

# Comprehensive care of patients with PWS – consensus, questions and future directions

Second Expert Meeting  
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*The following report is a comprehensive clinical overview of the Second Expert Meeting on PWS, held in Toulouse, 26–27 October 2006.*

## Session 1: Epidemiology

Chair: S Blichfeldt

### What can we learn from parents and associations?

J Heinemann

A medical survey (1471 patients) and reported deaths (178 patients with cause of death available from 152) from the PWS Association USA has revealed that the age of diagnosis has fallen over the past decade (from 1% to 40% diagnosed as infants), allowing earlier intervention to prevent morbidity, improve care and allow earlier initiation of GH replacement (67% less than 5 years old were on GH). Some interesting differences in the frequency of certain medical problems exist between patients of different age groups and genetic subtypes, and between those who have and have not received GH treatment. Deaths most often appear to be obesity related, but, as previously suggested, there are also cases of gastric rupture and necrosis (2%) and of choking (8%), highlighting factors that might be amenable to prevention (Wharton et al. 1997; Schrandt-Stumpel et al. 2004; Stevenson et al. 2007).

### Overview of spontaneous death reports in Japan

T Nagai

Reported deaths in patients with PWS not receiving GH treatment have been reviewed from 17 patients collected over 15 years from the Japanese PWS Association (some previously reported in Nagai et al. 2005). Sudden deaths of infants and young children were sometimes associated with milk aspiration or viral infection (especially rotavirus), or after a short history of fever and gastroenteritis. The deaths of adult patients were associated with complications secondary to morbid obesity and diabetes mellitus, including leg cellulitis and pulmonary embolism. Patients also died of drowning in a bath tub, perhaps related to poor respiratory control.

### What can we learn from the KIGS database?

P Wilton

The use of pharmacoepidemiological databases, such as the Pfizer International Growth Database KIGS, helps aid investigation of the long-term efficacy and safety of GH treatment, in rare diseases such as PWS. In a preliminary report (Craig et al. 2006) of 675 GH-treated PWS patients in KIGS, there were five cases of sudden death (age range 3–15 years). Three were obese (weight for height > 200%) and causes of death included bronchopneumonia, respiratory insufficiency and sleep apnoea.

Up to August 2006, there were 1242 patients with PWS in KIGS, 53% of whom were boys. The median GH treatment duration was 1.9 years, with data available for a total of 2538 treatment years. GH treatment improved height SDS from -1.6 to -0.6. Body mass index SDS was unchanged from +2.0 to +1.8, but the age at start of GH treatment ranged from 1.3 to 12.9 years, mean 6.4 years, and body composition analysis was not available. Predictors of first-year height velocity in multiple regression analysis were GH dose, body weight (positively correlated), height deficit from genetic target and chronological age (negatively correlated).

Adverse events totalled 184 per 1000 treatment years, with serious events 32 per 1000 treatment years, including six deaths in total and 35 patients developed scoliosis and 9 with respiratory problems including pneumonia and apnoea, with adverse drug reactions 4 per 1000 treatment years (5 patients with oedema, 2 with arthralgia, 2 with idiopathic intracranial hypertension and 1 with myalgia). The rate of adverse events and adverse reactions was similar to that in Turner syndrome, but the rate of serious adverse events was greater in PWS (70% due to scoliosis).

### **How to run an epidemiological study**

**J Whittington**

The Cambridge population survey run between 1998 and 2001 aimed to capture all cases of PWS within a well-defined geographical region in the United Kingdom. This case ascertainment study, with appropriate epidemiological considerations such as over-inclusive sourcing, genetic confirmation, and appropriate methods for dealing with non-responders, estimated the lower limit of birth incidence as 1 in 29 000, and population prevalence as 1 in 52 000 (Whittington et al. 2001). Similar lower limits for birth incidence have been obtained from studies in Belgium and Western Australia (Vogels et al. 2004; Thomson et al. 2006).

### **Multicentre studies, can they help?**

#### **European PWS Research Project**

**A Holland**

Ongoing prospective multi-centre studies will be able to subvert the problems associated with the rarity of PWS and the need to subdivide the population (e.g. by genetic subtype or age or GH

treatment) so as to collect data on sufficient numbers of patients. As part of the 6<sup>th</sup> Framework the European Union is funding a joint basic science and clinical study of PWS ([www.pwseu.eu](http://www.pwseu.eu)).

The European Research Database component allows reliable and lawful collection of clinical data across several European countries. This will be particularly valuable in assessing the responses to treatments such as GH, genotype-phenotype correlations and behavioural aspects of the syndrome such as those found from smaller epidemiological studies, for example the increased incidence of psychosis in patients with UPD (Boer et al. 2002; Whittington & Holland 2004).

### **Multicentre studies, can they help? NIH Rare Disorder Project**

**DJ Driscoll**

In the USA, the PWS and Early-onset Morbid Obesity (EMO) component (<http://rarediseasesnetwork.epi.usf.edu/arpwsc/takeaction/studies/pw-5202.htm>) of the Angelman, Rett, and Prader-Willi syndromes consortium of the NIH-funded Rare Diseases Clinical Research Network (RDCRN) seeks to collect and analyse detailed longitudinal data on the PWS and EMO populations. This will gather a better understanding of the natural histories of these conditions, from the neonatal period through adulthood, assess nutritional status, onset and progression of obesity, cognitive impairment, genotype-phenotype correlations and clinical comparisons between PWS and other children with EMO of unknown etiology (Miller et al. 2006; Hampton 2006).

## **Session 2: Psychiatric and behavioural disorders**

**Chair: LMG Curfs**

### **Overview and mechanisms**

**A Holland**

It is now well established that there is a characteristic profile of behavioural and psychiatric disorders associated with PWS. These include the following: a) a marked propensity to over-eat present from early childhood. This is universal in PWS and lifelong although may vary in intensity; b) repetitive and ritualistic behaviours and temper outbursts that may affect up to 80% of people with PWS; c) skin picking affecting approximately 50% of people with PWS; d) affective disorders including mood fluctuations, bipolar disorder, and affective psychotic illness. Affective disorders in general are very common in PWS, with affective psychotic disorders being significantly more prevalent in those with PWS due to maternal UPD compared to those with deletion (Boer et al., 2002 ; Vogels et al., 2003; Whittington et al., 2004).

Given the above epidemiological findings the focus of research is now directed at understanding the biological and other mechanisms that might account for the relationship between the PWS genotype and this characteristic phenotype. The identification of such mechanisms would potentially inform

the development of preventative strategies and treatment. There are different conceptual models that inform our understanding of maladaptive behaviours in people with intellectual disabilities in general. Such models indicate that multiple factors are likely to be important and distinctions need to be made between those biological, psychological, developmental or socially determined factors which predispose and maintain the propensity to such behaviours, on the one hand, and those which precipitate their occurrence, on the other. Key questions include: a) what is it about PWS that is specifically associated with the above cluster of behaviours? b) are similar or different mechanisms responsible for these various behaviours and the risk of affective disorder?; and c) how can we explain why such problems are not universal in PWS? Theoretical mechanisms proposed include specific deficits affecting hypothalamic feeding pathways, the effects of delayed development and therefore the abnormal continuation into adult life of behaviours that are common in childhood, and changes in the risk (or liability) for specific disorders as a result of hypothesized changes in brain function in PWS.

### **Is there a relationship between autism and PWS?**

**B Rogé**

The definition of autism has greatly extended still its first description. Given the diversity of the autistic disorders, the term of “autistic spectrum disorder” (ASD) had emerged. Prader-Willi syndrome is a developmental disorder which shared some behavioural features with other pathologies such as obsessional-compulsive disorders and autism.

In 2005, twelve studies regarding ASD in PWS and Angelman syndrome (AS) were reviewed by Veltman et al. It was noteworthy that among the genetically confirmed cases of PWS and AS, the prevalence of ASD was 25.3% in PWS and 1.9% in AS.

In 2006, Descheemaeker et al. investigated the co morbidity of pervasive developmental disorder (PDD) in 59 Prader-Willi syndrome individuals and in 59 non-specific mentally retarded controls. Nineteen percent of the PWS group did meet the full diagnostic criteria for PDD vs 15% in the “control” group. These results revealed that a higher IQ in PWS did not protect to develop genuine PDD and that uniparental disomy seemed to be an additional risk.

In 2006, Dimitropoulos et al. studied the compulsive behaviours in 68 children with PWS in comparison to 86 children with autism, and 57 children with developmental delays. Children, including young children, with PWS exhibited food- or non-food-related compulsive behaviours, more severe ritualistic behaviour than children with autism. Interestingly, the severity of non-food-related rituals was related to the severity of eating behaviour in PWS.

In 2006 Greaves et al. investigated the range of repetitive behaviour seen in individuals with PWS beyond food-related behaviours. Their findings emphasised the insistence on sameness, 'just right' behaviours, and the overlap with behaviour problems seen in children with autism.

Poor social adjustment has been reported in PWS. Koenig et al. (2004) reported that PWS children were comparable to a group of subjects with a PDD and performed significantly more poorly than subjects with comparable intellectual ability. Poor performance on this task by the PWS subjects suggests an underlying difficulty interpreting social information that is presented visually, which may be a critical factor in the impairment in social functioning in this population.

It was predicted that maternal UPD PWS cases would be more prone to ASD than Del PWS cases due to their duplicated maternally expressed genes. The results published by Veltman et al. in 2004 lend further support to the notion that abnormality in the expression of maternal imprinted 15q11-13 genes may confer a susceptibility to ASD.

In conclusion, there is a link between Prader-Willi syndrome and autistic spectrum disorder at the behavioural level, the social standing and cognitive level and at the genetic level.

### **Psychotic illness**

**S Soni**

There are now three published studies reporting that psychotic illness has a high prevalence in PWS due maternal UPD in comparison with those with deletion (Boer et al., 2002 ; Vogels et al., 2003; Whittington et al., 2004). The studies vary in the reported age of onset of the psychotic illness. There are undoubted cases where the onset is as early as 10 years of age but it is usually in early adult life. In a study of 119 adults with PWS investigating the phenomenology of the psychiatric illness (Soni, 2007) it was reported that mood fluctuations and mood disorders were very common in PWS, in general. It again confirmed that those with maternal UPD were most at risk for psychotic illness. This illness was predominately affective in nature, usually with a sudden onset, associated with delusions and hallucinations and sometimes atypically, for a mood disorder, associated with confusion. For those rarer examples of people with deletion and psychosis this was associated with a history of affective disorder in the mother in 50% of the 13 individuals meeting such criteria. In one study (Vogels 2004), life events (including physical symptoms) were found to precede or coincide with psychotic illness. It has been proposed that those with maternal UPD may be at risk for a more severe affective disorder (and therefore for psychotic phenomenology) and that theoretically this and the family history findings in those with deletion and psychotic illness are best explained by the unbalanced expression of a maternally expressed/paternally imprinted gene on chromosome 15.

### **Does GH improve cognitive and social development?**

**A Hokken-Koelega**

There are few data on GH effects in toddlers with PWS. Carrel et al in 2004 reported beneficial effect of GH treatment in 29 infants and toddlers with improved body composition and mobility acquisition when GH was started before 1.5 years of age.

The objectives of our study started in 2002 was to evaluate the effect of GH on height, weight, body composition, physical strength and activity level, psychomotor development and sleep-related breathing disorders. We included 43 children from 6 months to 3 years divided in 2 groups: one treated with GH for 2 years and the second been as a control 1 year and receiving GH for a subsequent year. Psychomotor development was scored with Bayleys scale and BSID II-NL.

All children were below normal particularly for mental development. Interestingly the group treated with GH during the first year had a greater increment in head circumference than the control group. Change in mental and motor development was significantly higher in children treated with GH. The benefit for mental development was even higher in the children with higher development delay at baseline, and the opposite was found with motor development.

A particular point dealing with the occurrence of Obstructive Sleep Apneas (OSAS) and psychomotor development was presented. Four children among 22 had OSAS and their mental development was significantly lower (65.5 vs. 74.9) with a trend for higher BMI (1.4 SDS vs. 0.6 SDS).

These preliminary data suggest that GH should be started early and that treatment of OSAS might significantly improve mental development.

### **Session 3: Breathing and sleeping abnormalities**

**Chair: M Ritzén**

#### **Prevalence of sleep related breathing disorders in children with PWS**

**DAM Festen**

Several cases of sudden death in GH-treated and non-GH-treated, mainly young, children with PWS were reported. For that reason, studies were performed to evaluate effects of GH on respiratory parameters (Festen et al, 2006; Miller et al, 2006; Nixon et al, 2002; Lindgren et al, 1999; Haqq et al, 2003). In 53 prepubertal, genetically confirmed PWS children (30 boys), with a median (interquartile range) age of 5.4 (2.1–7.2) years and body mass index of +1.0 SD score (–0.1 to 1.7), a polysomnography (PSG) was performed, before GH in 53 children and repeated after 6 months of GH treatment in 35 of them (Festen et al, 2006). Children were randomly assigned to either treatment with GH 1 mg/m<sup>2</sup>/day or no treatment for 1 year.

Before start of GH therapy, an increased Apnea Hypopnea Index (AHI) of 5.1 per hour (2.8–8.7) (normal 0–1 apnea per hour) was found, mainly due to an increased number of central apneas and hypopneas. The duration of the apneas was relatively short, 15.0 (13.0–28.0) seconds. AHI did not correlate with age or body mass index, but central apneas decreased with age ( $r = -0.34$ ,  $P = 0.01$ ). In the total group of mainly non-obese PWS children, obstructive apneas were rare, but they were found in four of the eight overweight patients. During 6 months of GH treatment, a non-significant decline in AHI was found from 4.8 (2.6–7.9) at baseline to 4.0 (2.7–6.2;  $P = 0.36$ ), mainly due to a

lower number of central apneas. During GH, no significant change in obstructive apneas was found. During upper respiratory disease, however, a marked increase in obstructive apneas (OSA) was found. Notably, one patient died unexpectedly during a mild upper respiratory tract infection (URTI), although he had a nearly normal PSG, indicating that a relatively normal PSG does not exclude the possibility of unexpected death during mild URTI.

In conclusion, most PWS children have sleep related breathing disorders (SRBDs) with a high AHI, mainly due to central apneas. BMI or age cannot explain the variability in the severity of the SRBD, although OSA was more prevalent in children with obesity than in normal-weight children. After 6 months of GH, a non-significant decrease in AHI was found. Thus, our data are reassuring with respect to the effects of GH on SRBDs. A normal PSG does, however, not exclude the possibility of unexpected death during mild URTIs. During URTI, AHI may rise and obstructive apneas may occur. Monitoring during upper respiratory tract infection in PWS children should be considered.

### **Sleep disorders: diagnosis and treatment, a practical approach**

**B Schlüter**

A multitude of sleep disorders is listed by the International Classification of Sleep Disorders (ICSD) and is systematically classified as dyssomnias, parasomnias and sleep disorders associated with physical and psychic diseases, respectively (Stores, 1999).

From early infancy to adolescence, physiologic sleep parameters (e.g. total sleep time (TST), sleep efficacy, proportions of non-REM and active-REM sleep) show marked age-related changes in normal children. The development of the circadian rhythm of sleep and wakefulness is influenced by brain maturation as well as social and environmental factors. Patients with hypothalamic dysfunction like subjects with PWS are at increased risk for disturbances of the rhythm of sleep and wakefulness (Schlüter, 2003).

In case of hypersomnia, too much sleep, pathologic causes are narcolepsy and sleep fragmentation due to sleep apnea or periodic leg movements during sleep. Sleep apnea is often accompanied by excessive sweating and other vegetative symptoms. Involuntary motor activity like periodic leg movements and restless legs can cause sleep fragmentation and a non-restorative sleep. Parasomnias, like night terrors and sleep walking, are examples of complex behavior, which are frequently observed in children. They have to be differentiated from sleep-related epilepsy. Disturbed sleep may not only lead to hypersomnia but also to abnormal daytime behavior with irritability, aggression, concentration deficit, resembling attention deficit hyperactivity disorder.

In PWS patients, hypersomnia is mainly the result of obstructive sleep apnea and of a sleep-wakefulness rhythm disorder due to the hypothalamic dysfunction. Advice about sleep hygiene is one of the main therapeutic interventions. "Ideal" sleep medication does not exist. Therapeutic

options with respect to sleep-disordered breathing include nasal continuous positive airway pressure (NCPAP), non-invasive positive pressure ventilation and tonsillectomy/adenoidectomy. Behavioral interventions are preferably used in children with difficulties of initiating and maintaining sleep. Sleep-wakefulness rhythm disorders may improve with light therapy.

In conclusion, PWS patients are at increased risk for sleep-related breathing disorders, disturbances of the circadian cycle of sleep and wakefulness and behavioral sleep disorders.

**Breathing abnormalities: is there any relationship with IGF-I levels?**

**JL Miller**

All individuals with PWS have some degree of sleep disordered breathing disorders (SRBD), mainly central apneas (Haqq et al, 2003; Miller et al, 2006; Festen et al, 2006). Most of them have also low serum levels of Insulin-like Growth Factor (IGF)-I, one of the major GH-dependent growth factors. GH treatment (1 mg/m<sup>2</sup>/day) improves SRBD, predominantly by reducing the number of central apneas (Haqq et al, 2003; Miller et al, 2006; Festen et al, 2006). At the same time, GH therapy induces a significant increase in serum IGF-I levels, into the mid-normal range. Central sleep apneas in PWS children are due to hypothalamic dysfunction, whereas obstructive apneas in PWS children can be due to hypotonia, obesity, large tonsils and large adenoid, gastroesophageal reflux, an upper airway respiratory infection or a combination of these factors. Supraphysiologic levels of IGF-I have been associated with onset or worsening of obstructive sleep apnea in children with GH-insensitivity syndrome, perhaps due to enlargement of tonsillar/adenoidal tissue (Backeljauw et al, 2001).

In case increased IGF-I levels would induce tonsillar and adenoidal hypertrophy in PWS children, the situation might be worse, as it might come on top of hypotonia and obesity. For that reason, it is strongly recommended to monitor serum IGF-I levels in individuals with PWS during GH therapy, thereby targeting dosing to achieve IGF-I levels within the normal range for age. Additionally, if GH-treated PWS children show signs of obstructive apnea, such as onset or worsening of snoring, breathing pauses greater than 5 seconds or increased daytime sleepiness, serum levels of IGF-I should be assessed, evaluation by an otolaryngologist should be performed and an overnight polysomnography (PSG) should be considered, even if the dose of GH has not been recently changed. In case of a slight or greater enlargement of the tonsils and/or adenoid, an adenotonsillectomy should be strongly considered.

In conclusion, all individuals with PWS have some degree of SRBD, which improves during GH therapy. GH dose should be targeted at IGF-I levels in the mid-normal range. If symptoms of SRBD emerge during GH therapy, IGF-I levels should be assessed, and a PSG and adenotonsillectomy should be considered.

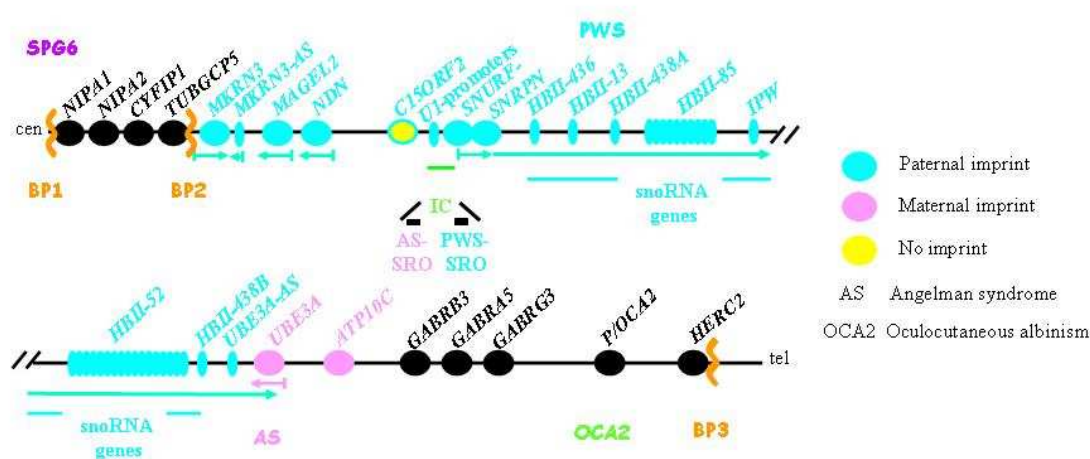
## Session 4: New insights on the genetics of PWS and their clinical relevance

Chair: F Muscatelli

### New genes involved in PWS

RD Nicholls

Segmental duplications in the human and mouse PWS genomic region lead to instability and incomplete genomic sequence. The origin of PWS deletions is believed to be homogeneous recombination between duplicated sequences of very high homology (>98.5 % identity). The initiating event is likely to be a double strand DNA break. Different mechanisms lead to different type of deletions, duplications, triplications, inversions and inversion duplications inside the PWS region (Amos-Landgraf, 1999). Major deletions (type I and II) derived from DNA breakpoints BP1, BP2 and BP3 (see figure).



Additional structural genomic variations could exist in the 15q11-q13 region (Chai, 2003; Zody, 2006) and particularly in the region between BP 1 and BP2 where the mean sequence assembly is a guess and the order of the 4 genes identified not sure. The NIPA1 gene encodes for the spastic paraplegia disorder (SPG6). It's a late (12-35 years) onset spastic paraplegia with progressive evolution leading to severe disability by the late forty's due to axonal degeneration in CNS especially corticospinal tracts to legs. This gene encodes for  $Mg^{2+}$  transporter with 9 transmembrane domains.

The mouse model (Stefan, 2005) of transgenic (Tg) PWS/Tg AS has been developed with LMP2A insertion leading to a deletion of PWS-AS region. The Tg PWS neonate mouse developed failure to thrive and neonatal lethality with impaired glucose metabolism (hypoglycemia starting the 2<sup>nd</sup> day post-natally with concomitant hypoinsulinemia and hypoglucagonemia). This is associated with perturbed islet development and function in this model. The severe hypoglycemia was possibly due

to failure to initiate gluconeogenesis following glycogenolysis in the newborn.  $\beta$ -cells and  $\alpha$ -cells were significantly decreased with a pancreatic insufficiency (Stefan, 2005). All imprinted genes are expressed in WT pancreas but not in Tg PWS pancreas (*Snurf-Snrpn*, *Ndn*, *Magel2*, *Mkrn3*, *Mirh1*). These genes were also expressed in “insulinoma”  $\beta$ -cells. The 1 Mb region between *Ndn* and the IC/*Snurf-Snrpn* includes a long non coding host gene for at least one class of micro RNA (miRNA).

A novel imprinted host gene encoding mi RNA was reported, *Mirh1 miR-344*, upstream the imprinting center. The *Mirh1* miRNA family is homologous to *miR-344*, a known rodent miRNA. At least 39 copies of related sequences for each putative premiRNA are encoded by *Mirh1*. The *miR-344* miRNA was shown to be brain imprinted in mouse. The promoter-exon 1 and one intronic region of the *Mirh1* host transcript are highly conserved in mammals, suggesting that a human ortholog(s) of the PWS miRNAs or other function exist. Many other imprinted PWS-region ESTs arise from unstable, low level transcribed pieces of the snoRNA host transcript (originating from *Snurf-Snrpn*) or the *Mirh1* miRNA host transcript. Several ESTs between *Mirh1* exon 59 and the IC encode potential polyGlu- or polySer- ORFs but functional evidence of polypeptides remains lacking. Alternative promoters of *U1-Snurf-Snrpn* were probably NRF-1 binding sites regulating most of the PWS imprinted loci. NRF-1 cluster was located between *Ndn* and *Snurf-Snrpn* genes. The PWS genomic domain of human and mouse shows a high level of genomic, genetic and epigenetic complexity. A complete knowledge of the protein and RNA coding functions in the PWS region is needed in order to understand PWS.

### **Does the genotype influence our care?**

**M Butler**

A paternal 15q11-q13 deletion is found in ~70% of subjects with Prader-Willi syndrome, ~25% have uniparental maternal disomy 15 (UPD), and the remaining 2% to 5% have imprinting defects. The proximal deletion breakpoint in the 15q11-q13 region occurs at one of two sites located within either of two large duplicons allowing for the identification of two deletion subgroups. The larger Type I (TI) deletion involves breakpoint 1 (BP1), which is close to the centromere, whereas the smaller Type II (TII) deletion involves breakpoint 2 (BP2) located ~500 kilobases distal to BP1. Breakpoint 3 (BP3) is located at the distal end of the 15q11-q13 region and is common to both typical deletion subgroups. Four genes have been localized in the region between BP1 and BP2 (Bittel and Butler, 2005):

- *NIPA1*, widely expressed in CNS, encodes for a  $Mg^{2+}$  transporter with 9 transmembrane domains; mutations are associated with autosomal dominant spastic paraplegia (SPG6).
- *NIPA2* encoding for a 360 amino acids protein with 9 transmembrane domains suggestive of transporter or receptor function

- *CYFIP1* interacts with *FMR1* protein which causes Fragile X syndrome
- *GCP5*, Gamma-tubulin complex component 5

Distinct differences have been reported between individuals with PWS having the typical deletion compared with UPD in physical, cognitive, and behavioral measures. For example, hypopigmentation and homogeneous clinical presentations are more often seen in individuals with PWS with a typical deletion compared with those having UPD or an imprinting defect. Higher verbal intelligence quotient (IQ) scores have been reported in PWS individuals with UPD compared with PWS subjects having the 15q deletion particularly in four subcategories of verbal testing: information, arithmetic, vocabulary, and comprehension. Similarly, Dykens and others also reported behavioral and cognitive differences in individuals with PWS including better jigsaw puzzle (spatial) skills and more skin picking in deletion subjects (Cassidy et al., 1997; Dykens et al., 1999). Those subjects with UPD have fewer maladaptive behaviors determined by internalizing, externalizing, and total domain scores. Conversely, visual processing of complex stimuli is significantly poorer in individuals with UPD compared with those having the deletion; however, they show more visual memory skills than deletion subjects. There also may be a tendency for improvement of behaviour in PWS subjects (deletion and UPD) with advancing age although psychotic problems may be more common in adult PWS subjects along with a tendency for autistic findings in the UPD group.

Analyses of the genetic subtypes to date have primarily compared individuals with typical deletions and uniparental maternal disomy 15 without grouping the individuals with a deletion into TI or TII. Recently, clinical differences have been reported in individuals with PWS categorized as having TI or TII deletions (Butler et al., 2004). For example, adaptive behaviour, obsessive-compulsive behaviors, and reading, math, and visual-motor integration assessment scores were generally poorer in individuals with PWS having the TI deletion compared with subjects with PWS and the TII deletion or with UPD. In addition, subjects with a Type I deletion had more compulsions and poorer adaptive behavior than subjects with Type II deletions. Their compulsions were particularly related to grooming and bathing and these compulsions were more disruptive of daily living than found in subjects with Type II deletions or UPD. Similarly, intellectual ability and academic achievement were poorer in subjects with Type I deletions. In general, subjects with Type I deletions have more behavioral and psychological problems than individuals with Type II deletions or UPD. Additionally, UPD subjects may be at risk for single gene conditions due to isodisomy 15 (genetic material from the same chromosome 15) by receiving two maternal copies of a single recessive 15q allele from a heterozygous mother. Similarly, X linked recessive conditions could occur in the UPD

subject due to extreme skewness of the X chromosome resulting from rescue of trisomy 15 cells leading to a small number of viable cells in early embryo development.

The four genes between BP1 and BP2 presumably involved with neurodevelopment and function become candidates for behavioral differences observed between TI and TII subjects. Therefore, we undertook gene expression studies using quantitative RT-PCR from mRNA isolated from lymphoblasts derived from 17 subjects with TI and TII deletions (Bittel et al., 2006). We compared expression levels (i.e., gene fold change) with cognitive and behavioral assessment scores and found a positive correlation with gene expression, particularly with *NIPA2*, in adaptive behavior and cognition and a negative correlation with maladaptive scores. The coefficient of determination suggested that expression of the four individual genes accounted for as much as 75% of the variation in scores. In addition, the joint impact of the four genes explained from 24% to 99% of the assessment scores obtained from the subjects further supporting their role in cognition and behavior in PWS.

In summary, evidence to date supports clinical differences between UPD and deletion subjects with PWS including behavioral differences in TI and TII subjects. Awareness of these clinical differences (strengths and weaknesses - e.g., higher verbal IQ and visual memory skills in UPD and more compulsions and self-injurious behavior in deletion subjects) and arranging appropriate care with better educational and treatment plans for PWS subjects based on their genetic subtype (UPD-heterodisomy vs. isodisomy or deletion -TI and TII) could lead to a better quality of life, prognosis and outcome for the person with PWS and their family.

### **Imprinting defects**

**B Horsthemke**

Imprinting defects is present in less of 1% in PWS patients. Imprinting center deletion screening in 85 unrelated patients showed that deletion occurred in only 11 (15%) patients: 5 familial deletions including 1 case with paternal somatic mosaicism and 6 *de novo* deletions or germ line mosaics (1 germ line mosaic proven). In the *de novo* deletion there is no theoretical high risk of recurrence but the presence of germ line mosaicism explains an increased recurrence risk in some cases. The risk of recurrence in the absence of deletion is found is theoretically 50% (El-Maarri et al., 2001).

The different theoretical steps leading to genomic imprinting included imprint erasure in primordial germ cells, imprint establishment in germ cells and imprint maintenance in embryo. In 19 families without IC deletion, the abnormal paternal allele was inherited by the grand-mother suggesting a failure of imprinting erasure in all the studied cases.

There was no significant association between haplotype and imprinting in 41 PWS families studied.

### **C/D snoRNAs and the Prader-Willi syndrome**

**J Cavallé**

Small RNAs of the C/D type (or C/D RNAs) represent one of the largest families of eukaryotic non-coding RNAs, with more than 100 different RNA species described so far. In mammals, most of them are involved in the biosynthesis of 2'-O-methylations of ribosomal RNAs (rRNAs) and/or spliceosomal RNAs (U-snoRNAs). They act through the formation of a specific RNA duplex (10 to 21 nt long) at each modification site in which the methylated nucleotide is systematically paired to the fifth position upstream from the D or D' box (5'-CUGA-3'), a conserved sequence motif found in all the C/D RNAs. Hence, these small RNAs have been also termed RNA methylation guides.

Pioneer RNomics approaches performed by A. Hüttenhofer and J. Brosius led to the identification of many novel C/D RNAs, with a few of them -MBII-13, MBII-52 and MBII-85- were found to be mainly (or exclusively) expressed in the mouse brain. In human, their orthologous genes (called HBII-13, HBII-85 and HBII-52) are clustered at the Prader-Willi locus between the *SNURF-SNRPN* and *UBE3A* genes and they are expressed from the paternal allele only (Cavallé et al., 2000; Runte et al., 2001). These C/D RNA genes are embedded within introns flanked by non-coding exons and they are predicted to be processed from a single very long primary transcript (>460 kb) believed to start at the *SNURF-SNRPN* promoter and overlapping the maternally-expressed *UBE3A* gene in the antisense orientation (Runte et al., 2001).

The paternally-expressed candidate gene(s) involved in PWS is (are) still largely unknown. Studies of knock-out mice models suggest that the candidate PWS gene(s) lie(s) between *Snrpn* and the MBII-52 gene cluster. Rare balanced translocations in human PWS patients have also enabled the identification of a minimal critical region that starts at intron 17 of *SNRPN* gene and extends to the end of the HBII-85 snoRNA gene cluster (Schule et al., 2005). This minimal critical region includes 47 copies of HBII-85 and HBII-438a. Since the mouse 7C imprinted locus lacks HBII-438, this latter is unlikely to play a significant role in PWS. On the contrary, HBII-85 snoRNA has been proposed to be, wholly or partly, at the origin of PWS-associated pathogenesis (Cavallé et al., 2000; Schule et al., 2005). Any major role of HBII-52 in contributing to PWS has been recently excluded as individuals carrying a deletion covering the entire HBII-52 cluster do not show any obvious clinical phenotype indicating that lack of expression of HBII-52 genes alone is not sufficient to promote PWS (Schule et al., 2005).

The functions of these imprinted C/D RNA genes are still unknown as they do not seem to be involved in rRNA or snRNA biogenesis (they are “orphan small RNAs” since they lack long complementarities to these RNAs). Interestingly, HBII-52 has a conserved, 18-nucleotide-long antisense element against a brain specific pre-mRNA, the serotonin receptor 5-HT<sub>2C</sub>. The 5-HT<sub>2C</sub> receptor is a trans-membrane signaling receptor coupled to heterotrimeric G proteins. Its mRNA possesses five A-to-I (adenosine to inosine) editing sites (A-E edited sites). A-to-I RNA editing is the site-specific modification of adenosine to inosine within precursor mRNAs that is catalyzed by members of the ADAR (adenosine deaminase acting on RNA) family. As translation occurs, an inosine is read as a guanine and consequently, a combination of 5-HT<sub>2C</sub> mRNA isoforms with different sequences exists and gives rise to sensitively different proteins, whose binding properties to G proteins differ. Importantly, the nucleotide predicted to be targeted by MBII-52 - the edited C-site - is known to play a pivotal role in regulating serotonergic signal transduction, suggesting that MBII-52 might specifically modulate the efficiency of RNA editing at this site. Indeed, we have recently shown that targeting the C-site for 2’O-methylation by MBII-52 significantly decreases the efficiency of RNA editing at that site specifically (Vitali et al., 2005).

## **Session 5: Unresolved questions on the endocrinology of PWS**

**Chair: G Chiumello**

### **Is ghrelin involved in the PWS phenotype?**

**BP Hauffa**

Ghrelin, the only known orexigenic hormone, may play a role in the development of hyperphagia in PWS. Ghrelin is expressed in the fetal pancreas; postnatally in the gastric fundus (X/A cells), but also in the hypothalamus, it crosses the blood-brain barrier and increases before meals to concentrations sufficient to stimulate hunger and food intake. Ghrelin is regulated by the state of energy homeostasis and involved in long-term body weight regulation, circulating in relation to energy stores and showing compensatory changes in response to body weight alterations. Ghrelin concentrations are inversely associated with BMI. Exogenous application of ghrelin increases hunger and calories eaten, and stimulates GH release.

In PWS, fasting and post-prandial levels of plasma ghrelin are greatly elevated (Cummings et al., 2002; DelParigi et al., 2002; Haqq et al., 2003, Goldstone et al., 2005). Ghrelin is suppressed by a mixed meal and by individual nutritional components as glucose in PWS (Paik et al., 2006). This appears to be at least partially explained by the preserved insulin sensitivity and hypoinsulinemia, compared to normal obese subjects (Goldstone et al., 2005). Although somatostatin suppresses

plasma ghrelin concentrations in PWS patients, appetite is not reduced (Tan et al., 2004). Ongoing studies are looking at the possible benefit of chronic administration of long acting somatostatin analogues in PWS. GH treatment has been found to lower total, but not acylated ghrelin in one study (Hauffa et al., 2007). Use of pharmacological agents that specifically abolish ghrelin action, as the spiegelmers, may be useful to further elucidate the role of ghrelin in PWS (Kobelt et al., 2006).

Any possible role for hyperghrelinemia in causing GH deficiency in PWS is difficult to investigate without such antagonists. Furthermore the role of endogenous ghrelin in controlling physiological GH secretion in humans is unclear.

### **PWS and peripheral satiety signals**

**AP Goldstone**

Appetite is controlled by a variety of peripheral signals that change in response to food intake, acting on the brain to alter feelings of hunger and satiety in order to determine meal initiation and termination. Satiety signals include a number of ascending neural inputs as vagal signals from the gut, as well as metabolic and hormonal changes, including plasma glucose, gastrointestinal release of anorexigenic hormones such as peptide YY (PYY), pancreatic polypeptide (PP), oxyntomodulin, glucagon-like peptide-1 (GLP-1) and cholecystokinin (CCK), suppression of ghrelin, and synergism with the anorexigenic adipocyte-derived leptin. PWS patients have delayed meal termination, earlier return of hunger after the previous meal with early meal initiation (Holland et al., 1993). Given free access to food, patients will consume approximately three times that of control subjects (Zipf and Berntson, 1987). This occurs despite delayed gastric emptying (Choe et al., 2005). There is no evidence for defects of leptin, CCK, PYY, or GLP-1 secretion in PWS that would contribute to hyperphagia, and gastrin, obestatin, glucagon and gastric inhibitory peptide levels are normal. In addition to elevated plasma ghrelin, pancreatic polypeptide deficiency and perhaps relative hypoinsulinemia may contribute to hyperphagia in PWS (Goldstone et al., 2005; Zipf et al, 1983).

It appears that in addition to hormonal abnormalities contributing to hyperphagia in individuals with PWS, there are overriding brain defects, including hypothalamic defects, which lead to resistance in peripheral satiety signals (Goldstone, 2004). Functional studies of the brain including PET and fMRI have revealed abnormal brain activation patterns in corticolimbic structures, at baseline and after food ingestion in PWS, suggesting altered reward and motivational responses to food, that may underlie the hyperphagia in PWS (Hinton et al., 2006; Shapira et al., 2005; Holsen et al. 2006; Miller et al., 2006).

Knowledge of the hormonal and structural defects will open the possibility of new therapies targeting hypothalamic and extrahypothalamic brain structures to reduce hyperphagia in PWS.

### **What is wrong with the hypothalamus?**

**DF Swaab**

Although hypothalamic dysfunction can explain many of the features of PWS, a unifying concept of disordered hypothalamic regulation and endocrine abnormalities has yet to be formulated.

At the hypothalamic level, the paraventricular nucleus (PVN) is decreased in volume by 28% and in total cell number by 38 % in PWS patients. This is due at least in part to a 42% reduction in number of the oxytocin neurons, involved in satiety control, and may contribute to the insatiable appetite and obesity of the patients (Swaab et al., 1995). The processing of the vasopressin precursor can be affected (Gabreels et al., 1998). The daytime hypersomnolence observed in some PWS patients may be related to disordered components of the circadian system (suprachiasmatic nucleus, pineal gland) as the number of hypocretin (orexin) neurons in the lateral hypothalamus is normal (Fronczek et al., 2005). Short stature affecting 95 % of PWS adults is due to a decreased height velocity in the first years of life and the lack of the pubertal growth spurt, the latter being caused by hypogonadism, the major part of which is of hypothalamic origin. Studies of GnRH neurons have yet to be reported in humans with PWS. Most patients with PWS fulfil the laboratory criteria of growth hormone deficiency (Burman et al., 2001). However, growth hormone releasing hormone (GHRH) cell number in the arcuate or infundibular nucleus (INF) was not found to be reduced in PWS patients compared to normal and obese individuals (Goldstone et al., 2003).

Animal studies have demonstrated the importance of neuropeptide Y (NPY) and agouti-related protein (AGRP) neurons for the stimulation of feeding, and of the anorexigenic action of proopiomelanocortin (POMC) containing neurons in the INF. Quantitative neuroanatomical studies of human tissue did not show abnormalities of NPY-, AGRP- or POMC-containing neurons in PWS patients compared to obese controls. NPY- and AGRP- immunostaining increased appropriately with pre-mortem illness duration, indicating that these neurons are capable of responding normally to alterations in peripheral signals (Swaab et al., 1995; Swaab, 1997; Goldstone et al., 2002). Anatomical defects of these neurons may therefore not explain the hyperphagia in PWS. Preliminary data have suggested that there may be an increased development of Alzheimer-like hyperphosphorylated Tau neurofibrillary tangles particularly in the PVN, temporal cortex and nucleus basalis of Meynert of adults with PWS.

**Early diagnosis and multidisciplinary care**

**M Tauber**

We analysed our recent practice in Prader-Willi syndrome (PWS) infants to describe and evaluate the impact of very early diagnosis and multidisciplinary care on the evolution and care of infants presenting with PWS. Nineteen infants diagnosed with PWS before the second month of life (median age 1 month [0.01-2]) were followed by our multidisciplinary team. Median age at the time of analysis was 3.1 yrs [range 0.4-6.5]. The data were compared with those collected in 1997 from 113 questionnaires filled out by family members of the French PWS association. The patients from this latter data set were 12.0 yrs [range 4 months-41 yrs] at the time of analysis, with a median age of 36 months at diagnosis, and had never been followed by a multidisciplinary team.

All infants of the present cohort presented with hypotonia at birth compared to 76% in the 1997 cohort. Diagnosis of PWS was made by neonatologists in 89.5% in the present cohort compared to 13.9% in the previous one. The duration of their hospitalisation time was significantly reduced from 30.0 [range 0-670] to 21 [range 0-90] days ( $p=0.043$ ). The duration of gastric tube feeding was significantly reduced from 30.5 [range 0-427] to 15 [range 0-60] days ( $p=0.017$ ). Questions about tube feeding remained accurate: should it be systematic? And if yes, what is the optimal duration? What could be the consequences of tube feeding on oral hypotonia, speech problems and on gut hormones abnormalities? Moreover, nutritional recommendations at this age are missing. Age at walking was not different (Median age 24 months). Early physiotherapy in this population need to be discussed regarding the type of the program its duration and its evaluation. GH testing and IGF-I evaluation have been performed in all infants showing a median GH peak of  $6.3 \pm 5.3$  mcg/L and low IGF-I levels  $46 \pm 41$  ng/mL. 38% of them presented a complete GH deficiency and 30.7 % a partial GH deficiency. More data are needed to precise the prevalence of GH deficiency and low IGF-I levels at this age. Growth hormone (GH) treatment was started at a mean age of  $1.9 \pm 0.5$  yrs in ten infants. There is not a consensus on when to start a GH treatment before of after the 1<sup>st</sup> year, at what dose, and how to monitor it. L-thyroxine was given in six infants due to low T4 levels. There are no clinical reports on the prevalence of hypothyroidism at this age. Only one infant became obese at 2.5 yrs, thus the prevalence of obesity in our present cohort was 0% at 1 year and 2 years and 11% at 3 years compared to 14.3, 12.2, and 29.6 % respectively in the 1997 cohort. Issues on parental guidance and support remained to be discussed i.e. impact of group educational sessions, how to avoid too much pressure on the parents.

Early diagnosis is possible and relies on neonatologists. Early diagnosis combined with multidisciplinary care decreases the hospitalisation time and duration of gastric tube feeding and prevents obesity in PWS infants.

### **Nutritional phases**

**DJ Driscoll**

PWS has been classically described as having 2 nutritional phases: 1) poor feeding and frequently failure to thrive (FTT) and 2) onset of hyperphagia leading to obesity (Butler et al, 2006; Cassidy, 2001).

Phase 1 occurs from birth to early infancy when infants with PWS have central hypotonia, a poor ability to suck and often require tube feeding. Phase 2 is described as beginning between age 1 and 6 years, usually between 2 and 4 years (Cassidy, 2001).

McCune et al, however, have longitudinally followed over 150 individuals with PWS for many years and suggest that a much more complex progression of the nutritional phases occur which greatly expands the traditionally accepted 2 nutritional phases (Mc Cune et al, 2005). Based on their observations there are four main phases, with sub phases occurring in the first two main phases.

In the first phase, the infant is hypotonic and not obese. Sub-phase 1a consists of feeding difficulties with or without FTT. FTT might even occur when the child receives adequate calories for age. In sub-phase 1b, the infant is growing steadily along a growth curve, at a normal rate.

In the second phase, the weight starts to increase, thereby crossing weight percentiles. This generally occurs between 18-36 months of age. In sub-phase 2a, the child's weight increases such that it crosses 1-2 or more weight percentiles without a significant increase in calorie intake. During this phase the children do not show an increased interest in food. This suggests that the early-onset obesity is not a result of hyperphagia but has a different etiology. In sub-phase 2b, the child increases its daily calorie intake and becomes overweight/obese. They have an abnormally increased interest in food, but they do not yet have the insatiable appetite and constant food seeking exhibited in phase 3. Nowadays, in some families the food and weight are so closely monitored that the child may not become obese in phase 2 or may not become obese at all. Thus, children in phase 2 become obese due to their increased interest in food, unless the family is able to restrict their food intake. Recently, growth hormone therapy has changed this phase for many children as it increases the child's metabolism, reduces appetite and increases lean body mass.

During the third phase, the typical hyperphagia starts, accompanied by aggressive food-seeking and a lack of satiety. The onset is quite variable as it may appear as early as 3 years of age or as late as 15 years. This is the classical phase that most people typically associate with PWS. A small minority of individuals with PWS never enter this phase. Although many individuals are obese in

this phase, some do not because their families and group homes monitor their food intake very closely.

In the fourth phase, an individual who was previously in phase 3 has no longer an insatiable appetite and can feel full. Families and care-takers note a significant improvement in appetite control. While individuals in this phase may still have a “greater than normal” appetite, it is not as aggressive and unrelenting as previously observed. McCune and Driscoll describe that this phase does not start until adulthood, between 21 and 50 years of age (McCune et al, 2005). They also reported that not all PWS individuals necessarily go through all the phases and sub-phases delineated above, but most do.

Further research is required because a better understanding of the various nutritional phases in PWS may help in the management of PWS individuals.

### **Motor training in PWS**

**MWG Nijhuis-van der Sanden**

Infants with PWS have infantile muscular hypotonia, decreased muscle mass and psychomotor delays (Schmidt et al, 2004). Their muscle fibres are different from controls as they have a greater variability in the size of both type I and II fibres, and atrophy of type II fibres. Type II fibre development is activity-dependent and it has been suggested that the lower number of type II fibers might be due to the reduced motor activity of PWS subjects.

Developmental processes are modified by activity in the developing central nervous system (CNS). The period during which experience can alter the neural representation defines the *sensitive period*. During *sensitive periods* children rapidly develop fundamental capabilities upon which subsequent development can be build. This development is influenced by biological and environmental factors and individual variations in resiliency and vulnerability. Lack of appropriate stimulation, consistent responsive care and opportunities to explore the immediate environment during the sensitive periods, may lead to less development of connections within the brain which should facilitate essential learning and lead to self-regulating skills. Not only the development of CNS but also the development of the muscle structure is activity-dependent.

During the early years of life, the hypotonic PWS infant is not able to overcome gravity while moving. These years are the sensitive period for motor development and skill acquisition. Decreased muscle force constraints the development of fundamental skills, eye-hand coordination and locomotion. These skills are necessary for cognitive and social development. GH therapy improves muscle mass in children with PWS but an additional effect was found of a simple training program in older children: muscle mass and spontaneous physical activity increased. In infants (age < 2 years), GH therapy had the greatest effect on locomotor performance and some effect on eye-

hand coordination (Eiholzer et al, 2004). Motor learning studies have demonstrated that training effects are highly task-specific and related to the number of repetitions (power law of practice).

Parents of PWS infants are uncertain how to stimulate their child. As a result, many PWS infants lack the appropriate stimulation and opportunities to explore the immediate environment and may fail to develop connections within the brain which are essential for development.

An age-specific, child-centered functional training program will enable the infant/toddler to learn skills while avoiding the disturbing influence of gravity (Ketelaar et al, 2001; De Kleine et al, 2006; Helders et al, 2003; Ekström et al, 2005). The number of repetitions is crucial for the effect of training. Parents learn how to stimulate their child and in which (adapted) circumstances their infant is able to play and perform the same motor activities as their peers. It stimulates parent-child interactions and contributes to a consistent pattern of daily life not only focusing on diet control but also activity control. By using an Internet based secured communication system, the developmental progress and readjustment of the training program is communicated with the parents and the local paediatric physical therapist.

In conclusion, an intensive, age-specific, child-centered functional training program will increase muscle mass, muscle composition (increase of type II fibres), motor development, motor learning capacity, parental interaction, enjoyment, self-esteem of the young child and its parents, and motivation for an active lifestyle, which will result in long-term changes in the care of young PWS children. GH treatment might further potentate the effects of the training program as it increases muscle mass.

### **Does GH improve morbidities associated with PWS?**

**AL Carrel**

GH treatment of children with PWS in a dose of 1.0 mg/m<sup>2</sup> body surface/day significantly improves growth rate and leads to a normalization of height standard deviation scores (SDS) (Lindgren et al, 1999; Eiholzer et al, 2000; Obata et al, 2003). In a randomized study, GH treatment also induced a significantly greater head growth (-0.9 to -0.1 SDS) over the first year of treatment compared to untreated PWS children (-0.5 to -0.2 SDS) (Carrel et al, 2004). During the first year of GH treatment, there was a significant decrease in percent body fat, followed by stabilization during the second year. Lean body mass increased significantly during 1 and 2 years of GH treatment (69 and 30%, respectively), compared to in untreated PWS children after 1 year (23%), whereas GH also significantly improves muscle strength and agility.

Infants with PWS typically display failure to thrive and decreased muscle mass with excess body fat for age. In a randomized study in 25 infants (ages 4-37 months), GH therapy induced faster head

growth, increased lean body mass accrual, and decreased percent body fat, as well as improved language ( $P=0.05$ ) and cognition ( $P=0.02$ ) in the treated PWS infants compared with the untreated ones after 1 year study (Eiholzer et al, 2004). A trend towards improved mobility and stability percentile rankings was noted with GH therapy, however, wide variability among PWS subjects was seen at all time points.

After the initial 2 years, GH therapy for two additional years had continued beneficial effects on growth velocity and body composition (decrease in fat mass and increase in lean body mass) when doses of 1.0 and 1.5 mg/m<sup>2</sup>/d were administered ( $P < 0.05$ ), but not with 0.3 mg/m<sup>2</sup>/d (Carrel et al, 2001; Carrel et al, 2002). This indicates that changes in body composition are GH dose dependent. Bone mineral density continued to improve at all doses of GH ( $P < 0.05$ ). Prior improvements in strength and agility that occurred during the initial 2 years were sustained, regardless of the GH dose.

GH therapy was well-tolerated and the safety profile proved to be reassuring (Lindgren et al, 1999; Eiholzer et al, 2000; Obata et al, 2003; Carrel et al, 2004; Eiholzer et al, 2004; Carrel et al, 2001; Carrel et al, 2002). However, long-term studies are required to evaluate the long-term effects of GH in cohorts of PWS children and to increase our understanding of the effects of GH therapy in PWS patients.

In conclusion, GH therapy in a dose of 1 mg/m<sup>2</sup>/day or more induces sustained changes in growth, body composition, physical function, agility and neurodevelopment. As greater benefits were seen in younger children, prompt referral to a pediatric endocrinologist for consideration of GH therapy is recommended for PWS children at an early age.

### **Scoliosis: epidemiology and management**

### **F Accadbled/T Odent**

Scoliosis is a frequent feature observed in children with PWS. The prevalence of the scoliosis varies between 30 to 70 % and is frequently associated with kyphosis. The high frequency of scoliosis in this disorder could be due to hypotonia, obesity or other endocrine disorders. A regular assessment is therefore required in these children. Obesity worsens scoliosis and increases the risk for additional kyphosis. In this report based on 139 children (mean age 10 years) the prevalence is 43%, and particularly high in young ages (15% before 5 years and 22% between 5 and 10 years). There is no difference between deletion and maternal disomy groups for scoliosis and kyphosis, and no sex ratio. The type of scoliosis is different from idiopathic scoliosis. Kyphosis seems to be a bad prognostic factor. Risk factors for scoliosis are BMI and possibly GH treatment. For the latter issue, it seems more an acceleration of scoliosis progression. Orthopaedic examination and systematic X-rays are mandatory on GH treatment.

Treatment basis are the same than in idiopathic scoliosis regarding modalities (bracing and surgery) and indications. Surgical treatment is indicated in scoliosis-kyphosis with severe early onset and in adolescents with near skeletal maturity. Surgical treatment requires expertise in neurological scoliosis and a multidisciplinary care for PWS. Complications are more frequent and severe than in idiopathic scoliosis with a high risk of paraplegia (20%) and 30% of major complications (deep infections, pneumonia, hook out).

In conclusion, early diagnosis and treatment are needed. Surgical team should have expertise on neuromuscular disease. Control of BMI could reduce the progression of scoliosis. Regular assessment on GH treatment is mandatory.

## **Roundtable B: Transition and adult care**

**Chair: L Gourash**

### **Sex steroid replacement and bone health**

**G Grugni**

In adolescents and adults with PWS, hypogonadism is very common but present to a variable degree (Burman et al. 2001; Crino et al. 2003; Eiholzer et al. 2006). Primary arguments for using sex steroid replacement in adults with PWS are known benefits to bone health, muscle mass metabolic protection and possible benefits to mental, emotional and physical well-being. Oestrogen levels in females with PWS may not reach the low post-menopausal levels perhaps related to aromatization of androgens from excess adipose tissue (Goldstone et al. 2001; Vestergaard et al. 2004).

Adolescents and adults with PWS have low bone mineral density (BMD) and are at risk of osteoporosis, related to sex steroid and GH deficiency and low muscular activity (Brambilla et al. 1997; van Mil et al. 2001; Butler et al. 2001; Burman et al. 2001; Hoybye 2004; Vestergaard et al. 2004). Biochemical markers of bone turnover are elevated suggesting the importance of hypogonadism. Reduced BMD in PWS is associated with high risk of fracture in the long bones as well as in the small bones of the hands and feet., with some patients sustaining multiple fractures (Butler et al. 2002; Kroonen et al. 2006).

These findings support the need for hormone therapy, particularly sex steroids replacement, from adolescence. However, neither standardized protocols for the prevention of osteoporosis nor systematic studies of sex hormone treatment in adolescents or adults with PWS are actually available. Oestrogen and androgen status could be monitored yearly during adolescence and adulthood and BMD assessed as indicated by dual-energy X-ray photon absorptiometry (DEXA). In

females with PWS, the use of gonadal hormone replacement could be considered if BMD becomes low-normal as well as in the presence of reduced oestradiol levels and/or amenorrhea or oligomenorrhea. Administration of low dose of testosterone may be useful in males with PWS starting from early adolescence. In adulthood, these patients should be treated as for any other hypogonadal subjects. Androgen therapy can be more physiologically administered using the new delivery systems of testosterone patches and gel preparations, which avoid the peaks and troughs of injections. However, sex steroid therapy should be ideally minimized and titrated based on individual assessment, BMD, quality of life, sexual and psychosocial issues, acceptability of menstruation, and potential side effects.

### **GH in transition and beyond**

**AC Lindgren**

Adolescence is a developmental period that involves biological, physical and environmental changes. Like most adolescents and even more dramatically, adolescents with chronic disease and disabilities struggle with issues of moving from childhood to adulthood encompassing the difficulties due to the changes of care givers and particularly physicians. Concerning GH treatment particularly, the goals of GH treatment implemented in childhood for PWS patients is to achieve a normal adult height consistent with target height and a normal or reasonably normal body composition.

Besides this goal, tissue maturation is also partly related to GH treatment as in GH deficient patients, i.e., achievement of a normal peak bone mass, improvement of muscle mass and strength, maintenance and improvement of carbohydrate metabolism and prevention of cardiovascular morbidity. What do we know in PWS children and adolescents? A poor number of studies related final height after GH treatment. Regarding lean body mass as assessed by DEXA, after 7 year of treatment, Lindgren reported a mean value of -1.7 SD ranging from -3.1 to -1.2 SD regarding body fat mass, Carrel et al (Carrel, 2002) in a dose effect study reported that patients receiving higher dose (1.5 mg/m<sup>2</sup>/day) maintained and even decreased their % of body fat after 4 years suggesting a better effect with higher dose. This point deserve to be confirmed by others studies. Lindgren reported her own experience; after reaching adult height, sexual substitution was started with steroid, GH dose was reduced to 0.2 mg following IGF-I levels, thyroid hormones and carbohydrates were monitored.

For the future, the following questions were raised: such studies investigating the effect of GH treatment in PWS during the transition period were strongly needed. The design of these studies (placebo? controlled?) requires discussion based on the data collected in adults treated with GH. We need also to define the individuals that could benefit most for GH treatment, the dose needed

for reaching and maintaining long term normal body composition and metabolism, the doses and type of sexual steroids.

Transition clinics adolescents, with competence in GH treatment and PWS will greatly help to manage these patients during this critical period. Help of psychologists and psychiatrists are compulsory. We should also emphasize for this specific population the long term preparation of transition and the need for collaborations between pediatricians and adult endocrinologists.

### **Prevention of obesity in adults (ethics and legalities)**

**C Hoybye**

Obesity management involves environmental control with early institution of a low-calorie, well-balanced diet, with regular exercise, rigorous supervision, restriction of access to food and money with appreciation of legal and ethical obligations, appropriate psychological and behavioural counselling of the patient and family (Dykens et al. 1996; Holland & Wong 1999; Dykens & Shah 2003; Eiholzer 2003). Group homes specifically designed for individuals with PWS have been particularly successful in management of these problems during adulthood (Ziccardi, 2006).

Anecdotally, pharmacological treatment, including available anorexigenic agents, has not been of benefit in treating hyperphagia, though there are few published placebo-controlled studies (Dykens et al. 1996; Dykens & Shah 2003; Goldstone 2004; Shapira et al. 2004). Study of any potential benefits of newer agents such as endocannabinoid antagonists are awaited in PWS. Restrictive bariatric surgery, such as gastric banding or bypass, have not been shown to reduce hyperphagia or achieve long-term weight reduction and are associated with unacceptable morbidity and mortality (Papavramidis et al. 2006). While some of the reports using biliopancreatic diversion have reported successful weight loss, there are with frequent complications from the resulting intestinal malabsorption (de Almeida et al. 2005; Papavramidis et al. 2006). Importantly it is unknown whether changes in the food environment might contribute to the outcomes following surgery.

Body composition studies show both increased body fat and reduced muscle in PWS (Brambilla et al. 1997; Goldstone et al. 2002). Magnetic resonance imaging has found a selective relative reduction in visceral adiposity in PWS adults of both sexes (Goldstone et al. 2001; Goldstone et al. 2003). This may explain the relative hypoinsulinaemia and lower triglyceride levels with preservation of insulin sensitivity and protective elevation in adiponectin levels in patients with PWS given their overall obesity (Tomita et al. 1989; Schuster et al. 1996; Goldstone et al. 2001; Hoybye et al. 2004; Goldstone et al. 2005; Kennedy et al. 2006).

Physical activity in PWS is significantly reduced (van Mil et al. 2000b), related to obesity,

hypersomnolence and persistent poor muscle strength. There is a reduced resting metabolic rate relative to body size, related to the abnormal body composition, which further contributes to a reduction in 24 hour energy expenditure (van Mil et al. 2000a; Goldstone et al. 2002). Increased physical activity and exercise programs are beneficial in improving body composition in PWS (Silverthorn & Hornak 1993).

### **Presentation and treatment of psychiatric disorders**

**A Vogels/D Thuilleaux**

Sound psychiatric and behavioural assessments are key to the effective management and treatment of behavioural and psychiatric disorders as they affect people with PWS. Distinctions need to be made between maladaptive behaviours that are common from childhood and may persist into adult life, such as the disruptive and repetitive and ritualistic behaviours, and those that develop during later childhood or adult life that are a manifestation of an underlying mood disorder or psychotic illness.

Assessments invariably require multidisciplinary approaches, applying different theoretical perspectives and identifying those developmental, biological, psychological and social factors that might contribute to the risk of such problems and to their occurrence. Part of this assessment is the identification of co-morbid physical and psychiatric disorders which may present as a deterioration in existing maladaptive behaviours, the onset of new problem behaviours, or with the appearance of new physical or psychiatric symptomatology. Presentations of both physical and psychiatric problems can be confusing with apparent physical symptoms (such as headache, incontinence) preceding the development of a psychotic illness, and a change in emotional state being indicative of a physical illness that may not in itself be obvious due to the lack of sensitivity to pain common in people with PWS.

Management and treatment approaches therefore require interventions designed to address those factors that might be contributing to the onset and continuation of the maladaptive behaviours or psychiatric illness as well as more symptomatic treatments. Where a co-morbid psychiatric illness is present, psychiatric medication may be effective. The medication used will depend on the exact psychiatric diagnosis of the co-morbid disorder. It should be started at low doses and sedation with benzodiazepine medications avoided. Preliminary findings indicate that antidepressant medication and antipsychotic medications (such as risperidone) are helpful, initially at low doses, where depression and psychotic illnesses are present. Preliminary findings from a follow-up study suggest that mood stabilizing medication may not be so effective. However, further studies are required.

Whilst the more entrenched behavioural problems associated with PWS require effective management in the environment where the person lives, the development of a severe co-morbid psychiatric illness may require hospitalization. Strategies to ensure a predictable, routine and relative low demand environment that optimize understanding and communication are helpful. Where depression is present suicidal ideation may occur and, in the case of bipolar disorder and/or psychotic illness, a person's behaviour may result in a risk to others. Under these circumstances a period of in-patient care may be indicated.

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