Pontocerebellar hypoplasia type 1

Clinical spectrum and relevance of *EXOSC3* mutations

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ABSTRACT

Objectives: Pontocerebellar hypoplasia with spinal muscular atrophy, also known as PCH1, is a group of autosomal recessive disorders characterized by generalized muscle weakness and global developmental delay commonly resulting in early death. Gene defects had been discovered only in single patients until the recent identification of *EXOSC3* mutations in several families with relatively mild course of PCH1. We aim to genetically stratify subjects in a large and well-defined cohort to define the clinical spectrum and genotype–phenotype correlation.

Methods: We documented clinical, neuroimaging, and morphologic data of 37 subjects from 27 families with PCH1. *EXOSC3* gene sequencing was performed in 27 unrelated index patients of mixed ethnicity.

Results: Biallelic mutations in *EXOSC3* were detected in 10 of 27 families (37%). The most common mutation among all ethnic groups was c.395A>C, p.D132A, responsible for 11 (55%) of the 20 mutated alleles and ancestral in origin. The mutation-positive subjects typically presented with normal pregnancy, normal birth measurements, and relative preservation of brainstem and cortical structures. Psychomotor retardation was profound in all patients but lifespan was variable, with 3 subjects surviving beyond the late teens. Abnormal oculomotor function was commonly observed in patients surviving beyond the first year. Major clinical features previously reported in PCH1, including intrauterine abnormalities, postnatal hypoventilation and feeding difficulties, joint contractures, and neonatal death, were rarely observed in mutation-positive infants but were typical among the mutation-negative subjects.

Conclusion: *EXOSC3* mutations account for 30%–40% of patients with PCH1 with variability in survival and clinical severity that is correlated with the genotype. ***Neurology*® 2013;80:438–446**

GLOSSARY

EXOSC3 = exosome component 3; **PCH** = pontocerebellar hypoplasia; **PCH1** = pontocerebellar hypoplasia with spinal muscular atrophy; **RRP40** = ribosomal RNA-processing protein 40; **SMA** = spinal muscular atrophy.

Pontocerebellar hypoplasia (PCH) denotes a clinically and genetically heterogeneous group of autosomal recessive developmental defects. The rare combination of PCH and anterior horn cell disease has also been referred to as amyotrophic cerebellar hypoplasia or cerebellar hypoplasia with Werdnig-Hoffmann disease.[1] The disorder was designated as PCH1[2] to distinguish from PCH2 without anterior horn cell disease; however, this designation did not fully capture the clinical and genetic

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Supplemental data at www.neurology.org

Supplemental Data

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heterogeneity within each group.[3] The most important differential diagnosis of PCH1 is infantile spinal muscular atrophy (SMA, OMIM 253300), which is a caused by mutations in the *SMN1* gene. In patients with PCH1, no mutations in *SMN1* have yet been found, thus PCH1 appears a distinct entity despite shared involvement of spinal motor neurons.

In PCH1, mutations were described in single patients only[4,5] up to the recent discovery of the *EXOSC3* gene.[6] *EXOSC3* encodes exosome component 3 (EXOSC3), also known as the ribosomal RNA–processing protein 40 (RRP40). EXOSC3 is a core component of the human RNA exosome complex that is evolutionarily conserved and important in RNA metabolism.

Although most patients with PCH1 reported to date conformed to the classical description of severe phenotype with onset prenatally or shortly after birth and death within the first year, some patients showed longer survival and a milder course.[7,8] We aim to investigate a large panel of unselected subjects for further genotype–phenotype correlation to ascertain the contribution of mutations in *EXOSC3* and to better define the clinical spectrum of PCH1.

METHODS We collected clinical and pathoanatomical details of 27 families with PCH1 recruited over many years from different countries worldwide. All families were compatible with autosomal recessive inheritance. *EXOSC3* gene analysis was performed by Sanger sequencing of all 4 exons and flanking introns as previously described.[6]

Standard protocol approvals, registrations, and patient consents. The study protocol was approved by the Institutional Review Board of the University Hospital RWTH Aachen. The inclusion criteria of at least one patient per family for genetic analysis were 1) presence of hypotonia, 2) evidence of neurogenic changes in the muscle by electromyogram, muscle biopsy, or anterior horn cell loss in the spinal cord by autopsy suggestive of spinal muscular atrophy, 3) symmetrical volume loss of the cerebellum by MRI or autopsy, and 4) exclusion of a biallelic deletion/mutation of the *SMN1* gene. A signed patient consent-to-disclose form was obtained for photographs of recognizable patients.

RESULTS **Mutation analysis.** Homozygous or compound heterozygous mutations of *EXOSC3* were detected in 15 patients of 10 families (table 1; figure 1). This corresponds to a diagnostic yield of 37% in the unselected cohort of 27 families. The most common mutation among all ethnic groups was c.395A>C, p.D132A, which was seen in 11 (55%) of the 20 mutated alleles.[6] A region of shared haplotypes of up to 1 cM observed in families harboring c.395A>C suggests an ancestral origin for this mutation (table e-1 on the *Neurology*® Web site at www.neurology.org). Family 9 had a compound heterozygous mutation including c.712T>C, p.W238R. The c.92G>C, p.G31A mutation was detected in the 2 Eastern European families (families 7 and 9); shared haplotype spanning *EXOSC3* in these 2 families is consistent with a founder mutation (table e-1). W238 and G31 are strictly evolutionarily conserved more than D132; all are located in functionally important motifs that alterations in these 3 residues are predicted to exert detrimental effects by in silico analysis as well as in their inability to rescue the mutant exosc3-knockdown phenotype in zebrafish embryos.[6]

We discovered 3 novel single nucleotide indel mutations: c.226dupG in family 4, c.155delC in family 8, and c.551delG in family 10. Furthermore, we observed a missense mutation c.2T>C, which alters the methionine-encoding start codon AUG, with the next in-frame AUG located more than 500 bases downstream, thereby expressing only the last 100 amino acids in this 275-residue protein. These mutations are predicted to result in null alleles.

Clinical features. The common clinical features of patients with *EXOSC3* mutations (table 1) include severe hypotonia, mostly absent motor or speech development, oculomotor dysfunction, and lack of fixation. Birth measurements were generally normal, and progressive microcephaly was documented in 6 of 10 patients where measurements were available (table 1). Head circumference regressed to mild to moderate microcephaly up to −3.5 SD but remained normal in some long-surviving patients (2-1, 3-1). Muscle tone was generally reduced but spasticity, increased tendon reflexes, and dystonic movements were recorded in 2 sibs of family 1, before flaccid pareses and reduced muscle tone was seen some years later. Epileptic seizures occurred in 3 patients and included absence epilepsy in patient 2-1 (responsive to valproic acid) and infantile spasms in 2 sisters (5-1, 5-2). Available autopsy specimens consistently demonstrated a loss of neurons in the cerebellum, parts of the midbrain, and the anterior spinal cord.

Despite the general consistency in all mutation-positive subjects in the clinical manifestations of hypotonia, progressive muscular atrophy, and global developmental delay, there was variation in the clinical presentation and lifespan that was correlated with the genotype:

Families 1–3 (mild PCH1) (figure 2): Patients appeared normal at birth but presented with psychomotor retardation within 6 months of life, had preserved respiratory function, showed little neurologic decline or regression, and survived beyond early infancy even into adulthood. Patients with mild PCH1 all shared the genetic feature of harboring homozygous c.395A>C, p.D132A mutations.

Table 1 Clinical features of 15 patients out of 10 families with mutations, arranged by decreasing age (at death) of the index patient

Patient (M/F)/origin	Mutations	Consanguinity	Age (death)	Age at onset	Pregnancy	Microcephaly (age: SD[a])	Feeding difficulties (age tube-fed)	Respiratory failure (age)	Motor development (age)	Speech development	Ophthalmologic findings	Neurologic findings	EMG/ muscle biopsy neurogenic	ENG normal	Brain imaging
1-1 (F)/ Turkey[b]	c.395A>C, p.D132A (homozygous)	+	20 y	3 mo	Normal	+ (5 y 3 mo: −2.5; 15 y: −3.5)	+	-	Head control, sat, crawled (5–8 y), few steps with support (6–8 y)	Single words	Strabismus	Spasticity, dystonia, increased reflexes, weakness and hypotonia from 8 y	+/+	+	PCH
1-2 (M)/ Turkey[b]	c.395A>C, p.D132A (homozygous)	+	16 y	3 mo	Normal	+ (7 mo: −1; 2 y: −2)	-	-	Head control	None	Strabismus	Spasticity, myoclonus, trunk hypotonia, increased reflexes	+/+	+	PCH
2-1 (F)/ Germany[7]	c.395A>C, p.D132A (homozygous)	-	18 y	<6 mo	Normal	− (4 y: 0, 18 y: 0)	+ (7 y)	+ (13 y)	?, Abduction pant until 6 mo (hip dysplasia)	None	Strabismus, nystagmus	Hypotonia, myoclonus, and absence epilepsy from 8 mo	+/+	+	CH
3-1 (F)/ Germany	c.395A>C, p.D132A (homozygous)	-	(5 y 4 mo)	3–6 mo	Normal	− (1 y 6 mo: 0; 4 y: −0.5)	+ (3 y 8 mo)	-	Head control, turned around (7–10 mo)	None	Strabismus, nystagmus	Hypotonia	+/+	+	PCH
4-1 (F)/ Norway[7]	c.395A>C, p.D132A; c.226dupG; p.D76fs	+ Remote	(2 y 3 mo)	Birth	Normal	+ (1 y 10 mo: −3.5)	-	-	None	None	Normal	Hypotonia	+/NA	NA	Cerebellar atrophy
4-2 (M)/ Norway[7]	c.395A>C, p.D132A; c.226dupG; p.D76fs	+ Remote	(4 mo)	Birth	Normal	NA	-	+ (4 mo)	None	None	Normal	Hypotonia	+/+	NA	PCH, mega cisterna magna
5-1 (F)/ Germany[7]	c.395A>C, p.D132A, c.2T>C	-	(2 y 2 mo)	Birth	Normal	− (2 y: +1)	+ (2 wk)	+ (2 y 2 mo)	None	None	Nystagmus	Hypotonia, infantile spasms <1 y	+/+	NA	CH, retrocerebellar cyst
5-2 (F)/ Germany[7]	c.395A>C, p.D132A, c.2T>C	-	(1 y 2 mo)	Birth	Normal	− (3 mo: 0)	NA	NA	None	None	Nystagmus	Hypotonia, infantile spasms from 4 mo	+/+	+	CH, retrocerebellar cyst
6-1 (F)/ Australia[c18]	c.395A>C, p.D132A; c.475-12A>G	-	(2 y 1 mo)	Birth	Normal	+ (2 y: −2)	+ (8 mo)	+ (2 y)	None	None	Normal	Hypotonia	+/NA	+	CH, retrocerebellar cyst
7-1 (M)/ Serbia	c.92G>C, p.G31A (homozygous)	-	(1 y 3 mo)	Birth	Normal	+ (4 mo: −2.5)	+ (2 wk)	+ (5 mo)	None	None	Normal	Hypotonia	+/+	NA	PCH, mega cisterna magna
8-1 (F)/ UK[25]	c.395A>C, p.D132A; c.155delC, p.52fs	-	(9 mo)	Birth	Normal	+	+ (1 mo)	+ (9 mo)	None	None	Normal	Hypotonia	+/+	NA	CH

Continued

Table 1 Continued

Patient (M/F)/origin	Mutations	Consanguinity	Age (death)	Age at onset	Pregnancy	Microcephaly (age: SD[a])	Feeding difficulties (age tube-fed)	Respiratory failure (age)	Motor development (age)	Speech development	Ophthalmologic findings	Neurologic findings	EMG/ muscle biopsy neurogenic	ENG normal	Brain imaging
8-2 (F)/ UK[25]	c.395A>C, p.D132A; c.155delC, p.52fs	-	(6 mo)	Birth	Normal	NA	+ (4 mo)	+ (6 mo)	None	None	Normal	Hypotonia	NA/+	NA	CH
9-1 (M)/ Czech Republic[d]	c.712T>C, p.W238R; c.92G>C, p.G31A	-	(8 mo)	Birth	Normal	NA	Birth	Birth	None	None	Normal	Hypotonia	+/NA	NA	PCH, mega cisterna magna
9-2 (F)/ Czech Republic[d]	c.712T>C, p.W238R; c.92G>C, p.G31A	-	(7 mo)	Prenatal	CH on ultrasound	NA	Birth	Birth	None	None	Normal	Hypotonia	NA/NA	NA	PCH
10-1 (M)/ Germany[7]	c.395A>C, p.D132A; c.551delG, p.184fs	-	(8 mo)	Prenatal	PH, fetal akinesia	NA	Birth	Birth	None	None	NA	Hypotonia, AMC, bone fractures	NA/+	NA	PCH

Abbreviations: + = present; – = absent; AMC = arthrogryposis multiplex congenita; CH = cerebellar hypoplasia; ENG = electroneurogram; NA = not available; PCH = pontocerebellar hypoplasia; PH = polyhydramnios.

[a] SD for head circumference according to WHO standards (http://www.who.int/childgrowth/standards/hc_for_age/en/index.html).

[b] Family 3 of reference 6.

[c] Family 6 of reference 6.

[d] Family 4 of reference 6.

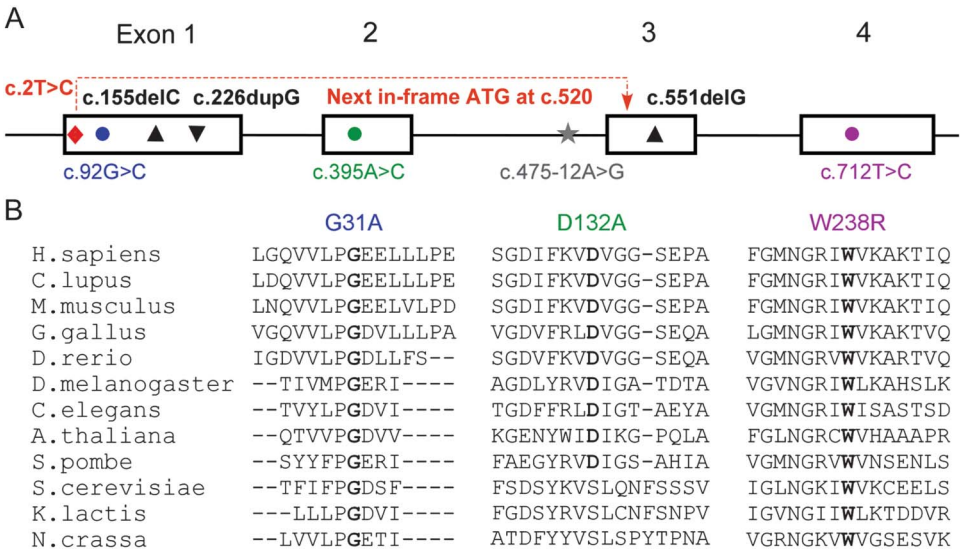
Families 4–6 (moderate PCH1): Patients showed a postnatal onset of rapidly progressive cerebellar volume loss and achieved no psychomotor developmental milestones. Respiratory failure was reported in late stages of the disease and was only rarely a cause of death, which occurred at age 1–3 years. Moderate PCH1 was correlated with c.395A>C in combination with an insertion mutation c.226dupG, a splice site mutation c.475-12A>G, and a mutation disrupting the start codon c.2T>C.

Families 7–10 (severe PCH1): Patients had a prenatal or congenital onset of cerebellar, pontine, and midbrain degeneration. They showed severe hypotonia and tongue fasciculations, while oculomotor dysfunction or other signs of cerebellar ataxia were not recorded. Most patients had postnatal respiratory failure leading to early death within the first year even under constant ventilation. Severe PCH1 was correlated with missense mutations distinct from c.395A>C (families 7 and 9) or with c.395A>C in combination with deletion mutations in families 8 and 10. Patient 7-1 with a homozygous missense mutation c.92G>C survived under permanent ventilation from the 5th month into the second year of life but had a clinical severity similar to family 8.

Brain imaging in mild PCH1 showed hypoplasia of the cerebellar hemispheres with preserved folia and a small but well-structured pons (figure 3, A and B). In moderate PCH1, larger structural defects in the posterior fossa like mega cisterna and retrocerebellar cyst were seen (figure 3, C and D). In severe PCH1, a more pronounced reduction of all cerebellar structures including pons and brainstem was documented (figure 3E). Progressive neurodegeneration results in a marked reduction of cerebellum, pons, and brainstem in severe and late stages of the disease (figure e-1, a and b). In patient 9-2, who was thoroughly examined during intrauterine development after the death of an affected sibling (patient 9-1), initial neuroimaging showed normal findings in the 22nd week of gestation, while visible cerebellar hypoplasia was documented in the 30th week of gestation (figure e-1, c–e).

In our cohort there were 23 patients of 17 families who did not show *EXOSC3* mutations (table e-2). Prenatal onset, congenital respiratory and feeding difficulties, joint contractures, and early death were rarely recorded in *EXOSC3* mutations but were typical features among the mutation-negative patients (table 2). Abnormal oculomotor function (strabismus, nystagmus, oculomotor apraxia) was commonly observed in patients surviving beyond infancy (>1 year) and was therefore more frequently documented in the mutation-positive group (29% vs 17%). In young infants who died early, it might have been difficult to detect oculomotor

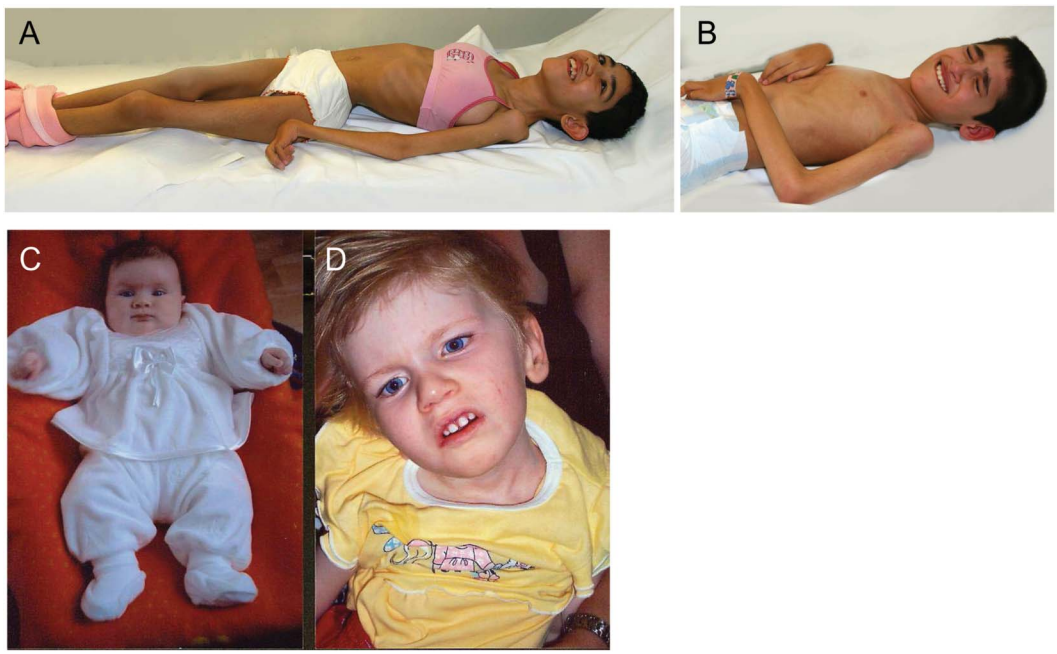
Figure 1 *EXOSC3* gene structure and mutations in PCH1



(A) Newly identified (bold) and previously reported mutations in 15 of 27 unrelated families with pontocerebellar hypoplasia with spinal muscular atrophy (PCH1) in *EXOSC3* spanning 4 exons (open boxes). ♦, shown in red, is a mutation in the start code that shifts the translation initiation 520 bases downstream, thus missing the first 174 amino acid residues. ●, missense mutations; ▲, deletion mutation; ▼, insertion mutation; star, splice site mutation. (B) Alignment of protein sequences encoded by *EXOSC3* in human and orthologous genes in other eukaryotes, showing alteration of evolutionarily conserved amino acid residues in bold; the altered amino acid residue and the corresponding missense mutation are represented by matching color.

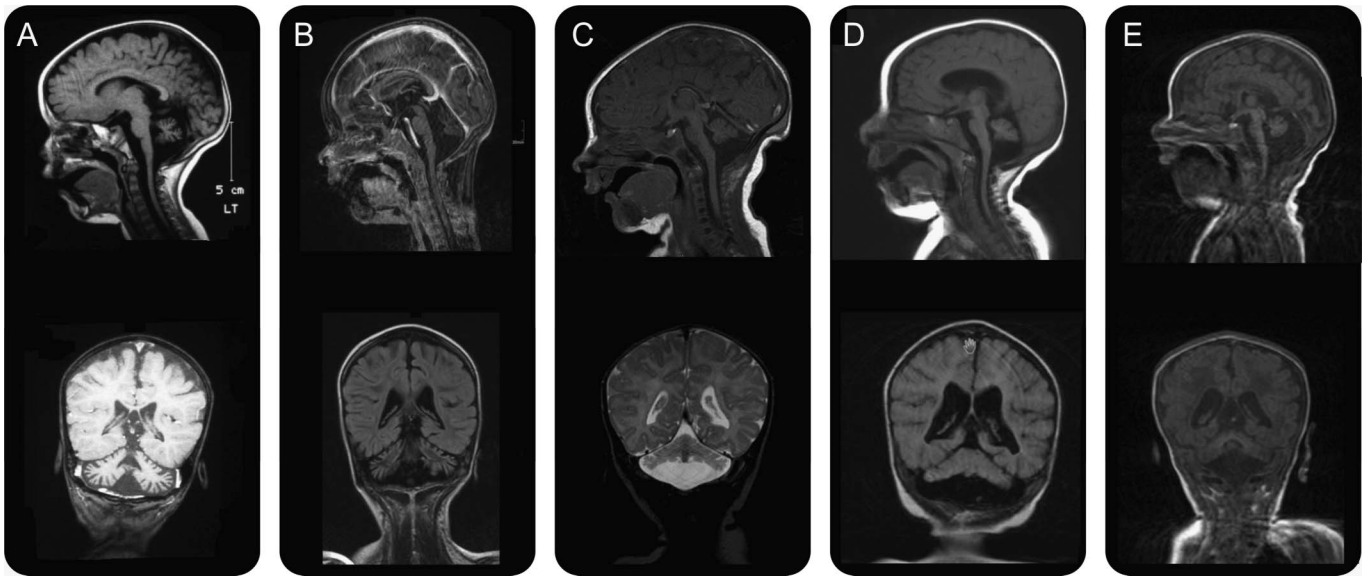
dysfunction. There was no evidence of a peripheral neuropathy in the mutation-positive group but in some patients without mutations. Motor nerve conduction velocities were markedly delayed (as low as 6 to 9 m/s in patient 17-1) or action potentials could not be elicited in motor or sensory nerves (patients 16-1, 17-2, 20-1). Sural nerve biopsy was only performed in patient 17-1 and did not give evidence of demyelination but showed loss of large myelinated axons as outlined previously.[7] Brain imaging and autopsy findings revealed

Figure 2 Clinical impression of the longest surviving patients in this cohort



(A) Patient 1-1 at 15 years. (B) Patient 1-2 at 11 years, demonstrating microcephaly, inability to direct gaze, and profound muscle atrophy. Patient 3-1 at different ages: (C) 3 months with normal appearance, (D) 4 years with generalized muscular hypotonia, no head control, and inability to track.

Figure 3 Brain imaging in *EXOSC3* mutation-positive patients



MRI of the brain in 5 patients with different subtypes of pontocerebellar hypoplasia with spinal muscular atrophy (PCH1) and confirmed *EXOSC3* mutations, mild PCH1 (A, patient 2-1, age 1 7/12 years; B, patient 3-1, age 4 4/12 years), moderate PCH1 (C, patient 6-1, age 2 weeks; D, patient 7-1, age 4.5 months), and severe PCH1 (E, patient 9-1, age 2 weeks). In A and B, there is cerebellar volume loss but all cerebellar lobules are present and the brainstem is relatively preserved. In C and D, MRI demonstrated larger structural defects in the posterior fossa compared to A and B. In E, there is marked volume loss of the cerebellar structures as well as the pons and brainstem. Top row: sagittal sections; bottom row: coronal sections.

Table 2 Clinical features of 15 patients with *EXOSC3* gene mutations in comparison to 23 patients without mutations[a]

Item	Patients with mutations (n = 15)	Patients without mutations (n = 23)
Median age at onset (range)	Birth (prenatal–6 mo)	Prenatal (prenatal–12 mo)
Median age at death (range)	9 mo (4 mo–5 y)	3 mo (2 h–4 y)
Longest survival, y	20	7
Pregnancy abnormal	2/15 (13)	11/23 (48)
Polyhydramnios	1/15 (7)	11/23 (48)
Fetal akinesia	1/15 (7)	1/23 (4)
Joint contractures	—	1/23 (4)
Arthrogryposis multiplex congenita	2/15 (13)	7/23 (30)
Progressive microcephaly	8/12 (67)	3/13 (23)
Failure to thrive from birth	3/14 (21)	12/19 (63)
Respiratory failure from birth	3/14 (21)	15/21 (71)
Absent motor development	11/14 (79)	19/23 (83)
Absent speech development	14/15 (93)	23/23 (100)
Nystagmus	4/14 (29)	0/12 (0)
Strabismus	4/14 (29)	2/12 (17)
Muscle tone abnormal	15/15 (100)	23/23 (100)
Hypotonia	13/15 (87)	21/23 (91)
Increased muscle tone/spasticity	2/15 (13)	2/23 (9)
Epilepsy	3/15 (20)	5/23 (22)

[a] Values are n (%).

similar pontocerebellar volume loss and nerve cell depletion in the mutation-positive and the mutation-negative group.

DISCUSSION Our PCH1 series demonstrates variability of clinical severity and broad spectrum of cerebellar and midbrain structural defects. PCH1 results from a profound underdevelopment of the neocerebellum and its adjacent structures, and also includes motor neuron degeneration in the spinal cord.[3] The hypoplastic cerebellum has a reduced number of folia, while the cerebellar shape is not altered. The developmental disturbance affects mainly the phylogenetically new parts of the cerebellum (i.e., the neocerebellum) with relative sparing of the vermis and the flocculonodular lobules. Since cerebellar development starts at about 5 gestational weeks and migration of neurons is not completed until 15 postnatal months,[9] this can explain why some patients with PCH1 display normal supratentorial and infratentorial brain structures by imaging methods at birth followed by a rapid loss of cerebellar hemispheres within weeks or months. Given the identical germinal zones, the association of pontine and cerebellar hypoplasia is common.[10] As such PCH is characterized by neuronal loss in the ventral pons, inferior olive, and cerebellum, with onset in utero but continuing after birth.[11] The cerebellum plays crucial roles in sensory integration and motor planning as well in higher cognitive processing such as attention and language acquisition.[12] This explains the profound psychomotor retardation in patients with PCH.

All genes so far identified as mutated in the other PCH subtypes (*TSEN*, *RARS2*, *SEPSECS*) are involved in tRNA processing,[3] and that maturing neurons are especially vulnerable for defects in protein synthesis has been hypothesized to account for the clinical phenotype. The clinical and pathoanatomic distinction among the different PCH subtypes can also be difficult, and in 1 patient with PCH1 mutations in *TSEN* were reported,[5] further complicating a reasonable classification of PCH. Of note, a child from a consanguineous family of Ashkenazi Jewish origin was diagnosed with PCH1 and had a homozygous nonsense mutation identified in the *VRK1* gene, which encodes a serine/threonine kinase that phosphorylates p53 and CREB and is essential for nuclear envelope formation.[4] The clinical presentation of this child and siblings (who were not genetically characterized) differed from PCH in that these patients had severe microcephaly at birth while mental development was normal or only mildly impaired. Ataxia and muscle weakness occurred later in the disease course and the onset of cerebellar degeneration remained unclear.[4]

The *EXOSC3* gene encodes a core component of the RNA exosomes that process and degrade RNA and thereby regulate the activity of gene expression. How mutations in *EXOSC3* lead to PCH1 has not yet been elucidated. As shown by the morpholino knockdown experiments in zebrafish, the missense mutations appear to be hypomorphic.[6] No patient has been observed to harbor biallelic null mutations; it is likely that homozygous null mutations of *EXOSC3* are incompatible with life and result in early embryologic lethality.[6] Patients harboring biallelic *EXOSC3* mutations demonstrate profound loss of cerebellar and spinal motor neurons while heterozygous mutation carriers are asymptomatic, suggesting that a minimum expression of EXOSC3 is essential to the development and survival of cerebellar and motor neurons.[6]

By comparing the clinical and pathoanatomic features of patients with PCH1 with and without *EXOSC3* mutations (table 2) and by surveying the literature, we make the following observations.

Birth measurements are normal in mutation-positive patients, i.e., primary microcephaly at birth as described in some patients[4,5,13] implies another genetic cause in PCH1. Two-thirds of patients with *EXOSC3* mutations develop progressive microcephaly ranging from mild to moderate (−2.0 to −3.5 SD). Psychomotor retardation is profound; no *EXOSC3* mutation-positive patient had motor or speech development above the level of a few-months-old infant. Initial normal psychomotor development or normal cognitive functions followed by cerebellar and spinal muscular atrophy as described in a handful of patients in the literature[4,6,14,15] are likely not associated with EXOSC3 dysfunction. Muscle tone and strength is generally reduced since birth, except in family 1 with initial spasticity followed by hypotonia and flaccid pareses later in the disease course. Thus, it may be useful to search for *EXOSC3* mutations in other PCH subtypes which are characterized by increased muscle tone and choreiform or dystonic movements. Epileptic seizures were uncommon in both the mutation-positive and -negative groups and did not follow a specific pattern. Abnormal oculomotor function (strabismus, nystagmus, oculomotor apraxia) was commonly observed in patients surviving beyond infancy. Additional organ involvement such as gonadal dysgenesis,[16] also seen in patient 23-2 (table e-2[8]), retinal degeneration or optic nerve atrophy,[5] hepatic fibrosis, and cystic kidney disease[17] are also not yet reported in patients with *EXOSC3* mutations. Mesangiocapillary glomeronephritis was diagnosed in a mutation-positive Australian patient[6] (patient 4 in reference 18). Since this is the only reported case to date, the kidney disease might have been a coincidental finding. EXOSC3-associated PCH1 shows a variable lifespan, ranging from a few months to adulthood. In particular, patients with homozygous c.395A>C mutations have a chronic course with minimal involvement of the pons, as noted in previously described families.[6] The clinical features

associated with "classical" PCH1 with prenatal onset, congenital respiratory and feeding difficulties, arthrogryposis multiplex congenita, and neonatal death are infrequent in patients with *EXOSC3* mutations but were most prevalent among the mutation-negative patients and also in the literature.[1,19–24]

Electromyographic examinations give evidence of neurogenic changes in the muscle in all patients based on the diagnostic entry criteria. Nerve conduction studies were normal in all patients with *EXOSC3* mutations and in the majority of patients without. In few patients without mutations motor nerve conduction velocities were delayed or action potentials could not be elicited in motor or sensory nerves. While the absence of an electric response by nerve conduction studies might be of technical nature in small children, markedly reduced conduction velocities, as also reported in some patients from the literature,[4,18,21,23,24] or reduction of demyelinated nerve fibers in sural nerve biopsies[19,24] might point toward distinct genetic entities.

Neuroimaging in *EXOSC3* mutation-positive patients shows a marked reduction of the cerebellar hemispheres giving rise to an enlarged fourth ventricle, sometimes with the impression of a mega cisterna magna, and a retrocerebellar cyst. In severe cases and as the disease progresses, pons and brainstem reduction become visible on MRI. Initially the cortical structures are normal, thus other brain malformations[18] or marked brain atrophy from the beginning giving rise to enlarged ventricles as seen in some nonmutated families suggest other disease causes.

Autopsy findings in patients with and without *EXOSC3* mutations did not reveal different cerebellar, brainstem, and spinal cord findings on the basis of the medical records made available. Generalized brain atrophy or white matter changes point toward entities involving more cerebral structures than seen in *EXOSC3* mutation-positive patients.

In this large series of PCH1 families, mutations in *EXOSC3* are an important cause of the disease and account for approximately one-third of the families. Although still affected with progressive neurogenic muscular atrophy and global developmental delay with marked cerebellar atrophy, homozygous c.395A>C mutations in *EXOSC3* are associated with relative sparing of the brainstem and survival beyond early childhood, with several individuals reaching teenage and early adulthood. The clinical manifestations in association with other *EXOSC3* mutations were more severe with early mortality, suggesting that these mutations may exert a more detrimental impact on the gene product than c.395A>C. The yield for mutation screening was low in those with primary microcephaly, primary hypoventilation, normal or mildly impaired psychomotor development, abnormal nerve conduction studies, or histologic evidence of peripheral neuropathy, and widespread neuroimaging abnormalities beyond the posterior fossa. Further *EXOSC3* mutation analyses may be considered in other PCH types or patients with milder olivopontocerebellar atrophy with or without anterior horn cell disease. Molecular genetic diagnosis is now possible for a large subgroup of patients with PCH1. Counseling and prenatal or preimplantation diagnosis can be offered in genetically characterized families.

AUTHOR CONTRIBUTIONS

Sabine Rudnik-Schöneborn: study concept and design, analysis and interpretation. Jan Senderek: analysis and interpretation, critical revision of manuscript. Joanna C. Jen: analysis and interpretation, critical revision of the manuscript. Gunnar Houge: acquisition of data. Pavel Seeman: acquisition of data, critical revision of manuscript. Alena Puchmajerová: acquisition of data. Luitgard Graul-Neumann: acquisition of data. Ulrich Seidel: acquisition of data. Rudolf Korinthenberg: acquisition of data. Janbernd Kirschner: acquisition of data. Jürgen Seeger: acquisition of data. Monique M. Ryan: acquisition of data. Francesco Muntoni: acquisition of data. Maja Steinlin: acquisition of data. Laszlo Sztriha: acquisition of data. Jaume Colomer: acquisition of data. Christoph Hübner: acquisition of data. Knut Brockmann: acquisition of data. Lionel Van Maldergem: acquisition of data. Manuel Schiff: acquisition of data. Andreas Holzinger: acquisition of data. Peter Barth: acquisition of data. William Reardon: acquisition of data. Michael Yourshaw: acquisition of data. Stanley F. Nelson: acquisition of data. Thomas Eggermann: acquisition of data. Klaus Zerres: study supervision.

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DISCLOSURE

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