CASE REPORT

Familial neurohypophyseal diabetes insipidus due to a novel mutation in the arginine vasopressin–neurophysin II gene

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Abstract

Background: Familial neurohypophyseal (central) diabetes insipidus (DI) is caused by mutations in the arginine vasopressin–neurophysin II (AVP–NP_{II}) gene. The majority of cases is inherited in an autosomal dominant way. In this study, we present the clinical features of a mother and her son with autosomal dominant neurohypophyseal DI caused by a novel mutation.

Case: A thirty-four-year-old woman and her three-year-old son were evaluated because of polyuria and polydipsia since the age of 1.5 years onwards. Both patients were subjected to a water deprivation test confirming the diagnosis of central DI. Magnetic resonance imaging of the brain of the mother showed a hypothalamus without apparent abnormalities and a relatively small neurohypophysis without a hyperintense signal. Mutation analysis showed a c.322G>T (p.?[?]/p.Glu108X) in Exon 2 of the AVP–NP_{II} gene in both mother and son.

Discussion: This study reports neurohypophyseal DI in a mother and her son due to a novel mutation in Exon 2 of the AVP–NP_{II} gene. Clinical and pathophysiological aspects of this disease are shortly reviewed and discussed.

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Introduction

Neurohypophyseal (central) diabetes insipidus (DI) is characterized by impaired secretion of the antidiuretic hormone arginine vasopressin (AVP) resulting in polyuria and polydipsia. Under normal circumstances, the precursor hormone prepro-AVP is produced by neurosecretory cells in the supraoptic and paraventricular nuclei of the human hypothalamus (1–3). During axonal transport to the pituitary gland, prepro-AVP is cleaved to AVP and finally stored in the posterior pituitary (neurohypophysis). When water deprivation causes the plasma osmolality to rise above 280–290 mOsmol/kg, AVP is released into the circulation. In the collecting ducts of the kidney AVP binds to the vasopressin type 2 (V₂) receptor, inducing up-regulation of the aquaporin II channels. This results in increased water retention with a rise in urine osmolality to a maximum of 1000–1200 mOsmol/kg and a restoration of plasma osmolality to within the reference range (4).

Familial neurohypophyseal DI accounts for ~5% of all cases of central DI (5) and is caused by mutations in the

AVP–neurophysin II (AVP–NP_{II}) gene on chromosome 20p13 (6). The majority of cases is inherited in an autosomal dominant way, while only two families with recessive types have been reported (7, 8).

In this study, we present the clinical features of two patients with neurohypophyseal DI, not related to the earlier described Dutch families with this disease (9, 10), caused by a novel mutation in the AVP–NP_{II} gene.

Case

A thirty-four-year-old woman was evaluated in the outpatient clinic of Endocrinology of the Academic Medical Centre because of polyuria and polydipsia, which had started at the age of 1.5 years. She drank an average of 8–10 l of water per day and had adjusted her life in such a way that she always had access to drinking water. Her medical history was otherwise unremarkable. She had no symptoms of neurological or pituitary dysfunction, nor a history of head trauma. Her menstrual cycle was regular. Her family history was unremarkable.

Figure 1 Results of the water deprivation test in the mother. The vertical dotted line represents the moment of desmopressin administration. (A) Plasma sodium and osmolality. The horizontal dotted line represents the upper limit of normal. (B) Urine volume and osmolality.

After exclusion of diabetes mellitus in childhood, she had not considered her drinking behavior to reflect a disease, assuming it was a peculiar habit, until she read an article in a Dutch newspaper on families with DI and recognized the symptoms. In addition, she also noticed that the older of her two sons, then 3 years old, had developed progressive polyuria and polydipsia over a period of 1.5 years. For instance, despite frequent changing of diapers his bed was wet every night and upon awakening the first thing he would ask for was water, often drinking 1 l immediately. Parallel to his large water intake, his food intake was low and he had a relatively low weight compared with his height. She contacted our outpatient clinic with a request for endocrine evaluation.

The mother underwent a water deprivation test, determining plasma osmolality, sodium and AVP concentration as well as urine volume and osmolality every hour (Fig. 1). After 6 h plasma sodium and osmolality had increased to 154 mmol/l and 299 mOsm/kg respectively. During the test she continued to have inappropriately dilute urine with an osmolality of 119 mOsm/kg and a diuresis exceeding 400 ml/h. Her body weight had decreased by 5.4%.

Plasma AVP (as determined, after solid phase extraction, by commercial RIA (Euro-Vasopressin, Euro-Diagnostica AB, Malmö, Sweden); detection limit 1.0 pmol/l; total assay variation 10–15% at 7.8 respectively 2.5 pmol/l) was undetectable at all time points. After intravenous administration of 2 µg minrin (desmopressin, or DDAVP), diuresis decreased to 0 and 125 ml/h during the last 2 h of the test respectively. Magnetic resonance imaging of the brain showed a relatively small neurohypophysis without a hyperintense signal, and no apparent abnormalities of the adenohypophysis or hypothalamus (Fig. 2).

Mutation analysis showed a heterozygous c.322G>T mutation in Exon 2 of the *AVP-NP_{II}* gene. Since position c.322 is the final base of Exon 2, it lies within the consensus donor splice site of intron 2. Splice site prediction programs indeed showed a marked decrease in the confidence scores of this site, ranging from a 30% decrease to complete abolishment of this splice site, depending on the used program. However, it is unclear how the splicing machinery will process the pre-mRNA and what the effect on the protein level would be, which results in the nomenclature p.? for the protein change. Abolishment of the intron 2 donor splice site would very likely not lead to nonsense-mediated decay of the mRNA, as the 50–55 nucleotide rule will apply (11). Instead, it will very likely lead to a read-through into intron 2. There is no putative stop codon in the 174 bp long intron 2, suggesting that the entire (in-frame) intron would be inserted into the mRNA leading to an insertion of 58 amino acids into the AVP protein. Alternatively, a cryptic donor splice site may be encountered, leading to a shorter (in-frame) insertion,

Figure 2 T1 midsagittal fat saturated MRI of the brain showing a relatively small and hypointense neurohypophysis.

or a frame shift of the protein leading to an alternative stop codon. Altogether, if splicing is affected, the first 107 amino acids of the AVP gene will be present, followed by a unknown C-terminus (p.?). If splicing of the mutant pre-mRNA is nevertheless not affected, the c.322G>T mutation causes to change codon 108 from encoding a glutamic acid into a premature termination codon (p.Glu108X). Since it was not experimentally tested whether splicing is (partially) affected, the effect on the protein level could be both, resulting in the description p.~/Glu108X. Either way, the c.322G>T mutation represents a pathogenic *AVP-NP_{II}* mutation causing central DI.

Treatment with oral desmopressin 0.1 mg 2–3 times daily was started. The patient now has a diuresis of 2–3 l/day and reports an improvement of her daily life that is no longer dominated by ascertaining sufficient access to drinking water.

Awaiting the gene mutation analysis of the mother, a water deprivation test was planned for her son. On the day of the test, the child spontaneously skipped his usual 1 l of drinking water on awakening. At entering the endocrine unit at 0900 h he weighed 7% less than a few days before and was clearly irritated. His plasma sodium concentration and osmolality were 153 mmol/l and 305 mOsm/kg, respectively, and urine osmolality was 118 mOsm/kg. As these results already confirmed a diagnosis of DI no further water deprivation test was performed. AVP was undetectable and *AVP-NP_{II}* gene analysis showed the same mutation as in the mother.

Treatment with intranasal desmopressin, 2.5 µg in the morning and afternoon and 5 µg before sleeping was started immediately. Polydipsia and polyuria disappeared with dry beds during the night, and food intake and growth improved. In 4 months he gained 2.2 kg.

Discussion

This study describes a mother and her son with neurohypophyseal DI due to a novel mutation in Exon 2 of the *AVP-NP_{II}* gene. They presented with a history of polyuria and polydipsia starting at the age of 1.5 years. The diagnosis was confirmed by a water deprivation test and DNA analysis.

At present, 62 mutations leading to neurohypophyseal DI have been reported (GenBank accession number M11166). All of these occur in the *AVP-NP_{II}* gene, which is composed of four moieties encoding a signal peptide, AVP, NP_{II}, and a glycoprotein of unknown function called copeptin (12). The majority of mutations, including those of the present family, have been found in the part of the gene encoding NP_{II}, which is an intracellular binding protein for AVP. Only a few mutations have been localized to the signal peptide or the AVP coding sequence, and none in the glycoprotein moiety. It is assumed that all known autosomal

dominant mutations cause defective folding or dimerization of the precursor (13).

Under normal circumstances the AVP protein forms homodimers, most probably in the endoplasmic reticulum (14). Mutant AVP-C67X also binds to the wild-type AVP, and the heterodimers are retained in the endoplasmic reticulum (14–17). It has been hypothesized that the accumulated mutant AVP heterodimers are cytotoxic, causing gradual but progressive loss of viable neurosecretory neurons, finally leading to AVP deficiency. In fact, a loss of neurons in the supraoptic and paraventricular nuclei of the hypothalamus, in particular of the magnocellular neurons, has been shown in transgenic mice (16) as well as in brain autopsy studies in cases of familial neurohypophyseal DI (18–20). The mechanism of cytotoxicity of the accumulated mutant AVP could explain the dominant inheritance pattern, the onset after the first year of life and the progressive nature of neurohypophyseal DI. In line with this, radiological studies show a gradual loss of the bright spot in the posterior pituitary as neurohypophyseal DI patients age (21). As suggested earlier, the c.322G>T mutation is expected to result in an AVP mutant with a different C-terminus (starting at codon 108) and/or in a truncated protein (p.~/p.Glu108X). It is, therefore, very likely that the p.~/p.Glu108X mutants are also able to heterodimerize with wild-type AVP considering that the C67X mutant is able to bind with wild-type AVP, indicating that the binding site resides within the first 66 amino acids of the AVP protein. The dominant inheritance in this family would be consistent with this reasoning.

Apart from its peripheral effects, AVP exerts central effects as a neuromodulator or neurotransmitter, mediated by centrally projecting AVP containing neurons in the paraventricular nucleus and in a number of additional hypothalamic nuclei such as the suprachiasmatic nucleus, the bed nucleus of the stria terminalis, the anteromedial subnucleus of the basal nucleus, and the diagonal band of Broca (1). These projections form the anatomical basis for central effects of AVP on mental functions including learning, memory, and social behavior (22). At present, it is not completely clear whether and how much these functions are affected in patients with central DI. A study on neuropsychological function in 23 neurohypophyseal DI patients, who were compared with non-affected family members and unrelated volunteers, did show minor disturbances in memory retrieval and sustained attention (23). The reason, why in patients with central DI abnormalities in central brain functions appear relatively small, compared with the major disturbance in osmoregulation, is not clear. One explanation could be that fewer parvocellular AVP neurons are lost compared with the magnocellular neurons of the hypothalamo-hypophyseal system (22), as a result of higher production rates of AVP in

the latter (24). Alternatively, the plasticity of the developing brain may be high enough to compensate for the loss of one particular neuropeptide, whereas the specific antidiuretic effect in the kidney via the V2 receptor cannot be compensated for by another peptide hormone.

Of interest is that the mother had noted two distinct periods during which the symptoms had changed in a major way. During pregnancy the symptoms had been worse, probably due to increased vasopressinase activity by the placenta, leading to increased degradation of the already sparse AVP (25). On the contrary, during labor, she had noticed a dramatic improvement of her polyuria and polydipsia. It is tempting to speculate that this is related to the increase in plasma oxytocin during late pregnancy and labor from ~17 to 45 pmol/l, which is 50 times higher than AVP concentrations (26). The sequence of oxytocin differs from AVP by only 2 amino acids, explaining why oxytocin has a mild antidiuretic effect via the V2 receptor in the rat kidney (27).

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.

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References

- 1 Swaab DF. *Handbook of Clinical Neurology; Human Hypothalamus: Basic and Clinical Aspects, Part I*. Series Eds Aminoff, Boller & Swaab. 2004.
- 2 Dierckx K & Vandesande F. Immunocytochemical localization of the vasopressinergic and the oxytocinergic neurons in the human hypothalamus. *Cell Tissue Research* 1977 **184** 15–27. (doi:10.1007/BF00220524)
- 3 Fliers E, Swaab DF, Pool CW & Verwer RW. The vasopressin and oxytocin neurons in the human supraoptic and paraventricular nucleus; changes with aging and in senile dementia. *Brain Research* 1985 **342** 45–53. (doi:10.1016/0006-8993(85)91351-4)
- 4 Robertson GL, Shelton RL & Athar S. The osmoregulation of vasopressin. *Kidney International* 1976 **10** 25–37. (doi:10.1038/ki.1976.76)
- 5 Baylis PH & Cheetham T. Diabetes insipidus. *Archives of Disease in Childhood* 1998 **79** 84–89. (doi:10.1136/adc.79.1.84)
- 6 Riddell DC, Mallonee R, Phillips JA, Parks JS, Sexton LA & Hamerton JL. Chromosomal assignment of human sequences encoding arginine vasopressin–neurophysin II and growth hormone releasing factor. *Somatic Cell and Molecular Genetics* 1985 **11** 189–195. (doi:10.1007/BF01534707)
- 7 Abu LA, Levy-Khademi F, Abdulhadi-Atwan M, Bosin E, Korner M, White PC & Zangen DH. Autosomal recessive familial neurohypophyseal diabetes insipidus: onset in early infancy. *European Journal of Endocrinology* 2010 **162** 221–226. (doi:10.1530/EJE-09-0772)
- 8 Willcutts MD, Felner E & White PC. Autosomal recessive familial neurohypophyseal diabetes insipidus with continued secretion of mutant weakly active vasopressin. *Human Molecular Genetics* 1999 **8** 1303–1307. (doi:10.1093/hmg/8.7.1303)
- 9 Bahnsen U, Oosting P, Swaab DF, Nahke P, Richter D & Schmale H. A missense mutation in the vasopressin–neurophysin precursor gene cosegregates with human autosomal dominant neurohypophyseal diabetes insipidus. *EMBO Journal* 1992 **11** 19–23.
- 10 Nijenhuis M, van den Akker EL, Zalm R, Franken AA, Abbes AP, Engel H, de Wied W & Burbach JP. Familial neurohypophysial diabetes insipidus in a large Dutch kindred: effect of the onset of diabetes on growth in children and cell biological defects of the mutant vasopressin prohormone. *Journal of Clinical Endocrinology and Metabolism* 2001 **86** 3410–3420. (doi:10.1210/jc.86.7.3410)
- 11 Maquat LE. Nonsense-mediated mRNA decay: splicing, translation and mRNP dynamics. *Nature Review. Molecular and Cellular Biology* 2004 **5** 89–99. (doi:10.1038/nrm1310)
- 12 Sausville E, Carney D & Battey J. The human vasopressin gene is linked to the oxytocin gene and is selectively expressed in a cultured lung cancer cell line. *Journal of Biological Chemistry* 1985 **260** 10236–10241.
- 13 Eubanks S, Nguyen TL, Deeb R, Villafania A, Alfadhli A & Breslow E. Effects of diabetes insipidus mutations on neurophysin folding and function. *Journal of Biological Chemistry* 2001 **276** 29671–29680. (doi:10.1074/jbc.M103477200)
- 14 Ito M, Yu RN & Jameson JL. Mutant vasopressin precursors that cause autosomal dominant neurohypophyseal diabetes insipidus retain dimerization and impair the secretion of wild-type proteins. *Journal of Biological Chemistry* 1999 **274** 9029–9037. (doi:10.1074/jbc.274.13.9029)
- 15 Davies J & Murphy D. Autophagy in hypothalamic neurones of rats expressing a familial neurohypophysial diabetes insipidus transgene. *Journal of Neuroendocrinology* 2002 **14** 629–637. (doi:10.1046/j.1365-2826.2002.00822.x)
- 16 Russell TA, Ito M, Ito M, Yu RN, Martinson FA, Weiss J & Jameson JL. A murine model of autosomal dominant neurohypophyseal diabetes insipidus reveals progressive loss of vasopressin-producing neurons. *Journal of Clinical Investigation* 2003 **112** 1697–1706. (doi:10.1172/JCI18616)
- 17 Si-Hoe SL, De Bree FM, Nijenhuis M, Davies JE, Howell LM, Tinley H, Waller SJ, Zeng Q, Zalm R, Sonnemans M, Van Leeuwen FW, Burbach JP & Murphy D. Endoplasmic reticulum derangement in hypothalamic neurons of rats expressing a familial neurohypophyseal diabetes insipidus mutant vasopressin transgene. *FASEB Journal* 2000 **14** 1680–1684. (doi:10.1096/fj99-0892fje)
- 18 Bergeron C, Kovacs K, Ezrin C & Mizzen C. Hereditary diabetes insipidus: an immunohistochemical study of the hypothalamus and pituitary gland. *Acta Neuropathologica* 1991 **81** 345–348. (doi:10.1007/BF00305879)
- 19 Braverman LE, Mancini JP & McGoldrick DM. Hereditary idiopathic diabetes insipidus. A case report with autopsy findings. *Archives of Internal Medicine* 1965 **63** 503–508.
- 20 Green JR, Buchan GC, Alvord EC Jr & Swanson AG. Hereditary and idiopathic types of diabetes insipidus. *Brain* 1967 **90** 707–714. (doi:10.1093/brain/90.3.707)
- 21 Mahoney CP, Weinberger E, Bryant C, Ito M, Jameson JL & Ito M. Effects of aging on vasopressin production in a kindred with autosomal dominant neurohypophyseal diabetes insipidus due to the DeltaE47 neurophysin mutation. *Journal of Clinical Endocrinology and Metabolism* 2002 **87** 870–876. (doi:10.1210/jc.87.2.870)
- 22 de Wied D, Diamant M & Fodor M. Central nervous system effects of the neurohypophyseal hormones and related peptides. *Frontiers in Neuroendocrinology* 1993 **14** 251–302. (doi:10.1006/frne.1993.1009)
- 23 Bruins J, Kovacs GL, Abbes AP, Burbach JP, van den Akker EL, Engel H, Franken AA & de Wied D. Minor disturbances in central

- nervous system function in familial neurohypophysial diabetes insipidus. *Psychoneuroendocrinology* 2006 **31** 80–91. (doi:10.1016/j.psyneuen.2005.06.001)
- 24 Burbach JP, Luckman SM, Murphy D & Gainer H. Gene regulation in the magnocellular hypothalamo-neurohypophysial system. *Physiological Reviews* 2001 **81** 1197–1267.
- 25 Durr JA, Hoggard JG, Hunt JM & Schrier RW. Diabetes insipidus in pregnancy associated with abnormally high circulating vasopressinase activity. *New England Journal of Medicine* 1987 **316** 1070–1074. (doi:10.1056/NEJM198704233161707)
- 26 Risberg A, Olsson K, Lyrenas S & Sjoquist M. Plasma vasopressin, oxytocin, estradiol, and progesterone related to water and sodium excretion in normal pregnancy and gestational hypertension. *Acta Obstetrica et Gynecologica Scandinavica* 2009 **88** 639–646. (doi:10.1080/00016340902919002)
- 27 Li C, Wang W, Summer SN, Westfall TD, Brooks DP, Falk S & Schrier RW. Molecular mechanisms of antidiuretic effect of oxytocin. *Journal of the American Society of Nephrology* 2008 **19** 225–232. (doi:10.1681/ASN.2007010029)

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