Growth retardation in untreated autosomal dominant familial neurohypophyseal diabetes insipidus caused by one recurring and two novel mutations in the vasopressin-neurophysin II gene

Cécile Brachet1,*, Julia Birk3,*, Catherine Christophe2, Sylvie Tenoutasse1, Brigitte Velkeniers4, Claudine Heinrichs1 and Jonas Rutishauser 3
1Pediatric Endocrinology and 2Pediatric Radiology, Hôpital Universitaire des Enfants Reine Fabiola-ULB, 1020 Brussels, Belgium, 3Biozentrum, Division of Biochemistry, University of Basel, Klingelbergstrasse 70, CH-4056 Basel, Switzerland and 4Endocrinology, Universitair Ziekenhuis Brussel, 1020 Brussels, Belgium
*(Correspondence should be addressed to J Rutishauser; Email: j.rutishauser@unibas.ch)

Abstract

Objective: Autosomal dominant familial neurohypophyseal diabetes insipidus (adFNDI), a disorder caused by mutations in the vasopressin (AVP)-neurophysin II (NPII) gene, manifests gradually during early childhood with progressive polyuria and polydipsia. Patients are usually treated with synthetic AVP analog. If unlimited access to water is provided, prognosis is usually good even in the absence of specific treatment.

In this study, we describe three families with adFNDI, in which growth failure was a prominent complaint, on the clinical and molecular level.

Design/methods: Histories from affected and unaffected family members were taken. Height and weight of index patients were recorded longitudinally. Patients underwent water deprivation tests, magnetic resonance imaging, and genetic analysis. One mutant was studied by heterologous expression in cell culture.

Results: A total of ten affected individuals were studied. In two of the three pedigrees, a novel mutation in the AVP-NPII gene was found. The index children in each pedigree showed growth retardation, which was the reason for referral in two. In these cases, water intake was tightly restricted by the parents in an attempt to overcome suspected psychogenic polydipsia and to improve appetite. Once the children were treated by hormone replacement, they rapidly caught up to normal weight and height.

Conclusions: Genetic testing and appropriate parent counseling should be enforced in adFNDI families to ensure adequate treatment and avoid chronic water deprivation, which causes failure to thrive.

European Journal of Endocrinology 164 179–187

Introduction

Autosomal dominant familial neurohypophyseal diabetes insipidus (adFNDI) is a rare disorder resulting from an inherited deficiency of arginine vasopressin (AVP), the antidiuretic hormone. Polydipsia and polyuria generally appear in early infancy. adFNDI is caused by mutations in the AVP-neurophysin II (NPII) gene, as reported for the first time in 1991 (1). Since then, more than 50 mutations in the AVP gene have been associated with adFNDI (2). The AVP gene is located on chromosome 20p13 and contains three exons that encode AVP and the N-terminal portion of NPII (exon 1), the central portion of NPII (exon 2), and the C-terminal portion of NPII and a glycopeptide called copeptin (exon 3) (3). Intracellular AVP transport and secretion depend on correct folding of the entire hormone precursor. Expression studies in cell cultures (4–11) have shown that dominant AVP mutants fail to exit the endoplasmic reticulum (ER), and a knock-in mouse model (12) supports the hypothesis that adFNDI results from the accumulation of retained mutant protein that is toxic to the AVP-producing magnocellular neurons of the supraoptic and paraventricular nuclei of the hypothalamus. Recently, we have shown that mutant AVP precursors causing adFNDI form ER-derived fibrillar aggregations when expressed in a neuronal cell line (13). Thus, the disease can be viewed as a neurodegenerative disorder confined to vasopressinergic nuclei.

Clinically, diabetes insipidus (DI) in childhood is characterized by polyuria and polydipsia and may present with failure to thrive, e.g. in nephrogenic DI (14). The age of disease onset in adFNDI is variable. Although there is no clear genotype–phenotype correlation, certain mutations may be associated with a later
disease onset (15) and less toxicity in the adFNDI knock-in mouse (12). In addition, intrafamilial variability of the age of onset of DI has been described, which may result from interactions with other genetic or environmental factors. The loss of AVP secretion is a gradual process with partial DI evolving towards complete AVP deficiency over time (16–18). According to the neurotoxicity hypothesis, the gradual accumulation of cytotoxic mutant AVP protein in the magnocellular neurons accounts for both the progressive nature and the dominant inheritance of the disease. Along with the development of DI, a gradual disappearance of the posterior pituitary hyperintense signal observed on T1-weighted magnetic resonance imaging (MRI) may be observed (17). Data on anterior pituitary morphology in adFNDI are scarce, but a reduction in size, as assessed by MRI, has been described in some patients (19).

This study describes three families with adFNDI, among which failure to thrive was an unusual finding in several affected individuals. We found a novel mutation in the AVP gene in two of the three family trees and characterized one of these by heterologous expression studies.

**Patients and methods**

**Patients**

We studied three adFNDI families with a total of ten affected individuals (Fig. 1), six of which were children. Written informed consent was obtained from the adults and the children’s parents. The study was approved by the local review board.

**Clinical and radiological assessment**

In each family, medical histories from both affected and unaffected family members were taken. All patients with a history of polyuria and polydipsia had one or more clinical assessments to confirm the diagnosis of DI, including i) self-report of the daily fluid intake and measurements of urine osmolality (uOsm), plasma osmolality (pOsm), and plasma AVP (commercially available ELISA, DiaSorin, Inc., Stillwater, MN, USA) while not under treatment with desamino-8-D-AVP (DDAVP) and on ad libitum fluid intake; ii) measurements of pOsm, uOsm, and plasma AVP concentrations during a standard water deprivation test (WDT) of sufficient duration to increase pOsm and plasma sodium above 300 mOsm/kg and 150 mM respectively; or to cause weight loss > 5% of total body weight. On a 1.5 T MRI scanner (Philips Intera, Eindhoven, Netherlands), sagittal and coronal images of the hypothalamus and pituitary were obtained with and without gadolinium enhancement in some patients to exclude structural abnormalities and to establish the presence or absence of the characteristic hyperintense (‘bright spot’) signal of the posterior pituitary observed on T1-weighted images of a normal posterior pituitary.

**Genetic analysis**

The AVP gene was amplified from genomic DNA by PCR in a Biometra T3 thermocycler using the Expand High Fidelity PCR System (Roche) and 10% DMSO. Two PCR products were generated, one consisting of exon 1, the other of exon 2/3 including intron 2. The following primers were used for amplification: TGCTGAAT-CACGTGACGCTGGGACC, GCTATGCGTGGCCCTG-AGATGGCCACAGTG (exon 1, forward and reverse respectively); TGCGTGCTCCCCCTCAACCCCTGAGAC-TCC, CCTCTCCCCCTTCCCTTCCCGCCAGAG (exon 2/3, forward and reverse respectively). PCR was carried out for 40 cycles of melting (94 °C for 1 min), annealing (55 °C for 1 min), and extension (72 °C for 3 min), with a final 15 min at 72 °C for extension. PCR products were run on 1% agarose gels, and bands were extracted...
from agarose using a commercial kit (NucleoSpin Gel extraction; Macherey-Nagel, Düren, Germany). Both strands of purified PCR products were sequenced on an automated sequencer using the following primers: TGCCTGAATCACTGCTGACCGCTGGGGACC, GCTATGGCTGCCCTGAGATGGCCCACAGTG (exon 1, forward and reverse respectively); TCGCTGCGTTCCCCTCCAACCCCGACTC, CGCCCCCCCCCAGGCCCGCCCCCGCGCGC (exon 2, forward and reverse respectively); CCCAGGC-GCCCGTGCTCACACGTCCTCCCG, CCTCTCTCCCCTTCTTCCCGCCAGAG (exon 3, forward and reverse respectively).

**Plasmids and constructs**

The cDNAs of the human wild-type AVP and the dominant mutant C59Y have been described previously (8, 13). The C92W mutant was generated by PCR. All cDNAs were subcloned in the pECE expression plasmid, and their correct sequences were confirmed by automated sequencing.

**Cell culture experiments**

Culture and transient transfection, metabolic labeling, immunoprecipitation, and immunofluorescence using COS-1 cells were done as described previously (13). In brief, subconfluent cells were transfected in six-well plates using polyethylenimine (Sigma) and analyzed 2 days after transfection. To analyze secretion of pro-AVP precursors, transfected cells were pulse labeled for 30 min with 100 μCi/ml [35S] protein labeling mix and chased with excess cysteine and methionine. Cell lysates and culture media were immunoprecipitated with polyclonal rabbit anti-NPII antibody (20), and immune complexes were analyzed by PAGE (SDS-PAGE) and autoradiography. For immunofluorescence studies, transfected cells were permeabilized and incubated with polyclonal anti-NPII antiserum and MAB against the endogenous ER marker, calnexin (BD Bioscence, Franklin Lakes, NJ, USA). Cy2-labeled goat anti-rabbit and Cy3-labeled goat anti-mouse secondary antibodies (Jackson Immunoresearch, West Grove, PA, USA) were used to stain the cells.

**Results**

Relevant medical histories as well as clinical and laboratory data of the affected family members are summarized in Table 1.

**Family A**

**Clinical studies** The pedigree of the family is shown in Fig. 1, panel A. The index case (Fig. 1, A/4) was a 2.6-year-old girl referred for failure to thrive since 20 months of age (Fig. 2, left and right upper panels). She was born to non-consanguineous Belgian parents of academic background. The father’s height was 173 cm (−0.5 s.d.), and the mother’s height was 156.7 cm (−1.2 s.d.). Pregnancy and delivery were uneventful. Polydipsia had been present since 6 months of age. Poor appetite and food refusal were prominent complaints. The parents had restricted the patient’s water intake since 15 months of age, assuming that this would improve her appetite. On physical examination, height was found to be 84 cm (−2 s.d.), weight 10.3 kg, and body mass index 14.6 kg/m² (−1.3 s.d.). The girl seemed sad and tired. WDT showed central DI (Table 1). Anterior pituitary function as assessed by glucagon.

![Table 1](image-url)

Table 1 Clinical and laboratory data of ten affected subjects from three kindreds with adFNDI

<table>
<thead>
<tr>
<th></th>
<th>Family A</th>
<th>Family B</th>
<th>Family C</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age of polyuria onset by history (years)</strong></td>
<td>0.5</td>
<td>1</td>
<td>0.7</td>
</tr>
<tr>
<td><strong>Clinical parameters at diagnosis</strong></td>
<td>2.6</td>
<td>1.75</td>
<td>0.7</td>
</tr>
<tr>
<td><strong>Height SDS</strong></td>
<td>-2</td>
<td>-1.2</td>
<td>ND</td>
</tr>
<tr>
<td><strong>BMI SDS</strong></td>
<td>-1.3</td>
<td>0.7</td>
<td>ND</td>
</tr>
<tr>
<td><strong>WDT</strong></td>
<td>2.6</td>
<td>N/A</td>
<td>70</td>
</tr>
<tr>
<td><strong>pOsm_max (mOsm/kg)</strong></td>
<td>304</td>
<td>296</td>
<td>303</td>
</tr>
<tr>
<td><strong>uOsm_max (mOsm/g)</strong></td>
<td>241</td>
<td>330</td>
<td>24</td>
</tr>
<tr>
<td><strong>uOsm_DDAVP (mOsm/kg)</strong></td>
<td>459</td>
<td>473</td>
<td>880</td>
</tr>
<tr>
<td><strong>Parental water restriction</strong></td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td><strong>Poor appetite</strong></td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td><strong>Start of DDAVP treatment (years)</strong></td>
<td>2.6</td>
<td>6</td>
<td>70</td>
</tr>
</tbody>
</table>

WDT, water deprivation test; pOsm_max, maximum plasma osmolality; uOsm_max, maximum urinary osmolality; uOsm_DDAVP, maximum uOsm under DDAVP treatment; N/A, not available.

*This patient refused DDAVP during the WDT but later responded very well clinically to treatment.
between 1 and 2 years of age. He continued to have urine or water from flowerpots. His weight stagnated she restricted his water intake so that he drank his own (Table 1). Although his mother also had polydipsia, A/1 had developed polydipsia at 8 months of age years of age. was started. Intranasal DDA VP was initiated only at 6 administration resulted in urinary concentration of 459 mosmol/kg. Catch-up growth was observed under DDAVP treatment (Table 1 and Fig. 2, top panels).

TRH, and GnRH tests was normal. MRI showed a Rathke’s pouch cyst and a normal anterior pituitary gland (height 3.3 mm), and a small but present posterior pituitary hyperintense signal. DDAVP administration resulted in urinary concentration of 459 mosmol/kg. Catch-up growth was observed under DDAVP treatment (Table 1 and Fig. 2, top panels).

The index patient’s father (Fig. 1, A/2) had a diagnostic work-up at 21 months of age for failure to thrive. Polydipsia had been present since 1 year of age. After central DI had been diagnosed by a WDT, his parents had refused pitressin treatment and restricted adequate water intake. His growth curves had shown weight stagnation, at which point carbamazepine was started. Intranasal DDAVP was initiated only at 6 years of age.

The paternal grandfather of the index case (Fig. 1, A/1) had developed polydipsia at 8 months of age (Table 1). Although his mother also had polydipsia, she restricted his water intake so that he drank his own urine or water from flowerpots. His weight stagnated between 1 and 2 years of age. He continued to have clinically severe DI until around 55 years of age, drinking ~10 l/day. At that time, polydipsia and polyuria diminished to ~3 l/day. At 57 years of age, he presented with an acute episode of headache and transient cranial nerve III palsy. He was diagnosed with severe panhypopituitarism 3 years later; an MRI showed an absent posterior pituitary T1 signal and a hypoplastic anterior pituitary gland. When hormonal substitution with hydrocortisone, testosterone, GH, and L-thyroxine was started, his polydipsia again became more significant, and DI was confirmed by a WDT at 70 years of age.

**Genetic analysis** Genomic DNA samples from three affected (Fig. 1, A/1, A/2, and A/4) and two unaffected (Fig. 1, A/3 and A/5) family members were amplified by PCR and sequenced. The affected individuals, but not the healthy family members, had a heterozygous point mutation of cytosine to a guanine at nucleotide 1873 (1873C>G) in exon 2 (Fig. 3, panel A). This mutation alters codon 92 of the precursor molecule from TGC to TGG, substituting cysteine by tryptophan. C92W, within the NPII moiety. No mutations were detected in exon 1 or 3.

**Expression studies** To analyze secretion of wild-type pro-AVP, the known dominant mutant C59Y (13) and the novel mutant C92W were expressed in COS-1 cells, pulse-labeled with [35S]methionine/cysteine, chased for 2 h, isolated from the cells and from the media by immunoprecipitation, and analyzed by SDS-PAGE and autoradiography (Fig. 4). In contrast to the wild-type, where a considerable fraction could be recovered from the medium (Fig. 4, lane 5), the mutants C59Y (Fig. 4, lane 6) and C92W (lane 8) were completely retained in the cells. These findings illustrate that the dominant mutant C92W is secretion deficient, as has been shown for C59Y and other known mutants causing adFNDI (8).

**Immunofluorescence** To characterize the intracellular localization of the C92W precursor, transfected COS-1 cells were analyzed by immunofluorescence staining 48 h after transfection (Fig. 5). In many cells, a strong reticular staining typical for ER-retained proteins was visible (panel A). However, a considerable number of cells also showed larger aggregations, typical for mutant pro-AVP (13) (panel B). These inclusions were of round, sometimes short tubular appearance, and co-stained positive for calnexin, confirming that they originated from the ER. Our findings thus demonstrate that the C92W mutant prohormone shares important characteristics with other mutants associated with adFNDI.

**Family B**

**Clinical studies** Figure 1, panel B, depicts the pedigree of the family. The index case (Fig. 1, B/3 and Table 1) was a 5-year-old boy referred for failure to thrive with a history of polydipsia since 14 months of age. He was
born to non-consanguineous Belgian parents living in socially deprived conditions. The father’s and mother’s heights were 170 cm (1 s.D.) and 149.7 cm (2.3 s.D.) respectively. Pregnancy and delivery were uneventful. Feeding difficulties were reported since 1 month of age, when the boy was weaned from breastfeeding. He had nocturnal enuresis. Unrestricted water intake amounted to 4.5 l/d, but water consumption was tightly restricted by the parents. He regularly drank from puddles and bins. Both poor appetite and polydipsia were attributed to behavioral problems, and the boy was followed by a psychologist. WDT confirmed central DI (Table 1), while MRI showed a small posterior hyperintense signal on T 1-weighted images. Catch-up growth occurred after the initiation of DDA VP treatment (Table 1 and growth curves in Fig. 2, left and right middle panels). At age 8, a 2 years’ delay in bone age (Greulich and Pyle method) was observed. Based on bone age, a predicted adult height of 171 cm was calculated (Bayley-Pinneau method), which is close to the target height based on parental heights (165.5 ± 8.5 cm).

A younger sister of the index case (Fig. 1, B/4) had slight polydipsia with a normal WDT at 2.5 years of age (uOsmmax 850 mosmol/kg). A diagnosis of psychogenic polydipsia was made. At 5.8 years of age, she was still polydipsic with poor appetite and showed growth retardation. Water intake was tightly restricted by the parents. WDT showed partially preserved renal concentration ability (Table 1), although the test had to be stopped after 5 h because of weight loss > 5% of initial body weight. DDAVP treatment was initiated.

A newborn sister (Fig. 1, B/5) of the index case showed polydipsia starting at 5.5 months of age. At 7.5 months of age, she was drinking 1.5 l/day; WDT, performed over 5 h and carefully monitored, was compatible with partial DI (Table 1). At 11 months of age, she had severe polydipsia (2 l/day) and was started on intranasal DDAVP.

The mother of the index case (Fig. 1, B/1) had been diagnosed with a defect in urine concentrating ability at the age of 6 years. She never had any treatment or medical follow-up. Her short stature (149.7 cm) was noteworthy, but being an adopted child, her parents’ heights were not known. She had seven uncomplicated pregnancies and had given birth to seven children (three affected by adFNDI) but also had two spontaneous abortions. Upon referral to us, WDT confirmed DI, and treatment was started (Table 1).

**Genetic analysis** Genomic DNA samples from four affected (Fig. 1, B/1, B/3, B/4, and B/5) and one unaffected (Fig. 1, B/2) family members were amplified by PCR and sequenced. In all four affected individuals, but not in the unaffected control, a heterozygous point mutation of cytosine to guanine at nucleotide 1720 (1720C > G) was found in exon 2 (Fig. 3, panel B).

![Figure 3](https://example.com/figure3.jpg)  
**Figure 3** Genomic DNA sequence analyses of the AVP gene in families A, B, and C. In families A and B, sequences from an unaffected (A/3 and B/2; top lanes) and affected (A/4 and B/3; bottom lanes) individuals are shown. In family C, DNA was available only from affected individuals (C/1, C/2, and C/3). The chromatogram of patient C/2 is shown. Arrows indicate heterozygous single base mutations at three different locations within the NPII moiety of the gene, resulting in double peaks in the chromatogram. The predicted changes in amino acid sequences are indicated for each of the three point mutations.

![Figure 4](https://example.com/figure4.jpg)  
**Figure 4** Retention of mutant vasopressin precursors in transfected COS-1 cells. Wild-type (wt) and mutant (C92W and C59Y) prepro-vasopressin were expressed in COS-1 cells. The cells were labeled for 30 min with [35S]methionine/cysteine and chased for 0 (lanes 1–3) and 2 h (lanes 4–9). The collected medium (M) and the lysed cells (C) were subjected to immunoprecipitation using anti-NPII antiserum. Immunoprecipitates were analyzed by gel electrophoresis and autoradiography. The approximate molecular mass (kDa) of the immunoprecipitated proteins is indicated at the left side of the gel.
Family C

Clinical studies The index case (Fig. 1, C/2) was a 2.9-year-old boy referred for polydipsia. He was born to non-consanguineous Moroccan parents. The family was well integrated socially. The father’s height was 172 cm (−0.7 SDS), and the mother’s height was 163 cm (−0.1 SDS). Pregnancy and delivery were uneventful. Polydipsia was present from 8 months of age, when he was weaned from breastfeeding. Current water intake was 4 l/d. Poor appetite was a prominent complaint and was attributed to polydipsia, but water intake was only slightly restricted by his mother. Slight growth retardation was present since 24 months of age, which normalized under DDAVP treatment that was initiated after a diagnostic WDT (Table 1 and Fig. 2, left and right lower panels).

The younger sister of the index case (Fig. 1, C/3) started to have polydipsia and polyuria at 1 year of age. WDT, performed at age 1.5 years, had to be stopped after 5 h because of loss of >5% of initial body weight, although it only indicated partial DI (Table 1).

The mother of the index case (Fig. 1, C/1) had shown polyuria and polydipsia since 1 year of age; severe polydipsia was reported in five of her eight siblings.

Genetic analysis Genomic DNA samples from three affected (Fig. 1, C/1, C/2, and C/3) family members were amplified by PCR and sequenced. While no mutations were detected in exons 1 and 3, affected individuals had a heterozygous point mutation of guanine to adenine at nucleotide 1829 (1829G>A), altering codon 78 within NPII from GAG to AAG (Fig. 3, panel C). This point mutation predicts the substitution of glutamic acid by lysine, E78K, and has been previously described in a Japanese pedigree (21).

Discussion

We report on three families with adFNDI, caused by two novel and one recurring mutations of the AVP gene. These families illustrate several remarkable clinical aspects of the disease. All three index patients showed growth retardation, which was more severe and therefore the reason for referral in families A and B. Growth retardation was also found in several other affected members of the three families. Before diagnosis, poor appetite, along with polydipsia, was a prominent complaint in several affected children. In an attempt to increase their child’s appetite, parents – even if suffering from DI themselves – imposed chronic water restriction on their affected children. Free access to water was also restricted during school hours. Chronic parent-imposed water restriction, encouraged in the case of family A by a psychologist’s misdiagnosis of psychogenic polydipsia, further enhanced imperative thirst and led to extreme behavior such as drinking from bins or puddles. This was then mistaken as obsessive drinking. The misinterpretation of their children’s symptoms by affected parents is striking, and to our knowledge, comparable patient care has not been described in families with adFNDI. Interestingly, the social background of the families did not seem to clearly influence the parents’ behavior, as only members of family B showed a low level of education and depended on social welfare.

Growth failure in adFNDI may be associated with absence of DDAVP treatment and unmet fluid needs. In family C, where water restriction by the parents was minor, the index patient showed less pronounced growth retardation. In fact, we have observed that even if no hormone replacement therapy is given, children affected with adFNDI may thrive normally, provided they have unlimited access to fluids (22). In contrast, Nijenhuis et al. (10) described a large adFNDI pedigree of which two boys presented with failure to thrive and growth retardation, which were rapidly corrected by DDAVP treatment. Maghniet et al. (19) in their study of children and young adults with neurohypophyseal DI of various origins, found growth retardation in 13 of 79 patients (one of five with adFNDI). Growth retardation is also observed in patients with nephrogenic DI (14, 23). Several factors could predispose patients with untreated DI to growth failure, such as decreased calorie intake due to predominant fluid consumption or loss of appetite.
because of an activated hypothalamic–pituitary axis (HPA) stress axis in chronic hyperosmolality (24). Upon treatment of the AVP deficiency by DDAVP, a catch-up growth over several months occurred in all our patients. Thus, the clinical response to DDAVP administration was excellent both in terms of quality of life and gain in weight and height.

Intrafamilial variability of age of onset of DI severity is illustrated by family B. Two affected children (Fig. 1, B/3 and B/5) presented with early onset of severe DI, whereas in a third sibling (Fig. 1, B/4) with a normal WDT at 2.5 years of age, only a repeated WDT at 5.8 years of age indicated partial DI. Variable age of disease onset has been previously described within adFNDI families (22). In adFNDI, repetitive WDTs during childhood have shown a progressive decrease in AVP secretion (16–18). Since partial DI is particularly difficult to diagnose, genetic testing is of great value in these cases. Indeed, patients C/3 and B/4, despite significant weight loss during WDT, showed partial DI, which was discordant with the severe symptoms presented by other affected family members. These patients also illustrate that criteria commonly used during WDT, such as target pOsm or sodium concentration, may not be strictly applicable in clinical practice, particularly when partial DI is present.

MRI is often used in the work-up of central DI. Typically, the posterior pituitary in healthy persons shows a hyperintense signal (‘bright spot’) on native T1-weighted images and lacks this signal in patients with neurogenic DI, e.g. adFNDI (22). However, the presence of a normal posterior pituitary on MRI does not exclude neurohypophyseal DI, and the absence of the ‘bright spot’ does not prove it, particularly in the elderly (25). Interestingly, we found a small but present posterior pituitary hyperintense signal in two index cases with complete DI (Fig. 1, A/4 and B/3). Maghnie et al. in their cohort of children and young adults identified an abnormal posterior pituitary in the majority of patients but found a ‘bright spot’ in 6%, which disappeared after a median follow-up of 1.5 years (19). In this respect, our study is limited by the lack of longitudinal MRI data in patients A/4 and B/3. Despite this shortcoming, it is important to note that MRI studies cannot substitute for biochemical and/or genetic work-up if neurohypophyseal DI is suspected in children.

Interestingly, the index patient’s grandfather in family A (Fig. 1, A/1) presented with panhypopituitarism and a hypoplastic anterior pituitary gland at 57 years of age, whereas the index case in family A (Fig. 1, A/4) had a small Rathke’s pouch cyst with normal anterior pituitary size and function. Other cases of neurohypophyseal DI associated with an abnormally small anterior pituitary have been described in the literature. Maghnie et al. (19) reported this finding in one of five children with adFNDI. We have reported on one case of partial empty sella among ten patients with adFNDI from three families (26). Melo et al. described a child with adFNDI and a hypoplastic anterior pituitary with normal anterior pituitary function (27). Considering the pathogenesis of adFNDI – selective degeneration of magnocellular vasopressinergic neurons – MRI or functional abnormalities of the anterior pituitary are likely to be unrelated findings.

During the period of untreated panhypopituitarism, patient A/1 showed a remarkable transient remission of his severe DI. Irrespective of anterior pituitary function, reduction of polyuria has been described in aging adFNDI patients (22), the reason of which is not entirely clear. On the other hand, if hypocortisolism is present, DI may be masked due to limited renal ability to excrete free water; consequently, initiation of hydrocortisone replacement therapy was probably a main factor leading to reappearance of symptoms. Thus, although the association of anterior pituitary failure with adFNDI may have been fortuitous, this and previous reports support the notion that a work-up of anterior pituitary function may be warranted in adFNDI if changes in the polyuric phenotype occur.

More than 50 mutations in the AVP gene have been described in adFNDI families, the majority in the NPII sequence. None of the known mutations are located within the glycopeptide moiety. We found a novel missense mutation in two of the three adFNDI families. A Cys 92→Tyr mutation has been described in an unrelated US American adFNDI family of European ancestry (28). Replacement of Cys 92 by Trp, as described in family A in this study, can be expected to produce the same clinical phenotype; in fact, replacement of one of the 14 cysteine residues present in the NPII moiety has been found in a number of adFNDI cases, including the mutation found in family B in this report. As shown in Fig. 5, the ER-retained Cys 92→Trp mutant prohormone forms large ER-derived aggregates visible by immunofluorescence. In several other adFNDI mutants, we have recently described the formation of these large aggregates developing from disulfide-linked oligomers (13). One of these is the ΔGlu78 mutant, first described in 1993 (29), which affects the same amino acid residue as the mutation found in family C. The mechanism of how these intracellular aggregates relate to the neurotoxicity elicited by the mutant molecules, and whether they are causally involved in cell damage or rather have a protective effect on vasopressinergic neurons presently remains unclear.

In conclusion, we report on three families with adFNDI due to one recurrent and two novel mutations in the AVP gene. The most striking clinical aspect in these families was growth retardation of the index children and other affected individuals, obviously in conjunction with lack of treatment and parent-imposed water restriction. Age of disease onset and severity varied considerably among members of the same family. These findings underscore the importance of parental guidance and genetic testing of infants in adFNDI...
families. Knowledge of the genetic status in all newborn family members could effectively prevent the deprivation of adequate treatment.

Declaration of interest
The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

Funding
This work was supported by grant 31003A-113319 from the Swiss National Science Foundation to J Rutishauser.

References


Received 1 November 2010
Accepted 16 November 2010