definite diagnosis and therapy of a Leydig cell tumor. Using a fast hormone assay intra-operative hormone recording is a valuable tool in cases of not clearly circumscribable sites of autonomous hormone production in ovaries.

**P2-d3-529** Endocrine Oncology 1

Implementation of endocrine follow up in a childhood cancer survivor unit

Paula Casano-Sanchez1; Fred Cavallio2; Susana Rives2; Andrea Parareda2; Carmen Vallis2; Ruben Diaz-Nadeni2

1 Sant Joan de Deu Hospital, Pediatric Endocrinology, Barcelona, Spain; 2 Sant Joan de Deu Hospital, Hematology, Barcelona, Spain;

The improvement in oncology therapies supposes that 1:800 adults will be a childhood cancer survivor. Endocrine disorders are the most common among the late side effects reported; accordingly, international guidelines have encouraged endocrinologists to prospectively evaluate the survivors. To describe the prevalence and to identify under-diagnosed endocrine disorders; To assess sequelae depending on treatments received; To evaluate gonadal toxicity according to pubertal stage at diagnosis. Prospective longitudinal study -from 2006-2010 of endocrine sequelae in childhood cancer survivors <25 y after chemotherapy, radiation or autologous stem cell transplantation (ASCT).

Evaluation of clinical and analytical data were performed depending on the tumor, treatment and recommendations of Children’s Oncology Group Long-Term Follow-Up Guidelines. We followed 194 patients; age 10 y ± 6.8 yr. Oncology treatments were finished 5.2 years before follow-up (range 1 -12 y). 65% per cent of survivors showed endocrine disorders that required specialized care. Almost 30% of those were seen 4 y after the end of therapy were found to have under-diagnosed endocrine abnormalities. The most affected axes were: gonadotrophic (25/84 at pubertal age), thyroid (8%) and somatotrophic (4.6%). The sequelae were classified by treatments, with the highest prevalence in those patients who received abdominal radiotherapy and ASCT (93% and 75%, respectively with gonadal dysfunction). Prepuberty did not protect against gonadal toxicity, as 68% of patients with gonadal dysfunction were prepubertal at diagnosis (age, 4.8 ± 3.3 yr). The deficiencies of a prospective follow-up program may result in the under-diagnosis of some endocrine disorders; In patients receiving pelvic RT to ASCT fertility preservation before treatment start should be considered; Prepubertal stage does not protect against gonadal toxicity; Given the high prevalence of endocrine sequelae, we recommend evaluation of survivors by paediatric endocrinologists as part of a multidisciplinary team of long-term sequelae follow-up.

**P2-d3-530** Endocrine Oncology 1

Unusual pediatric endocrine tumor (glucagonoma) presenting as hypercortisolism and catch-down growth

Gabriella Pozzobon; Giuseppe Cannarile; Chiara Maria Damia; Giesset Garbetta; Maria Piera Ferrarelli; Chiara Degradi; Berardo di Natale; Giovanna Weber; Giuseppe Chiumento

Vita Salute San Raffaele University, Pediatric Endocrine Unit, Milan, Italy

Background: Deflection in growth velocity might be the presenting symptom of pediatric oncologic disease. Pancreatic neuroendocrine tumors (PNET) are unusual during childhood (incidence 0.01;10000); they are functionally silent in 53% of cases and early diagnosis is difficult.

Objective and hypotheses: Female (8.8 yr) presented catch-down growth (growth velocity -2 SDS in 1 y) with no other symptoms. Laboratory screenings evidenced persistent hyperthyrotopinemia, L-Thyroxine therapy was immediately started. Blood glucose monitoring was normal. Growth hormone (GH) provocative tests showed normal GH incretion, with adequate IGF1 levels. Bone age delay (6,5 yr). Brain MRI: normal hypothalamic-pituitary region. Karyotype: 46 XX. Hypothalamic-pituitary-adrenal axis: loss of circadian rhythm with high urinary and salivary cortisol levels; at dexamethasone suppression test: adequate inhibition of cortisol and ACTH.

Methods: To evaluate hypercortisolism (bilateral inferior petrosal sinus sampling) she was referred to our Hospital: height -2.56 SDS, midparental height 2 SDS, no cushing phenotype. Abdomen ultrasound, MRI e CT showed a hypervascular solid formation of 5 cm diameter at pancreatic tail. Tumor and neuroendocrine markers analysis were positive for glucagon (118 pMoli/L 17-51). After adequate treatment with Octreotid, a subtotal pancreatectomy was performed.

Results: Lesion histology: well-differentiated PNET. Immunophenotype: sinaptofis, chromogranin, neuron-specific enolase positive. Glucagon positive (80%). Insulin, somatostatin, polypeptide pancreatic and ACTH negative. During follow up (2 y) we observed normalization of serum markers, catch-up growth (+1,5 SDS) and no recurrence or onset of new lesions. Mild elevation of hemoglobin glycated (6%).

Conclusions: PNETs, such as glucagonoma, may present with reduction in growth velocity with no further symptoms. Prompt diagnosis and appropriate therapy can reduce comorbidities and allow adequate catch-up growth. The etiopathogenesis of hypercortisolism remains unclear.

**P2-d3-531** Endocrine Oncology 1

Timing of menarche in childhood cancer survivors: results of a nationwide survey

Theda Wessel1; Magdalena Balcerak2; Simone Reinmuth2; Cynthia Hohmann3; Thomas Keil3; Guenter Henze4; Anja Borgmann4

1Charité University Children's Hospital, Paediatric Endocrinology, Berlin, Germany; 2Charité University Children's Hospital, Paediatric Haematology and Oncology, Berlin, Germany; 3Charité University Children's Hospital, Institute of Social Medicine, Epidemiology and Health, Berlin, Germany

Background: Cure rates of childhood cancer have improved dramatically over the past decades. However, side effects of treatment are still causing significant health problems.

Objective: The aim of the present analysis was to evaluate the influence of tumour localisation, radiotherapy, and chemotherapy on the age of menarche.

Methods: 4689 adult former paediatric oncology patients, diagnosed 1980-2004, were contacted in collaboration with the German Childhood Cancer Registry.

Results: 1036 out of 1461 female participants reported their age at menarche and had oncological diseases diagnosed before menarche. The median age at menarche was 13 years (mean 13.0 years, SD 1.6), compared to 12.8 years in the general population. Relative to the localisation of brain tumours shifts towards earlier (cerebrum, median 11 years) and later time points (epiphysis, median 16 years) were observed. A significant delay of menarche was seen in patients with hypophyseal radiation doses of ≥ 30 Gy (mean 13.6 years, SD 2.2) compared to girls receiving < 30 Gy (mean 12.5 years, SD 1.4, p=0.05). Patients who received hypophyseal radiation of ≥ 30 Gy and additional spinal radiation were even older at menarche (mean 14.4 years, SD 2.5). Only few dosage groups among chemotherapeutic agents were associated with a slight menarcheal delay of < 1 year. Menarche never occurred in five of 1338 patients.

Conclusions: Most female childhood cancer survivors reach menarche at a normal age. Chemotherapeutic agents showed only a small or no influence on the age of menarche. A hypophyseal radiation dosage of ≥ 30 Gy, spinal and pelvic radiotherapy were associated with a moderate delay in the occurrence of menarche. These patients should be monitored particularly closely to ensure normal pubertal development.
P2-d3-532 Endocrine Oncology 1
Paediatric craniopharyngioma: a 30 year retrospective analysis of the West of Scotland experience
Diana Mcintosh1; Hanna Minhas1; Humayun Khadja1; Jennifer Brown1; Jerome St George1; Dermot Murphy1; Milind Ronghe1; Jairam Sastry1; Malcolm Donaldson1; S. Faisal Ahmed1; Fiona Cowie1; Richard Jones1; M Gutter Shakti2
1Royal Hospital for Sick Children, Paediatric Oncology, Glasgow, United Kingdom; 2Royal Hospital for Sick Children, Paediatric Endocrinology, Glasgow, United Kingdom; 3Aga Khan University, Department of Paediatrics and Child Health, Karachi, Pakistan; 4Southern General Hospital, Paediatric Neurosurgery, Glasgow, United Kingdom; 5Gartnaval General Hospital, Clinical Oncology, Glasgow, United Kingdom

Background: Craniopharyngioma accounts for 10% of all paediatric brain tumours and is classified as benign. These patients are at significant risk of long term morbidity relating to tumour site and insidious onset of symptoms leading to frequent diagnostic delay and treatment related effects.

Objective and hypotheses: This study aims to ascertain presenting features, symptom duration, treatment received and the incidence of immediate and long term complications.

Methods: Retrospective case note analysis from children diagnosed with craniopharyngioma over the last 30 years in the West of Scotland.

Results: Between 1981 and 2010, 27 patients aged ≤16.0 years were diagnosed, (15 male). Median age at presentation was 8.5 years (IQR 5.5-10.1) Median symptom duration was 3 months (IQR 1.8-6.0). Mean height, weight and BMI SDS at diagnosis were -1.31, 0.41, 1.71 respectively.

Presenting Features Frequency (%)
Headache 81.4
Lethargy 70.3
Visual Disturbance 51.9
Hydrocephalus 50.0
Cranial nerve palsy 48.1
Weight loss 11.0
Weight gain 11.0
Nausea and vomiting 19.5

Most patients (n=22) were treated with surgery and 11 required further surgery and radiotherapy for recurrence. 5 were lost to follow up. Common long term sequelae include growth hormone deficiency (77.7%), hyperphagia (40.7%), memory problems (33.3%). Visual problems were present in 70% post treatment compared with 54% pre-treatment.

Conclusions: Clinical presentation of craniopharyngioma and symptom duration in our population was highly variable. The majority presented with headaches, cranial nerve palsy and reduced visual acuity. There were significant endocrine, neurological, neurocognitive and ophthalmological morbidity at presentation and in the long term. Recurrence rates are high. It is difficult to compare outcomes over time due to changes in management during the 30 year period. All children with craniopharyngioma require lifelong surveillance to manage disease, treatment related complications and monitoring for potential late effects. This should be in a multidisciplinary paediatric craniopharyngioma clinic.

P2-d3-533 Endocrine Oncology 1
Insulinomas as the first sign of MEN1 syndrome in children
Maria Melikyan1; Yulia Averyanova3; Larisa Gurevich3; L Friis-Hansen4; Valentina Peterkova1; Henrik Christensen4
1Endocrinology research center, Paediatrics, Moscow, Russian Federation; 2Russian Children’s Clinical Hospital, Abdomen surgery, Moscow, Russian Federation; 3M. F. Vladimirsky regional clinical institute, Hystopathology, Moscow, Russian Federation; 4Copenhagen University Hospital, Clinical genetics, Copenhagen, Denmark; 5Odense University Hospital, Paediatrics, Odense, Denmark

Background: Insulinomas are extremely rare tumors in children. Insulinomas can develop as a part of the Multiple Endocrine Neoplasia syndrome type 1 (MEN1 syndrome), which however usually presents with parathyroid gland tumors. Early clinical and genetic diagnosis is very important for appropriate medical assessment and family counseling.

Objective: To investigate the clinical and genetic characteristics of Russian children with primary pancreatic insulinomas.

Materials and methods: Insulinoma was diagnosed biochemically and by imaging and was verified histopathologically. Sequencing of the MEN1 gene was performed in all patients. Families of the mutation carriers were studied after the molecular genetic verification. Follow up included screening for signs of the MEN1 syndrome (hormonal, imaging) and metastases in the cases of malignant tumors.

Results: Eight children aged 8-16 years were diagnosed to have pancreatic insulinoma. Three of the 8 children were found to have mutations in MEN1 gene and developed hyperparathyroiditis and hyperprolactinemia during next 10 years. In two of them, clinical testing of their relatives revealed the spectrum of MEN1 components, but with no clinical symptoms. Two patients had malignant insulinomas. One patient with wild type MEN1 gene developed hyperparathyroiditis.

Table 1. Clinical features and MEN1 gene investigation.

<table>
<thead>
<tr>
<th>Age (yrs)</th>
<th>Patient</th>
<th>Number of pancreatic tumors</th>
<th>Malignancy</th>
<th>Treatment</th>
<th>MEN1 sequencing</th>
<th>Family history</th>
<th>Results of clinical testing of relatives</th>
<th>Age (yrs) and findings at follow up</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>8</td>
<td>1 B</td>
<td>Pilocytic</td>
<td>c.1547T&gt;C</td>
<td>Gastric cancer in grandmother, thyroid cancer in aunt.</td>
<td>ND</td>
<td>21. Hyperprolactinemia</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>11</td>
<td>3 M</td>
<td>Pilocytic</td>
<td>c.830G&gt;C</td>
<td>PTH adenoma, liver + mesogastic mts</td>
<td>ND</td>
<td>21. HPTH, PTH adenoma, liver +</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>11</td>
<td>2 B</td>
<td>Pilocytic</td>
<td>c.936delC</td>
<td>Father: HPT, prolactinoma, parathyroid, pancreas adenoma</td>
<td>ND</td>
<td>19. PTH, PHTH adenoma, prolactinoma, adrenal nodular hyperplasia</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>11</td>
<td>1 B</td>
<td>Insulinoma</td>
<td>wt</td>
<td>Rectum cancer in grandfather</td>
<td>ND</td>
<td>15. None</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>13</td>
<td>1 B</td>
<td>Insulinoma</td>
<td>c.936delC</td>
<td>None</td>
<td>ND</td>
<td>18. None</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>13</td>
<td>1 B</td>
<td>Insulinoma</td>
<td>wt</td>
<td>Paternal line: thyroid cancer in patient</td>
<td>ND</td>
<td>17. None</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>16</td>
<td>1 M</td>
<td>Insulinoma</td>
<td>wt</td>
<td>Paternal line: thyroid cancer in aunt</td>
<td>ND</td>
<td>14. None</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>12</td>
<td>1 B</td>
<td>Insulinoma</td>
<td>wt</td>
<td>Paternal line: rectum cancer in grandfather</td>
<td>ND</td>
<td>14. None</td>
<td></td>
</tr>
</tbody>
</table>

B-benign, M-malignant, HPTH - hyperparathyroiditis, NDT-not done, wild type.

Conclusion: MEN1 syndrome should be suspected in all cases of pediatric insulinomas. Familial MEN1 gene sequencing is informative, even in cases with no clinical features of other MEN1 components and absence of suspicious family history. Insulinomas in children may be malignant, which stresses the need for histopathological investigations. Presumably sporadic cases with wild type MEN1 gene may develop to multiple tumor growth and should require specific and permanent medical assessment as well as genetically verified MEN1 cases.

P2-d3-534 Endocrine Oncology 1
Noonan syndrome and hematopoietic anomalies
Isabel Gonzalez Casado1; Ana Coral Barreda Bonis1; Luis Salamanca Fresno1; Ana de la Puente Arévalo1; Beenhia Zribi Zubicaray1; Julio Guerrero-Fernández1; Ricardo Gracia Bouthelier1
1La Paz Children’s Hospital, Endocrinology Unit, Madrid, Spain; 2Gregorio Marañón Hospital, Biochemical Laboratory, Madrid, Spain

Background: Noonan syndrome(NS) is characterized by short stature, facial, skeletal and cardiovascular abnormalities. PTPN11 mutations account for 50% of cases and promote non-receptor protein tyrosine phosphatase(SHP-2) gain of function through the RAS/MAPK pathway, which has been implicated in a

50th Annual Meeting of the ESPE
Horm Res 2011;76(suppl 2) 167
wide variety of cancers. NS patients are prone to develop malignancies, most commonly hematologic ones (juvenile myelomonocytic leukemia), and other bleeding diatheses anomalies (55%).

**Objective and hypotheses:** The aim of the study was to describe bleeding and oncological defects and if there was a genetic relationship.

**Methods:** Reported patients were collected from our Pediatric Endocrinology Hospital in agreement with van der Burgt’s criteria.

**Results:** 27 subjects were reviewed (74% male; 26% female), aged between 0.1-10 years at diagnosis. 9 patients (33%) had some hematologic-oncologic disorder (3 subjects hemostatic defects; 1 oncological and 5 both of them):
- Hemostatic defects involved: FVIII (2 cases) and FVIII (1 deficiency), thrombocytopenia (3), thrombopathy (1) and prolonged bleeding times (3), some of them coexisted and were mild.
- Oncological defects were: intracranial tumors (2 cases), acute lymphoblastic leukemia (1), hyperesinophilia (2), juvenile Xanthogranuloma (1) and unexplained hepatosplenomegaly-adrenalinsomnia (3). They were unaggressive. PTPN11 mutations were studied in 21 subjects, which were positive in 11 (52.4%); T42A, N558, D61N, Y65C, Q79K, E139D, Q256K, F285, N308D. 21C7 polymorphism in intron 7 was found in 3 cases. Our results showed a statistically significant association between hemostatic and oncological disorders, and near significant for hemostatic defects and PTPN11 mutations. No correlation was found between malignancies and PTPN11 mutations. Age and gender were not either significant.

**Conclusions:** We found a statistical association between hemostatic and oncological disorders in Noonan patients. Further studies are necessary in order to clarify genotypic-phenotypic correlation.

---

**P2-d1-536 Fat Metabolism, Obesity 1**

**Association between metabolic syndrome in children and early-onset atherosclerosis**

*Maria Dolores Martinez-Jimenez*; Ramón Carreño; Mercedes Gil-Campos; Concepción Aguilar; Josune Olza; Miguel Valle; Angel Gil

1Hospital Universitario Reina Sofia, Unidad De Endocrinologia Pediátrica, Servicio De Pediatría, Imibic, Hrs, Universidad De Córdoba, Córdoba, Spain; 2Universidad de Granada, Departamento De Bioquímica Y Biología Molecular 2, Granada, Spain; 3Hospital Valle de los Pedroches, Servicio de Análisis Clínicos, Pozoblanco, Spain

The metabolic syndrome (MS) favours the development of cardiovascular disease and diabetes mellitus type 2 in obese children. Childhood obesity favours a chronic proinflammatory and prothrombotic state which contributes to the establishment and development of cardiovascular dysfunctions.

**Aim:**
To measure arterial blood pressure and several biomarkers of endothelial activation, as in vitro markers for atherosclerosis, in obese prepubertal children.

**Material and methods:** A randomised, prospective, case-control study of 381 prepubertal children: 167 obese (body mass index [BMI] Z-score 3.71 ± 0.11; age 8.83 ± 0.15); 72 overweight (BMI Z-score 1.17 ± 0.05; age 9.24 ± 0.17) and 142 controls (BMI Z-score -0.25 ± 0.48; age 9.14 ± 0.12). The following were measured:
- anthropometric parameters (weight, height and waist girth [WG]; arterial blood pressure [BP]; and the endothelial activation biomarkers intercellular adhesion molecule-1 (sICAM-1), vascular cell adhesion molecule-1 (sVCAM-1), endothelial selectin (E-se) and plasminogen activator inhibitor-1, both active and total (PAI-1a, PAI-1t). These biomarkers were measured using specific monoclonal antibodies and Xmap technology (Luminex 200). Means were compared using ANOVA for independent groups, and bivariate correlations by Pearson’s coefficient; binary logistic regression analysis was performed for MS. Results: In the obese group, 13.2% of children met the criteria for MS. The same children had higher BP and higher levels of all endothelial activation biomarkers, except sVCAM-1, than the other groups. 94% of children with MS also had high blood pressure. Correlations were observed between these pathologies (obesity and MS) and both E-se and PAI-1a, while E-se, a specific biomarker for endothelial activation, was associated with a relative risk of 1.07 (p = 0.005) for MS. Conclusions: Obesity and MS correlate in a gradual manner with certain endothelial activation biomarkers, especially E-se and PAI-1a, and also with BP. This may indicate greater risk of inflammation and vascular damage at an early age.

---

**P2-d3-535 Endocrine Oncology 1**

**Endocrine, hypothalamic and neuro-developmental outcomes following treatment of craniopharyngioma**

*Esieza Ikazoboh*; Mehul Dattani; Helen Spoudeas

1UCL Institute of Child Health, Developmental Endocrinology, London, United Kingdom; 2University College London Hospital, London Centre for Paediatric and Adolescent Endocrinology, London, United Kingdom

**Background:** Craniopharyngiomas (CP) are rare benign pituitary-related tumours whose surgical excision and invasive nature may cause significant neuroendocrine morbidity and premature mortality.

**Objective:** To determine whether an age- and risk-based conservative surgical management approach avoiding further hypothalamic compromise has improved hypothalamic, endocrine and neuro-developmental outcomes at our centre in the last decade.

**Methods:** Retrospective case-note analysis of 33 patients (13 males) of median age 8.13 (2.0 – 15.8) years with confirmed CP diagnosed between 2012 and 2015. Patients had either debulking (open) surgery +/- radiotherapy (DXT) (GpA N11) at diagnosis. Patients had either debulking (conservative) cystic aspiration +/- DXT (GpC N11) at diagnosis. 20/33 patients in total had DXT. Endocrine, Hypothalamic (including Obesity) and Neuro-developmental morbidity (MS) was scored out of a maximum of 15, according to DeVile et al 1996 for historical CP comparison, higher scores implicating worse outcomes.

**Results:** 14 (42.4%) patients recurred, 5-year EFS was 50%. MS score rose significantly in obese group compared to those of healthy controls. When obese patients were hyperphagic, 5 sleep disordered, 2 had temperature dysregulation, and oncological disorders, and near significant for hemostatic defects and PTPN11 mutations. No correlation was found between malignancies and PTPN11 mutations. Age and gender were not either significant.

**Conclusions:** We found a statistical association between hemostatic and oncological disorders in Noonan patients. Further studies are necessary in order to clarify genotypic-phenotypic correlation.

---

**P2-d1-537 Fat Metabolism, Obesity 1**

**P300 auditory event related potentials in children with obesity: Is childhood obesity related with impairment in cognitive functions?**

*Mehmet Emre Taslarci*; Doga Turkahraman; Oguzhan Oz; Mehmet Yucel; Mustafa Taskesen; Ibrahim Eker; Ayhan Abaci; Rusen Dundaroz; Umit Hicir Ulus

1Gulhane Military Medical Academy, Pediatric Endocrinology, Ankara, Turkey; 2Gaziantep Children Hospital, Pediatric Endocrinology, Gaziantep, Turkey; 3Gulhane Military Medical Academy, Neurology, Ankara, Turkey; 4Gulhane Military Medical Academy, Pediatrics, Ankara, Turkey; 5Dokuz Eylul University, Faculty of Medicine, Pediatric Endocrinology, Izmir, Turkey

**Background:** Some studies recently show that obesity can be related with cognitive dysfunction in adolescent and adults. Little is known about how obesity can affect cognitive function in these patients.

**Objective:** To investigate alterations in P300 auditory event related potentials in children with obesity in order to detect changes in cognitive functions.

**Methods:** 50 children with obesity and 23 age and sex-matched healthy control subjects were included in the study. Laboratory tests were performed to detect dislipidemia and insulin resistance (IR). The latencies and amplitudes of P300 waves were measured in healthy and obese subjects with or without IR. The oddball paradigm was used in recordings of P300 auditory event related potentials.

**Results:** A significant difference was observed between groups regarding latency and amplitude of P300 component obtained from Cz electrode. The grand means of P300 latency was longer, and amplitude was decreased significantly in obese group compared to those of healthy controls. When obese group divided into two different subgroups; those with IR and without IR,
the grand means of P300 latency was longer, and amplitude was decreased significantly in subjects with IR compared to those of without IR.

**Conclusions:** Both decreased amplitude and prolonged latency of P300 is associated with IR in children. Moreover, in children with obesity, which demonstrates the impairment of neural activity associated with sensory and cognitive information processing in these children. Further studies are necessary to strengthen the current findings, and to determine the exact mechanism of cognitive impairment in obese children.

**P2-d1-538 Fat Metabolism, Obesity 1**

**Chemerin serum levels are associated with parameters of vascular inflammation in obese children**

Kathrin Landgraf1; Daniela Friebe2; Juergen Kratzsch3; Kathrin Dittrich4; Gunda Herbert5; Wieland Kiss5; Sandra Erbs6; Antje Koerner7

1 University of Leipzig, Hospital for Children and Adolescents, Leipzig, Germany; 2 University of Leipzig, Institute of Laboratory Medicine, Leipzig, Germany; 3 UFZ Helmholtz Centre for Environmental Research, Department of Environmental Immunology, Leipzig, Germany; 4 University of Leipzig, Heart Centre, Department of Cardiology, Leipzig, Germany

**Background:** The new adipokine chemerin plays a role in adipocyte differentiation and is associated with parameters of obesity and metabolic syndrome in adults.

**Objective and hypotheses:** In this study, we aimed to analyze chemerin serum levels and their association to measures of obesity and early-onset metabolic and vascular sequelae in children.

**Methods:** Serum levels of chemerin were quantified by ELISA in 69 lean and 105 obese children of the Leipzig Atherobesity childhood cohort.

**Results:** We revealed highly significant associations of chemerin serum concentrations with obesity-related parameters, such as BMI SDS (r=0.57, p<0.001), leptin (r=0.39, p<0.001), and skinfold thickness (r=0.54, p<0.001).

Mean serum chemerin concentrations were significantly higher in obese compared to lean children (117.82±26.35 ng/mL vs. 89.75±16.08 ng/mL, p<0.001). Moreover, chemerin levels were associated with parameters of glucose and insulin metabolism, as fasting plasma insulin levels (r=0.38, p<0.001) and Matsuda ISI (r=0.40, p<0.001). However, after adjustment for BMI SDS and age in partial correlation analyses, all significant correlations of chemerin with metabolic parameters were lost indicating an underlying association with obesity and fat mass. Furthermore, we identified significant BMI-independent correlations with general measures of inflammation, as hsCRP (r=0.50, p<0.001) and white blood cell count (r=0.30, p<0.001), as well as the parameters of endothelial activation, ICAM-1 (r=0.33, p<0.001) and E-selectin (r=0.30, p<0.001). Multiple regression analyses revealed that chemerin is the strongest predictor of ICAM-1 and E-Selectin serum concentrations independent of BMI SDS.

**Conclusions:** Chemerin serum levels are associated with obesity and metabolic syndrome in children. Moreover, in children chemerin is strongly associated with parameters of inflammation and endothelial activation suggesting a potential role of chemerin in vascular inflammation as an early stage of atherogenesis.

**P2-d1-539 Fat Metabolism, Obesity 1**

**Declining prevalence rates for overweight and obesity in German children starting school**

Ana Mosel1; Jochen Klenk2; Klaus Simon3; Heinrun Thaiss4; Thomas Reinherz5; Martin Wabitsch6

1 University of Ulm, Division of Pediatric Endocrinology, Diabetes and Obesity Unit, Department of Pediatrics and Adolescent Medicine, Ulm, Germany; 2 University of Ulm, Institute of Epidemiology and Medical Biometry, Ulm, Germany; 3 Institute for Health and Work of the German state North Rine-Westphalia, Düsseldorf, Germany; 4 Ministry for Labour, Social Affairs and Health of Schleswig-Holstein, Kiel, Germany; 5 Vestische Childrens- and Adolescents Hospital, Division of Pediatric Endocrinology, Diabetes and Nutritional Medicine, Datteln, Germany

**Background:** To estimate the development of prevalence rates for overweight and obesity in children starting school in Germany data for children’s height and weight out of the compulsory school enrolment examinations (SEE), conducted annually in every German federal state, were available. A former analysis of these data showed a marked increase of prevalence of overweight and obesity until 2004 (Moll, A. et al. 2007).

**Objective:** Aim of this project was to give an updated overview on the development of prevalence rates for overweight and obesity in children upon school entry by including recent data until 2008.

**Methods:** Data on measured height and weight from the yearly conducted SEE were obtained from all 16 German federal states. Overweight and obesity were defined by BMI > 90th and BMI > 97th age and gender related percentiles of German reference values, respectively.

**Results:** In 2008 the prevalence for overweight varied from 8.4 % in Saxony to 11.9 % in Bremen and Thuringia. The current prevalence rates for obesity ranged from 3.3 % in Brandenburg and Saxony till 5.4 % in Saarland. Interestingly, the current data from SEE show by the majority of the individual states that the prevalence for both overweight and obesity did not increase any more after 2004 and is even declining in some states compared to the former data inquiry. Absolute decrease of prevalence rates were up to 3% for overweight and 1.8% for obesity.

**Conclusion:** The current data from the SEE of individual German states are based on census and show by the majority that the prevalence of overweight and obese children starting school did not increase anymore and even declined in the last 4 years, respectively. It is supposed that the in the 1990s initiated and afterwards implemented measures for prevention have contributed to this positive development in Germany.

**P2-d1-540 Fat Metabolism, Obesity 1**

**Early risk factors of obesity in a French overweight pediatric population**

Heloise Perry1; Anne-Marie Bertrand2; Sylvain Quinart3; Pierre Rohrlich1; Veronique Negre4

1 RePPOP-FC, Pediatrie, Besancon, France; 2 CHU, Pediatrie, Besancon, France; 3 CHU, Pediatrie, Besancon, France

**Background:** In 2006, 17.8% of French children aged 3 to 17 were overweight and 3.5% of them were obese. Since the care of these children is difficult, especially if early onset and severe obesity, prevention should be initiated as early as possible.

**Objective and hypotheses:** To identify the early markers of childhood obesity during pregnancy of mothers and first year of life of overweight children included in a primary care program.

**Methods:** 341 children and adolescents (51.6% girls), entered a therapeutic program for overweight during 2006-2008. Median age was 11.5 (4-18). 62.5% were obese and 37.5% overweight. BMI mean Z-score was 3.45. Family and patient history were collected as well as child’s anthropology, and biological parameters.

**Results:** Prevalence of prepregnancy maternal overweight was 40.8% as compared to 12-14% in the general French women population, with a significant difference according to the overweight degree (46% of obese and 31% of overweight children, p=0,006). Gestational diabetes history was present in 18.1% of obese and 8.8% of overweight children (p=0.02). Prepregnancy maternal overweight and gestational diabetes linked were (p<0.0001). Overweight and obese children were more prone to macrosomy at birth (17.9% versus 4.4% to 9.6% in global french population) and were less breastfed (40.6% versus 73% in 2008 in the same region). The breastfeeding duration was short: mean of 1.6 months and less than 3 months in 79.6%. Metabolic syndrome incidence according to a child-adapted definition of the adult NCEP-ATPIII classification was 11.4%. Macrosomy at birth was a risk factor for metabolic syndrome (incidence 18.9% versus 9.7%, p=0.04).

**Conclusions:** This study in a French cohort confirms that maternal obesity and gestational diabetes are prenatal risk factors for childhood obesity. Perinatal factors include macrosomy at birth and short breastfeeding. Macrosomy is a risk factor for metabolic syndrome among overweight children. Identification of these risk factors should help to promote individualized prevention of childhood obesity.
Longitudinal association between insulin resistance and ambulatory blood pressure in obese children

Loredana M. Marcorvecchio; Rosanna Fucci; Valentina Chiaravalloti; Tommaso de Giorgi; Ebe D’Adamo; Francesco Chiarelli; Angelika Mohn
University of Chieti, Department of Paediatrics, Chieti, Italy

Background: Decreased insulin sensitivity is a common finding among obese children and is associated with an adverse cardiovascular-metabolic profile. It is alarming that this association can be already detected in prepubertal children, as it is likely that it will worsen during puberty due to physiological insulin resistance.

Objectives and hypotheses: Our aim was to assess changes in ambulatory blood pressure (ABP) from the prepubertal to pubertal period and their association with variations in insulin sensitivity and body mass index (BMI) in obese children.

Methods: Thirty obese children (15 males) underwent a first assessment when they were prepubertal (visit 1, age 9.0±1.2years) and were re-evaluated during puberty (visit 2, age 14.1±1.4years). At both visits anthropometric parameters were assessed, blood samples were collected for fasting insulin and glucose measurements, and a 24-hour ABP monitoring was performed. HOMA-IR was calculated as an index of insulin sensitivity.

Results: A significant increase from visit 1 to visit 2 was detected in 24-hour systolic BP standard deviation score (SBP SDS) (0.38±0.70 vs. 0.88±0.68, p<0.001) and nighttime SBP SDS (1.05±0.80 vs. 1.35±0.76, p=0.005). Mean BMI SDS did not significantly change from visit 1 to visit 2 (2.2±0.4 vs. 2.2±0.6), whereas both fasting insulin (12.2±5.4 vs. 21.2±8.0 mIU/L, p=0.001) and HOMA-IR (2.6±1.2 vs. 4.3±1.6, p=0.005) significantly increased. At both visits BMI SDS was significantly correlated with SBP SDS and HOMA-IR.

Conclusions: In this longitudinal study decreased insulin sensitivity was significantly related to ABP already during prepuberty, and the pubertal decline of fat storage by WAT could indicate a relative impairment of WAT fat storage capacity, subsequently favouring liver fat deposition.

Weight loss switches off the effect of PNPLA3 rs738409 polymorphism on liver enzymes in childhood obesity

Alessandra Amato; Emanuele Miraglia del Giudice; Anna Grandone; Grazia Cirillo; Anna Di Sessa; Grazia Cantelmi; Vincenzo Fierro; Gianluca Della Rotonda; Roberta Romano; Laura Perrone
Seconda Università di Napoli, Department of Pediatrics, Napoli, Italy

Background: Recently, a single-nucleotide polymorphism (rs738409) in the patatin-like phospholipase 3 gene (PNPLA3) has been associated with hepatic steatosis and increased liver enzyme levels in obese children and adolescents.

Objective and hypotheses: We evaluated, in a group of obese children, the effect of a weight loss program on the association between PNPLA3 rs738409 variant and liver enzyme levels.

Methods: We enrolled 129 Caucasian obese children and adolescents (66 girls), from 3 to 17 years. At baseline, all participants underwent physical examination, and liver enzymes were evaluated. All patients were genotyped for PNPLA3 rs738409 C to G variant, underlying the 1148M substitution, that has been detected by PCR and enzymatic (FOKI) digestion. All patients consumed a nutritionally balanced self-selected diet of common foods (60% of the recommended dietary energy allowances for age and sex) and underwent lifestyle modifications. After a reduction of BMI-SDS at least of 0.5 anthropometric and biochemical evaluations were repeated.

Results: Fifty-three patients were homozygous for the wild type allele (CC), 51 were heterozygous (CG) and 25 were homozygous for the minor allele (GG). At baseline circulating ALT and AST levels were increased significantly in children carrying the 148M allele (p<0.03 and p<0.04 respectively), adjusting for age, gender, pubertal stage, and BMI-SDS. After weight loss a significant decrease in serum ALT (p<0.0001) and AST concentration (p<0.002) was observed in all patients, but a greater reduction was found in children homozygous for the minor allele compared with those carrying the other genotypes (p<0.02 and p<0.04 respectively). At re-evaluation no differences among genotypes were found for serum ALT and AST levels.

Conclusions: Our findings suggest that, in obese children and adolescents, the effect of PNPLA3 1148M variant on liver enzymes is switched off by weight loss.

The presence of liver steatosis is associated with reduced body fat and high-molecular-weight adiponectin levels in obese children

Gabriel Angel Martos-Moreno1; Vicente Barrios2; Sara Sirvent3; Guillermo Martínez4; Federico Hawkins5; Jesús Argente6
1Hospital Infantil Universitario Niño Jesús, Universidad Autónoma, ISCIII, Instituto de Investigación La Princesa, Endocrinology, Madrid, Spain; 2Hospital Infantil Universitario Niño Jesús, Universidad Autónoma, ISCIII, Instituto de Investigación La Princesa, Endocrinology, Department of Pediatrics, CIBERobn, Madrid, Spain; 3Hospital Infantil Universitario Niño Jesús, Universidad Autónoma, ISCIII, Instituto de Investigación La Princesa, Endocrinology, Department of Pediatrics, CIBERobn, Madrid, Spain; 4Hospital Infantil Universitario Niño Jesús, Radiology, Madrid, Spain; 5Hospital Universitario 12 de Octubre, Universidad Complutense de Madrid, Endocrinology, Madrid, Spain

Background: Normal development and function of white adipose tissue (WAT) are essential for the prevention of ectopic fat deposition. The presence of liver steatosis from early ages could be indicating some degree of impairment of fat storage by WAT.

Objective and hypotheses: We aimed to compare serum metabolic profiles, body fat content and distribution, and leptin, leptin soluble receptor (sOB-R), total and high-molecular-weight (HMW) adiponectin levels between obese children with or without liver steatosis.

Methods: Eighty-three obese children (11.8±2.9 years; 41.1±1.5 BMI-SDS; 43 females/40 males) were studied. Liver steatosis was assessed by ultrasonography. Body composition (DXA) and abdominal MRI were performed. Lipid profile and uric acid, glucose and insulin levels were measured through an oral glucose tolerance test (OGTT). Leptin and total adiponectin were measured by RIA (Millipore®), and sOB-R and HMW-adiponectin by ELISA (BioVendor®; Millipore®, respectively).

Results: The 16 patients with liver steatosis (LS) had a similar mean age (12.1±2.6 years) and BMI (3.59±1.32) as the remaining 67 (no-LS; 11.8±3.0 years and 4.15±1.47 SDS). MRI analysis of abdominal WAT found no difference in the proportions of subcutaneous and visceral fat between groups. In contrast, DXAs revealed that LS patients had a lower percentage of body fat (36.62±5.33 vs. 40.51±4.46; p=0.05). This was associated with a lower leptin/sOB-R ratio (1.08±0.59 vs. 1.99±1.71; p=0.001), reduced levels of HMW-adiponectin (3.89±1.74 vs. 4.99±2.72 µg/ml; p=0.04) and a larger area under the curve (AUC) for insulin in the OGTT (184.0±108.8 vs. 132.6±89.5; p=0.005). No differences were observed in the remaining studied parameters.

Conclusions: The higher insulin levels following the OGTT in LS patients could be associated with the deposition of ectopic hepatic fat and the reduced HMW-adiponectin levels. Their lower percentage of body fat and leptin levels could indicate a relative impairment of WAT fat storage capacity, subsequently favouring liver fat deposition.
**P2-d1-544 Fat Metabolism, Obesity 1**

**Regulation of retinol binding protein 4 expression in human adipocyte by estrogen and dihydrotestosterone**

**Primož Kotnik; Daniel Tews; Martin Wabitsch; Pamela Fischer-Posovszky**

University of Ulm, Department of Pediatrics and Adolescent Medicine, Ulm, Germany

**Background:** Retinol binding protein 4 (RBP4), a novel adipokine suggested to link obesity and whole body insulin resistance/type 2 diabetes, is more abundantly expressed in female than male adipose tissue.

**Objective and hypotheses:** To determine the role of the two main sex hormones acting on adipose tissue, estradiol (E) and dihydrotestosterone (DHT), on the expression of RBP4 in human adipocytes in vitro.

**Methods:** Human SGBS adipocytes were stimulated with either pooled 10% female or male serum, E (10^{-11} - 10^{-7} M) or DHT (10^{-12} - 10^{-8} M) for 48 hours. Relative gene expression was determined using quantitative real time-PCR. Data was analyzed by a comparative 2−^ΔΔCT method. Results from 3 independent experiments were presented as mean ± SEM. For statistical comparison between groups Student t-test or ANOVA and post hoc Dunnett’s test were used. P < 0.05 was considered statistically significant.

**Results:** RBP4 mRNA expression significantly increased during adipogenic differentiation (p < 0.01) and SGBS adipocytes respond to both estrogen and androgen receptors, validating them as an appropriate model to test our hypothesis. Human serum significantly decreased RBP4 expression compared to the control (p < 0.05); female serum however tended to decrease RBP4 expression to a lesser degree than male (.50 ± .14 vs .38 ± .07; ratio of control ± SEM; n.s.). Both E and DHT resulted in increased RBP4 mRNA expression when compared to the control (E (10^{-10} M) 1.40 ± .04*; DHT (10^{-10} M) 1.34 ± .05*; ratio of control ± SEM; *P < 0.05).

**Conclusions:** Both E and DHT stimulate RBP4 mRNA expression to a comparable extent. Local differences in their concentrations and in the expression of their receptors could have a role in the regulation of RBP4 expression in adipocytes and could thereby at least in part explain the gender difference in RBP4 expression in adipose tissue.

**P2-d1-545 Fat Metabolism, Obesity 1**

**Fat mass and obesity associated (FTO) interferes with insulin action in human adipocytes**

**Daniel Tews; Pamela Fischer-Posovszky; Martin Wabitsch**

University Medical Center Ulm, Dept. of Pediatrics and Adolescent Medicine, Ulm, Germany

**Background:** The association between gene variants in FTO (fat mass and obesity associated) has been shown in different genome-wide association studies, FTO encodes a 2-oxoglutarate dependent demethylase and is expressed ubiquitously. FTO deficient mouse models point to a participation of this gene in the energy metabolism. However, its precise role in adipocyte metabolism has not been elucidated so far.

**Objective and hypotheses:** To examine the role of FTO concerning proliferation, differentiation and metabolism of human white adipocytes.

**Methods:** By using lentiviral-mediated expression of shRNA, we generated FTO deficient SGBS pre- and adipocytes. Successful knockdown was monitored by qPCR and Western Blot. Differentiation rate was determined by microscopic counting and by measuring triglyceride content. Insulin stimulated glucose uptake and de novo lipogenesis was examined by using radioactive labeled glucose.

**Results:** In human SGBS preadipocytes and adipocytes we reached a transduction efficiency of >90%. This led to an inhibition of FTO mRNA expression by 73% and to a total repression of FTO protein expression. Proliferation and differentiation rate of FTO deficient cells were not affected. Repression of FTO led to a significant inhibition of insulin stimulated glucose uptake by 12%. Simultaneously, insulin stimulated de novo lipogenesis was reduced by 30%. Expression of key enzymes of the lipogenic pathway was not affected.

**Conclusions:** In this study we could show that knockdown of FTO in human adipocytes results in a decline of insulin-dependent processes. This reduction is not based on an inhibition in the expression of key lipogenic enzymes but may rely on a reduced insulin responsiveness of FTO deficient SGBS cells.

**P2-d1-546 Fat Metabolism, Obesity 1**

**Therapeutic strategies of nonalcoholic fatty liver disease (NAFLD) in children and adolescents: a systematic review**

**Maris Rosaria Licenziati; Selvaggia Lenta; Pietro Vajo**

1AORN Santobono, Dept. of Pediatrics, Naples, Italy; “Università di Napoli “Federico II”, Dept. of Pediatrics, Naples, Italy

**Background:** NAFLD is the most common cause of chronic liver disease both in children and adults.

**Objective and hypotheses:** A systematic review of randomized controlled trials regarding therapeutic strategies of NAFLD.

**Methods:** A Pub Med search for NAFLD therapy in children up to January 2011 has been performed. Ten relevant pediatric articles concerning treatment have been selected.

**Results:** The first open-label pediatric trial [Lavine, 2000] with Vitamin E in children affected by NAFLD showed improvement of serum transaminases levels, without changes of BMI values and of bright liver at ultrasonography. Three studies with Vitamin E [Vajro, 2004; Nobili ,2006; Wang ,2008] did not find a higher efficacy of this treatment vs diet only. Currently an international large randomized double blind placebo controlled trial (TONIC) is under investigation [Lavine, 2010]. A pilot study with metformin conducted with biopsy-proven NASH and elevated ALT levels showed improvement in serum ALT levels and reduction in hepatic steatosis assessed with Magnetic Resonance Spectroscopy [Schwimmer, 2005]. A subsequent study [Nobili, 2008] did not show more benefit of metformin when compared with lifestyle intervention. Nadeau, 2009 showed a significant improvement of bright liver prevalence and severity, and of fasting insulin with metformin compared to placebo in obese insulin resistant adolescents. A RCT in obese children with NAFLD [Vajro, 2000] did not show any beneficial effect of UDCA on improvement in serum ALT levels or fatty liver. A double-blind RCT [Nobili, 2011] conducted in children with biopsy-proven NAFLD have demonstrated that dietary supplementation with docosahexaenoic acid (DHA) improves liver steatosis (as detected by ultrasonography) and insulin sensitivity after 6 months of treatment.

**Conclusions:** Available data still show that up to now no pharmacological agent has been consistently shown to be effective in obese children with NAFLD unable to lose weight. NAFLD prevention however still remains the primary target and requires a multidisciplinary approach.

**P2-d1-547 Fat Metabolism, Obesity 1**

**YKL-40 levels are associated with Ghrelin but not Leptin in obese prepubertal children**

**Ioannis Kyriopoulos; Charilaos Stylianou; Assimina Galli-Tsinooupolou**

Medical School, Aristotle University of Thessaloniki, 4th Department of Pediatrics, Thessaloniki, Greece

**Background:** Relationship between YKL-40 and peptides that regulate weight gain has not been studied before.

**Objective and hypotheses:** To further elucidate hormonal profile in childhood obesity and its interaction with inflammation markers we evaluated YKL-40, ghrelin and leptin levels and investigated their possible inter-associations.

**Methods:** Forty-one obese prepubertal children and 41 age- and gender-matched lean controls were included. Obesity was defined based on pooled international data for body mass index (BMI) linked to the adult cut-off point of 30 kg/m2. BMI linked to the adult cut-off point of 25 kg/m2 was used to define the controls. Insulin resistance was estimated using homeostasis model assessment for insulin resistance (HOMA-IR) index. Anthropometric, metabolic measurements as well as serum YKL-40, ghrelin and leptin levels in the fasting state were determined.

**Results:** Obese children had higher YKL-40 (p=0.008), leptin (p=0.001) and lower ghrelin levels (p=0.002) compared with controls. Insulin resistant individuals also showed higher YKL-40 (p=0.027) and leptin levels (p=0.014) compared with non-insulin resistant after adjusted for age, gender and BMI z-score whereas no significant difference in ghrelin levels was observed between these subgroups (p=0.199). Moreover, serum YKL-40 levels were significantly correlated with ghrelin (r=−0.359, p=0.014) but not leptin levels (r=0.169, p=0.261). Significant negative correlation between ghrelin and leptin levels was also found (r=−0.276, p=0.041). These findings remained unchanged when analyses were done separately for obese and non-obese subjects.
Conclusions: Results suggest that regulation of YKL-40 secretion may be a consequence of reduced ghrelin concentrations and not a consequence of total body fat accumulation associated with elevated leptin concentrations.

**P2-d1-548 Fat Metabolism, Obesity 2**

**Evaluation of body therapy sessions in a programme of therapeutic education for obese adolescents**

*Beatrice Jourde*

Jean-Luc Sudrez*

Marie Dupuy*

Farida Ghrib*

Hélène Degardin*

Isabelle Hubert*

Mélanie Glattard*

Catherine Arnaud*

Gwenaëlle Diene*

Maitê Tauber*

*CHU Toulouse, Hôpital des Enfants, Unité Endocrinologie, Toulouse, France; CERPP, Université de Toulouse le Mirail, UFR de Psychologie, Toulouse, France; RePEPOP Toulouse Midi-Pyrénées, Unité Endocrinologie, Toulouse, France; INSERM US58, Université Paul Sabatier Toulouse III, Epidémiologie et Santé Publique, Toulouse, France*

**Background:** Somatopsychic profile is an important aspect to take into account for obesity care.

**Objective and hypotheses:** To evaluate impact of body therapy sessions in a programme of therapeutic education on the somatopsychic profile of obese adolescents.

**Methods:** 56 adolescents (22 boys; age (median [interquartile range]): 13.5 [12-6-14.7] years) followed in our pediatrics department were enrolled in the study and divided into two groups: control group (n=30) and experimental group (n=26). Both groups benefited from the same therapeutic education program (5 sessions focused on food balance and physical activity). The experimental group received in addition sessions of body therapy and inter-sessions coaching by phone. Self-questionnaires were administered at the beginning (T0) and the end (T1) of the program, to explore body image (Body Prominence, Figure Rating Scale, Questionnaire of Body Preoccupation), self-esteem (Self Esteem Scale), anxiety (Revised children Manifest Anxiety Scale) and depression (Center for Epidemiologic studies – Depression Scale). Evolution between T0 and T1 was analysed in each group using Wilcoxon and Mac Nemar tests.

**Results:** In the control group, an improvement of self-esteem was observed (18.5 [14-23] in T0 vs 22 [17-25] in T1; P=0.0025). In the experimental group, a reduction of body consciousness (3[1-4] vs 2 [0-3]; P=0.0417) and social desirability (3.5 [1-6] vs 3 [1-5]; P=0.0340) was noted as well as an increase of self-esteem (19 [13-24] vs 23 [16-26]; P=0.0225). Moreover, a decrease of body preoccupation was noticed concerning “thorax” (69.2% vs 34.6%; P=0.0067) and also a trend for “hip” (65.4% vs 42.3%; P=0.10) and “muscular strength” (65.4% vs 42.3%; P=0.10) whereas no significant evolution was found in the control group.

**Conclusions:** Our multidisciplinary program of therapeutic education seems to generate an improvement of self-esteem in obese adolescents while body fat accumulation associated with elevated leptin concentrations.

**P2-d1-550 Fat Metabolism, Obesity 2**

**Does preterm birth influence fat mass and lipid profile in young adults?**

Petra Braukhoff*

Gerthe Kerkhof*

Ruben Willemsen*

Anita Hokken-Koelega*

ErasmusMC-Sophia Children’s Hospital, Paediatric Endocrinology, Rotterdam, Netherlands

*1CHU Toulouse, Hôpital des Enfants, Unité Endocrinologie, Toulouse, France; 2CERPP, Université de Toulouse le Mirail, UFR de Psychologie, Toulouse, France; 3RéPPOP Toulouse Midi-Pyrénées, Unité de Psychologie, Toulouse, France; 4INSERM U558, Université Paul Sabatier Toulouse III, Epidémiologie et Santé Publique, Toulouse, France*

**Background:** Dyslipidemia is one of the major risk factors for CVD and is characterized by raised levels of TC, LDLc, HDLc, TG, apoA-I, apoB, and Lp(a). Several studies reported on an association between size at birth and components of the lipid profile in later life. Most studies focused on subjects with a lower birth weight and either did not have information on gestational age, did not correct for it, or only included subjects born at term. It is, therefore, difficult to know whether the effect of a small size at birth on cardiovascular risk factors in later life was due to a small size for gestational age (SGA) or due to prematurity. Very few studies report on the specific effect of prematurity on fat mass and lipid levels in later life, and the results are inconclusive.

**Objective:** We investigated the effect of prematurity on fat mass and lipid levels in young adulthood.

**Methods:** 455 healthy subjects, aged 18 to 24 years were included. Total fat mass, trunk fat mass and limb fat mass were evaluated by DXA and fasting levels of TC, LDLc, HDLc, TG, apoA-I, apoB, and Lp(a) were measured. Differences between preterm and term subjects were assessed using multiple linear regression analyses, adjusted for age and gender. In addition, adjustments were made for other possible confounders in the analysis of fat mass and lipid levels.

**Results:** Differences in clinical characteristics, fat mass, and lipid levels between preterm and term subjects are shown in Table 1. Young adults born preterm tended to have more total body fat, trunk fat mass and limb fat mass than those born at term. Preterm subjects had a lower TC, LDLc, HDLc, TG, apoA-I, and apoB, and a higher ApoA-I than subjects born at term.

**Conclusions:** Young adults born preterm tend to have more fat mass, but their lipid profile is more favourable than those born at term.
is to evaluate the association of obesity in children with maternal obesity during pregnancy.

**Objective:**

To prevent child obesity, the purpose of the current study is to evaluate the association of obesity in children with maternal obesity during pregnancy.

**Methods:**

In this retrospective cohort study, we reviewed data collected from all births of obese mothers (BMI > 30 reported before pregnancy or measured during pregnancy). We determined blood glucose (BG) levels >140 mg/dl at 30th minute on oral glucose tolerance test (OGTT) of some obese patients. We aimed to evaluate importance of this finding. For this reason, we compared lipid profile, HOMA-IR, and systemic inflammatory markers in these children to those in obese children with impaired glucose tolerance (IGT) and normal glucose tolerance.

**Results:**

None of anthropometric parameters were found significantly different among three groups (p=0.01, p<0.004, and p<0.004, respectively). There was a moderately positive correlation between Lp-PLA2 levels and BMI Standard Deviation Score (SDS) in group 3 (r:0.461, p<0.05). IL-6, neopterin and Lp-PLA2 levels were associated with high blood glucose levels of 30th minute on OGTT (B:0.01, p<0.02, B:2.2, p<0.008 and B:0.01, p<0.01, respectively).

**Conclusions:**

In obese children with high glucose levels at 30th minute of OGTT, systemic inflammatory markers were even higher than obese children with IGT. As these markers were thought to be predictors of atherosclerosis or visceral adiposity, high glucose levels at 30th minute of OGTT may be an alerting sign for presence or onset of atherosclerosis and visceral adiposity.

<table>
<thead>
<tr>
<th></th>
<th>Preterm (n=167)</th>
<th>Term (n=288)</th>
<th>p-value</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender (male/female)</td>
<td>84/83</td>
<td>114/174</td>
<td>0.026</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>20.7 (1.7)</td>
<td>20.8 (1.6)</td>
<td>0.531</td>
<td></td>
</tr>
<tr>
<td>Gestational age</td>
<td>32.0 (2.2)</td>
<td>39.2 (1.6)</td>
<td>&lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Birth length SDS</td>
<td>-1.3 (1.9)</td>
<td>-1.5 (1.4)</td>
<td>0.201</td>
<td></td>
</tr>
<tr>
<td>Birth weight SDS</td>
<td>-0.5 (1.6)</td>
<td>-1.2 (1.4)</td>
<td>&lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Adult height SDS</td>
<td>-0.4 (1.9)</td>
<td>-1.1 (1.4)</td>
<td>&lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Adult weight SDS</td>
<td>-0.3 (1.2)</td>
<td>-0.7 (1.4)</td>
<td>0.002</td>
<td></td>
</tr>
<tr>
<td>BMI SDS</td>
<td>0.1 (1.2)</td>
<td>0.0 (1.3)</td>
<td>0.746</td>
<td></td>
</tr>
</tbody>
</table>

**Objective and hypotheses:**

We determined blood glucose (BG) levels=140 mg/dl at 30th minute in OGTT of some obese patients. We aimed to evaluate importance of this finding. For this reason, we compared lipid profile, HOMA-IR, and systemic inflammatory markers in these children to those in obese children with impaired glucose tolerance (IGT) and normal glucose tolerance.

**Methods:**

The study involved 80 obese [Body Mass Index (BMI)>95th percentile for age and sex] adolescents (48 female, 32 male), between 11-16 years of age. OGTT was performed to all participants. Depending on OGTT results, patients were divided into three groups as normal tolerance (group 1), IGT (group 2) according to International Diabetes Federation criteria, and BG levels=140 mg/dl at 30th minute (group 3). Lipid profile and systemic inflammatory markers [C-reactive-protein (CRP), interleukin-6 (IL-6), neopterin, fibrinogen, and Lipoprotein Associated Phospholipase A2 (Lp-PLA2)] were assessed in all adolescents.

**Results:**

None of anthropometric parameters were found significantly different among three groups (p=0.01, p<0.004, and p<0.004, respectively). There was a moderately positive correlation between Lp-PLA2 levels and BMI Standard Deviation Score (SDS) in group 3 (r:0.461, p<0.05). IL-6, neopterin and Lp-PLA2 levels were associated with high blood glucose levels of 30th minute on OGTT (B:0.01, p<0.02, B:2.2, p<0.008 and B:0.01, p<0.01, respectively).

**Conclusions:**

In obese children with high glucose levels at 30th minute of OGTT, systemic inflammatory markers were even higher than obese children with IGT. As these markers were thought to be predictors of atherosclerosis or visceral adiposity, high glucose levels at 30th minute of OGTT may be an alerting sign for presence or onset of atherosclerosis and visceral adiposity.
Methods: Biochemical parameters and IGF1, IGFBP3 and leptin levels were measured. mRNA expressions of IGF1, IGFBP3 and leptin genes were evaluated by qRT-PCR and 2-ΔΔCT IGFBP3 gene polymorphism by PCR-RFLP methods.

Results: Obese children had significantly higher HOMA-IR. IGFBP3 SDS was lower in obese children than controls, while IGF-1 SDS was not different. IGFBP3 SDS levels were negatively correlated with serum leptin levels (r=0.348, p<0.003, r=-0.341, p<0.001). We found a significantly higher serum leptin levels in obese children than controls. Furthermore, serum leptin levels were positively correlated with BMI SDS (r=0.272, p<0.01). IGF-1 ve IGFBP-3 gene expressions were lower in obese children, and leptin gene expression was found to be significantly higher in obese children compared to controls. The mRNA expression levels of IGF-1 was positively correlated with the mRNA expression levels of IGFBP-3 and leptin (r=0.587, p<0.003, r=0.474, p<0.01).

Conclusions: The decreased mRNA levels of IGFBP3 and IGF-1 along with IGFBP3 SDS might indicate that those children are at high risk of developing insulin resistance later in life. The positive and significant correlation between IGF-1, IGFBP-3 and leptin indicates the crosstalk between IGF and leptin signaling pathways.

Table 1: Anthropometric, laboratory findings and molecular analysis of the study groups.

<table>
<thead>
<tr>
<th></th>
<th>Obese(n=90)</th>
<th>Controls(n=84)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>12.5±3.5</td>
<td>11.8±3.4</td>
<td>0.000</td>
</tr>
<tr>
<td>BMI SDS</td>
<td>2.5±0.6</td>
<td>1.0±0.8</td>
<td>0.003</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>4.6±5.6</td>
<td>1.6±1.3</td>
<td>0.000</td>
</tr>
<tr>
<td>IGFBP3 SDS</td>
<td>-0.7±1.6</td>
<td>0.2±1.9</td>
<td>0.002</td>
</tr>
<tr>
<td>IGF-1 SDS</td>
<td>0.4±1.9</td>
<td>0.7±1.5</td>
<td>0.363</td>
</tr>
<tr>
<td>Leptin (ng/ml)*</td>
<td>19.1±11.5</td>
<td>0.5±2.9</td>
<td>0.000</td>
</tr>
<tr>
<td>IGFBP3 expression (AU)*</td>
<td>0.004±0.04</td>
<td>0.06±2.5</td>
<td>0.000</td>
</tr>
<tr>
<td>IGF-1 expression (AU)*</td>
<td>0.08±1.66</td>
<td>1.29±5.91</td>
<td>0.001</td>
</tr>
<tr>
<td>Leptin expression (AU)*</td>
<td>0.24±6.17</td>
<td>0.006±5.95</td>
<td>0.01</td>
</tr>
<tr>
<td>IGFBP3 polymorphism(%)</td>
<td>AA(wt):28.3</td>
<td>AC:42.4, CC:29.3</td>
<td>AA(wt):28.6, AC:32.9, CC:38.6</td>
</tr>
</tbody>
</table>

Continuous variables are presented as mean ±SD and were compared by t-test. p<0.05 is significant. AU, arbitrary units. Values were log transformed and given geometric mean±SD.

P2-d1-555 Fat Metabolism, Obesity 2

Lower YKL-40 levels in children conceived after ICSI in comparison to age matched naturally conceived controls: an early indicator of a favorable metabolic profile?

Alexandra Gkourogianni1; Alexandra Margeli1; Ioanna Kostera1; Maria Konsta1; Dimitrios Loutradis2; George Mastorakos2; Ioannis Papassotiriou3; Christina Kanaka-Gantenbein1; George P. Chrousos1

1Athens University Medical School, First Department of Pediatrics, Athens, Greece; 2Agia Sophia Children’s Hospital, Department of Clinical Biochemistry, Athens, Greece; 3Athens University Medical School, Division of In Vitro Fertilization, First Department of Obstetrics and Gynecology, Athens, Greece; 4University of Oxford, Nuffield Department of Clinical Medicine, Oxford, United Kingdom

Background: We previously reported that children conceived with conventional in vitro fertilization (IVF) have increased arterial blood pressure and circulating triglycerides, both early components of the metabolic syndrome (MS).

Objective and hypotheses: No discrete studies have been done in children conceived with intracytoplasmic sperm injection (ICSI). YKL-40 is mainly produced by macrophages and neutrophils. Recently, it was proposed that YKL-40 might be implicated in the low-grade inflammation of atherosclerosis, produced by macrophages and neutrophils.

Methods: The presence of metabolic syndrome parameters, including circulating YKL-40 levels, in children conceived by ICSI and naturally conceived (NC) children was compared.

Results: The main findings are: a) No significant differences in anthropometric and biochemical parameters between the two groups of children studied, while ICSI children had significantly lower SBP and hs-CRP values than NC children (15.0±6.8 vs. 27.5±15.4 ng/mL, p<0.001, c) YKL-40 correlated positively with BMI-z score in NC (p<0.01), while in ICSI children it correlated positively with SBP (p<0.03).

Conclusions: These findings indicate that children conceived with ICSI, in contrast to those conceived with conventional IVF, have a favorable metabolic profile. Whether this discrepancy is due to the different assisted reproduction method employed and/or because these children were studied at a younger, pre-pubertal age, is not yet known. Prospective follow-up studies will be informative.
P2-d1-556 Fat Metabolism, Obesity 2

The efficacy of laparoscopic sleeve gastrectomy (LSG) in adolescents with morbid obesity

Zohar Landau1; Gideon Karpius2; Aaron Hanukoglu1; Shirlit Abiri1; Anat Levy1; Francis Serour2

1Wolfson Medical Center, Pediatric Endocrinology, Holon, Israel; 2Wolfson Medical Center, Pediatric Surgery, Holon, Israel

Background: Laparoscopic sleeve gastrectomy (LSG) is still a controversial procedure as a single modality for morbid obesity. This procedure may be advantageous in adolescents since does not require foreign body placement and carries no risk of lifelong malabsorption.

Objective: To determine the short- and mid-term efficacy of LSG as treatment for morbid obesity in adolescents.

Methods: All patients (n=7, all female) participated in a weight loss program for at least 6 months without success. At referral, the mean age was 16.2 years (range 13.8-18), mean body mass index (BMI in kg/m²) was 44.4 (range 38.9-55.2). All suffered from various co-morbidities of obesity: type 2 diabetes, insulin treated (n=1), hypertension (n=5), fatty liver (n=2), obstructive sleep apnea (n=2) and pseudotumor cerebri (n=1). The decision for bariatric surgery was taken unanimously by the parents, patient, and the multi-disciplinary obesity team (pediatric endocrinologist, pediatric surgeon, dietitian and clinical psychologist).

Results: There were no intra- or postoperative complications. After a mean follow-up of 13.6 months (range 6-26 months), all patients but one had reduced BMI (mean BMI to 32.55). Throughout follow-up ruled out malnutrition or vitamin deficiency. In all subjects who lost weight, remission of the co-morbidities was noted.

Conclusion: In this preliminary study with 13.6 months of follow up LSG proven to be a safe and effective option for bariatric surgery in adolescents, resulting in a significant weight loss and remission of co-morbidities. We suggested LSG might be considered as single intervention for morbid obesity in adolescents. Long-term studies are needed to compare the results of LSG operation with other procedures.

P2-d1-557 Fat Metabolism, Obesity 2

The association of fatty liver with carotid intima media thickness in obese children

Leyla Akin1; Selim Kurtoglu1; Ali Yuklimaz2; Mumtaz Mustafa Mazicioglu3; Mustafa Kendirci3; Mustafa Akin4

1Erciyes University Faculty of Medicine, Pediatric Endocrinology, Kayseri, Turkey; 2Erciyes University Faculty of Medicine, Pediatric Radiology, Kayseri, Turkey; 3Erciyes University Faculty of Medicine, Family Medicine, Kayseri, Turkey; 4Erciyes University Faculty of Medicine, Pediatrics, Kayseri, Turkey

Background: The prevalence of nonalcoholic fatty liver disease (NAFLD) in children is becoming to increase which is parallel to the increased childhood obesity. Carotid intima media thickness (IMT) is a noninvasive indicator of subclinical atherosclerosis and well known to be strongly associated with risk of cardiovascular diseases. There is only a few study concerning c-IMT values in obese children with fatty liver disease.

Aim: We wished to investigate the presence of association between NAFLD and subclinical atherosclerosis in obese children.

Patients and methods: The study population consisted of 157 obese children (78 males and 79 females, mean age: 11.3±2.6, age range: 6-16 years). Anthropometric and body fat measurements (BFM) were performed. Fasting blood glucose, serum sialic acid, C-reactive protein, insulin, serum lipids and transaminases (ALT, AST) were measured. Oral glucose tolerance test was performed to all subjects. Elevated liver enzymes defined as serum ALT and/or AST levels above normal values for age. Subjects with elevated liver enzymes were referred for evaluation and causes other than obesity, including hepatitis B, hepatitis C, alpha-1-antitrypsin deficiency, Wilson disease, autoimmune hepatitis were excluded. The diagnosis of NAFLD was based on ultrasound scan. The c-IMT was measured by ultrasonography in the far (deeper) wall of left distal common carotid artery.

Results: There was statistically significant difference between the groups with or without NAFLD for the following parameters: BMI, waist circumference, middle arm circumference, neck circumference, systolic blood pressure, BFM, ALT and AST levels (p<0.05). Mean cIMT measurements were higher in the group with NAFLD (0.48 mm and 0.45 mm respectively, p<0.000).

Conclusion: We showed that cIMT, an early marker of atherosclerosis was higher in obese children with NAFLD than those in without NAFLD. Obese children with fatty liver should be treated urgently as they may have an increased risk of cardiovascular diseases.

50th Annual Meeting of the ESPEDr. John SmithHorm Res 2011;74(suppl 2)175

P2-d1-558 Fat Metabolism, Obesity 2

Genetic study of heterozygous familial hypercholesterolemia in pediatric patients

Sara Berrada1; Mirentxu Oyarzabal; Maria Chueca; Silvia Souto2; Teresa Molins Complejo Hospitalario de Navarra, Endocrinologia Paediatric, Pamplona, Spain

Introduction: Heterozygous familial hypercholesterolemia (FH) is an alteration of lipoprotein metabolism that associates increased risk of early cardiovascular disease. Its prevalence among the general population is between 1/200 and 1/500 individuals. Its pattern of inheritance is autosomal dominant and it is caused by alterations in the LDL receptor (LDLR) gene located on chromosome 19 and on the apolipoprotein B (ApoB) gene, with more than 700 different mutations having been reported. There are very few international references to the pediatric age group.

Material and methods: Retrospective, descriptive study of the medical records of 64 patients under the age of 15 years (50% males and 50% females); mean age at diagnosis: 7.7±3.5 years, being followed at the Pediatric Endocrinology Clinic belonging to a tertiary hospital for heterozygous FH. The analysis of FH-related mutations of the LDLR and apolipoprotein B was performed by means of a molecular biology DNA-array or biochip technique, called Lipochip®.

Results: In 92.3% there was a family history of FH with early vascular disease in up to 45%, which was what motivated their referral to our clinic. Total cholesterol was 294±46.3 mg/dL and LDL-cholesterol was 217±47.5 mg/dL. The genetic study revealed mutations in 47 cases (73%); 2 cases were pending, and the remaining were negative. Up to 16 different mutations have been observed, all on the LDLR gene, the most common one (45%) being M025+M080, located on exon 3. No mutations on ApoB have been found. Two patients are of particular interest with a null allele caused by a large deletion associated with more severe forms of the disease, one of whom had no prior diagnosis in the family.

Conclusions: We present a broad study conducted in children that enables us to intensify treatment and prevent early vascular disease. Furthermore, this study has made it possible to characterize relatives who had not been diagnosed.

P2-d1-559 Fat Metabolism, Obesity 2

FABP4 serum levels and gene expression in childhood obesity

Ardaç Yaman1; Fatmahan Atalar2; Hulya Gunoz2

1Istanbul Medical School, Pediatrics Department, Istanbul, Turkey; 2Istanbul Medical School, Department of Pediatric Endocrinology, Istanbul, Turkey

Background: Fatty acid binding proteins (FABP) are a family of cytoplasmic lipid chaperons binding to hydrophobic molecules reversibly and with high affinity. FABP4, a member of the family, was first isolated from adipocytes.

Objective and hypotheses: Reports linking FABP4 to obesity and metabolic syndrome have been increasing in number. Our aim was to investigate role of FABP4 in childhood obesity.

Methods: We compared 42 obese children, aged 12.7 ± 2, having a body mass index greater than 95th percentile to a control group consisting of 40 lean children (BMI < 85th percentile) aged 12 ± 1.9. Measurements for weight, height, BML, wrist circumference, hip circumference, weight to hip ratio were taken, standart deviation scores (SDS) calculated. Following a night fast, venipuncture was performed for serum FABP4 levels and FABP4 gene expression in peripheric leucocytes. Enzyme linked immunosorbent assay (ELISA) method was used to evaluate FABP4 serum levels, and PCR, Ct (2-ΔΔCt) method for gene expression.

Results: Using Mann-Whitney U test, serum FABP4 levels and FABP4 gene expression were both found to be higher among obese subjects. (Z = -2.029, p = 0.042; Z = -2.213, p = 0.027) Spearman’s ranked correlation coefficient
method used to evaluate correlations. A positive correlation between BMI SDS, waist circumference, and serum FABP4 levels was found; \( r = 0.329, p = 0.003 \); \( r = 0.372, p = 0.001 \) while no correlation existed between serum FABP4 levels and height SDS, waist to hip ratio; \( r = 0.023, p = 0.838; r = 0.154, p = 0.194 \). FABP4 gene expression in peripheral leukocytes were positively correlated to BMI SDS, waist SDS and waist to hip ratio; \( r = 0.272, p = 0.015; r = 0.346, p = 0.002; r = 0.269, p = 0.019 \).

Conclusions: Serum FABP4 levels and peripheral FABP4 gene expression found out to be related to obesity formation and further studies with larger study groups warranted.

P2-d1-560 Fat Metabolism, Obesity 3
Early nutritional changes induce sexually dimorphic long-term effects on bodyweight gain and the response to sucrose intake in adult rats
Esther Fuente-Martín1; Miriam Granado1; Cristina García-Cáceres1; Miguel A. Sánchez-Garrido2; Francisca Díaz2; Laura Maria Frago4; Manuel Tena-Sempere2; Jesús Argente2; Julie Chowen2
1Hospital Infantil Universitario Niño Jesús-Universidad Autónoma de Madrid, Ciberobn, ISS-Princesa, Pediatrics-Endocrinology, Madrid, Spain; 2University of Córdoba. Instituto de Investigaciones Biomédicas de Córdoba (IMIBIC)-Ciberobn, Cell Biology, Physiology and Immunology, Córdoba, Spain; 3Hospital Infantil Universitario Niño Jesús Ciberobn, ISS-Princesa, Endocrinology, Madrid, Spain; 4Hospital Infantil Universitario Niño Jesús-Universidad Autónoma de Madrid, Ciberobn, ISS-Princesa, Pediatrics-Endocrinology, Madrid, Spain

Background: Early nutrition can have long-lasting effects on body weight (BW) and modify the response to nutritional challenges in adulthood, with males and females often responding differently.

Objective and hypotheses: We hypothesized that both neonatal under- and over-nutrition affect BW and the response to a diet rich in sucrose in adulthood and that these effects are sexually dimorphic.

Methods: To induce changes in neonatal food intake, litters were adjusted to 4 (L4), 12 (L12) or 20 pups (L20), on the day of birth with equal numbers of males and females in each litter and with no difference in mean BW between groups.

Results: When pups were weaned, rats from L4 (n = 16 for each sex) weighed more, and those from L20 (n = 18 for each sex) less, than rats from normal size litters (L12; n = 20 for each sex; ANOVA = p<0.0001 for both sexes). Mean daily food intake (ANOVA = p<0.02) and weight gain (ANOVA = p<0.05) during the five weeks post-weaning were inversely related to litter size in males, but unaffected in females. When fed a high sucrose diet (2 weeks of a 33% sucrose solution instead of water in half of each group starting on PND50) all groups of both sexes increased total energy intake (ANOVA = p<0.0001), but had either normal (L12) or decreased (L4 and L20) weight gain (ANOVA = p<0.0001). Yet, sucrose intake increased serum leptin levels in L4 and L20 males (ANOVA = p<0.0001), with no effect of sucrose or litter size on serum ghrelin or insulin levels.

Conclusion: In conclusion, the long-term response to early nutritional changes differed between the sexes, with females being more capable of maintaining a normal BW under some conditions. Furthermore, the weight gain response to increased energy intake in the form of sucrose differed as a result of neonatal nutritional status.

P2-d1-561 Fat Metabolism, Obesity 3
Metabolic syndrome risk is increased with coexistence of elevated alanine aminotransferase and obesity in adolescents
Shin Hye Kim1; Sang Shin Park2; Mi-Jung Park1
1Ankara University School of Medicine, Pediatric Endocrinology, Ankara, Turkey; 2Ankara University School of Medicine, Pediatric Molecular Genetic, Ankara, Turkey

Background: Elevation of alanine aminotransferase (ALT), a surrogate marker of nonalcoholic fatty liver disease, has been reported as an indicator of insulin resistance.

Objective and hypotheses: We aimed to investigate the association between ALT levels and metabolic risk factors among adolescents.

Methods: Data of 5164 adolescents (2709 boys and 2455 girls; aged 10 to 18 yrs) from the Korean National Health and Nutrition Examination Surveys 1998–2009, were analyzed for the association of ALT and metabolic risk factors.

Results: The prevalence of elevated ALT (>40 U/L) was increased with obesity level in both genders (21.7% in obese boys, 10.4% in overweight boys, 1.4% in normal-weight boys, P<0.0001; 9.5% in obese girls, 2.1% in overweight girls, 0.5% in normal-weight girls, P<0.0001). Waist circumference, body mass index, blood pressure, triglyceride, homeostasis model assessed and prevalence of metabolic syndrome (MetS) were increased, whereas HDL cholesterol was decreased from the first quartile to the fourth quartile for ALT. Subjects with highest ALT quartile had a higher risk of MetS than those in the lowest quartile after adjusting for age [odds ratio (95% confidence interval): 5.7(3.5-9.3) for boys, 3.2(2.0-5.3) for girls]. In the lowest ALT quartile and normal-weight subjects, the prevalence of MetS was 2.7%, whereas in the highest ALT quartile and obese subjects, the prevalence of MetS was 49.7%. (P<0.0001).

Conclusions: The prevalence of MetS was dramatically increased with coexistence of elevated ALT and obesity. Further cohort studies are needed to determine the usefulness of ALT levels to predict the development of MetS in adolescents.

P2-d1-562 Fat Metabolism, Obesity 3
Evaluation of hypercoagulability in obese children with thrombin generation test and microparticle release: effect of metabolic parameters
Zeynep Siklari1; Gonul Ocalt; Merih Berberoglu1; Bulent Hacihamdoolgolu1; Senay Savas Erdeve1; Yonca Egin2; Nejat Akar2
1Ankara University School of Medicine, Pediatric Endocrinology, Ankara, Turkey; 2Ankara University School of Medicine, Pediatric Molecular Genetic, Ankara, Turkey

Background: Obesity is known as associated with a hypercoagulable state. Insulin resistance which is settled in the center of obesity, is not known how exactly affects the coagulation system. Thrombin is one of the central enzymes in blood coagulation and measurement of a subject’s capacity to generate thrombin is very useful as a reflection of a thrombotic phenotype compared to conventional coagulation tests. Microparticles are membrane nanofragments with procoagulant and proinflammatory properties.

Objective and hypotheses: Thrombin generation test (TGT) and microparticle levels were not studied in obese children extensively. It is aimed to determine if any differences in the coagulation system between obese and normal weighed children exist with the use of TGT and microparticles release.

Methods: Hundred-twenty obese (BMI >95th) and 38 healthy children were included to the study. For evaluate microparticle release, plasma samples studied by using STA-PROCOAG-PPL Kit (DIAGNOSTICA STAGO SAS), and STAR48. Thrombin Generation Test measured by Thrombin Generation Kits including Trombin Calibrator, PPP-Reagent 5pM, FluCa-Kit. Glucose metabolism, insulin, lipids levels were evaluated by routine methods. In obese children, the relationship between TGT parameters, microparticles release and metabolic parameters were analysed.

Results: An increase of thrombin generation and microparticles levels reflecting as hypercoagulability were found in obese children. A significant negative correlation was found between microparticles release time and BMI SDS. Unexpectedly, hyperinsulinaemia could not find a risk factor for hypercoagulability and non of the parameters of TGT has been showed to be related to parameters of metabolic syndrome in this age group.

<table>
<thead>
<tr>
<th>Obese Group</th>
<th>Control Group</th>
<th>p values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>120</td>
<td>38</td>
</tr>
<tr>
<td>Age (years)</td>
<td>11.4 ± 3.2</td>
<td>11.2 ± 3.2</td>
</tr>
<tr>
<td>Microparticles release time (sec)</td>
<td>26.41 ± 8.03</td>
<td>90.36 ± 37.07</td>
</tr>
<tr>
<td>Lag time (min)</td>
<td>3.01 ± 4.98</td>
<td>5.31 ± 2.41</td>
</tr>
<tr>
<td>ETP (Mx min-1)</td>
<td>1903.36 ± 536</td>
<td>1701.32 ± 555.87</td>
</tr>
<tr>
<td>Peak (mm)</td>
<td>418.62 ± 25.7</td>
<td>321.58 ± 111</td>
</tr>
<tr>
<td>Tt peak (min)</td>
<td>5.54 ± 0.98</td>
<td>8.54 ± 2.56</td>
</tr>
<tr>
<td>Start tail</td>
<td>18.48 ± 3.83</td>
<td>26.43 ± 8.58</td>
</tr>
</tbody>
</table>

Conclusions: This study emphasized that obese subjects are under the risk of
“hypercoagulability beginning from childhood period” independent of metabolic syndrome parameters. Management of obesity would lead to improvement of hypercoagulability, and this will reflect to cardiovascular health.

**P2-d1-563 Fat Metabolism, Obesity 3**

**Bone mineral density in prepuberalt and pubertal obese children**

Shokery Awadalla

Hospital San Jose, Pediatric Endocrinology, Bogota, Colombia

**Background:** Obese children are at more risk of fractures that can be due to more impact on growing bones or reduced quality of bones. Reports about bone mineral density in children with obesity are conflicting, and this could be due to the time of study of children whether pubertal or prepubertal.

**Objective and methods:** To evaluate the effect of obesity on bone health we studied 40 children with obesity (22 girls and 18 boys) with body mass index (BMI) > 95 percentile, and 32 normal weight children (20 girls and 12 boys) with BMI < 85 percentile. Each group was subdivided into prepubertal and pubertal. Patients with endocrine or bone diseases or receiving medication that might affect bone health was excluded.

**Results:** In group 1, 18 (10 girls, 8 boys) were prepubertal with Tanner stage 2-3, and in group 2, 14 (8 girls, 6 boys) were pubertal with Tanner stage 2-3. Age in both groups was between 7 and 12 years.

Bone Mineral Density (BMD) was assessed by dual energy x-ray absorptiometry (DEXA) in lumbar vertebra and values were expressed in Z score and were adjusted for weight.

Pubertal children in both groups had similar BMD with Z score of 0.7 in group 1, and 0.65 in group 2 with no sex differences. There was a significant difference between groups in prepubertal children 0.45 in group 1 and 0.6 in group 2 (p < 0.05).

This difference may explain the increased incidence of fractures in this group.

**Conclusions:** Children with obesity have lower BMD than normal weight before puberty and this difference disappear with puberty. The effect of sex steroids in addition to the stress by the overweight on the bones may help to improve BMD and compensate the lack of exercise.

**P2-d1-564 Fat Metabolism, Obesity 3**

**A study on beta-cell function and insulin sensitivity in obese children with and without acanthosis nigricans**

Feng Xiong, Li-Xia Si, Min Zhu, Pei-Yun Lei, Lei-Li Deng

Children’s Hospital of Chongqing Medical University, Pediatric Endocrinology, Chongqing, China

**Objective and hypotheses:** To investigate the beta-cell function and insulin sensitivity in obese children with and without acanthosis nigricans (AN).

**Methods:** From Jul. 2006 to Mar. 2010, 187 obese children were recruited in endocrine clinic of Children’s Hospital of Chongqing Medical University. All of them were divided into three degrees by WHO weight for height standard, 11 of mild obesity, 84 of moderate and 92 of severe. According to whether appearing AN, obese children were divided into obesity with AN group (60 cases) and non-AN group (127 cases). An Oral glucose tolerance test and insulin releasing test were performed in the obese children above moderate degree obesity. The insulin resistance and insulin sensitivity were assessed by HOMA-IR and insulin sensitivity index (ISI). The pancrea beta-cell function were assessed by HOMA-beta which labels the basic insulin release, 1-phase and 2-phase Stumvoll insulin release index and index of insulin increment to glucose increment at 30 min.

**Results:** (1) The total incidence of AN in obese children is 32%, in which mild obese group is zero, 19% in moderate group and 47.8% in severe group, respectively.

(2) The FINS, INS180, HOMA-IR and HOMA-beta in AN group were significantly higher than non-AN group, P < 0.05. Meanwhile, ISI in AN groups is lower than non-AN group, P < 0.05.

While there is no significant differences in plasma glucose levels and insulin levels in half hour, one hour and two hours and index of insulin increment to glucose increment at 30 min, P < 0.05. Although the 1-phase and 2-phase Stumvoll insulin release indexes are different, but it is no statistical differences.

**Conclusions:** AN is more frequently associated with the severity of obesity.

Obese children with AN have higher insulin resistance and pancrea beta-cell dysfunction, the latter was displayed the characteristics of high levels of basal insulin release: in fasting insulin and 3 hours insulin.

**P2-d1-565 Fat Metabolism, Obesity 3**

**FGF23 - not only the marker of late cardiovascular complications, but a novel factor contributing to metabolic syndrome**

Małgorzata Wojcik1; Katarzyna Domek-Oltarzewska1; Dominika Janus1; Dorota Drozdzi2; Krystyna Sztetko2; Jercy Starcyko1

1Jagiellonian University Collegium Medicum, Department of Pediatric and Adolescent Endocrinology, Krakow, Poland; 2Jagiellonian University Collegium Medicum, Department of Pediatric Nephrology, Krakow, Poland; 3Jagiellonian University Collegium Medicum, Department of Biochemistry, Krakow, Poland

**Background:** Fibroblast growth factor-23 (FGF23) is a hormonal regulator of circulating phosphate and vitamin D levels. In recent years it has been noticed, that besides playing a key role in the pathogenesis of calcium-phosphorus disorders, FGF23 may be an indicator of cardiovascular complications. Its potential role in the development of metabolic syndrome is discussed.

**Objective and hypotheses:** The aim of the study was to examine for a possible correlations between FGF23 serum level and body composition, blood pressure, and selected parameters of glucose, insulin and fat metabolism in adolescents with simple obesity.

**Methods:** In 68 (35 girls/33 boys) adolescents (mean age 13.9 years) with simple obesity [mean BMI SDS 4.9 (5% CI 4.4-5.4)] the levels of FGF23, total cholesterol, LDL cholesterol, HDL cholesterol and triglycerides were measured. Standard oral glucose tolerance test was performed with the assessment of fasting and after 120° post-load of glucose and insulin levels; the insulin resistance index HOMA-IR was calculated.

**Results:** Regardless of gender, there was a significant negative correlation between the FGF23 level and fasting insulin level [r= (-)0.3, as well as HOMA-IR [r= (-)0.29]. The correlation between FGF23 level and lipid fractions was noticed, but not significant.

**Conclusions:** FGF23 seems to be a novel factor contributing to insulin resistance. Further investigations are needed to define it’s role in the development of metabolic syndrome.

**P2-d1-566 Fat Metabolism, Obesity 3**

**A low glycemic diet prevents glucose intolerance and diabetes in a mouse model of neonatal overnutrition**

Marta Ramon Krauel1; Thais Pentinat1; Judith Cebria1; Ruben Diaz1; Josep C. Jimenez-Chillariz1

1Hospital Sant Joan de Déu, Pediatric Endocrinology, Esplugues Llobregat, Barcelona, Spain; 2Fundació Sant Joan de Déu, Endocrinology, Esplugues Llobregat, Barcelona, Spain; 3Hospital Sant Joan de Déu, Endocrinology, Esplugues Llobregat, Barcelona, Spain

**Background:** Epidemiological and clinical data show that rapid weight gain early in life is strongly associated to obesity and type2 diabetes.

**Hypothesis:** A low glycemic diet (LGD) designed to reduced insulin resistance and post-prandial hyperglycemia, could be a safe and effective way to prevent this phenotype in children that exhibit rapid weight gain in early life.

**Objective:** To prevent the diabetic phenotype in adult life using a mouse model of neonatal overfeeding (ON) and accelerated growth rate.

**Research design and methods:** We have previously described a mouse model of neonatal overnutrition and accelerated growth rate by culling offspring at 4 days of age. Here, we will randomly assigned ON mice onto two groups: ON (n=16) and LGD (n=16). LGD mice were maintained on a standard chow diet (SD) or LGD, provided ad libitum, from weaning until age 5 months.

**Results:** Growth velocity on the ON group is greater than controls (C) during the first 2 weeks of life (ON=2.79±0.2 vs C=2.59±0.35g/week; p<0.01) and, thus, ON mice are significantly heavier than C by age 2 weeks (ON=12.44±0.70 vs C=10.01±0.95g; p<0.01). Accelerated weight gain may be partly explained by the transitory increased food intake (ON=0.061±0.012 vs C=0.045±0.017g/ body weight; p<0.01). As we have previously published.
that maintained a weight loss of >1 SD of BMI (WL, n=25) and those without weight loss (NWL, n=7) and a control group of the same pubertal stage (CG, n=8).

Results: In WL and NWL there was an increase (p<0.01) in the L/A and L/sObR indexes compared to CG at baseline. Both indexes decreased in the WL group as puberty progressed (p>0.05), with no changes in CG and NWL. There was a positive correlation between leptin and BMI, PC, and % TBF and a negative correlation with sObR. Finally, we observed a positive correlation between L/A and HOMA index and L/A and L/sObR with % TBF in WL and NWL throughout the study period.

Conclusions: The leptin/adiponectin index is a good marker of insulin resistance and the percentage of total body fat in obese patients during puberty.

P2-d1-569 Fat Metabolism, Obesity 3

Obesity and insulin sensitivity in Down syndrome - possible roles of adiponectin and leptin
Asa Myrelid1; Per Frisk1; Mats Stridsberg2; Göran Annerén1; Jan Gustafsson1; 1Uppsala University Hospital, Women's and Children's Health, Uppsala, Sweden; 2Uppsala University Hospital, Medical Sciences, Uppsala, Sweden; 3Uppsala University Hospital, Genetics and Pathology, Uppsala, Sweden

Background: Overweight and obesity (BMI >25 and >30 kg/m2, respectively) are common clinical characteristics in several genetic disorders including Down syndrome (DS). The mechanisms behind obesity in DS are unknown, but there are several possible contributory factors, e.g. endocrine alterations, reduced physical activity, hypotonia, and eating behaviour. In the general population insulin resistance with its metabolic consequences, such as cardiovascular disease, is closely related to obesity. Previous results from our group indicate a relative peripheral and hepatic insulin resistance in DS subjects. However, obesity and regional fat deposition seems to be of limited importance in development of cardiovascular disease, since this is relatively uncommon, in DS.

Objective: In the present study we hypothesized that insulin resistance in DS is related to abnormal secretion of adiponectin and leptin.

Methods: We examined serum levels of leptin and adiponectin, insulin sensitivity and body composition in ten young adults (24-32 years of age, 5 F) with DS and compared the data with those of eighteen healthy matched controls.

Results: The DS subjects had a higher BMI, meeting the criteria for overweight/obesity in 9 out of 10 subjects. The fat mass proportion and HOMA index were increased in comparison with controls. There were no differences between the groups regarding levels of adiponectin, neither as absolute figures nor in relation to fat mass. Levels of leptin were markedly increased in the DS subjects both as absolute (34.2 vs. 8.6 µg/L, p<0.008) and relative figures (1.28 vs. 0.37 µg/L/kg fat, p<0.004).

Conclusions: The normal levels of adiponectin in the DS subjects, despite overweight/obesity, indicate an altered regulation of adiponectin secretion in this population. The hyperleptinemia, which was further enhanced after adjustment for fat mass, is possibly an effect of leptin resistance. Consequently, hyperleptinemia, in the absence of hypoadiponectinemia, may be involved in the development of decreased insulin sensitivity in DS.
NAFLD in children studies. However, gender and pubertal development have been found to be relevant factors in children and adolescents with this disease. **Objective and hypotheses:** The aim of this study was to identify potential differences between the gender and pubertal development with respect to clinical and laboratory variables studied in adolescents with NAFLD. **Methods:** Two hundred twenty obese including 99 males and 121 females between 4 and 18.6 years olds were evaluated. The diagnoses of liver steatosis were made by liver ultrasonography. Pubertal developments were evaluated as prepubertal, pubertal and post pubertal stage by gender. Anthropometric and serum biochemical variables were measured. **Results:** NAFLD prevalence in the study sample was 34.5 %. According to pubertal development NAFLD were determined to be prepubertal 7.1%, pubertal 30.8% in girls and prepubertal 37.9%, pubertal 48.8% in boys. Biochemical parameters (AST, ALT, GGT, Triglyceride, HDL, HOMA-IR) were different between gender and pubertal development. These values were shown Table 1 and 2. **Conclusions:** NAFLD was found different between gender and pubertal development. Prepubertal and pubertal obese boys have been higher NAFLD risk than girls. NAFLD is most common in pubertal boys. A possible explanation for the differences between genders and pubertal stage could be that sexual hormones may be mediators of NAFLD start. The limitation of this study, is that sex steroid levels weren’t evaluated. Further research should be conducted to determine the differences in metabolism and sex steroid levels between males and females in the NAFLD.

---

**P2-d2-572 Fat Metabolism, Obesity 4**  
**Metabolic syndrome in Egyptian adolescents**  
Mona Hafez1; Isis Ghal1; Nermine Salab1; Mona Mamdouh2;  
Shereen Gaffar1; Abbeer Atef2; Ghada Anwar2; Hend Megawed2;  
Fatma El Mougy1  
1Cairo University, Diabetes Endocrine Metabolic Pediatric Unit,  
Children Hospital, Cairo, Egypt; 2Diabetes Endocrine Metabolic Unit,  
Children Hospital, Cairo, Egypt

**Background:** Despite that obesity and overweight are becoming a problem among Egyptian youth, limited studies addressed the problem of metabolic syndrome in Egyptian children and adolescents. **Objective:** To detect the prevalence of metabolic syndrome in a group of obese, overweight and lean Egyptian adolescents and to explore the relationship between adiposity and individual metabolic risk factors. To assess which of the fat compartments is related to metabolic complications. **Study design:** This is a prospective study which included 600 adolescents (ages 10 to 16 years) divided into 400 obese (BMI> 95 percentile), 100 overweight (BMI> 85 and <95 percentile) and 100 lean (BMI< 85 percentile) adolescents. **Methods:** Baseline measures included: height, weight, BMI, waist circumference (WC), blood pressure, pubertal staging according to Tanner, lipid profile (TG, HDL). Insulin resistance (IR) was determined using a HOMA model. For 65 of obese patients, TFM was measured by (DEXA) and abdominal VFM and SCFM were measured by MRI. **Results:** The prevalence of MS in Egyptian adolescents in this study is (18%) in obese, 6-6 % in overweight versus 2% in lean adolescents. Among obese patients all fat compartments were strongly associated with a parental history of obesity. Boys have more VFM than girls after adjustment for age, family history of obesity and BMI. For the same degree of obesity, adolescents with more VFM were more at risk of developing MS. WC is the best predictor of VFM. Factor analysis showed that obesity and impaired glucose metabolism, dyslipidemia, and increased blood pressure explained 69 % of the total variance in the data. Linear logistic regression analysis using HOMA IR as the dependent variable showed that acanthosis nigricans and WC are the only independent predictive factors of insulin resistance. **Conclusions:** A close relation is noticed between parameters of adiposity and blood pressure, lipid profile and glucose metabolism. Worsening obesity has significant deleterious effects on each component of MS.

---

**P2-d1-571 Fat Metabolism, Obesity 3**  
**Symptoms of ADHD and depression in a clinical population of obese children: relations with BMI and metabolic syndrome**  
Panagiota Pervanidou1; Alexandra Gkourogianni1; Ioannis Syros2;  
Christina Kanaka-Gantenbein1; George P. Chrousos1  
1Childhood Obesity Clinic, First Department of Pediatrics, Athens  
2University Medical School, “Aghia Sophia” Children’s Hospital, Athens, Greece

**Background:** High rates of behavioural and emotional symptoms have been reported in obese children. Both behavioural and neuroendocrine pathways contribute to high rates of comorbidity between emotional and behavioural disorders and obesity. **Objective and hypotheses:** The aim of our study was to assess the prevalence of symptoms of Attention Deficit Disorder (ADD) and depression among a clinical population of children followed at the Childhood Obesity Clinic of our Department. Furthermore, to examine relations between such symptoms and BMI z-scores and parameters of the metabolic syndrome. **Methods:** A total of 74 (51 females) obese children, aged 7-16, were examined for symptoms of ADD and depression. Parents were asked to complete an ADHD questionnaire, based on DSM-IV diagnostic criteria. Children completed the Children’s Depression Inventory (CDI). **Results:** Eleven out of 74 (14.8%) obese children presented with a high level (over the diagnostic cut-off point) of symptoms of ADD. Sixteen children (21.6%) presented with diagnostic symptoms of depression. Seven additional children (9.5%) had comorbid diagnostic symptoms of ADD and depression. The group with ADD symptoms had significantly higher BMI z-scores compared to the total non-ADD group (p=0.002). Furthermore, children with ADD symptoms had higher BMI z-scores compared to obese children with symptoms of depression and obese children with comorbid symptoms. However, the group with comorbid symptoms had the higher prevalence of parameters of the metabolic syndrome compared to the other groups. **Conclusions:** A high prevalence of symptoms of ADD and depression is noted in a clinical population of obese children compared to the general population. Children with ADD symptoms, have a higher BMI z-score while children with comorbid ADD and depression have a greater number of parameters of Metabolic Syndrome compared to the other groups.
Background and aims: Food intake is related to the release of satiety hormones of the gastrointestinal-endocrine cells. The physiology of these hormones during normal meal intake remains unclear. Further it is known that impaired insulin sensitivity and diabetes mellitus typ 2 is a consequence of overweight. This study was designed to compare the responses of three gut hormones (GLP-1, PYY and ghrelin) in response to meal intake between lean and obese adolescents. Alongside we investigated the insulin sensitivity of these children.

Methods: A total of 16 obese (8 males, 8 females; mean BMI 29.7 ± 0.68 kg/m²) and 14 lean (5 males, 9 females; mean BMI 20.7 kg/m²) adolescents, aged 8-16 years, consumed a mixed 500 kcal meal (bread, butter and chocolat milk) during which plasma measurements of ghrelin, GLP-1 and total PYY were obtained. Fasting glucose and insulin levels were used to calculate insulin sensitivity using the HOMA index.

Results: Fasting ghrelin tended to be lower in obese than in lean subjects (not significant); after meal intake ghrelin levels fell in both groups. Fasting GLP-1 and PYY levels were similar in both groups; after meal intake PYY and GLP-1 levels were significantly attenuated in the obese group (p<0.05, respectively). Obese adolescents had higher fasting and postprandial glucose and insulin levels (p<0.05, respectively). Postprandial glucose levels in the lean subjects returned to baseline by 120 min after the start of the meal. Glucose levels in the obese subjects still remained on average higher than their baseline at 120 min. The HOMA index was as expected significantly higher in obese compared to lean subjects (p<0.01) suggesting insulin resistance.

Conclusions: 1. Meal-related changes in ghrelin, GLP-1 and PYY in obese subjects show a significantly different hormone release compared to normal-weight subjects. This disturbed release could contribute to the development or worsening of obesity. 2. Obese adolescents have a increased insulin resistance.

P2-d2-574 Fat Metabolism, Obesity 4

Serum leptin level and its association with bone mineral density in obese children
Mariya Vishnevskaya1; Anzhalika Solntsava2
1Belarusian State Medical University, Pediatrics, Minsk, Belarus; 2Belarusian State Medical University, 1st Department of Childrens Disease, Minsk, Belarus

Background: Total body composition by dual energy X-ray absorptiometry (DEXA) can be used as a specific method of measuring body composition for identify the individual risk of metabolic complications in children.

Objective and hypotheses: To investigate serum leptin level and its relationship with bone mineral density in obese children.

Methods: 91 children with obesity (male / female = 52/39, mean age 12.81 ± 0.5 years) were involved. DEXA was used to determine bone mineral density and body composition. BMI were average 95 percentile for age and sex. Serum leptin was measured using ELISA by “DRG Diagnostics” (USA). All the data were performed non-parametric (ANOVA, test of Mann-Whitney U) and parametric (t-Student criterion) methods.

Results: The mean BMI m/f (29.7 ± 0.68/28.3 ± 0.52 kg/m²) (p < 0.05). DEXA data (m / f): bone mineral density-1.12 ± 0.02 / 1.06 ± 0.03 g / cm² (p<0.01), Android-51.05 ± 0.93 / 53.98 ± 0.58 (p<0.05), Glucose-46.93 ± 0.61 ± 0.55 (p<0.05), A/G-1.09 ± 0.02 / 1.05 ± 0.01 (p<0.05), total body-42.5 ± 0.71 / 46.5 ± 0.54 (p<0.05), fat mass-3595.9 ± 2228.08 ± 34/216 ± 1374.2 g (p<0.01), lean mass, 47,804 ± 2,421,72 / 38790,33 ± 1451,34 g (p<0.05), leptin, 24,574,93 ± 2,422,83/41343,36 ± 14966,6 g (p<0.05), HOMA index was as expected significantly higher in obese compared to lean subjects (p<0.01) suggesting insulin resistance.

Conclusions: 1. Serum leptin level was positively correlated with mineral density (m / f) puberty (r = 0.3 / 0.46), early puberty (r = 0.32 / 0.49), puberty (r = 0.4 / 0.58) groups, more expressed in girls (p<0.01). 2. Serum leptin level was positively correlated with mineral density. A lean mass was significantly correlated with mineral density in boys.
Long lasting positive effects of a multidisciplinary intervention program to treat obesity in preschool children

Gianni Bocca1; Ronald Stolk1; Pieter Sauer2
1Beatrix Children’s Hospital, University Medical Center Groningen, Pediatric Endocrinology, Groningen, Netherlands; 2University Medical Center Groningen, Epidemiology, Groningen, Netherlands; 3Beatrix Children’s Hospital, University Medical Center Groningen, Pediatrics, Groningen, Netherlands

Background: Childhood obesity has increased, especially in the younger age groups. Little is known about lifestyle interventions programs to treat obesity in preschool children, notably about the effects after the program has finished.

Objective and hypotheses: To assess the effect of a multidisciplinary intervention program for the treatment of overweight and obese preschool children.

Methods: This study was a three months randomized multidisciplinary intervention program in overweight and obese children, aged 3 – 6 years. The intervention group followed a multidisciplinary program including dietary interventions, exercise sessions and behavioural therapy for parents. Primary outcome was the change in body mass index (BMI) directly after the intervention period, and nine months later. Secondary outcome were a change in waist- (WC) and hip circumference (HC), % body fat (%BF) and subcutaneous (SF) and visceral fat (VF) assessed by ultrasound.

Results: The results are shown in the table.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Baseline</th>
<th>3 months</th>
<th>12 months</th>
</tr>
</thead>
<tbody>
<tr>
<td>BMI (kg/m²)</td>
<td>(mean (SD))</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>21.2 (2.9)</td>
<td>20.4 (3.1)</td>
<td>20.6 (3.6)</td>
</tr>
<tr>
<td>Usual care</td>
<td>21.1 (2.7)</td>
<td>20.6 (2.6)</td>
<td>21.2 (2.8)</td>
</tr>
<tr>
<td>z-BMI (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>2.7 (1.0)</td>
<td>2.3 (1.0)</td>
<td>2.2 (1.0)</td>
</tr>
<tr>
<td>Usual care</td>
<td>2.7 (1.0)</td>
<td>2.4 (0.9)</td>
<td>2.4 (0.8)</td>
</tr>
<tr>
<td>WC (cm) (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>64.6 (7.1)</td>
<td>64.4 (8.3)</td>
<td>66.2 (10.1)</td>
</tr>
<tr>
<td>Usual care</td>
<td>65.2 (8.0)</td>
<td>66.6 (8.0)</td>
<td>66.0 (7.1)</td>
</tr>
<tr>
<td>z-WC (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>2.7 (1.0)</td>
<td>2.4 (1.2)</td>
<td>2.4 (1.2)</td>
</tr>
<tr>
<td>Usual care</td>
<td>2.7 (1.0)</td>
<td>2.8 (1.0)</td>
<td>2.4 (0.9)</td>
</tr>
<tr>
<td>HC (cm) (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>69.0 (7.9)</td>
<td>67.6 (8.1)</td>
<td>70.2 (8.6)</td>
</tr>
<tr>
<td>Usual care</td>
<td>68.7 (7.2)</td>
<td>68.1 (6.9)</td>
<td>71.5 (6.6)</td>
</tr>
<tr>
<td>%BF (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>29.0 (7.8)</td>
<td>28.1 (7.9)</td>
<td>27.7 (8.6)</td>
</tr>
<tr>
<td>Usual care</td>
<td>28.6 (6.3)</td>
<td>28.9 (6.2)</td>
<td>29.8 (5.8)</td>
</tr>
<tr>
<td>SF (cm) (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>1.8 (0.7)</td>
<td>1.7 (0.7)</td>
<td>1.7 (0.8)</td>
</tr>
<tr>
<td>Usual care</td>
<td>1.7 (0.7)</td>
<td>1.6 (0.7)</td>
<td>1.7 (0.8)</td>
</tr>
<tr>
<td>VF (cm) (mean (SD))</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multidisciplinary</td>
<td>4.4 (1.4)</td>
<td>4.0 (1.0)</td>
<td>3.8 (0.9)</td>
</tr>
<tr>
<td>Usual care</td>
<td>4.3 (0.8)</td>
<td>4.0 (0.8)</td>
<td>4.4 (0.9)</td>
</tr>
</tbody>
</table>

In total, 75 children were included in the study. Both groups lost weight during the study, but children in the usual care group regained most weight after the intervention period, in contrast to the multidisciplinary treatment group. Δ-BMI (12 months vs. baseline) = -0.6 and 0.1 for the multidisciplinary and usual care group, respectively. Δ-z-BMI was -0.5 and -0.3 for the multidisciplinary and usual care group, respectively. %BF decreased with 1.3% in the multidisciplinary group, in contrast to an increase of 1.3% in the usual care group. VF decreased with 0.6 cm in the multidisciplinary group and increased with 0.1 cm in the usual care group.

Conclusions: A multidisciplinary obesity treatment program in preschool children results in lasting weight loss, compared to usual care treatment. Also, %BF and VF decrease during multidisciplinary treatment, in contrast to an increase observed with usual care treatment.
to avoid diagnostic and therapeutic errors. Medical approach has to take in account the peculiarity of this syndrome: one have to search systematically the hypventilation, to pay attention in prescribing growth hormone and to avoid oxygen in case of hypercapnia.

**P2-d2-580 Fat Metabolism, Obesity 4**

Metabolic syndrome in obese Hispanic and caucasian pediatric population: the influence of ethnicity

**Patricia Erroz; Sinzi Stanescu; Noelia Alvarez; Maria Martín-Frias; Milagros Alonso; Raquel Barrio**

Ramon y Cajal Hospital, Pediatric Endocrine and Diabetes Unit, Madrid, Spain

Background: Spain has experienced marked increases in the prevalence of childhood obesity in the last decade. Hispanic immigrant population seems to be a group with special risk for obesity.

Objective and hypotheses: Analyze the influence of ethnicity in the prevalence of metabolic syndrome (MS) in obese Caucasian and Hispanic pediatric populations.

Methods: Retrospective study of 616 obese children and adolescents [BMI>2SD (Hernández-2004)], 142 Hispanics and 474 Caucasians. Mean age: 11.03±2.8. MS was defined by modified Cook criteria-2003: obesity and 2 or more of the following features: HDL-cholesterol <40 mg/dl, triglycerides (TG) >110 mg/dl, systolic or diastolic blood pressure >90 (Task Force 2004) or impaired glucose metabolism (ADA). We also assessed: hepatic function (ALT, GGT and liver ultrasounds), family history of MS, Hba1c and insulin resistance (IR) (HOMA-IR=[3 ml/m(2) in pubertal and >2.4 in prepubertal subgroups], basal insulin <15 mU/ml and 10.5, respectively, or insulin during oral glucose tolerance test >150 mg/ml or >75 2h). Statistical analysis: SPSS-12.0 (multivariate analysis, logistic regression).

Results: The two populations were comparable regarding mean age, pubertal status and BMI-SD. MS was present in 30% of Hispanics vs 15% in Caucasians [OR=2.4 (IC95%:1.5-3.8) p<0.005, and OR=2.5 adjusting for sex, BMI-SD and pubertal stage] at puberty: 41% vs 20% (p=0.005) and prepubertally: 22% vs 11% (p<0.05). Hispanics also had more IR (57% vs 43% p=0.005).

<table>
<thead>
<tr>
<th></th>
<th>Hispanics</th>
<th>Caucasians</th>
</tr>
</thead>
<tbody>
<tr>
<td>1BP (%)</td>
<td>19.3</td>
<td>24.5</td>
</tr>
<tr>
<td>[HDL]/TG (%)</td>
<td>45/34.8</td>
<td>23/<em>15.1</em>*</td>
</tr>
<tr>
<td>IFG/GTT/INDET (%)</td>
<td>2.2/5.6/0.7</td>
<td>4.6/2.7/2.5</td>
</tr>
<tr>
<td>Diabetes (%)</td>
<td>0.7</td>
<td>0.6</td>
</tr>
<tr>
<td>Hba1c (%)</td>
<td>5.8±0.49</td>
<td>5.2±0.48*</td>
</tr>
<tr>
<td>Acanthosis (%)</td>
<td>62</td>
<td>25*</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>3±2.4</td>
<td>2±2.1*</td>
</tr>
<tr>
<td>ALT/GGT (mg/dl)</td>
<td>31.2±28.9/22.3±21.4</td>
<td>20.5±9.8/16.1±5.9*</td>
</tr>
<tr>
<td>Hepatic steatosis ultrasound (%)</td>
<td>30.7</td>
<td>25.7</td>
</tr>
<tr>
<td>Family history MS (%)</td>
<td>78</td>
<td>87*</td>
</tr>
</tbody>
</table>

*p<0.05 **p<0.005 IFG: impaired fasting glucose. IGT: Impaired glucose tolerance. INDET: indeterminated glycemia.

Conclusions: MS in obese Hispanic children and adolescents doubles the prevalence in their Caucasian counterparts, representing higher risk for metabolic and cardiovascular disease.

**P2-d2-581 Fat Metabolism, Obesity 4**

Is infantile obesity a risk for obesity or metabolic abnormality at 12 years?

**Satomi Koyama1; Go Ichikawa; Yuzuru Yamazaki2; Akiko Kanbara; Naoto Shimura; Toshihiro Sairenchi; Osamu Arisaka1**

1Dokkyo Medical University, Department of Pediatrics, Mibu, Tochigi, Japan; 2Dokkyo Medical University, Department of Public Health, Mibu, Tochigi, Japan

Background: The age of adiposity rebound (AR), when body mass index (BMI) starts to rise after infancy, is thought to be an origin of obesity in later life. We have already reported that children who exhibited an earlier AR were associated with the higher BMI and atherogenic metabolic status at 12 years of age. We have also reported that weight gain during infancy is not associated with the timing of AR, suggesting that infantile obesity does not relate to childhood obesity.

Objective and hypotheses: We investigated if infantile obesity is a risk for obesity or metabolic abnormality at 12 years.

Methods: A total of 296 children (157 boys and 139 girls) in the community were enrolled in the study. Serial measurements of BMI from 4 months to 12 years were carried out prospectively. We calculated the age of AR, defined as the age which the lowest BMI occurred during this period. The subjects were divided into 2 groups according to BMI at 8 months. There were no significant differences of the data of BMI (mean 21.4 (SD 3.3) in obese group vs 19.8 (3.5) in non-obese group), LDL-cholesterol (98 (26) vs. 92 (22) mg/dl), HDL-cholesterol (65 (10) vs. 65 (11) mg/dl), triglyceride (62 (27) vs. 64 (27) mg/dl), atherogenic index (1.6 (0.4) vs. 1.6 (0.5)), LDL particle size (26.6 (0.4) vs. 26.7 (0.4)mm), systolic blood pressure (110 (11) vs. 108 (10)mmHg) and diastolic blood pressure (60 (8) vs. 61 (7)mmHg) in the each groups.

Results: There was no relationship between the BMI at 8 month and the age of AR. None of the subjects belonging to the obese group showed higher BMI (over 90 percentile) at 12 years. There were no significant differences of the data of BMI (mean 21.4 (SD 3.3) in obese group vs 19.8 (3.5) in non-obese group), LDL-cholesterol (98 (26) vs. 92 (22) mg/dl), HDL-cholesterol (65 (10) vs. 65 (11) mg/dl), triglyceride (62 (27) vs. 64 (27) mg/dl), atherogenic index (1.6 (0.4) vs. 1.6 (0.5)), LDL particle size (26.6 (0.4) vs. 26.7 (0.4)mm), systolic blood pressure (110 (11) vs. 108 (10)mmHg) and diastolic blood pressure (60 (8) vs. 61 (7)mmHg) in the each groups.

Conclusions: None of the obese infant at 8 months shows obesity and the risks of metabolic abnormality at 12 years of age. It seems infantile obesity is not a risk for obesity or metabolic abnormality in the future.
in the regulation of body weight.

**Objective and hypotheses:** Genetic variations at the LEPR locus were screened in a selection of 50 Russian overweight children, then an association study between genotypes and obesity phenotypes was performed in these overweight patients who were prescribed a low calorie diet.

**Methods:** We examined 50 subjects aged 3-18 y who gained an average of 15.8 kg (range 10-52 kg) during 2.8-10 y with frequency of occurrence of several polymorphisms in candidate genes of obesity and some lifestyle factors. Control group consisted of 50 non-obese children. Polymorphisms were determined in the LEPR-gene (LEPR Gin223Arg) and in the PPARGC1 (PGC-1 alpha) gene.

**Results:** The genotype and allele frequencies of the LEPR Gin223Arg polymorphism were significantly different between normal-weight and overweight plus obese groups. A trend was observed for homozogotes individuals with higher mean BMI compared to heterozygotes individuals presenting intermediate mean BMI. However, individuals bearing the heterozygote variant of PPARGC1 presented a higher BMI than wild-type homozgyotes.

**Conclusions:** The study population consisted of 40 obesity (Group 1:20 obesity with insulin resistance and Group 2: 20 obesity without insulin resistance) and 15 healthy control children. Whose age, sex, height, weight, BMI, and puberty status are matched. Immediately after sampling for blood glucose, insulin and GLP-1. The subjects were administered a standard oral glucose tolerance test. Another sampling was performed for plasma glucose, insulin and GLP-1 levels at 0, 60 and 120 minutes.

**Results:** Body mass index were different in obesity and healthy control group children. Plasma GLP-1 levels both 60,120 min after OGTT; were different between groups 1-2 and groups 1-3 (p<0.05). Plasma GLP-1 levels were not correlated with endogenous insulin secretion, in either group. Serum GLP-1 showed no significant correlations between anthropometric data, glucose, insulin, lipid profile and HOMA-IR in the obese groups (Group 1 and Group 2).

**Conclusions:** The concentrations fasting and postprandial plasma GLP-1 reduced in obesity with insulin resistance after OGTT. This decreased levels of GLP-1 study results suggest a role for GLP-1 in the obesity pathogenesis.
intima-media thickness (IMT), an in vivo marker for atheromatosis, in children.

Aim: To assess carotid IMT and various characteristic comorbidities (dyslipidaemia, insulin resistance and arterial hypertension) in obese prepubertal children.

Material and methods: A randomised, prospective, case-control study of 381 prepubertal children: 167 obese (body mass index [BMI] Z-score 3.71 ± 0.11; age 8.83 ± 0.15); 72 overweight (BMI Z-score 1.17 ± 0.05; age 9.24 ± 0.17) and 142 controls (BMI Z-score -0.25 ± 0.48; age 9.14 ± 0.12). The following were measured: anthropometric parameters (weight, height and waist girth [WG]; arterial blood pressure (BP); plasma triglycerides (TG); total cholesterol and fractions (LDLc and HDLc); glucose and insulin. In a subsample (62 obese, 11 overweight and 29 controls) carotid IMT was measured by high-resolution ultrasonography. Means were compared using ANOVA for independent groups, and bivariate correlations by Pearson’s coefficient.

Results: In obese prepubertal children, an association was observed between comorbidities such as insulin resistance, higher BP, and a range of dyslipidaemias, elevated TG and HDLc hypocholesterolaemia. Carotid IMT tended to be greater in obese children than in the other groups.

Conclusions: Insulin resistance, dyslipidaemia and high BP are characteristic findings in obese prepubertal children; since these may be linked to early-onset atherosclerosis, increased carotid IMT should be evaluated in obese children.

Conclusion: The presence of elevated transaminases as a surrogate marker of NAFLD in obese children and adolescents is strongly associated with metabolic syndrome. Patients meeting criteria for MS should be evaluated for NAFLD, not only with liver enzyme measurements but with liver ultrasonography as well.

Background: Higher cortisol levels and hyperactivity of the hypothalamic-pituitary adrenal axis might play a role in the development of MS. Pediatric data are scanty.

Objective and hypotheses: To evaluate ACTH and cortisol association with MS, its components, family history of metabolic derangements and birth weight in pediatric obesity.

Methods: Cross-sectional design. 271 Caucasian obese children and adolescents (age range: 1.8-18.0 yrs) were evaluated for: family and birth history, clinical examinations, fasting morning ACTH, cortisol, glucose, insulin, lipid profile and OGTT test. We divided patients according to pediatric NCEP criteria for MS. Overnight dexamethasone tests were performed when needed.

Results: 62.4% of the subjects had MS (73.9% hypertension, 58.4% HDL-cholesterol <10th percentile, 23.1% triglycerides > 90th percentile, and 8.2% IFG, IGT or type 2 diabetes). Children and adolescents with MS presented higher ACTH (mean±SEM: 31.1±2.0 vs 21.9±1.5 pg/ml; p<0.001) and cortisol levels (13.1±0.6 vs 11.0±0.4 µg/dl; p<0.003) than those without MS. Increasing features of MS were associated with higher ACTH, but not cortisol, also when adjusted for confounding factors (p<0.001). Subjects with altered glucose or HDL-cholesterol or triglycerides had higher ACTH, but not cortisol levels (p=0.01). Subjects with hypertension had increased levels of both ACTH and cortisol levels (p=0.02) if the cut-off was 95th percentile, and only cortisol (p=0.05) if the cut-off was reduced to 90th percentile. ACTH and cortisol levels were associated with insulin resistance. Acanthosis index and striae rubrae were not dependent by them. Subjects with a family history of hypertension (p=0.003) or low birth weight had higher cortisol, but not ACTH levels. The other family histories are independent by ACTH and cortisol.

Conclusions: In obese pediatric subjects, MS is associated with higher ACTH and cortisol levels. The features of MS could be differently modulated by ACTH and cortisol. Cortisol secretion seems mainly involved in hypertension and its family history.

Background: Non-alcoholic fatty liver disease (NAFLD), the most common cause of liver disease in children is associated with obesity and insulin resistance. Therefore a relationship with metabolic syndrome (MS) has been anticipated and NAFLD has been proposed to be a component of MS.

Objective and hypotheses: The aim of the study was to determine the association between NAFLD and the presence of MS in obese children.

Methods: The data from a cohort of 114 obese children and adolescents (BMI >95th centile) with elevated (>40U/L) serum alanine (ALT) or aspartate (AST) aminotransferases were compared to the data of 114 obese children with normal aminotransferase levels, well matched in age, sex and BMI SDSs.

Results: The components of the metabolic syndrome were significantly worse in the group of obese children with elevated ALT and/or AST than in the group with normal transaminases. Children with presumed NAFLD had higher fasting glucose, insulin, total cholesterol, triglycerides, waist circumference, blood pressure and lower high density lipoprotein cholesterol levels than obese children without NAFLD. Children with NAFLD were significantly more likely (46% vs 23%, p<0.01), to have MS than obese children without NAFLD. The odds ratios for elevated transaminases in children with MS was 6.1% (95%CI: 2.5 to 10.3).

Conclusion: The presence of elevated transaminases as a surrogate marker of NAFLD in obese children and adolescents is strongly associated with metabolic syndrome. Patients meeting criteria for MS should be evaluated for NAFLD, not only with liver enzyme measurements but with liver ultrasonography as well.
Fat Metabolism, Obesity 5

Conclusions: An increase in lean mass (p<0.05), whereas abdominal MRI revealed that weight loss resulted in a decrease in body fat percentage (p<0.001) and total and HMW-adiponectin (both p<0.05) and decreased insulin levels (B:15.21±9.33 vs. 6M:10.04±5.22 µg/ml; p<0.05). Patients achieving a BMI reduction >1.5 SDS also increased total and HMW-adiponectin (both p<0.001) and HMW-adiponectin levels (B:4.71±2.75 vs. 6M:6.13±3.37 µg/ml; p<0.001) and an increase in total (B:10.59±4.80 vs. 6M:14.03±6.68 µg/ml; p<0.001) and LDL-cholesterol levels were measured. Serum total and HMW-adiponectin levels were measured by RIA and ELISA (Millipore®), respectively at baseline (B, n=100) and after 6 months (6M; n=53). Body composition (DXA) and abdominal MRI were performed in all patients at B, and at 6M in patients achieving a BMI reduction over 1.5 SDS (n=17).

Results: At 6M, 45 of the 53 patients attained a reduction in BMI-SDS (-0.93±0.55 BMI-SDS). This was associated with a decrease in uric acid (p<0.001) and an increase in total (B:10.59±4.80 vs. 6M:14.03±6.68 µg/ml; p<0.05) and HMW-adiponectin levels (B:4.71±2.75 vs. 6M:6.13±3.37 µg/ml; p<0.001). Patients achieving a BMI reduction >1.5 SDS also increased total and HMW-adiponectin (both p<0.05) and decreased insulin (B:15.21±9.33 vs. 6M:10.04±5.22 µU/ml; p<0.05) and LDL-cholesterol levels (B:93.55±19.12 vs. 6M:84.13±21.57 mg/dl; p<0.05). DXA scans showed that weight loss resulted in a decrease in body fat percentage (p<0.001) and an increase in lean mass (p<0.05), whereas abdominal MRI revealed that visceral and subcutaneous fat stores were reduced in parallel (both p<0.001), without altering their ratio.

Conclusions: Weight reduction in obese children affects both the visceral and the abdominal fat stores in a similar fashion. Although slight BMI reduction can modify the circulating adipokine profile, greater weight loss is necessary for the improvement of carbohydrate and lipid metabolism derangements.

P2-d2-592 Fat Metabolism, Obesity 5

Gastric banding in early adolescence: preliminary results of a pilot study

Background: Morbid obesity showed a major increase in the last ten years in European children. It is clear for all pediatricians that the traditional approach based on dietary and behavioural interventions (DBI) is unsuccessful in most of these children.

Objective: To evaluate in a population of obese adolescents (12 to 18 years), the impact of gastric banding procedure (GBP) on the evolution of weight (BMI Z score) and comorbidities, during short (1 to 5 years) and long (10 years and more) follow up periods. To find psychological, behavioural, socioeconomic and physical predictive factors of success or failure of surgical approach.

Method: We randomly selected 230 obese patients who were morbidly obese, among whom 44 M and 50 F had a BMI > 40.5 and a mean weight gain of 9.6 and 10.8 kg/year respectively. Only 5/44 males and 1/50 were able to diminish or stabilize their weight during the previous 24 months of observation. Considering the good benefit/risk ratio reported in the adult and pediatric literature, we started a pilot trial of the GBP in this group of patients. Inclusions started April 2009 with GBP starting an year later. Selection criteria were a BMI Zscore ≥ 3.5 in patients older than 12 yrs with common obesity showing no tendency to improvement after two years of intensified DBI.

Results: GBP was performed in 14 patients (10F/4M), aged 15.8 +/- 1.9 years (12-18.6), having a BMI Zscore of 4.3 +/- 0.5 (3.7-5.2). Mean weight loss after one month was 4.6 +/- 2.3 kg (3-8) with a BMI Zscore variation of °C 0.2 +/- 0.06 and after two months - 8.9 +/- 2.1 kg (6-11.1), BMI Zscore (-0.37 +/- 0.03). No significant side effects were observed.

Conclusions: Our preliminary results support that GBP may be an alternative to the classical approach in young obese. More patients, a longer follow-up, and a statistical analysis are needed to assess efficacy, safety, predictive factors and quality of life in operated patients.

P2-d2-591 Fat Metabolism, Obesity 5

Analysis of the potential cause of obesity in children with melanocortin receptor 4 (MC4R) mutations

Background: The melanocortin system consists of agonists (α-MSH, β-MSH, γ-MSH, and ACTH), antagonists (Agouti & Agouti-related peptide) and receptors (MC1R to MC5R). The melanocortin-4 receptor (MC4R; 18q21.3) is an intron-less gene encoding a 332 amino acid protein and more than 150 mutations of this gene associated with monogenic obesity have been identified to date.

Objective and hypotheses: As MC4R mutations are the most common cause of monogenic obesity, the MC4R gene was screened for mutations in 77 obese children.

Methods: Sequence analysis and functional studies were performed.

Results: We identified 3 sequence variants, all of them in heterozygosity, in 3 unrelated patients whose obesity began in the first year of life: two novel changes (N74I and P272L) and the third (I251L) previously suggested to confer protection against obesity. The N74I variant was present in a male (BMI +5.6 SDS) born from consanguineous parents with several dysmorphic features (polydactyly in four limbs, motor delay, retnitits pigmentaria and microcropsis). The male carrying the P272L variant (BMI +6.5 SDS) presented overgrowth, accelerated bone-age and increased adrenal androgen production throughout infancy, with normal pubertal development and predicted adult height above his target height. His mother was also a carrier and presented obesity. The third sequence variant (I251L) was present in a female (BMI +3.59 SDS) with no dysmorphic features or metabolic derangements. Functional studies showed that P272L conferred poor protein expression at the membrane despite normal cAMP generation in response to ligand. No abnormalities in cell localization or response to ligand were observed for the other two variants.

Conclusions: Our data add to the current knowledge of MC4R mutations associated with obesity and question the protective effect of the I251L variant.
**Conclusions:** The onset of puberty was at the age of 9 years, mental development by D. Wechsler and corresponded with the normal range target (171.5 cm). Intelligence Quotient (IQ) in patient was assessed by WAIS and correlates positively with the daytime systolic blood pressure levels (p < 0.05). Furthermore, nocturnal systolic blood pressure is directly related to the relative left ventricular wall thickness (p > 0.05). The nocturnal mean blood pressure is negatively correlated to HOMA index (p < 0.05).

**Methods:** Prospective study of 64 obese children and adolescents (BMI > 30 Kg/m² for age and sex according to IOTF, 2000; mean age 11.6±2.3 SDS. Patients were monitored for 24 hours blood pressure (Diasys Integra®). Statistical analysis is conducted of the possible relationship of ABPM data, anthropometric variables (BMI, waist and hip circumference) and echocardiographic parameters. Results: Physiological dip of nocturnal systolic blood pressure is not objective in 33.4% of individuals. This dipping is neither observed in diastolic or mean blood pressure in the 25.86% and 29% of patients respectively. Waist circumference is the only anthropometric parameter related to blood pressure and correlates positively with the daytime systolic blood pressure levels (p < 0.05). Night values of systolic and diastolic blood pressure are positively related to basal insulin levels (p < 0.05). Furthermore, nocturnal systolic blood pressure is directly related to the relative left ventricular wall thickness (p < 0.05). The nocturnal mean blood pressure is negatively correlated to HOMA index (p < 0.05).

**Conclusions:** These data suggest that echocardiographic and blood pressure by ABPM is not invasive and it can bring cardiovascular risk data useful for monitoring of obese children.

**Background:** Oxidative stress increases in obesity can increase the risk of cardiovascular morbidity and mortality in adulthood. Oxidative stress seems to be involved in the pathophysiology of atherosclerosis, diabetes and cardiovascular complications in obesity. Objective and hypotheses: The aim of our study was to evaluate the level of oxidative stress markers in obese children comparing to the lean control group.

**Methods:** Oxidative stress markers (TOS – total oxidative capacity, oxy-LDL, leptin and adiponectin were determined in 42 obese children and 40 healthy controls. Nutritional status by BMI and waist/height ratio calculation and body composition analysis (Tanita BC-418) was assessed in all children.

**Results:** TAC was significantly (p < 0.0001) lower, but oxy-LDL level was significantly (p < 0.05) higher in obese than in healthy children. TOC was significantly correlated with fat percentage (r = 0.494 p<0.05) measured by bioimpedance and with leptin level (r = 0.518, p<0.01) and adiponectin/leptin ratio (r = 0.481 p<0.05) in obese children.

**Conclusions:** High level of oxy-LDL together with low antioxidant capacity detected in obese children, and significant relation of nutritional status to total oxidative stress indicate imbalance in oxidative/antioxidative status in obese children. This situation can leads to higher risk of atherosclerotic and diabetic complications in the future.
P2-d2-597 Fat Metabolism, Obesity 6
Low self esteem and low socioeconomic status is associated with depressive mood in over weight and obese Israeli adolescents
Michal Yakobovitch-Gavriel1; Revital Masyh-Tamir1; Nessia Nagelberg1; Moshe Phillip2; Joseph Meyerovitch1
1Schneider Children's Medical Center of Israel, Institute for Endocrinology and Diabetes, Tel Aviv University, Sackler Faculty of Medicine, Petah Tiqwa, Israel; Schneider Children's Medical Center of Israel, Institute for Endocrinology and Diabetes, Petah Tiqwa, Israel; 2Schneider Children's Medical Center of Israel, Institute for Endocrinology and Diabetes, Rehabilitation Workshop, Petah Tiqwa, Tel Aviv, Israel

Background: Studies relating depression to weight status in adolescents have inconsistent results. It has been argued that this inconsistency is due to the influence of socio-demographic mediators which vary across samples.

Objective and hypotheses: The aim of the study was to compare depressive mood between over-weight and obese as compared to normal weight Israeli adolescents, and to explore factors associated with depression in each weight-group.

Methods: 30 normal-weight (BMI percentile 5-84.99) and 60 over-weight and obese (BMI percentile >85) adolescents (mean age 14.3±1.9 years) were included in the study. All participants were measured for height and weight, and completed Beck depression inventory (BDI) (Beck, 1961), personal details questionnaire, family climate questionnaire (Moos, 1976), body image questionnaire (Stunkard et al, 1983) and self esteem questionnaire (Rosenberg, 1973).

Results: BDI score were marginally higher in over-weight and obese as compared to normal-weight adolescence (median (IQR): 4 (0, 8.8) & 0 (0, 5) accordingly, p=0.053). No difference in BDI scores were found between boys (n=49) and girls (n=41). Linear regression models for variable associated with BDI score revealed different pattern for over-weight and obese (BMI percentile >85) and girls (n=41). Linear regression models for variable associated with BDI score were marginally higher in over-weight and obese as compared to normal-weight adolescence (median (IQR): 4 (0, 8.8) & 0 (0, 5) accordingly, p=0.053). No difference in BDI scores were found between boys (n=49) and girls (n=41). Linear regression models for variable associated with BDI score revealed different pattern for over-weight and obese (BMI percentile >85) and girls (n=41).

Conclusions: Over-weight and obese adolescents especially with low self esteem and from lower socioeconomic families are more prone to depressive mood as compared to normal-weight adolescents. Psychological assessment and treatment for depressive sings is crucial as integral component of the treatment of adolescents’ obesity.

P2-d2-599 Fat Metabolism, Obesity 6
Utilization of bioelectrical impedance in the diagnosis and treatment obesity in the child
Ignacio Díez-Lopez1; Ainhoa Sarasua Miranda; Isabel Lorente Blazquez
1Txagorritxu Hospital, Paediatric Endocrinology Unit, Vitoria, Spain

Background: Children’s obesity has spectacularly increased in last years. The development of new physio-medical technologies, allows the professionals to have helpful tools at monitoring and evolution follow-up of this type of patients.

Objective and hypotheses: Patients typed as hexogen obesity, with basic biochemical test. Somatometic evaluation of weight, height, BMI, folds and perimeters done always by only one watcher. Reference tables Orbezogo 2004. Evaluation of puberty (Tanner) and ethnic origin. Utilisation of impedance meter TANITA BEF 350 of 4 channels. Education material: “Nens. in movement” program (PhD A. Carrascosa). Clinic follow up at 1,3,6,9,12 15 and 18 months, with evaluation of impedance, weight and height. In this study we’ve separated the patients in groups depending on sex, pubertal stage, ethnic origin, and basal metabolism (Kcal/Kg of weight). Statistical study SPSS 17.0 for WIN.

Methods: A total of 150 patients were studied. Average age at the beginning of the study was of 11.5 years (SDS 3,8) (6% F). 95% of the patients had started puberty (Tanner II). 45% was born out of Spain (68/150), the major part of them proceeded from Latin -America countries (50/68). Average weight was of + 2,58 SDS and BMI of + 2,75 SDS. Basal metabolism (initial data), gave an average of 1310 Kcal (24,7 Kcal/Kg). The initial fat mass was of 32% (±5.7) with an excess of 4,8 Kg. At the end of our study, fat mass average was of + 2,18 SDS p=0.03 and BMI of +2,15 p=0.02; fat mass was of 28% (±0.12) with excess of 4.5 Kg. There are significant differences among the groups. Pre-pubertal girls with a lower BMI respond better (showing a larger loss of fat mass). Immigrant children show initially a larger obesity, a lower metabolism and worst results at the end of the treatment. Cases with a higher basal metabolism show better results.

Conclusions: We do recommend the use of new technologies in obesity, to decide, depending on the characteristics of each patient, the best therapeutic option.

P2-d2-600 Fat Metabolism, Obesity 6
Twelve common genetic variants explain a substantial part of the variation in BMI of obese children
Henriette Delemarre-van de Waal1; Marije Wetzels2; Peter Henneman3; Egbert Bakker4; Ko Willems van Dijk1; Linda Van de Berg1
1Leiden University Medical Center, Pediatrics, Leiden, Netherlands; 2Leiden University Medical Center, Human Genetics, Leiden, Netherlands

Background: Results of twin and family studies suggest that up to 80% of the variation in body mass index (BMI) in children can be explained by genetic factors. A recent study (Diabetes 59:2980-2988) investigated 12 single nucleotide polymorphisms (SNPs) that are robustly associated with adult BMI in a large pediatric population. Direction-consistent associations with BMI were found for all of these variants in children. However, the risk factors explained a disappointing 1% of the population variation in BMI when taken together.
Hypotheses: We hypothesized that these 12 SNPs also explain variation in BMI in obese children.

Methods: We studied 64 patients from our pediatric obesity clinic to test this hypothesis (39% boys; mean age:13.2; age range: 6 to 19; all children of North-West European descent). BMI-standard deviation scores (BMI-SDS) were used as a measure of body fat percentage of the children. BMI-SDS indicates how many standard deviations the child is away from the mean of a reference population of the same age and sex.

Results: Mean BMI-SDS of the study group was +2.8 (range: +1.9 to +3.8). The SNPs were genotyped using pre-designed TaqMan assays. We calculated a genetic predisposition score for each subject, i.e. the number of BMI-increasing alleles (theoretical range: 0 to 24). The genetic predisposition score significantly predicted BMI-SDS in this group of obese children (p=0.02). The score explained 8.4% of the variation in BMI-SDS in the group. The largest effect size was found for the fat mass and obesity associated gene (FTO, beta=−0.2).

P2-d2-603 Fat Metabolism, Obesity 6
The relationship between levels of serum visfatin, vaspin, adiponectin and metabolism disturbance in children with obesity of different types
Feng Xiong1; Jie Yu; Min Zhu; Pei-Yun Lei; Lei-Li Deng
Children's Hospital of Chongqing Medical University, Pediatric Endocrinology, Chongqing, China

Objective: To study the levels of serum visfatin, vaspin and adiponectin in children with different fat distribution, and to investigate the association of concentration of these adipocytokines with abdominal obesity and metabolism disturbance.

Methods: 78 children with abdominal obesity, 18 children with non-abdominal obesity and 75 normal children were recruited, anthropometric variables, blood-fat, serum glucose and insulin were measured. Serum visfatin, vaspin concentrations and serum adiponectin of children in every group were examined by enzyme-linked immunosorbent assay (ELISA).

Results: (1) The levels of serum visfatin of abdominal, non-abdominal, normal group were respectively 68.91±11.67, 21.85±1.18 and 20.06±2.34ug/ml, respectively. (2) Serum visfatin concentrations in children with abdominal obesity and metabolism disturbance. (3) Serum adiponectin of children in every group were examined by enzyme-linked immunosorbent assay (ELISA).

P2-d2-604 Fat Metabolism, Obesity 6
T.A.F.F. – Telephone-based Adiposity prevention
For Families: socio-demographic description of participants and barriers to participation
Jana Markert1; Franziska Alff2; Ruth Gausche3; Wieland Kiess1; Susann Blüher1
1University of Leipzig, Women and Child Health, Hospital for Children and Adolescents, IFS Adiposity Diseases, Leipzig, Germany; 2University of Leipzig, Women and Child Health, Hospital for Children and Adolescents, Leipzig, Germany; 3University of Leipzig, CrescNet GmbH, Leipzig, Germany

Background: The high prevalence of childhood obesity requires effective prevention, but participation rates in intervention programs are rather low. Objective and hypotheses: T.A.F.F. aims to prevent further increase in BMI-SDS in children. Here we (a) characterized participating families socio-demographically and (b) analyzed participation barriers.

Methods: 4005 overweight (OV)/obese (OB) children aged 3–17 years were screened via the CrescNet-database. (a) Participants (n=303) were described socio-demographically (standards of German federal office for statistics) and compared to the 2009 Saxonian microcensus. Anthropometric measures were compared to the 2009 Saxonian microcensus.
compared between P and the entire screening group; differences were tested by chi2 and t-test. (b) For participation barriers, 433 families completed identical questionnaires on lifestyle, eating patterns, socio-demographic/eco-nomic factors, anthropometric parameters and quality of life (241 participants (P); 192 non-participants (Np)). Factors associated with non-participation were calculated by univariate analyses/binary logistic regression.

Results: (a) P included more children 3-10 years, less children 14-17 years, less OV and more OB children compared to the screening group (all p<0.001). P significantly differed from the control group regarding number of single parents, paternal education, household-net-income, number of persons per household (p<0.05). (b) Reasons for non-participation were: 1.) Weight status: prevalence of OV was higher in Np and of OB was higher in P (p<0.001). 2.) Eating patterns: P had regular breakfasts (p=0.008)/five regular meals per day (p=0.003); 3.)NP had a higher subjective physical well-being (p=0.018); 4.) Household income was significantly lower in Np (p<0.001).

Conclusions: The perception of obesity is still underestimated. Setting-related approaches are needed to support families in their change of lifestyle behaviour, especially single parents and families with low household income. For adolescents, other intervention strategies apart from family settings are required.

P2-d2-606 Fat Metabolism, Obesity 6
Sub-clinical inflammation in overweight and obese children with and without metabolic syndrome. Is there a rationale for distinction?
Konstantinos Kitsios1; Maria Papadopoulou1; Nikolaos Kadoglou2; Alkistis Kapelouzou3; Dimitrios Chatzidimitriou4; Fani Chatzopoulou4; Konstantina Kosta1; Maria Papagianni3; Nikolaos Malisiovas2
1Aristotle University of Thessaloniki, Hippokration General Hospital, Third Department of Pediatrics, Thessaloniki, Greece; 2Foundation of Biomedical Research, Academy of Athens, Athens, Greece; 3Aristotle University of Thessaloniki, Medical School, Laboratory of Microbiology, Thessaloniki, Greece

Background: It’s still controversial whether clustering cardiovascular risk factors as metabolic syndrome (MS) adds more information in cardiovascular risk stratification.

Objective: We assessed the serum levels of well-established markers of sub-clinical inflammation and atherogenesis in overweight and obese children with MS compared to their counterparts without MS.

Methods: 54 consecutive obese (BMI>95PC) and 50 overweight (85pc≤BMI<95PC) children and adolescents (6-17 years old), attending the obesity clinic, were screened for NAFLD by using liver ultrasound and Liver Function Tests (LFTs). MS was diagnosed according to the Cook criteria, modified for an upper normal limit of fasting plasma glucose<100mg/dl. 40 normal-weight, age-matched children were enrolled as control group. Interleukin-6 (IL-6), high-sensitivity CRP(hsCRP), TNFα, fibrinogen, White Blood Cells count (WBC), adiponectin and homocysteine serum levels were measured. One-way ANOVA and Tuckey post-hoc test were used for statistical analysis (p<0.05).

Results: Obese children appeared with significantly higher levels of IL-6, hsCRP, fibrinogen, WBC, homocysteine and lower levels of adiponectin compared to control group (p<0.001). Compared to overweight, obese children had significantly higher levels of fibrinogen(p=0.014), IL-6(p=0.014) and WBC (p=0.019) and lower levels of adiponectin (p=0.005). Concerning the aforementioned parameters, overweight children had significantly higher levels of fibrinogen, hsCRP, homocysteine (p=0.001) and IL-6 (p=0.027) compared to control group. Moreover, MS was diagnosed in 21 obese and overweight children (31.5% of the obese and 8% of the overweight children). The latter MS-children had significantly higher levels of IL-6 (2.9±1.9 vs 1.7±1.5pg/ml, p=0.013) and fibrinogen (446±91 vs 389±94mg/dl, p=0.016) compared to obese and overweight children without MS.

Conclusion: Obese and overweight children with MS seem to have increased levels of pro-inflammatory and pro-atherogenic markers compared to their counterparts without MS. Children with MS may run an additional risk for cardiovascular disease beyond that of simple obesity.

P2-d2-606 Fat Metabolism, Obesity 6
Metabolic characteristics and markers of inflammation in obese children with and without non-alcoholic fatty liver disease
Konstantinos Kitsios1; Maria Papadopoulou1; Nikolaos Kadoglou2; Dimitrios Chatzidimitriou3; Fani Chatzopoulou4; Alkistis Kapelouzou3; Konstantina Kosta1; Kiriaki Tsiroukidou2; Nikolaos Malisiovas2
1Aristotle University of Thessaloniki, Hippokration General Hospital, Third Department of Pediatrics, Thessaloniki, Greece; 2Foundation of Biomedical Research, Academy of Athens, Athens, Greece; 3Aristotle University of Thessaloniki, Medical School, Laboratory of Microbiology, Thessaloniki, Greece

Background: Non-Alcoholic Fatty Liver Disease (NAFLD) may represent independent risk factor for cardiovascular disease(CVD). Data about its prevalence in childhood and its relationship with sub-clinical inflammation are still insufficient.

Objective: To assess the prevalence of NAFLD in overweight and obese children and to explore its association with other metabolic abnormalities, such as metabolic syndrome (MS), prediabetes and insulin resistance, and with markers of sub-clinical inflammation.

Methods: 54 consecutive obese (BMI>95PC) and 50 overweight (85pc≤BMI<95PC) children and adolescents (6-17 years old), attending the obesity clinic, were screened for NAFLD by using liver ultrasound and Liver Function Tests (LFTs). MS was diagnosed according to the Cook criteria, modified for an upper normal limit of fasting plasma glucose<100mg/dl. Prediabetes was diagnosed according to the American Diabetes Association criteria. Insulin resistance was defined as HOMA-IR>3.1 (HOMA-IR=homostasis model assessment-insulin resistance). Interleukin-6 (IL-6), high-sensitivity CRP (hsCRP), TNFα, fibrinogen, White Blood Cells count (WBC), adiponectin and homocysteine were measured. Student’s t-test was used for statistical analysis (p<0.05).

Results: Based on liver ultrasound examination NAFLD was diagnosed in 44.6% of the obese and 18% of the overweight children. However, only 29.2% of children with NAFLD had normal LFTs. Insulin resistance, MS and prediabetes were diagnosed in 79.6%, 16.7% and 29.2% of the obese children with NAFLD, respectively. Obese and overweight children with NAFLD had significantly higher levels of TNFα (2.7±1.1pg/ml vs 1.9±1.0pg/ml, p<0.005) and hsCRP (0.78±1.4mg/dl vs 0.34±0.31mg/dl, p=0.016) compared to those without NAFLD.

Conclusions: NAFLD is prevalent amongst obese and overweight children and adolescents and is often associated with insulin resistance, MS and prediabetes. Obese children with NAFLD have elevated levels of markers of sub-clinical inflammation, such as TNFα and hsCRP, and may run additional to the obesity risk for CVD.

P2-d2-607 Fat Metabolism, Obesity 6
Increased BMI in association with breast feeding and birth weight among primary school children in Latvia
Sarmite Limeza; Igrida Rumba-Rozenfeld; Ilva Daugule University of Latvia, Faculty of Medicine, Riga, Latvia

Background: Childhood obesity and overweight is an increasing problem. Breast feeding has been suggested a protective factor against increased weight.

Objective and hypotheses: To detect the prevalence of elevated body mass among primary school children and association with birth weight and breast feeding.

Methods: A prospective, cross-sectional study was performed in 2010 in primary schools, 6-9 years old children. Anthropometric measurements were performed, parents were asked to fill out a questionnaire. The body mass index (BMI) was calculated and BMI >25kg/m2 was considered as elevated. Statistical method: chi2 test, logistic regression analysis, one-way ANOVA test. Result: In total, 465 children were included (mean age 7.5). Elevated children were included (mean age 7.5). Elevated BMI was detected in 39%(180/465). The mean birth weight was significantly higher among children with elevated BMI compared to control group: 3.66kg(SD=0.69) vs. 3.46kg (SD=0.5), p=0.001. Birth weight >4kg was more observed in a group of patients with elevated BMI: 23% (41/175) vs. 12%(34/272), OR=2.195%CI: 1.3-3.6; p=0.02; birth weight <2.6kg was observed less often in a group of patients with elevated BMI 0.6%(1/174)
Protein intake of toddlers in Greece and its nutritional consequences

Andrianna Vazaiou1; Alexandra Papadopoulou2; Lampros Fotis1; Katerina Kouti2; Panagiota Botonis3; Maria Stathopoulou2; Maria Skouroliakou1
1P and A Kyriakou Children’s Hospital, 1st Department of Pediatrics, Athens, Greece; 2Iaso General Hospital, Department of Dietetics, Athens, Greece; 3Athens University, Faculty of Physical Education and Sports Science, Athens, Greece

Background: Excessive protein intake during the first years predisposes to obesity later in life.

Objective and hypotheses: The aim of the study was to evaluate the dietary intake of Greek toddlers and its impact on their nutritional status.

Methods: Three hundred and one toddlers (mean [SD] age (months) 24.4 (7.6); 141 males) were studied. A 4-day diet assessment was recorded. Food records were analyzed using the USDA Food Search for Windows version 1, database SR18 software. Macro- and micronutrient intake was compared to dietary reference intake (DRI). One hundred eighty five toddlers [mean (SD) daily caloric intake was 1059.7(192.1) Kcal. Mean (SD) protein intake was 48.9 (11.2) g/day, exceeding the recommended by WHO and USDA average intake of protein (13g/day) by 373%. Polyunsaturated fat protein intake was 48.9 (11.2) g/day, exceeding the recommended by WHO utilization of anemia significantly increased serum concentration of IGF-I but does not affect the increase of IGF-I in response to GH stimulation.

Conclusions: In opposite to other studies, decreased birth weight was not associated with overweight later in childhood. Higher birth weight detected in the group of patients with increased BMI could point to risk factors in development of overweight (congenital as well as environmental) acting already in the first year of life in the Breast feeding per se was not a protective factor against increased weight in our study population.

Methods: We measured serum concentrations of insulin like growth factor-I (IGF-I) in 20 thalassemic males with short stature (height SDS <-2) and/or slow growth velocity (GV) < -1 SD, throughout their childhood and adolescence and performed IGF-I generation before and after blood transfusion (Btx).

Results: 1 No statistical difference in age, HSDS, target height SDS or bone age was observed between patients with BTM with growth hormone deficiency (GHD) versus those with normal GH secretion (GHS). 2) The age-related changes in serum total IGF-I in thalassemic males showed significantly decreased concentrations from early childhood to 18 years of age compared to normal data. Thalassemic males with GHD did not show any significant peak of IGF-I level till 18 years of age. Whereas thalassemic males with normal GH response to provocation (GHS) achieved a significant peak level of IGF-I that was attenuated and late (at 18 years of age) compared to normal males. A significant increase in the circulating basal IGF-I concentrations from 56 +/- 40 ug/l to 82.6 +/- 39 ug/L was achieved with increasing Hb concentration after blood transfusion. The peak IGF-I response to GH injections did not differ before versus after blood transfusion. The % increment of IGF-I levels generated after GH injections were higher in thalassemic patients with GHD vs those with GHS both before and after blood transfusion.

Conclusions: Our results showed that age-related serum IGF-I concentrations were significantly lower in short thalassemic patients, with and without GHD, during childhood and adolescence, compared to normal standards. Correction of anemia significantly increased serum concentration of IGF-I but does not affect the increase of IGF-I in response to GH stimulation.

The growth hormone/IGF-I axis and growth hormone receptor mutations in idiopathic short stature

Mohamed El Kholy1; Patrizia Meli2; Mona Rashad3; Fabio Buzz4; Sally Zahra2; Heba Elsawy5
1Ain Shams University, Paediatrics, Cairo, Egypt; 2University of Brescia, Institute of Molecular Medicine "Angelo Nocivelli", Brescia, Italy; 3University of Brescia, Paediatrics, Brescia, Italy

Background: It was hypothesized that some children with idiopathic short stature (ISS) may have partial insensitivity to GH.

Objectives and hypotheses: In this study analysis of the GH/IGF-I axis as well as GH receptor (GHR) gene was done in children with ISS to determine the possible underlying factor(s) to their short stature.

Methods: Forty-eight patients with a diagnosis of ISS were studied; 33 boys and 15 girls aged 13.6±3.7 years. Molecular analysis of the GHR was performed and GH sensitivity was tested by the IGF-I generation test.

Results: Basal IGF-I levels were <2SD in 22.9% and 53.5% showed an IGF-I response below 40% (0-38%) to GH stimulation. Growth hormone binding protein (GHBP) levels were below the normative mean in almost all patients. Mutations in the region of the GHR gene that codes for the extracellular domain of the receptor were found in 15.5%, one newly described mutation was formed and GH sensitivity was tested by the IGF-I generation test. The % increment of IGF-I levels after blood transfusion did not differ before versus after blood transfusion. The peak IGF-I response to GH injections did not differ before versus after blood transfusion. The % increment of IGF-I levels generated after GH injections were higher in thalassemic patients with GHD vs those with GHS both before and after blood transfusion.

Conclusions: The results of our study suggest that some children with ISS might have partial resistance to GH. Testing children with ISS for their GH sensitivity would be helpful in optimizing treatment with GH, IGF-I or both.

The age-related changes in serum concentrations of insulin-like growth factor-I (IGF-I) in patients with beta thalassemia major (BTM) and IGF-I generation before and after blood transfusion

Ashraf Solimão1; Ahmed Abushahin2; Khaleel Abohezeima3; Hany Khalafallah4; Ashraf Adel5; Ahmed Elawwa6; Naima Elmulla6
1Hamad Medical Center and University of Alexandria, Pediatrics, Doha, Qatar; 2Hamad Medical Center, Pediatrics, Doha, Qatar

Background: Growth and maturational delay are striking features of beta-thalassemia major (BTM). Hypotheses: Alterations in IGF-I regulation may provide an explanation for BTM growth impairment.

Methods: We measured serum concentrations of insulin like growth factor-I (IGF-I) in 20 thalassemic males with short stature (height SDS <-2) and/or slow growth velocity (GV) < -1 SD, throughout their childhood and adolescence and performed IGF-I generation before and after blood transfusion (Btx).

Results: 1 No statistical difference in age, HSDS, target height SDS or bone age was observed between patients with BTM with growth hormone deficiency (GHD) versus those with normal GH secretion (GHS). 2) The age-related changes in serum total IGF-I in thalassemic males showed significantly decreased concentrations from early childhood to 18 years of age compared to normal data. Thalassemic males with GHD did not show any significant peak of IGF-I level till 18 years of age. Whereas thalassemic males with normal GH response to provocation (GHS) achieved a significant peak level of IGF-I that was attenuated and late (at 18 years of age) compared to normal males. A significant increase in the circulating basal IGF-I concentrations from 56 +/- 40 ug/l to 82.6 +/- 39 ug/L was achieved with increasing Hb concentration after blood transfusion. The peak IGF-I response to GH injections did not differ before versus after blood transfusion. The % increment of IGF-I levels generated after GH injections were higher in thalassemic patients with GHD vs those with GHS both before and after blood transfusion.

Conclusions: Our results showed that age-related serum IGF-I concentrations were significantly lower in short thalassemic patients, with and without GHD, during childhood and adolescence, compared to normal standards. Correction of anemia significantly increased serum concentration of IGF-I but does not affect the increase of IGF-I in response to GH stimulation.
IGF1&BP3 and chelator treatment on long term basis necessary. Profile in β TA pts present; Investigation of possible correlation between very low IGF-1 SDS (more than -2) in 21.1% and BP3 in 12% of all pts. Significant lower IGF-1 &BP3-SDS compared to controls (p<0.0001); subset (p=0.03) and 20-30 yrs (p=0.003); in males 9-12 yrs (p<0.0001) and 20-30 yrs during all age periods; c) especially affected IGF 1 in females 15-20 yrs TA pts: a) preserved age-related IGF1&BP3 dynamic; b) lower IGF1&BP3 followed by a decline; Earlier IGF1 increment in girls compared to boys; β below 2 yrs, gradual permanent increase to maximal values during puberty.

Results: Cross-sectional study (2008-2010, pts treated in two outpatient centers in Sofia, below and over 18 yrs). Serum IGF1 and BP3 (Elisa, IBS) measured in 215 controls (136f, 79m, 9 age groups) and 92 pts (38f, 54m, from 3-40 yrs); Statistical Analysis by SPSS.

Results: Characteristic IGF1&BP3 pattern in the controls: low concentrations below 2 yrs, gradual permanent increase to maximal values during puberty followed by a decline; Earlier IGF1 increment in girls compared to boys; β TA pts: a) preserved age-related IGF1&BP3 dynamic; b) lower IGF1&BP3 during all age periods; c) especially affected IGF 1 in females 15-20 yrs (p=0.03) and 20-30 yrs (p=0.003); in males 9-12 yrs (p=0.001) and 20-30 yrs (p=0.05); BP3 in females 21-30 yrs (p=0.003), in males 9-11 yrs (p=0.05); d) Significant lower IGF1-1&BP3-SDS compared to controls (p<0.0001); subset of very low IGF-1 SDS (more than -2) in 21.1% and BP3 in 12% of all pts.

Conclusions: Evidence for reduced magnitude of IGF1&BP3 age specific profile in β TA pts present; Investigation of possible correlation between IGF1&BP3 and chelator treatment on long term basis necessary.

P2-d1-612 GH and IGF Physiology 1
IGF-I and IGFBP3 levels in patients (pts) with βThalassaemia (β TA)
Iva Stoeva1; Rosadlava Emilova1; Denka Stoianova2; Mirella Rangelova1
1University Pediatric Hospital Sofia, Screening and Functional Endocrine Diagnostics, Sofia, Bulgaria; University Pediatric Hospital Sofia, Rheumatology and Thalassaemias, Sofia, Bulgaria; National Center Transfusional Hematology, Thalassaemias and pathological Hemoglobinias, Sofia, Bulgaria

Background: Short stature has persisted in β TA pts despite major advances in the complex treatment. Adult shortness is associated with subnormal growth (fetal to puberty) rather than any one single phase.

Objective and hypotheses: Description of the character of circulating IGF1&BP3 in β TA pts. Aim: Evaluate whether basal IGF1&BP3 levels in β TA pts longitudinally.

Methods: Cross-sectional study (2008-2010, pts treated in two outpatient centers in Sofia, below and over 18 yrs). Serum IGF1 and BP3 (Elisa, IBS) measured in 215 controls (136f, 79m, 9 age groups) and 92 pts (38f, 54m, from 3-40 yrs); Statistical Analysis by SPSS.

Results: Characteristic IGF1&BP3 pattern in the controls: low concentrations below 2 yrs, gradual permanent increase to maximal values during puberty followed by a decline; Earlier IGF1 increment in girls compared to boys; β TA pts: a) preserved age-related IGF1&BP3 dynamic; b) lower IGF1&BP3 during all age periods; c) especially affected IGF 1 in females 15-20 yrs (p=0.03) and 20-30 yrs (p=0.003); in males 9-12 yrs (p=0.001) and 20-30 yrs (p=0.05); BP3 in females 21-30 yrs (p=0.003), in males 9-11 yrs (p=0.05); d) Significant lower IGF1-1&BP3-SDS compared to controls (p<0.0001); subset of very low IGF-1 SDS (more than -2) in 21.1% and BP3 in 12% of all pts.

Conclusions: Evidence for reduced magnitude of IGF1&BP3 age specific profile in β TA pts present; Investigation of possible correlation between IGF1&BP3 and chelator treatment on long term basis necessary.

P2-d1-614 GH and IGF Physiology 1
Growth impaired children with epidermolysis bullosa have increased serum markers of inflammation and reduced circulating IGF-1
Arati Raj1; Jemima E Mellendorf1; Melanie Sklar1; Caroline Briar2; Jeremy Algrove3; Ian R Sanderson3; Anna E Martinez3
1Barts and the London School of Medicine and Dentistry, Queen Mary University of London, Centre for Digestive Diseases, London, United Kingdom; 2Great Ormond Street Hospital for Children NHS Trust, Departments of Paediatric Dermatology and Endocrinology, London, United Kingdom; 3Barts and the London Children's Hospital, Royal London Hospital, Department of Paediatric Endocrinology and Diabetes, London, United Kingdom; 4Great Ormond Street Hospital for Children NHS Trust, Great Ormond Street, Departments of Paediatric Dermatology and Endocrinology, London, United Kingdom

Background: Epidermolysis bullosa (EB) is a group of rare but devastating diseases in which patients have inflammation of the skin with epithelial fragility and recurrent blistering. Children with the severe types of EB have poor linear growth, which is only partially reversed by nutritional supplementation. Pro-inflammatory cytokines impair growth in children with inflammatory bowel disease by inhibiting signal transduction from growth hormone (GH) to insulin-like growth factor-1 (IGF-1). We hypothesised that a similar mechanism occurred in EB.

Objective and hypotheses: To examine growth impairment and IGF-1 levels in a cohort of patients with severe generalised recessive dystrophic EB (RDEB).

Methods: 18 children with RDEB managed at a Paediatric EB centre were identified. Height and height velocity (HV) standard deviation scores (SDS), inflammatory markers and serum IGF-1 and IGFBP3-3 (IGFBP-3) were measured.

Results: The median age (range) of the children was 12.42 years (2.43-17.62). 10 were female, 8 male. All of the girls were premenarcheal, and the boys prepubertal. 17 of 18 had a negative height SDS (mean -2.43, SD 1.33) and 16 of 18 had an increased HV SDS (mean -2.79, SD 2.51). The impaired growth occurred despite nutritional supplementation (16 of 18 had a gastroscopem in situ, 2 had oral supplementary feeds). All children had elevated inflammatory markers (mean [SD] CRP 78.27 [39.18], ESR 106.9 [30.72]). All had low circulating IGF-1 (mean SDS -2.53 [SD 0.39]) and IGFBP-3 (mean SDS -2.84 [SD 0.68]). IGF-1 levels did not correlate directly to HV SDS (p=0.58).

Conclusions: Children with severe generalised RDEB have poor growth and low circulating IGF-1 and IGFBP-3. This is likely to be due, in part, to inflammation. Correcting growth retardation will require non-steroidal therapies targeted at reducing inflammation.
Background: Idiopathic Short Stature (ISS) and Short children born Small for Gestational Age (SGA) are frequently investigated in paediatric endocrinology. In recent studies, up to 20% of the Short non GH deficient (non-GHD) children were considered as primary IGF1 deficiency. SGA are usually non-identified in these studies.

Objective and hypotheses: Determination of IGF1 levels in prepubertal children with ISS and SGA and comparison with reference values.

Methods: Monocentric cohort of children, seen between Jan 2007 and Dec 2009: aged over 2 years and prepubertal, height below -2 SD without GHD. Children with identified causes of growth failure (inflammatory, nutritional, endocrine disease or syndrome) were excluded. Results were analyzed for all patients and by subgroup (ISS and SGA) by t test or Chi2 test when appropriate.

Results: 77 out of 245 tested children were included. The mean IGF1 level was significantly lower than our reference values. ISS had mean IGF1 level significantly lower than the SGA.

<table>
<thead>
<tr>
<th>All patients</th>
<th>SGA</th>
<th>ISS</th>
<th>SGA vs ISS</th>
</tr>
</thead>
<tbody>
<tr>
<td>(n=77)</td>
<td>(n=41)</td>
<td>(n=36)</td>
<td>p</td>
</tr>
<tr>
<td>GA (WA)</td>
<td>3.8±2.5</td>
<td>38.2±3.0</td>
<td>39.3±1.7</td>
</tr>
<tr>
<td>BW (SDS)</td>
<td>-1.0±1.0</td>
<td>-1.6±0.9</td>
<td>-0.4±0.6</td>
</tr>
<tr>
<td>BL (SDS)</td>
<td>-1.9±1.0</td>
<td>-2.6±0.6</td>
<td>-1.1±0.7</td>
</tr>
<tr>
<td>TH (SDS)</td>
<td>-0.9±0.8</td>
<td>-1.0±0.8</td>
<td>-0.7±0.9</td>
</tr>
<tr>
<td>Age/test (y)</td>
<td>8.5±3.5</td>
<td>7.5±3.7</td>
<td>9.6±3.0</td>
</tr>
<tr>
<td>H/test (SDS)</td>
<td>-2.7±0.5</td>
<td>-2.8±0.6</td>
<td>-2.5±0.4</td>
</tr>
<tr>
<td>BMI (SDS)</td>
<td>-0.7±1.0</td>
<td>-0.7±1.0</td>
<td>-0.6±1.0</td>
</tr>
<tr>
<td>IGF1 (SDS)</td>
<td>-1.2±1.2</td>
<td>-0.9±1.2</td>
<td>-1.6±1.0</td>
</tr>
<tr>
<td>IGF1 ≤-1 SDS n (%)</td>
<td>51/77 (66.2)</td>
<td>20/41 (48.8)</td>
<td>31/36 (86.1)</td>
</tr>
<tr>
<td>IGF1 ≤-2 SDS n (%)</td>
<td>18/77 (23.4)</td>
<td>6/41 (14.6)</td>
<td>12/36 (33.3)</td>
</tr>
</tbody>
</table>

Background: Poor growth in utero combined with early catch-up postnatally confers an increased relative risk of later life morbidity. These growth patterns are influenced by ethnic background. The mechanism for translating this early life programming into later life risk is unknown. Changes to hormonal axes have been implicated. This is explored in the present study.

Objective and hypotheses: To study the influence of the IGF system on growth, adiposity and BP over the first 4 years among British born South Asian (SA) and White European (WE) children.

Methods: Healthy SA (M 45, F 22) and WE (M 73, F 64) infants were followed prospectively from birth to age 4 years. Mixed linear regression (MLR) modelling adjusting for age, sex and ethnicity was used to study the effect of IGF-1 and IGFBP-3 levels on anthropometry and systolic BP (SBP) over all time points.

Results: At birth, SA were shorter and lighter than WE (P<0.05) but not significantly different in length by 1y, or in weight and BMI by 2y in girls and 4y in boys. Both groups had similar triceps (Tr) skinfold thickness (SFT), subscapular (Ss) SFT, IGF-1 and IGFBP-3 levels at birth. There was a trend for IGF-1 and IGFBP-3 to be higher in SA than WE boys beyond 1y, and the difference became significant for IGF-1 at 3y (P=0.046). Over the 4 years, MLR modelling showed that ethnicity was significantly associated with height SDS, weight SDS, BMI SDS and SBP (all P values <0.01); IGF-1 with weight (P=0.001) and BMI SDS (P=0.003); IGFBP-3 (and not IGF-1) with SkSFT (P<0.001) and SBP (P=0.006); and SBP with age (P=0.009) and TrSFT (P=0.037).

Conclusions: Over the first 4 years, ethnicity was significantly associated with height, weight and SBP. From a hormonal perspective, IGFBP-3 but not IGF-1 was associated with truncal adiposity and systolic BP.