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PAK3 Related Mental Disability: Further Characterization of the Phenotype

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We report clinical, neuropsychological and molecular findings in affected males and carrier females in the fourth reported family with mental retardation caused by mutation in the *PAK3* gene (Xq22.3-q23), W446S. In contrast to previous reports, carrier females manifested learning problems and mild mental disability. Skewed X-inactivation was observed here for the first time in carriers of *PAK3* mutation. Neuropsychological tests in affected males and carrier females suggested a common neuropsychological profile of impaired spatial cognitive abilities and defects in attentional and executive functions. The five affected males examined herein had a proportionally small head size or microcephaly, large ears, oral motor hypotonia with drooling and inarticulate speech and short attention span, anxiety, restlessness, and aggression. Brain imaging showed signs of chronic non-progressive hydrocephalus in one patient who manifested psychosis and fluctuant gait deterioration, while two other patients showed no abnormalities. EEG recordings were available from four affected males and one carrier female, and all showed similar posterior slow wave activity without epileptic discharges. Only one affected male in the family suffered from epilepsy. When comparing the affected males in this family and the three previously reported families with mental retardation due to a *PAK3* mutation, similarities in their characteristics were small head size or microcephaly, large ears, speech defects, behavioral abnormalities, and psychiatric disease. © 2007 Wiley-Liss, Inc.

Key words: *PAK3*; mutation; mental retardation; X-linked; mental disorder; behavioral symptoms; carrier symptoms

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INTRODUCTION

X-linked mental retardation (XLMR) is clinically and genetically heterogeneous with some 60 causative genes identified to date [Kleefstra and Hamel, 2005; Raymond and Tarpey, 2006]. It is divided into syndromic and non-syndromic forms, the latter referring to cases which lack recognizable dysmorphic features or other distinguishing symptoms or signs. The boundary between these two, however, has been repeatedly blurred by discovery of pleiotropic phenotypes with shared monogenic background [Kleefstra and Hamel, 2005; Raymond and Tarpey, 2006].

The search for X-chromosomal genes related to mental retardation in non-syndromic cases has revealed a mutation in the *PAK3* gene in three previous families each carrying a different rearrangement [Donnelly et al., 1996; des Portes et al., 1997; Allen et al., 1998; Bienvenu et al., 2000; Gedeon et al., 2003]. We report here the evaluation of five retarded males and four carrier females of a family who underwent thorough clinical and neuropsychological examination. We also compare these patients to previous families to delineate the *PAK3* mutation related phenotype.

M. Peippo, A.M. Koivisto, and T. Särkämö contributed equally to the work.

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Case Report

Five mentally disabled males (II-1, II-4, III-2, III-3, and IV-1, Fig. 1) and four of their female relatives (I-1, II-2, II-3, and III-1) were available for this study. We reviewed their medical history, performed personal interviews, clinical examinations and molecular studies. Each had an unremarkable pre- and perinatal history. The cause of the mental disability in males had not been revealed by chromosomal studies in all of them including high-resolution karyotype (550 band resolution) in one, subtelomere FISH in one, search for FRAXA mutation in three, FRAXE mutation in one, or urinary metabolic screening tests in three of the five males.

As reported by several relatives, individual II-10 is mentally retarded but neither he nor his mother were available for the study. Several relatives reported individuals I-6 and II-6 to have been mentally subnormal, unable to go to school or to live independently. They had died at ages 88 and 17 years respectively. Individual I-2 died in early childhood due to congenital malformations. Six family members (I-4, II-5, III-4, III-5, III-6, and III-11) with a normal educational and professional history consented to molecular studies and were interviewed by telephone.

The ethical committee of the Department of Obstetrics and Gynaecology at the Helsinki University Central Hospital, Helsinki, Finland had approved the study. The use of anonymous blood donors as control samples was approved by the Ethical Committee of the Finnish Red Cross Blood Transfusion Services.

Molecular Genetic Studies

DNA was extracted from 10 ml of peripheral blood using genomic DNA purification kit (PureGene®, Gentrasystems) according to the manufacturer's instructions. Genotypes for 88 evenly spaced X-chromosomal microsatellites (average interval of 2 cM) were generated using standard fluorescent-detection–based semi-automatic technology [Kong et al., 2002]. The allele frequencies for the data were estimated using DOWNFREQ 2.1 program. Haplotypes shared by affected males were analyzed manually.

PCR amplification of the candidate genes were performed as described elsewhere [Forsman et al., 2003] and the specificity of the PCR products was assessed by 1.5% agarose gel electrophoresis. The sequence reactions were performed from the purified PCR products using the BigDye 3.1 terminator kit (Applied Biosystems) according to manufacturer's instructions. PCR and sequencing primers can be obtained from the authors at request. The products were separated on an ABI 3730 DNA Analyzer (Applied Biosystems) and base calling was performed by using Sequencing Analysis 5.2 software (Applied Biosystems). Reference sequences were obtained from the UCSC Human Genome Browser (http://www.genome.uscs.edu; PAK3: NM_002578.2) and sequence analysis was performed by using Sequencher 4.1.4. software (Gene Codes). The possible functional impact of the identified amino acid change was predicted by the PolyPhen program (http://www.tux.embl-heidelberg.de/ramensky/polyphen/cgi). ClustalW (http://

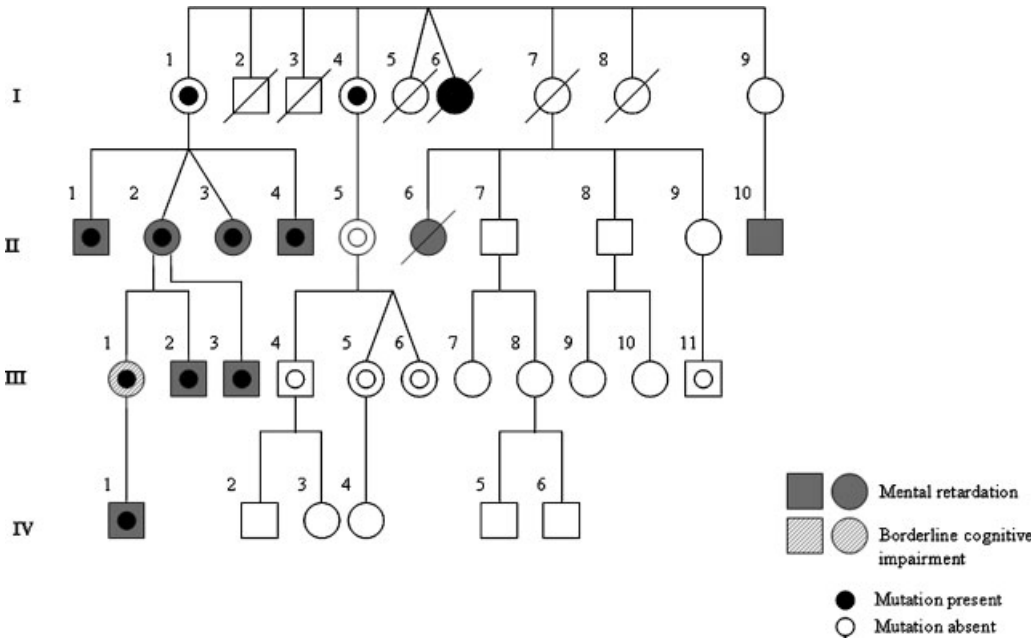

FIG. 1. Pedigree of the family showing the phenotypes and the results of the molecular studies.

www.ebi.ac.uk/clustalw) was used for multiple alignment of the different human p-21 activated kinase (PAK) proteins and orthologs.

X-inactivation was examined in females I-1, I-4, II-2, II-3, and III-1 by the methylation status of the polymorphic *AR* locus (Xq12) as described elsewhere [Allen et al., 1992; Plenge et al., 1997].

Neuropsychological Assessment

Neuropsychological examination of males II-1, II-4, III-2, and III-3, and females I-1, II-2, II-3, and III-1 was performed to assess the degree of mental retardation and neuropsychological impairment. Individual IV-1 had previously been evaluated clinically for proper choice of school type. The level of intelligence and reasoning abilities was evaluated with Wechsler Adult Intelligence Scale III (WAIS-III). Based on WAIS-III total IQ score the degree of mental retardation was classified according to ICD-10 criteria (mild: IQ 50–69, moderate: IQ 35–49, severe: IQ 20–34, profound: IQ < 20). The following cognitive domains were also evaluated (test methods indicated in parenthesis): verbal comprehension (Verbal Comprehension Index of WAIS-III), verbal expression (Verbal fluency test, Boston naming test), reading and writing, visuospatial perception (Clock test), constructional function (Block Design subtest of WAIS-III, Copying of geometric designs, Rey Complex Figure Test, Bicycle drawing test), verbal memory (Digit Span and Logical Memory subtests of Wechsler Memory Scale-revised (WMS-R), Word span and Word list memory tests), visual memory (Visual Reproduction subtest of WMS-R, Rey Complex Figure Test), attentional and executive functions (Trail Making Test, Digit Symbol subtest of WAIS-III, Verbal fluency test, Digit Span subtest of WMS-R), motor speed (Finger Tapping Test), and fine motor function (Luria's test). Performance in these cognitive domains was classified as normal, mildly impaired, moderately impaired or severely impaired based on clinical experience and normative data (normal = ≤1 SD, mild = ≤2 SD, moderate = ≤3 SD, severe >3 SD below normative mean).

RESULTS

Molecular Genetic Findings

The affected males were found to share a haplotype at Xq21.31q24 spanning about 30 cM. The region contained three known non-syndromic XLMR genes, namely p-21 activated kinase 3 (*PAK3*), CoA synthetase long-chain family member 4 (*ACSL4*) and angiotensin II receptor gene (*AGTR2*). All three genes were sequenced for mutations by direct sequencing of the open reading frame and flanking introns. We identified a novel missense mutation c.1337G > C in exon 7 of the *PAK3*, which predicts p.W446S (Fig. 2) [Berg et al., 2002]. The mutation was present in all five affected males and absent in the two unaffected male relatives. Each mother of an affected male was found to be carrier of the mutation. Thus the segregation of the mutation was compatible with X linked inheritance implicated by the pedigree (Fig. 1). The mutation was not detected in any of the 200 anonymous, unrelated, normal Finnish controls.

The PolyPhen program predicted the mutation to have an influence on the structure and function of the *PAK3* encoded protein by causing an alteration in its kinase domain. This is a highly conserved region among as well humans as several other species including *Caenhorhabditis elegans* as shown by the alignment results (Fig. 3).

In studies of the *AR* locus a skewed X-inactivation pattern was identified in the phenotypically normal females I-1 (90%:10%) and I-4 (91%:9%) as well as in the borderline female III-1 (100%:0%). Females II-2 and II-3 (dizygotic twins; data not presented) were homoallelic at the *AR* locus and thus uninformative.

Clinical Features of the Affected Males

Individual *II-1* is a 46-year-old man with moderate mental retardation but fairly good self-help skills. He has stooping posture, shuffling steps, normal tendon reflexes and no spasticity. In childhood he drooled copiously. Still his mouth tends to remain open, he speaks unclearly and hastily and uses short sentences. His recurring behavioral problems have been resistant to all trials of pharmacotherapy. He lives in a residential home and goes to a sheltered job. Figure 4 shows the facial features of the males at various ages

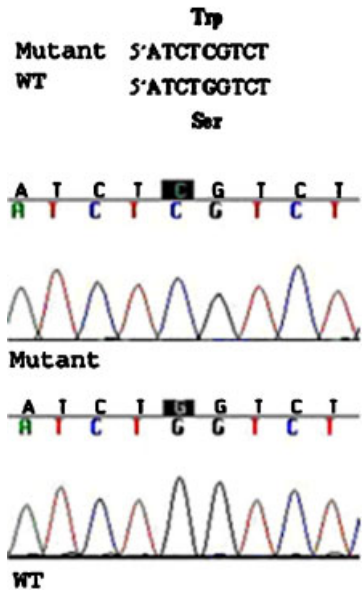

FIG. 2. The *PAK3* mutation detected in the family is c.1337G > C, which predicts p.W446S. The figure presents as an example the mutated sequence of IV-1 in the pedigree. [Color figure can be viewed in the online issue, which is available at http://www.interscience.wiley.com.]

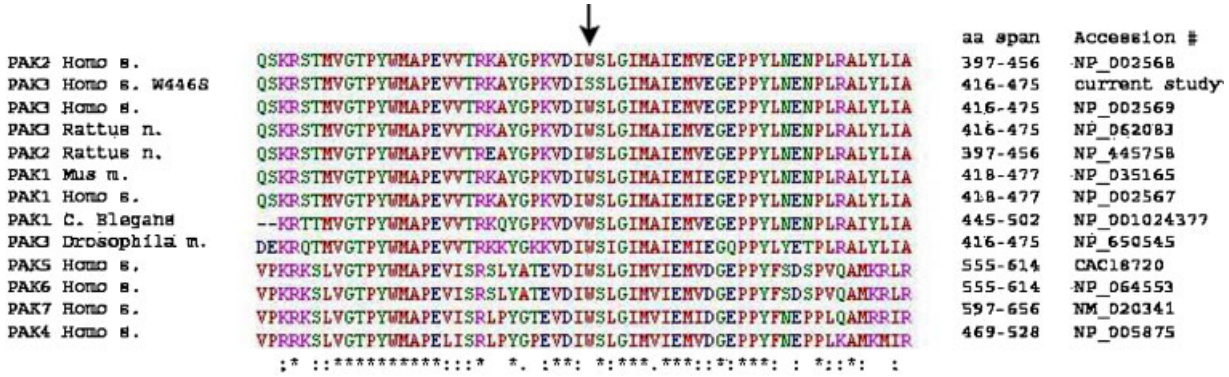

FIG. 3. Alignment of different human PAK proteins and orthologs showing the sequence similarity of the region flanking the p.W446S mutation. [Color figure can be viewed in the online issue, which is available at http://www.interscience.wiley.com.]

(except for individual IV-1 where permission to publish photos was denied). A summary of their main clinical features is presented in Table I. A cranial CT scan was normal at age 44 years. His EEG recording showed posterior slow waves without epileptic discharges.

Individual *II-4* is a 42-year-old man with moderate mental retardation. He has had paranoid psychosis

FIG. 4. Photos of the affected males. Patient II-2 at ages 46 (A1), 20 (A2), and 10 years (A3). Patient II-4 at ages 42 (B1) and 16 years (B2). Patient III-2 at ages 21 (C1) and 6 years (C2). Patient III-3 at ages 16 (D1) and 7 years (D2) and 1 year (D3). Note large ears, high bridged nose, and thin upper lip; sloping and low forehead (A, B, C); deep frontal furrows (A, B); unusual hair patterning (B, C). [Color figure can be viewed in the online issue, which is available at http://www.interscience.wiley.com.]

TABLE I. Clinical Features in the Affected Males

Patient and age	II-1, 46 years	II-4, 42 years	III-2, 22 years	III-3, 16 years	IV-1, 6 years
MR	Moderate	Moderate	Mild	Mild	Moderate
Height	179.0 cm (0 SD)	175 cm (−0.6 SD)	178.6 cm (0 SD)	166.0 cm (−1 SD)	118.0 cm (0 SD)
OFC	55.5 cm (−1.8 SD)	55.5 cm (−1.8 SD)	53.7 cm (−2.5 SD)	55.0 cm (−1 SD)	49.9 cm (−2.5 SD)
Age of independent walking	28 Months	27 Months	3 Years	22 Months	21 Months
Age of first words	30 Months	3 Years	5 Years	28 Months	21 Months
Ear lengths	7.8/7.8 cm (>+3 SD)	6.9/6.7 cm (>+3 SD)	7.8/7.9 cm (>+3 SD)	7.2/6.8 cm (+3 SD)	6.0/6.0 cm (>+1.5 SD)
Forehead	Low; deep furrows; heavy orbital ridges	Low; deep furrows; heavy orbital ridges	Low	Ordinary	Ordinary
Eyes	Deep set	Deep set	Ordinary	Ordinary	Ordinary
Nose	High bridged	High bridged	High bridged	High bridged	High bridged
Mouth	Thin upper lip	Thin upper lip	Thin upper lip	Thin upper lip	Thin upper lip
Palate	High vaulted	High vaulted	Narrow high vaulted	Narrow high vaulted	Narrow high vaulted
Fingertips	Fetal pads	Fetal pads	Fetal pads	Fetal pads	Fetal pads
Hair	Ordinary	Unusual pattern	Unusual pattern	Ordinary	Ordinary
Testicular size	Normal	NA	Normal	NA	Normal
Oral motor features	Mouth remains open; indistinct speech	Mouth remains open; hasty, mildly indistinct speech	Mouth remains open; indistinct speech; copious drooling	Ordinary	Mouth remains open; hasty, indistinct speech; drooling
Behavior and mental features	Aggression; inattention; agitation; anxiety; clinging	Aggression; inattention; agitation; paranoid psychosis	Aggression; inattention; insomnia, obsessions	Aggression; inattention; temper tantrums; restlessness	Inattention; high hyperactivity
Neurological symptoms	Stooping posture; short steps	Stooping posture; short shuffling steps; chronic non-progressive hydrocephalus	Childhood hypotony; motor clumsiness; epilepsy (general tonic-clonic seizures, absences)	Childhood hypotony	Early childhood hypotony; motor clumsiness; occasional febrile convulsions
Brain imaging (age)	CT normal (44 years)	CT hydrocephaly (32 and 41 years)	ND	CT normal (8 months)	ND

MR, mental retardation; NA, not available; ND, not done.

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since his late twenties causing recurrent hospitalizations. Since 32 years of age he has complained pains in lower extremities and has walked with a stiff shuffling gate, which fluctuates regardless of antipsychotic medication. His deep tendon reflexes are normal, there is no spasticity, and no progression of motor signs has been observed at repeated neurological examinations. He lives in a residential home, has persistent hallucinations, but yet most days goes to a sheltered job. A cranial CT scan both at the age of 32 and 41 years showed enlarged lateral and 3rd ventricles with normal sized 4th ventricle and flat cortical gyri; the CT images were not available for publication. The etiology of the hydrocephaly remains obscure. His EEG recordings show a similar pattern of slow waves like in his brother II-1.

Individual *III-2* is a 22-year-old man with mild mental retardation. His developmental delay was noticed at the age of 8 months. Epileptic seizures started at the age of 7 years, they are usually short absences and sometimes generalized tonic-clonic seizures and occur a few times a year. Behavioral problems manifested also at the age of 7 years and an effective medication or other treatment has been difficult to find. Oral motor hypotonia and copious drooling continue. He speaks using only single word expressions. He has completed elementary school in special education, goes to a sheltered job, and lives in a residential home. Brain imaging studies have not been performed. His EEG recordings also show an excess of slow posterior waves but no epileptic discharges.

Individual *III-3* is a 16-year-old schoolboy with mild mental retardation. His developmental delay was obvious at 8 months of age. He has asthma and frequent respiratory infections. An idiopathic scoliosis was operated at the age of 15 years with a satisfactory result. His only epileptic seizure occurred at the age of 9 years. His sudden outbursts of aggression were suspected to be caused by epileptic mechanisms and were treated by oxcarbazepine for 4 years. He has normal hearing, vision and ophthalmologic findings. He had oral motor dysfunction which has alleviated with age. His self-help skills are good but he needs continuous supervision. His cranial CT scan was normal at the age of 8 months. His EEG also showed posterior slow waves but no epileptic discharges.

Individual *IV-1* is 6 years old with moderate mental retardation. He suffers from asthma and frequent respiratory infections. His developmental delay was diagnosed at the age of 5 months when he was exceptionally weepy, passive, and limp with normal tendon reflexes. At the age of 34 months, his expressive speech corresponded to 13–15 months. Earlier he had oral hypotonia with drooling and tendency to keep his mouth open. Presently he is extremely hyperactive, stubborn and talkative with slightly inarticulate speech. Brain imaging studies or EEG recordings have not been done to him.

Clinical Features of the Carrier Females

Individual *I-1* is a 76-year-old pensioner, who after elementary school worked as a cleaning lady and a factory worker. She has always taken good care of her family and gives impression of a person with determination. Clinical examination of her was not feasible. Figure 5 shows the facial features of I-1, II-2, II-3 at various ages, III-1 denied permission to publish photos.

Individual *II-2*, twin sister of II-3, is 44 years old. Her height is 153 cm (−2 SD), OFC 53.5 cm (−1.2 SD), ear lengths 6.0 cm and 6.3 cm (∼0 SD) and her palate is narrow and high-vaulted. She has gone to special school, has no occupational education and has been unemployed for years. She lives independently with her son (III-3).

Individual *II-3*, twin sister of II-2, is 44 years old. Her height is 157 cm (−1.5 SD), OFC 56.7 cm (+1.6 SD) and ear lengths 6.0 cm and 6.3 cm (+0.8 SD). Her palate is high-vaulted, palpebral fissures slant up, neck is short and nose high bridged. In early childhood, she was hypotonic with normal deep tendon reflexes. She learned to walk at 16 months of age. Ophthalmologic examination at 1.5 years due to right exotropia, revealed small papillae and large oval chorioretinal scars in both fundi with right macular damage; re-examination at the ages of 9 and 44 showed no progression. Vision in the left eye was 20/28 (logMAR 0.15) with the best correction (−3.5 D cyl +0.5 D × 130) and counting fingers 1 m in the right. Recurrent blotchy alopecia healed in her teens. Hypothyroidism as a sequel of childhood thyreoiditis is compensated by thyroxin medication. She went to special school and had an adapted occupational training in textile work. Due to hand eczema, she is retired since the age of 31 years. She lives independently with help in some practical maters. Minor intracranial calcifications on the skull X-ray were seen at the age of 1.5 years. Sabin–Feldman dye test was positive at age 1.5 years and toxoplasmosis was suspected, but 8 years later the dye test was negative. Her EEG at the age of 6 years showed occipital slow waves. No brain imaging study has been performed.

Individual *III-1* is a 25-year-old woman, whose height is 157 cm (−1.5 SD), OFC 54.0 cm (−1 SD), ear lengths 6.4 cm and 6.5 cm (+1 SD), palate is high-vaulted and nose-high bridged. She learned to walk at 18 months and had normal speech development. Due to immaturity, hyperactivity, and short attention span she started school 1 year late. She attended basic school in special education, dropped out from vocational education, and is presently unemployed. She needs extensive social support to raise her son (individual IV-1).

Neuropsychological Findings

Neuropsychological findings in males II-1, II-4, III-2, and III-3 and in carrier females I-1, II-2, II-3, and

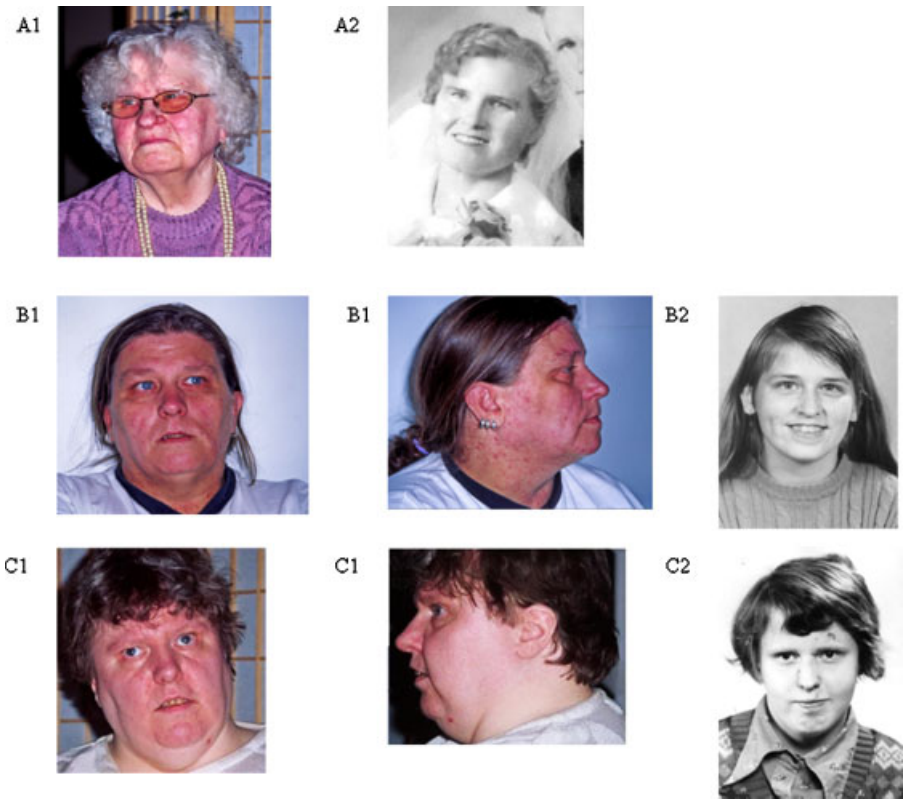

FIG. 5. Photos of the carrier females. Female I-1 at ages 76 (A1) and 28 years (A2). Female II-2 at ages 42 (B1) and 12 years (B2). Female II-3 at ages 42 (C1) and 12 years (C2). Note thin upper lip, high bridged nose; proportionally large ears (B, C); upward slant of palpebral fissures (C). [Color figure can be viewed in the online issue, which is available at http://www.interscience.wiley.com.]

III-1 are presented in Table II. The males had mild to moderate mental retardation, moderately to severely impaired visuospatial and constructional skills, severe visual memory impairment and severe impairment of attentional and executive functions. Their writing was limited at best to short words and their fine motor performance was mildly to moderately impaired. Individuals II-1 and III-2 had moderately to severely- and II-4 and III-3 mildly to moderately impaired verbal expression, comprehension, reading, and memory.

Females II-2, II-3, and III-1 had borderline or mild mental retardation, while I-1 had an overall cognitive ability of low average level. Females II-2, II-3, and III-1 had normal to mildly impaired verbal expression, comprehension, and memory. They were rather fluent readers whereas their constructional skills, visual memory and attentional and executive functions were mildly to severely impaired. They all reported having always had difficulties in everyday tasks requiring spatial cognition such as drawing, weaving or doing needlework or navigating routes, whereas they did not report analogous verbal difficulties. All four females had mild dysgraphia with many spelling errors and letter omissions, and normal to mildly impaired fine motor skills. Female I-1 with advanced age and low average IQ, had mild impairments in language and visual skills and moderate deficits in memory.

DISCUSSION

Our report describes the fourth family with mental retardation cosegregating with a mutation in the *PAK3* gene. The p.W446S mutation is novel and was detected in all affected males as well as in obligate carrier females, but was absent from normal male relatives and from 200 anonymous normal control individuals. Like the mutations in two previous families [Allen et al., 1998; Gedeon et al., 2003], p.W446S is also a missense mutation while the third previously reported family had a truncating mutation. All four *PAK3* mutations reside in the functionally important kinase domain of this evolutionary conserved gene. The function of *PAK3* in man is still unknown but in mice it is implicated as a partner in the Rho signaling pathway that regulates synaptic function and cognition [Meng et al., 2005].

In the present family, thorough neuropsychological assessment was done for the males, and to our knowledge, this is the first report of such testing in carrier females. Unlike carriers in previously

TABLE II. Neuropsychological Findings in the Affected Males and Carrier Females

Patient, age	II-1, 46 years	II-4, 42 years	III-2, 22 years	III-3, 16 years	I-1, 76 years	II-2, 44 years	II-3, 44 years	III-1, 25 years
Gender	Male	Male	Male	Male	Female	Female	Female	Female
Intelligence (IQ)	Moderate MR (42)	Moderate MR (49)	Mild MR (60)	Mild MR (55)	Low average (80)	Mild MR (61)	Mild MR (68)	Borderline (72)
Language skills								
Verbal expression	Moderate I	Mild I	Moderate I	Mild I	Mild I	Normal	Normal	Normal
Verbal comprehension	Severe I	Moderate I	Moderate I	Mild I	Mild I	Mild I	Mild I	Mild I
Reading	Single letters	Slow	Single letters	Short words	Fluent, some errors	Slow	Slow	Fluent
Writing	Unable	Short words	Short words	Short words	Mild I	Mild I	Mild I	Mild I
Visual skills								
Visuospatial perception	Severe I	Severe I	Severe I	Severe I	Mild I	Normal	Mild I	Normal
Constructional function	Severe I	Severe I	Moderate I	Moderate to severe I	Mild I	Moderate I	Moderate I	Mild I
Memory								
Verbal memory	Severe I	Moderate I	Severe I	Mild	Moderate I	Average	Mild I	Mild I
Visual memory	Inassessable	Severe I	Severe I	Severe I	Moderate I	Severe I	Severe I	Severe I
Attentional and executive functions	Severe I	Severe I	Severe I	Severe I	Normal	Mild I	Severe I	Mild I
Fine motor skills								
Speed	Moderate I	Moderate I	Mild I	Moderate I	Mild I	Normal	Mild I	Mild I
Function	Moderate I	Moderate I	Mild I	Mild I	Normal	Normal	Mild I	Normal

MR, mental retardation; I, impairment.

reported PAK3 retardation families, the carriers in the present case showed deficiencies of cognitive capacity. X-inactivation studies were undertaken to further examine this finding although no data on X-inactivation was reported in the prior three cases. X-inactivation studies were uninformative in two of the mildly retarded or borderline females II-2 and II-3, but interestingly, skewed X-inactivation ($\geq$80%:20%) was seen in two normal carrier females (individuals I-1 and I-4) and in the borderline retarded female (individual III-1). The skewed X-inactivation is in keeping with carrier status of an X-linked deleterious mutation and identifies this phenomenon also in *PAK3*-related XLMR [Plenge et al., 2002]. It is hypothesized that the mutated X is usually inactivated since the mutation would render a growth disadvantage to rapidly proliferating cells like blood cells [Willard, 2000; Plenge et al., 2002], but also a positive growth advantage for the mutant X has been reported [Migeon et al., 1981]. The results in female III-1 could thus imply either a positive growth effect for the *PAK3* mutation or that the proportions of active normal and mutated X-chromosomes differ in brain cells from that in blood cells. Additionally, variations in other genes of the Rho signaling pathway could modify the resulting phenotype.

Table III summarizes the clinical features of all 28 affected males with a *PAK3* mutation [Donnelly et al., 1996; des Portes et al., 1997; Gedeon et al., 2003] and the present study. Interestingly, the mental retardation seems to be more severe in the family of des Portes et al. [1997], which had a truncating mutation [Bienvenu et al., 2000] as compared with families with a missense mutation. Typical symptoms in the males were mild to moderate mental retardation, small head size or microcephaly, large ears and tendency to oral motor hypotonia with drooling and inarticulate speech. Short attention span, anxiety, restlessness, and aggression were frequent in the patients reported here and these features have been encountered also in two of the previously reported families. *PAK3* mutations also appear to be associated with psychiatric morbidity. Cranial imaging findings have not been reported in previous patients. In two of three cases reported here, CT scan findings were normal whereas one showed chronic non-progressive hydrocephalus. This latter patient also had psychosis and fluctuating non-progressive motor symptoms. An EEG recording was available from four male patients and one mildly retarded carrier female. All recordings showed a similar pattern of posterior slow waves without epileptic discharges. Among the 28 males with a *PAK3* mutation only two have epilepsy, suggesting that epilepsy is an infrequent manifestation in *PAK3* related mental retardation.

All four personally examined affected adult males showed moderate to severe impairment in visuospatial and constructional abilities, visual

TABLE III. Review of Clinical Presentation in Previous and Present Male Patients With a Mutation in *PAK3*

References	Donnelly et al. [1996] and Allen et al. [1998]	des Portes et al. [1997] and Bienvenu et al. [2000]	Gedeon et al. [2003]	Present patients
Mutation	Missense (R67C) c.199C-T	Truncating (R419X) c.1255C-T	Missense (A365E) c.1094C-A	Missense (W446S) c.1337G-C
Number of patients examined	4	6	13	5
Mental retardation	Mild in 4/4	Moderate in 5/6 Severe in 1/6	Borderline to mild in 13/13	Mild in 2/5 Moderate in 3/5
Stature	Normal range	Normal range	Normal range	Normal range
Small head size	in 4/4	NR	NR	in 4/5
Appearance	Long ears in 1/4	Unspecific	Long ears in 7/13 High palate in 1/13 Prominent nose in 1/13	Long ears in 4/5 High palate in 5/5 High bridged nose in 5/5 Thin upper lip in 4/5 Fetal pads in fingertips in 5/5
Oral motor features	NR	Poor and inarticulate speech in 4/6	Inarticulate speech in 1/13	Poor and inarticulate speech in 4/5 Open mouth appearance in 4/5 Prolonged/persistent drooling in 5/5
Behavioral/psychological problems	in 2/4	None	in 3/13	in 5/5
Neurologic symptoms	None	None	Epilepsy in 1/13	Epilepsy in 1/5 Chronic hydrocephaly in 1/5 Shuffling gate in 2/5
Neuropsychological profile	ND	Unspecific	ND	Impaired spatial cognitive skills and deficits in attentional and executive functions in 4/4
EEG	NR	NR	NR	Done in 4: posterior slow waves in 4/4
Brain imaging	ND	ND	ND	Done in 3: normal CT in 2/3, chronic hydrocephaly in CT in 1/3

NR, not reported; ND, not done.

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memory, attentional and executive functions and writing, whereas only two of them had verbal impairments of comparable severity. All had also mild to moderate fine motor impairment. Impaired visuospatial abilities were also found in two out of three neuropsychologically assessed patients in a previous report [des Portes et al., 1997]. These results would suggest that visuospatial deficits are characteristic of the *PAK3*-related cognitive phenotype.

A new finding detected in the present study is the low normal to borderline and mildly retarded cognitive level in carrier females. They shared a similar neuropsychological profile with the affected males, with markedly impaired visuoconstructional ability, visual memory, attentional, and executive functions and deficits in writing, but relatively spared verbal skills. Interestingly, the carrier women also shared some physical similarities with their retarded male relatives such as a high-vaulted palate and relatively large ears (Figs. 2 and 3).

PAK3 has been functions in neuronal synapse formation and hippocampal plasticity [Boda et al., 2004; Meng et al., 2005; Zhang et al., 2005]. As the hippocampus has a well-known function in spatial perception and memory [Squire et al., 2004] the spatial deficits observed in our patients are consistent with the role of hippocampal plasticity in the cognitive impairment in patients with a *PAK3* mutation. In the future, the possible link between spatial deficits and hippocampal function in patients with *PAK3* mutations might be studied by using detailed neuropsychological methods with structural and functional brain imaging.

Including the present patients, a total of 28 mentally retarded males with a *PAK3* mutation have so far been reported in detail. A typical physical, behavioral, and cognitive phenotype emerges with small head size or microcephaly, large ears, oral motor hypotonia with prolonged or persistent drooling and inarticulate speech, and tendency to behavioral abnormalities, psychiatric symptoms, and impaired spatial cognitive abilities. Patients who present with these findings should be considered candidates for analysis of the *PAK3* gene. The present family also shows that female *PAK3* mutation carriers can share some physical features with the affected males, manifest learning problems, or be mildly mentally disabled. Further families and careful examination of the female carriers are needed to clarify the frequency of these characteristics in PAK3-related mental retardation.

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