Minireview

Childhood ataxia with CNS hypomyelination/vanishing white matter disease—A common leukodystrophy caused by abnormal control of protein synthesis

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Abstract

Mutations in eukaryotic initiation factor 2B (eIF2B) cause one of the most common leukodystrophies, childhood ataxia with CNS hypomyelination/vanishing white matter disease or CACH/VWM. Patients may develop a wide spectrum of neurological abnormalities from prenatal-onset white matter disease to juvenile or adult-onset ataxia and dementia, sometimes with ovarian insufficiency. The pattern of diffuse white matter abnormalities on MRI of the head is often diagnostic. Neuropathological abnormalities indicate a unique and selective disruption of oligodendrocytes and astrocytes with sparing of neurons. Marked decrease of asialo-transferrin in cerebrospinal fluid is the only biochemical abnormality identified thus far. Eukaryotic translation initiation factor 2B (eIF2B) mutations cause a decrease in guanine nucleotide exchange activity on eIF2-GDP, resulting in increased susceptibility to stress and enhanced ATF4 expression during endoplasmic reticulum stress. eIF2B mutations are speculated to lead to increased susceptibility to various physiological stress conditions. Future research will be directed towards understanding why abnormal control of protein translation predominantly affects brain glial cells.
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Keywords: Leukodystrophy; Protein translation; Initiation of protein translation; Oligodendrocyte; Astrocyte; Glia; Brain; ER stress; Myelin

Introduction

Leukodystrophies are neurogenetic disorders that predominantly affect the white matter of the brain. Since the etiology cannot be found in a substantial proportion of patients with leukodystrophies, an effort has been made in recent years to define specific leukodystrophy syndromes and identify their cause [1]. As a result, a number of new leukodystrophy syndromes have recently been described [2,3]. The etiology of some of them has been discovered and their molecular defects contribute to our understanding of myelin development and mainte-

nance [4]. Childhood ataxia with CNS hypomyelination (CACH) was first identified in 1992 [5]. Similar patients later described as vanishing white matter disease (VWM) or myelinopathia centralis diffusa [6–8]. Despite extensive pathological and biochemical analysis [9], the etiology of this genetically heterogeneous disease was only found, thanks to astute use of molecular genetics in a population of a limited geographical region [10,11]. The identification of mutations in any one of the subunits of eukaryotic translation initiation factor 2B (eIF2B) as the cause of CACH/WWM propelled this obscure disease into the center of molecular and cellular biology. As often happens, the identification of the genetic defect of CACH/VWM led to the recognition of a wider clinical spectrum.

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Clinical manifestations

CACH/VWM is a panethnic autosomal recessive disease [10,12]. An autosomal dominant form of the disease of yet unknown cause has recently been described [13]. Early development may be initially normal, whereas other patients have speech and cognitive delay [7,9,14]. New-onset ataxia is the most common initial symptom between ages 1 and 5 years [9,14]. Some patients develop coma or dysmetric tremor [7,15]. These can be spontaneous or follow a mild head trauma or febrile illness and apparently even acute fright [16]. Subsequently, deterioration is generally progressive with increasing difficulty in walking, cerebellar tremor and dysmetria, spasticity, hyperreflexia, dysarthria, and seizures [14,17]. At any phase of the illness, patients may remain stable for years. Head circumference has been normal in almost all patients [9,14]. Dysphagia and optic atrophy develop late in the disease; the peripheral nervous system is usually unaffected [9]. Death typically occurs during the first or second decade of life [14]. Some patients, especially those who develop symptoms after 5 years of age, have a more slowly progressive spastic diplegia, relative sparing of cognitive ability, and a likely longer survival [18–20]. Other forms of CACH/VWM exist including congenital forms with manifestations in organs besides the brain [21]. The abnormalities found in these infants included oligohydramnios, intrauterine growth retardation, cataracts, pancreatitis, hepatosplenomegaly, hypoplasia of the kidneys, and ovarian dysgenesis [21]. A rapidly fatal infantile form of this syndrome was described; it was found to be allelic to the more common form of CACH/VWM with the 3q27 locus [22]. A subacute variant occurs in the indigenous population of northern Canada [12]. These patients have an onset of neurologic deterioration in the first 6 months of life and die by 2 years of age [23,24]. On the other end of the clinical spectrum are patients with a slowly progressive neurologic disorder, onset after age 5 years that is often associated with ovarian insufficiency (dysgenesis) that we termed ovarioleukodystrophy syndrome [25,26]. Adult-onset disease is not rare, particularly with the most common R113H EI2B5 mutation [14,27].

Laboratory evaluation

The most important ancillary test in a patient suspected of having a leukodystrophy is an MRI of the head using at a minimum T1- and T2-weighted as well as fluid attenuated inversion recovery (FLAIR) images [7,9,18,28]. The MRI in CACH/VWM is distinctive with diffusely decreased signal in the white matter on T1-weighted images MRI and corresponding increase signal intensity in T2-weighted images that usually involves the subcortical white matter (Figs. 1A and B) [7,9]. This MRI abnormality is present even in asymptomatic children [14]. Cystic breakdown of the white matter or cavitation is best seen on the proton density or FLAIR sequences (Figs. 1C and D)

[7,18]. There is no gadolinium enhancement of the lesions on MRI. These FLAIR abnormalities may not be present in mild, relatively nonprogressive cases. Unlike many other leukodystrophies, in typical CACH/VWM patients cortical atrophy and ventricular dilation are typically absent, even in rather advanced cases [9]. On the other hand, in adults with long-standing disease, including in ovarioleukodystrophy patients, lateral ventricles may become dilated over time, and subcortical white matter may be appear normal [26]. Prenatal-onset CACH/VWM may have a wider variety of abnormalities of the white matter with marked dilation of the lateral ventricles [21]. Proton MR spectroscopy typical shows global reduction of all metabolites occasionally combined with a lactate peak [6,7,9,15]. These abnormalities are thought to be related to rarefaction of the white matter (Fig. 2A) [7,15].

Routine studies of blood, urine, and cerebrospinal fluid (CSF) are normal [9]. The first biochemical abnormality identified in CACH/VWM patients was found recently using a variety of proteomic techniques [29]. It consisted of a marked reduction in asialo-transferrin in the CSF, with normal transferrin profile in the blood [29]. This form of transferrin is thought to be produced exclusively in the brain, possibly by olidgodendrocytes and astrocytes, and its reduction may reflect disturbance in the function of these cells [29–32]. If this finding is confirmed, it will be useful as a diagnostic tool and possibly also as a marker for therapeutic trials. Biochemical analysis of brain material shows reduction of myelin proteins and lipids, thus confirming the impression of hypomyelination but these abnormalities are nonspecific [9]. Proton-decoupled phosphorous magnetic resonance spectroscopy studies indicated that the eIF2B defect in CACH/VWM causes an abnormality of myelin membrane synthesis or myelin membrane transport in vivo [33]. The authors found a decrease in glycerophosphorylethanolamine and an increase in phosphorylethanolamine that suggested a defect in plasmalogen metabolism [33]. It would be important to determine how this abnormality directly relates to the basic cellular defect in this disease.

Neuropathology

Gross examination of the brain reveals normal consistency of the cortical gray matter in marked contrast to the white matter of the centrum semiovale that is softened, atrophic, and gelatinous [7,34]. Light microscopy demonstrates rarefaction of the white matter with relative sparing of axons and subcortical U-fibers (Fig. 2A) [7,9,18,34,35]. There is moderate to severe vacuolation of the white matter. The hallmark of CACH/VWM is the presence of foamy oligodendrocytes, not seen in any other leukodystrophy [34]. Increased numbers of oligodendrocytes were described by two groups [35,36]. The "foamy oligodendrocytes of CACH/VWM stain with a combination of Alcian blue/periodic acid Schiff suggesting an abnormality of glycosylation (Fig. 2B) [12]. However,
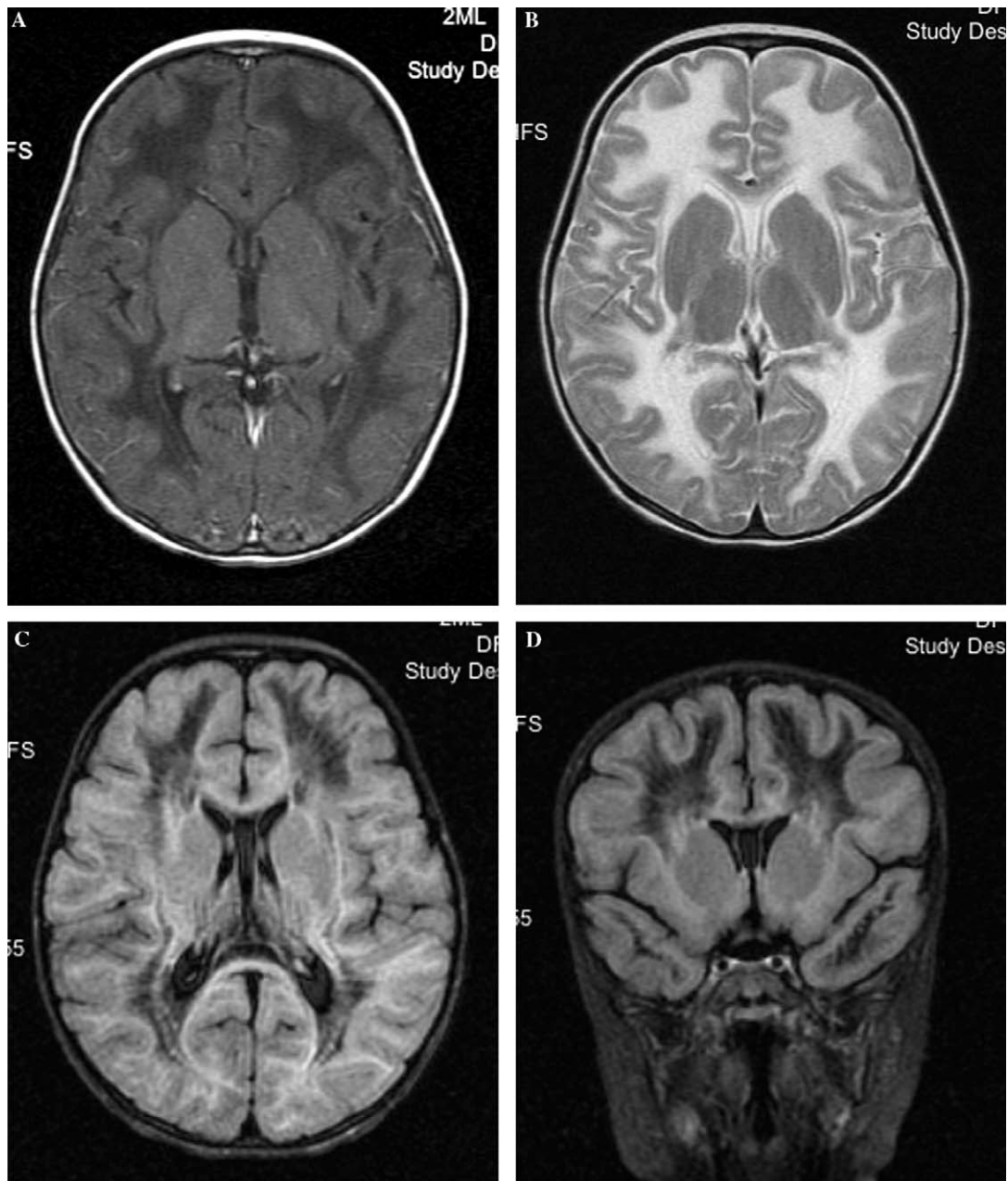
Fig. 1. Typical MRI of a 2-year-old patient with CACH/VWM showing the white matter to be diffusely hypointense on T1-weighted images (A) and hyperintense on T2-weight image (B). Note the small lateral ventricles and absence of cortical atrophy. Axial (C) and coronal (D) FLAIR images in the same patient show widespread secondary breakdown of the abnormal white matter that leaves visible bundles of nerve fibers crossing the subcortical region.

such abnormal glycoforms were not found [37]. Ultrastructure analysis showed abnormal foamy oligodendroglial cells with abundant cytoplasm-containing membranous structures and numerically increased and morphologically abnormal mitochondria [34]. These oligodendrocytes apparently have increased rate of apoptosis [8,35]. Gliosis with abnormally shaped coarse astrocytes is present and glial fibrillary acidic protein-positive glial fibers were seen aligning capillaries (Fig. 2C) [9,18,25]. In the severe forms, there is a reduction of the number of astrocytes and possible also astrocyte progenitors, but not of oligodendrocyte progenitors [22,38]. Sural nerve pathology is almost always normal [9]. In one patient who died at 15 years of age, there was significant neuropathic changes (Fig. 2D). Based on histopathological and biochemical criteria, CACH/VWM appears to be a hypomyelinating glial disease affecting predominantly oligodendrocytes and astroctyes with relative preservation of axons [9,22,34,38]. As in almost all myelin disorders, the clinical deterioration in patients with this leukodystrophy is directly linked to secondary axonal loss seen on MRI images and neuropathological materials [39].
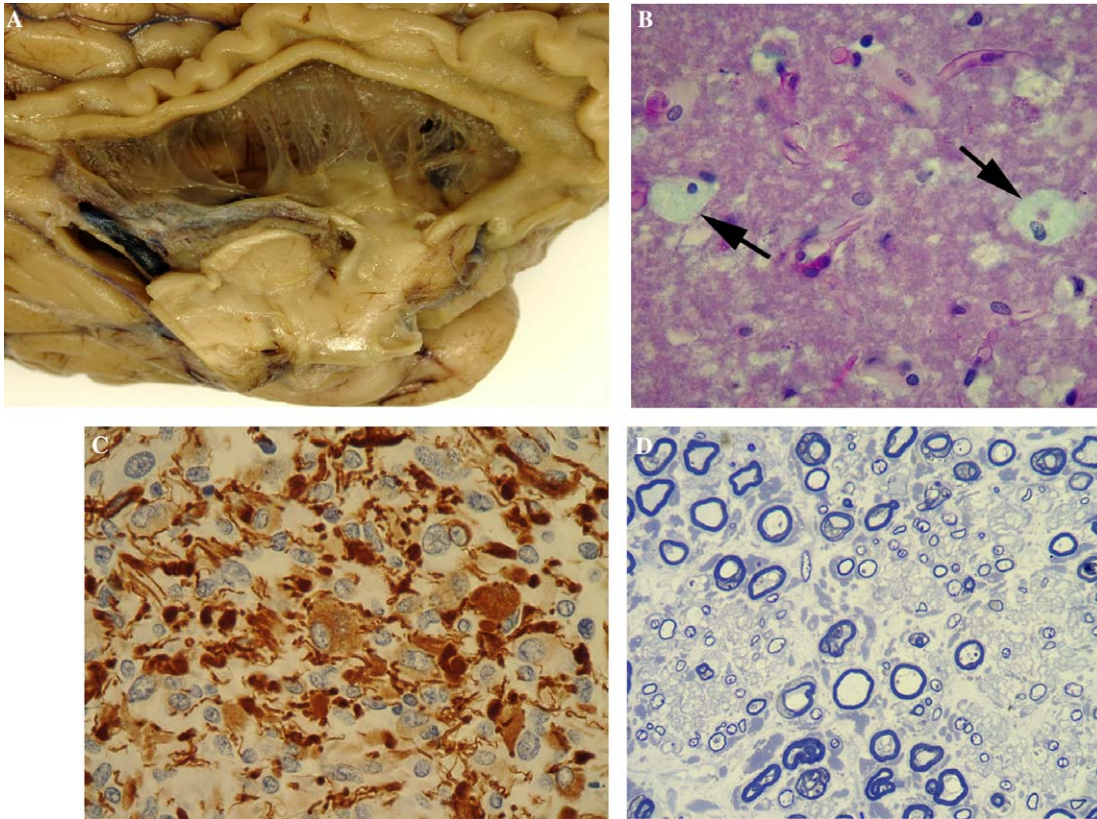
Fig. 2. Characteristic pathological finding in CACH/VWM. (A) Gross pathology: although the cortex looks largely intact, all is left of the white matter is thin and fragile strands tissue. (B) Alcian blue-periodic acid Schiff staining shows large "foamy" oligodendrocytes (arrows) X100. (C) GFAP staining showing grossly coarse and atypical astrocytes X100. (D) Sural nerve of a 15-year-old male with CACH/VWM showing mild loss of large myelinated axons with a greater loss of small myelinated axons. The unmyelinated axons appear swollen with greater than normal cross sectional diameter. There are scattered severely, atrophic axons and occasional degenerating axons. All axons had a thinner than normal myelin sheath but 'onion bulbs' were not identified. X100. All panels are courtesy of Kondi Wong, M.D.

Etiology

Despite the early evidence that this leukodystrophy syndrome is genetically heterogeneous, linkage to a region at chromosome 3q27 was found by recognition of a geographic clustering in the eastern part of the Netherlands of a small number of families, some consanguineous [10]. Subsequently, the gene in this region was identified as eIF2B5, which codes for the ε-subunit of eIF2B [11]. The same group later found that this disorder is indeed genetically heterogeneous and that it can be caused by mutations in any of the five subunits of eIF2B [40]. Mutations in EIF2B1, EIF2B2, EIF2B3, EIF2B4, and EIF2B5 have been found in about 90% of individuals with clinical and MRI presentation of CACH/VWM by use of mutation scanning or sequence analysis of the coding regions and splice sites [14]. Over 80 eIF2B mutations have been described (The Human Gene Mutation Database at the Institute of Medical Genetics in Cardiff, Wales: http://www.hgmd.cf.ac.uk/hgmd0.html) [41]. About 90% of mutations are missense [14,42]. Affected individuals are homozygotes or compound heterozygotes for mutations within the same gene [14,40]. A nonsense mutation on one allele is always associated with a missense mutation in the other allele [14,40]. Mutations have been found in affected individuals of all ethnic origins [42]. Sixty-seven percent of eIF2B mutations occur in the ε-subunit _(eIF2B5)_, 16% in β-subunit _(eIF2B2)_, 12% in δ-subunit _(eIF2B4)_, 4% in γ-subunit _(eIF2B3)_, and no more than 1% in the α-subunit _(eIF2B1)_ [14,40,42]. A limited genotype phenotype correlation exists [14,27]. Certain _EIF2B5_ homozygous mutations, such as R113H, never gives rise to the infantile type, and in the homozygous state is to our experience always mild with slow progression into adulthood [14,27]. In the homozygous state, certain _EIF2B5_ mutations, such as V309L and R195H are predictably associated with a severe disease [14]. Patients with mild mutations have a significantly later onset of CACH/VWM compared to those with all other mutations [14]. On the other hand, the phenotype of individuals homozygous for the T91A mutation in _EIF2B5_, may vary from childhood onset to adults with no symptoms [11,42]. In a small proportion of patients with typical clinical and radiological characteristics of CACH/VWM, no eIF2B mutations can be found, including one of the original 4 patients [9].

Pathogenesis

eIF2B is a 5-subunits guanine nucleotide exchange factor (GEF) that exchanges GDP for GTP on the 3-subunits translation initiation factor eIF2, to form eIF2-GTP complex [43]. The active eIF2-GTP binds aminoacylated initiator methionyl-tRNA (Met-tRNAi^{Met}), forming eIF2·GTP·Met-tRNAi^{Met} termed ternary complex (Fig. 3), which is then loaded onto the 40S ribosomal subunit prior to each round of translation initiation step. The formation of ternary complex is a central regulation point of global translation rate according to the needs of the cell [44]. This important regulatory mechanism of protein synthesis is based on phosphorylation of Ser 51 of the α-subunit of eIF2 under stress conditions [43]. Phosphorylation of eIF2α is catalyzed by four protein kinases GCN2, PKR, PERK, and HRI [44]. Physiological conditions which result in eIF2α phosphorylation through activation of one of these kinases include amino acid deprivation, accumulation of unfolded proteins in the ER, virus infection, heat shock, changes in intracellular calcium, UV irradiation, oxidative stress, and iron deficiency [44]. As a result, the interaction of the phosphorylated
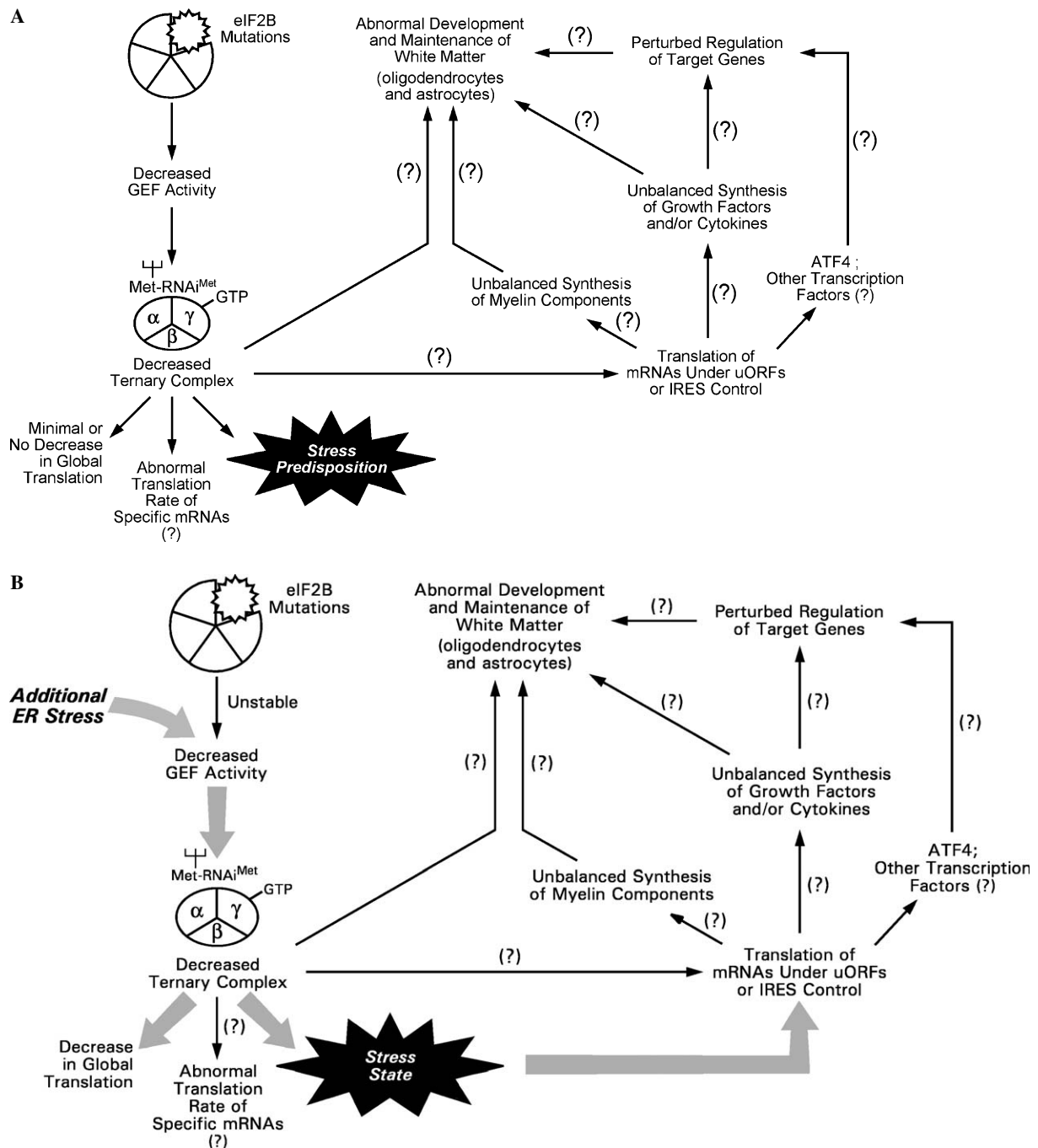

Fig. 3. Hypothetical pathogenetic mechanism of disease caused by eIF2B mutations at rest (A) and during ER stress (B).

eIF2 with eIF2B becomes inhibitory, and given that eIF2B is less abundant than eIF2, even low levels of phosphorylated eIF2 effectively inhibit eIF2B activity resulting in decreased translation initiation by up to 90% and therefore a significant reduction in protein synthesis in the cell. However, under conditions of low ternary complex level, the translation of some mRNAs is activated due to specific regulatory sequences within their 5′-untranslated regions (5′UTRs), specifically designed to confer translational advantage under stress conditions. Short upstream open reading frames (uORF) within the 5′UTR are one example. Under normal conditions, the uORF are normally translated and repress initiation at the major downstream ORF. However, under conditions of decreased availability of ternary complexes, leaky scanning and delayed re-initiation event make the translation of the major ORF more favorable. The best characterized examples for such translational regulation are the mRNAs of yeast GCN4 [45] and its mammalian homologue ATF4 [46,47]. Both mRNAs encode transcription factors, therefore their translational activation lead to transcription activation of numerous target genes necessary for appropriate cell growth and survival under stressful conditions [48–50]. Internal ribosomal binding sites (IRES elements) are another example for 5′UTR regulatory sequences. IRES elements enable ribosomes to bind internally to the mRNA rather than to its 5′cap structure. IRES-bearing mRNAs encode major regulatory proteins including transcription factors, growth factors and their receptors, cell cycle, and apoptosis regulators [51]. Interestingly, IRES elements often confer translational advantage under stress conditions that are associated with increased eIF2 phosphorylation [52,53]. Since proteins encoded by IRES-bearing mRNAs are often involved in proliferation, differentiation, and cell death, the activation of their synthesis gives rise to major cellular decisions [54].

Mutations in any of the five subunits of eIF2B of both alleles cause a partial deficiency of GEF activity of this protein [55–57]. Obligate carriers of such mutations have completely normal GEF activity, suggesting that when there are sufficient wild type subunits, the mutated ones are "crowded out" and do not interfere with the formation of adequate levels of normal eIF2B complexes. Complete abolition of eIF2B activity is considered lethal. Based on in vitro studies in yeast and human cellular models, the most common mechanism of reduction in GEF activity resulting from eIF2B mutations is reduced amount of intact eIF2B complexes and unstable protein complexes associated with exclusion of the mutated subunit from the holocomplexes [55,56]. Other identified mechanisms are impairment substrate binding and enhancement of eIF2 binding [55]. Decreased global protein synthesis and viability are seen in the most severe mutations of the yeast model [56]. Parallel studies reported partial loss of basal eIF2B GEF activity in EBV-transformed lymphocytes [57] and primary fibroblasts from CACH/VWM patients [58]. The current view is that most of CACH/VWM mutations are loss of function mutations.

How is the reduction in eIF2B GEF activity is related to clinical disease? Disease severity was significantly correlated with age of onset [14]. There was limited correlation between the age of onset of the disease and the GEF activity in transformed lymphoblasts of patients [57]. In cultured primary skin fibroblasts on the other hand, there was no difference in eIF2B GEF activity between the mildest genotype (eR113H/eR113H) and the severe infantile mutation of the Cree Leukoencephalopathy (eR195H/eR195H) [58]. Therefore, the mild reduction in the basal activity of the mutated eIF2B cannot explain the disease pathology, unless eIF2B activity in the brain is significantly lower than its activity in lymphocytes and fibroblasts. In such case, which is still an open question, it is possible that basal translation rate of certain mRNAs specific for brain white matter is altered, leading to phenotypic consequences (Fig. 3). In addition, there may be abnormalities associated with eIF2B function under physiological stress conditions. Such a scenario is appealing since the symptoms of the disease often appear or worsen upon stress. This explanation should also help understand why brain glial cells are particularly susceptible to eIF2B hypofunction although it is an essential factor in all cell types. Oligodendrocytes are extremely sensitive to infections, brain trauma, oxidative stress, and hypoxia/ischemia—stresses that in many cases end up in myelin damage [59,60]. The hypersensitivity of oligodendrocytes may be the consequence of their being extremely prone to ER-stress. This is because coordinated expression of lipids and proteins is a unique feature of myelinating cells, not possessed by other cell types that are engaged with intensive ER function. Myelin biogenesis is a highly regulated process requiring coordination of numerous events that include tight regulation of translation, specific mRNA transport, protein synthesis in and out of the ER, protein folding, glycosylation, vesicular transport, and lipid synthesis [61–63]. ER-stress is a normal physiological phenomenon dealing with temporal ER overload, which elicits cellular response known as unfolded protein response (UPR) [50]. The UPR is further activated by metabolic, thermal, or physical stress conditions that impair protein folding and/or processing in the ER. Translation attenuation, one of the tripartite arms of the UPR, is elicited by PERK activation which occurs within minutes after stress, resulting in eIF2 phosphorylation [64,65]. In parallel to global translational attenuation, the translationaly activated ATF4 is responsible for the transcriptional activation of numerous target genes. Depending on the balance between the downstream effectors, the UPR develops into a prosurvival or proapoptotic process. One of the ATF4 targets is C/EBP homologous protein (CHOP) known to be involved in ER stress-mediated apoptosis and in neurodegenerative diseases [66]. Another ATF4 target is GADD34 which is necessary for translational recovery towards survival. Perfect timing of translational recovery along the stress response is crucial for stress resistance [67,68]. It is hypothesized that oligodendrocytes expressing mutated

eIF2B genes are even more predisposed to stress than control oligodendrocytes that are innately highly sensitive anyway. In support with the stress predisposition hypothesis is the finding that primary fibroblasts from CACH/VWM patients had significantly greater increase in ATF4 compared to controls when exposed to similar ER-stress [58]. Such hyper-ATF4 synthesis suggests that less inactivation of eIF2B is needed to achieve the adequately low eIF2B activity required for ATF4 translational induction. The decreased expression of specific eIF2B subunits in some CACH/VWM lymphoblasts following exposure to heat shock further demonstrates the increased susceptability of eIF2B-mutated cells to stress [70]. It seems that important factors leading to the disease in addition to eIF2B mutations are intrinsic to each cell type according to its nature. These parameters include the basal level of eIF2B activity and the stress level each cell type normally experiences. In support of the above concept is the active UPR state which was recently demonstrated in glia of patients with CACH/VWM [69].

We hypothesize that the translation level of mRNAs of other genes, yet to be discovered, is abnormal under stress conditions in addition to the known ATF4 mRNA. Such candidates may include mRNAs directly encoding myelin components (Fig. 3). Lack of close correlation of mRNA levels and actual synthesis of HMG-CoA reductase (the enzyme catalyzing the rate-limiting step in cholesterol biosynthesis), ceramide galactosyltransferase (the rate-limiting step in cerebroside biosynthesis), and myelin basic protein points to a possible post-transcriptional control of these mRNAs [71–73]. Abnormal synthesis of cerebroside and cholesterol may affect the level of their metabolites, if they are not being shielded from cytoplasmic catabolic enzymes by being rapidly integrated into deep layers of myelin [74]. For example, 24S-hydroxycholesterol is a brain specific primary oxidation product of cholesterol that induces the expression of various genes in neural cells and may contribute to the pathogenesis of the disease [75]. Other equally likable mRNAs candidates that may be hypertranslated under stress conditions may include uORFs- or IRES-containing mRNAs encoding for transcription factors and other regulators that impact myelin maintenance. The blend of induced transcription factors (ATF4 included) may have a major influence on transcriptional activity of key target genes leading to direct or indirect effect on myelination (Fig. 3). Additional scenarios are also possible. Of special interest is the notion that the IRES of FGF-2 is highly active in the brain [76], and that FGF-2 converts mature oligodendrocytes to a different phenotype morphologically and biochemically as they lose their myelin markers [77]. Obviously, induced translation of IRES-controlled proapoptotic genes may be detrimental [8,35]. Animal models for the CACH/VWM will enable to confirm the hypothesis that unbalanced shift of translation towards specific mRNAs in the brain leads to the abnormal phenotypic consequences.

Epidemiology

The incidence of CACH/VWM is unknown. Although it is a rare disease, in our experience about half of the patients with previously unclassified leukodystrophies have been recently determined to be CACH/VWM. In a study of unclassified leukodystrophies in childhood, CACH/VWM was the most frequent [78]. In some countries, its incidence is close to that of metachromatic leukodystrophy (van der Knaap, personal communication). We estimate that more than 200 patients with CACH/VWM have been identified to date.

Management and prevention

Specific treatment is not yet available for CACH/VWM. One can speculate that therapy is a possibility since all patients have significant residual eIF2B GEF activity that in theory should be correctable to full activity. Supportive therapy includes physical therapy and rehabilitation for motor dysfunction. Ankle-foot orthoses were useful in some patients with hypotonia and weakness of dorsiflexion. Carbamazepine was useful for seizures in our patients. There are anecdotal reports that corticosteroids may be helpful during acute deterioration. We have no personal experience with such a success. Prenatal diagnosis when the mutations of the eIF2B subunit are known is currently possible.

Acknowledgments

References

[1] M.S. van der Knaap, J. Valk, N. de Neeling, J.J. Nauta, Pattern recognition in magnetic resonance imaging of white matter disorders in children and young adults, Neuroradiology 33 (1991) 478–493.
[2] R. Schiffmann, O. Boespflug-Tanguy, An update on the leukodystrophies, Curr. Opin. Neurol. 14 (2001) 789–794.
[3] R. Schiffmann, M.S. van der Knaap, The latest on leukodystrophies, Curr. Opin. Neurol. 17 (2004) 187–192.
[4] M.J. Noetzel, Diagnosing "undiagnosed" leukodystrophies: the role of molecular genetics, Neurology 62 (2004) 847–848.
[5] R. Schiffmann, B.D. Trapp, J.R. Moller, E.M. Kaye, C.C. Parker, R.O. Brady, N.W. Barton, Childhood ataxia with diffuse central nervous system hypomyelination, Ann. Neurol. (1992) 84.
[6] F. Hanefeld, U. Holzbach, B. Kruse, E. Wilichowski, H.J. Christen, J. Frahm, Diffuse white matter disease in three children: an encephalopathy with unique features on magnetic resonance imaging and proton magnetic resonance spectroscopy, Neuropediatrics 24 (1993) 244–248.
[7] M.S. van der Knaap, P.G. Barth, F.J. Gabreels, E. Franzoni, J.H. Begeer, H. Stroink, J.J. Rotteveel, J. Valk, A new leukoencephalopathy with vanishing white matter, Neurology 48 (1997) 845–855.
[8] W. Bruck, J. Herms, K. Brockmann, W. Schulz-Schaeffer, F. Hanefeld, Myelinopathia centralis diffusa (vanishing white matter

disease): evidence of apoptotic oligodendrocyte degeneration in early lesion development, Ann. Neurol. 50 (2001) 532–536.

 [9] R. Schiffmann, J.R. Moller, B.D. Trapp, H.H. Shih, R.G. Farrer, D.A. Katz, J.R. Alger, C.C. Parker, P.E. Hauer, C.R. Kaneski, et al., Childhood ataxia with diffuse central nervous system hypomyelination, Ann. Neurol. 35 (1994) 331–340.

[10] P.A. Leegwater, A.A. Konst, B. Kuyt, L.A. Sandkuijl, S. Naidu, C.B. Oudejans, R.B. Schutgens, J.C. Pronk, M.S. van der Knaap, The gene for leukoencephalopathy with vanishing white matter is located on chromosome 3q27, Am. J. Hum. Genet. 65 (1999) 728–734.

[11] P.A. Leegwater, G. Vermeulen, A.A. Konst, S. Naidu, J. Mulders, A. Visser, P. Kersbergen, D. Mobach, D. Fonds, C.G. van Berkel, R.J. Lemmers, R.R. Frants, C.B. Oudejans, R.B. Schutgens, J.C. Pronk, M.S. van der Knaap, Subunits of the translation initiation factor eIF2B are mutant in leukoencephalopathy with vanishing white matter, Nat. Genet. 29 (2001) 383–388.

[12] A. Fogli, K. Wong, E. Eymard-Pierre, J. Wenger, J.P. Bouffard, E. Goldin, D.N. Black, O. Boespflug-Tanguy, R. Schiffmann, Cree leukoencephalopathy and CACH/VWM disease are allelic at the EIF2B5 locus, Ann. Neurol. 52 (2002) 506–510.

[13] P. Labauge, A. Fogli, G. Castelnovo, A. Le Bayon, L. Horzinski, F. Nicoli, P. Cozzone, M. Pages, C. Briere, C. Marty-Double, O. Delhaume, A. Gelot, O. Boespflug-Tanguy, D. Rodriguez, Dominant form of vanishing white matter-like leukoencephalopathy, Ann. Neurol. 58 (2005) 634–639.

[14] A. Fogli, R. Schiffmann, E. Bertini, S. Ughetto, P. Combes, E. Eymard-Pierre, C.R. Kaneski, M. Pineda, M. Troncoso, G. Uziel, R. Surtees, D. Pugin, M.P. Chaunu, D. Rodriguez, O. Boespflug-Tanguy, The effect of genotype on the natural history of eIF2B-related leukodystrophies, Neurology 62 (2004) 1509–1517.

[15] G. Tedeschi, R. Schiffmann, N.W. Barton, H.H. Shih, S.M. Gospe Jr., R.O. Brady, J.R. Alger, G. Di Chiro, Proton magnetic resonance spectroscopic imaging in childhood ataxia with diffuse central nervous system hypomyelination, Neurology 45 (1995) 1526–1532.

[16] G. Vermeulen, R. Seidl, S. Mercimek-Mahmutoglu, J.J. Rotteveel, G.C. Scheper, M.S. van der Knaap, Fright is a provoking factor in vanishing white matter disease, Ann. Neurol. 57 (2005) 560–563.

[17] P.A. Leegwater, J.C. Pronk, M.S. van der Knaap, Leukoencephalopathy with vanishing white matter: from magnetic resonance imaging pattern to five genes, J. Child Neurol. 18 (2003) 639–645.

[18] M.S. van der Knaap, W. Kamphorst, P.G. Barth, C.L. Kraaijeveld, E. Gut, J. Valk, Phenotypic variation in leukoencephalopathy with vanishing white matter, Neurology 51 (1998) 540–547.

[19] A. Fogli, F. Gauthier-Barichard, R. Schiffmann, V.H. Vanderhoof, V.K. Bakalov, L.M. Nelson, O. Boespflug-Tanguy, Screening for known mutations in EIF2B genes in a large panel of patients with premature ovarian failure, BMC Womens Health 4 (2004) 8.

[20] K. Prass, W. Bruck, N.W. Schroder, A. Bender, M. Prass, T. Wolf, M.S. Van der Knaap, R. Zschenderlein, Adult-onset Leukoencephalopathy with vanishing white matter presenting with dementia, Ann. Neurol. 50 (2001) 665–668.

[21] M.S. van der Knaap, C.G. van Berkel, J. Herms, R. van Coster, M. Baethmann, S. Naidu, E. Boltshauser, M.A. Willemsen, B. Plecko, G.F. Hoffmann, C.G. Proud, G.C. Scheper, J.C. Pronk, eIF2B-related disorders: antenatal onset and involvement of multiple organs, Am. J. Hum. Genet. 73 (2003) 1199–1207.

[22] P. Francalanci, E. Eymard-Pierre, C. Dionisi-Vici, R. Boldrini, F. Piemonte, R. Virgili, G. Fariello, C. Bosman, F.M. Santorelli, O. Boespflug-Tanguy, E. Bertini, Fatal infantile leukodystrophy: a severe variant of CACH/VWM syndrome, allelic to chromosome 3q27, Neurology 57 (2001) 265–270.

[23] I.A. Alorainy, Y.G. Patenaude, A.M. O'Gorman, D.N. Black, K. Meagher-Villemure, Cree leukoencephalopathy: neuroimaging findings, Radiology 213 (1999) 400–406.

[24] D.N. Black, F. Booth, G.V. Watters, E. Andermann, C. Dumont, W.C. Halliday, J. Hoogstraten, M.E. Kabay, P. Kaplan, K. Meagher-Villemure, et al., Leukoencephalopathy among native Indian infants in northern Quebec and Manitoba, Ann. Neurol. 24 (1988) 490–496.

[25] R. Schiffmann, G. Tedeschi, R.P. Kinkel, B.D. Trapp, J.A. Frank, C.R. Kaneski, R.O. Brady, N.W. Barton, L. Nelson, J.A. Yanovski, Leukodystrophy in patients with ovarian dysgenesis, Ann. Neurol. 41 (1997) 654–661.

[26] A. Fogli, D. Rodriguez, E. Eymard-Pierre, F. Bouhour, P. Labauge, B.F. Meaney, S. Zeesman, C.R. Kaneski, R. Schiffmann, O. Boespflug-Tanguy, Ovarian failure related to eukaryotic initiation factor 2B mutations, Am. J. Hum. Genet. 72 (2003) 1544–1550.

[27] M.S. van der Knaap, P.A. Leegwater, C.G. van Berkel, C. Brenner, E. Storey, M. Di Rocco, F. Salvi, J.C. Pronk, Arg113His mutation in eIF2Bepsilon as cause of leukoencephalopathy in adults, Neurology 62 (2004) 1598–1600.

[28] A. Gallo, M.A. Rocca, A. Falini, C. Scaglione, F. Salvi, A. Gambini, L. Guerrini, M. Mascalchi, J.C. Pronk, M.S. van der Knaap, M. Filippi, Multiparametric MRI in a patient with adult-onset leukoencephalopathy with vanishing white matter, Neurology 62 (2004) 323–326.

[29] A. Vanderver, R. Schiffmann, M. Timmons, K.A. Kellersberger, D. Fabris, E.P. Hoffman, J. Maletkovic, Y. Hathout, Decreased asialo-transferrin in cerebrospinal fluid of patients with childhood onset ataxia and central nervous system hypomyelination/vanishing white matter disease, Clin. Chem. 51 (2005) 2031–2042.

[30] P. Davidsson, R. Ekman, K. Blennow, A new procedure for detecting brain-specific proteins in cerebrospinal fluid, J. Neural. Transm. 104 (1997) 711–720.

[31] A. Hoffmann, M. Nimtz, R. Getzlaff, H.S. Conradt, 'Brain-type' N-glycosylation of asialo-transferrin from human cerebrospinal fluid, FEBS Lett. 359 (1995) 164–168.

[32] G. Keir, A. Zeman, G. Brookes, M. Porter, E.J. Thompson, Immunoblotting of transferrin in the identification of cerebrospinal fluid otorrhoea and rhinorrhoea, Ann. Clin. Biochem. 29 (Pt 2) (1992) 210–213.

[33] S. Bluml, M. Philippart, R. Schiffmann, K. Seymour, B.D. Ross, Membrane phospholipids and high-energy metabolites in childhood ataxia with CNS hypomyelination, Neurology 61 (2003) 648–654.

[34] K. Wong, R.C. Armstrong, K.A. Gyure, A.L. Morrison, D. Rodriguez, R. Matalon, A.B. Johnson, R. Wollmann, E. Gilbert, T.Q. Le, C.A. Bradley, K. Crutchfield, R. Schiffmann, Foamy cells with oligodendroglial phenotype in childhood ataxia with diffuse central nervous system hypomyelination syndrome, Acta Neuropathol. (Berl) 100 (2000) 635–646.

[35] K. Van Haren, J.P. van der Voorn, D.R. Peterson, M.S. van der Knaap, J.M. Powers, The life and death of oligodendrocytes in vanishing white matter disease, J. Neuropathol. Exp. Neurol. 63 (2004) 618–630.

[36] D. Rodriguez, A. Gelot, B. della Gaspera, O. Robain, G. Ponsot, L.L. Sarlieve, S. Ghandour, A. Pompidou, A. Dautigny, P. Aubourg, D. Pham-Dinh, Increased density of oligodendrocytes in childhood ataxia with diffuse central hypomyelination (CACH) syndrome: neuropathological and biochemical study of two cases, Acta Neuropathol. (Berl) 97 (1999) 469–480.

[37] M. Sutton-Smith, H.R. Morris, P.K. Grewal, J.E. Hewitt, R.E. Bittner, E. Goldin, R. Schiffmann, A. Dell, MS screening strategies: investigating the glycomes of knockout and myodystrophic mice and leukodystrophic human brains, Biochem. Soc. Symp. (2002) 105–115.

[38] J. Dietrich, M. Lacagnina, D. Gass, E. Richfield, M. Mayer-Proschel, M. Noble, C. Torres, C. Proschel, EIF2B5 mutations compromise GFAP+ astrocyte generation in vanishing white matter leukodystrophy, Nat. Med. 11 (2005) 277–283.

[39] S. Bonavita, R. Schiffmann, D.F. Moore, K. Frei, B. Choi, M.N. Patronas, A. Virta, O. Boespflug-Tanguy, G. Tedeschi, Evidence for neuroaxonal injury in patients with proteolipid protein gene mutations, Neurology 56 (2001) 785–788.

[40] M.S. van der Knaap, P.A. Leegwater, A.A. Konst, A. Visser, S. Naidu, C.B. Oudejans, R.B. Schutgens, J.C. Pronk, Mutations in each of the five subunits of translation initiation factor eIF2B can cause leukoencephalopathy with vanishing white matter, Ann. Neurol. 51 (2002) 264–270.

[41] A. Ohlenbusch, M. Henneke, K. Brockmann, M. Goerg, F. Hanefeld, A. Kohlschutter, J. Gartner, Identification of ten novel mutations in patients with eIF2B-related disorders, Hum. Mutat. 25 (2005) 411.

[42] R. Schiffmann, A. Fogli, M.S. van der Knaap, O. Boespflug-Tanguy, 2003, Childhood Ataxia with central nervous system hypomyelination/vanishing white matter. Copyright 1997–2003. In: GeneReviews at GeneTests: Medical Genetics Information Resource [database online], University of Washington. Available at http://www.gene-tests.org., Seattle.

[43] S.R. Kimball, Eukaryotic initiation factor eIF2, Int. J. Biochem. Cell Biol. 31 (1999) 25–29.

[44] T.E. Dever, Gene-specific regulation by general translation factors, Cell 108 (2002) 545–556.

[45] A.G. Hinnebusch, K. Natarajan, Gcn4p, a master regulator of gene expression, is controlled at multiple levels by diverse signals of starvation and stress, Eukaryot Cell 1 (2002) 22–32.

[46] K.M. Vattem, R.C. Wek, Reinitiation involving upstream ORFs regulates ATF4 mRNA translation in mammalian cells, Proc. Natl. Acad. Sci. USA 101 (2004) 11269–11274.

[47] P.D. Lu, H.P. Harding, D. Ron, Translation reinitiation at alternative open reading frames regulates gene expression in an integrated stress response, J. Cell Biol. 167 (2004) 27–33.

[48] H.Y. Jiang, R.C. Wek, Phosphorylation of the alpha-subunit of the eukaryotic initiation factor-2 (eIF2alpha) reduces protein synthesis and enhances apoptosis in response to proteasome inhibition, J. Biol. Chem. 280 (2005) 14189–14202.

[49] M.J. Clemens, Translational control in virus-infected cells: models for cellular stress responses, Semin. Cell Dev. Biol. 16 (2005) 13–20.

[50] H.P. Harding, M. Calfon, F. Urano, I. Novoa, D. Ron, Transcriptional and translational control in the Mammalian unfolded protein response, Annu. Rev. Cell Dev. Biol. 18 (2002) 575–599.

[51] M. Stoneley, A.E. Willis, Cellular internal ribosome entry segments: structures, trans-acting factors and regulation of gene expression, Oncogene 23 (2004) 3200–3207.

[52] Y.K. Kim, S.K. Jang, Continuous heat shock enhances translational initiation directed by internal ribosomal entry site, Biochem. Biophys. Res. Commun. 297 (2002) 224–231.

[53] G. Gerlitz, R. Jagus, O. Elroy-Stein, Phosphorylation of initiation factor-2 alpha is required for activation of internal translation initiation during cell differentiation, Eur. J. Biochem. 269 (2002) 2810–2819.

[54] M. Holcik, N. Sonenberg, R.G. Korneluk, Internal ribosome initiation of translation and the control of cell death, Trends Genet. 16 (2000) 469–473.

[55] W. Li, X. Wang, M.S. Van Der Knaap, C.G. Proud, Mutations linked to leukoencephalopathy with vanishing white matter impair the function of the eukaryotic initiation factor 2B complex in diverse ways, Mol. Cell Biol. 24 (2004) 3295–3306.

[56] J.P. Richardson, S.S. Mohammad, G.D. Pavitt, Mutations causing childhood ataxia with central nervous system hypomyelination reduce eukaryotic initiation factor 2B complex formation and activity, Mol. Cell Biol. 24 (2004) 2352–2363.

[57] A. Fogli, R. Schiffmann, L. Hugendubler, P. Combes, E. Bertini, D. Rodriguez, S.R. Kimball, O. Boespflug-Tanguy, Decreased guanine nucleotide exchange factor activity in eIF2B-mutated patients, Eur. J. Hum. Genet. 12 (2004) 561–566.

[58] L. Kantor, H.P. Harding, D. Ron, R. Schiffmann, C.R. Kaneski, S.R. Kimball, O. Elroy-Stein, Heightened stress response in primary fibroblasts expressing mutant eIF2B genes from CACH/VWM leukodystrophy patients, Hum. Genet. (2005) 1–8.

[59] J.E. Merrill, N.J. Scolding, Mechanisms of damage to myelin and oligodendrocytes and their relevance to disease, Neuropath. Appl. Neurobiol. 25 (1999) 435–458.

[60] R.D. Folkerth, R.L. Haynes, N.S. Borenstein, R.A. Belliveau, F. Trachtenberg, P.A. Rosenberg, J.J. Volpe, H.C. Kinney, Developmental lag in superoxide dismutases relative to other antioxidant enzymes in premyelinated human telencephalic white matter, J. Neuropathol. Exp. Neurol. 63 (2004) 990–999.

[61] N. Baumann, D. Pham-Dinh, Biology of oligodendrocyte and myelin in the mammalian central nervous system, Physiol. Rev. 81 (2001) 871–927.

[62] J.N. Larocca, A.G. Rodriguez-Gabin, Myelin biogenesis: vesicle transport in oligodendrocytes, Neurochem. Res. 27 (2002) 1313–1329.

[63] A.G. Rodriguez-Gabin, G. Almazan, J.N. Larocca, Vesicle transport in oligodendrocytes: probable role of Rab40c protein, J. Neurosci. Res. 76 (2004) 758–770.

[64] H.P. Harding, Y. Zhang, D. Ron, Protein translation and folding are coupled by an endoplasmic-reticulum-resident kinase, Nature 397 (1999) 271–274.

[65] H.P. Harding, I. Novoa, Y. Zhang, H. Zeng, R. Wek, M. Schapira, D. Ron, Regulated translation initiation controls stress-induced gene expression in mammalian cells, Mol. Cell 6 (2000) 1099–1108.

[66] S. Oyadomari, M. Mori, Roles of CHOP/GADD153 in endoplasmic reticulum stress, Cell Death Differ. 11 (2004) 381–389.

[67] Y. Ma, L.M. Hendershot, Delineation of a negative feedback regulatory loop that controls protein translation during endoplasmic reticulum stress, J. Biol. Chem. 278 (2003) 34864–34873.

[68] M. Boyce, K.F. Bryant, C. Jousse, K. Long, H.P. Harding, D. Scheuner, R.J. Kaufman, D. Ma, D.M. Coen, D. Ron, J. Yuan, A selective inhibitor of eIF2alpha dephosphorylation protects cells from ER stress, Science 307 (2005) 935–939.

[69] J.P. van der Voorn, B. van Kollenburg, G. Bertrand, K. Van Haren, G.C. Scheper, J.M. Powers, M.S. van der Knaap, The Unfolded Protein Response in Vanishing White Matter Disease, J. Neuropathol. Exp. Neurol. 64 (2005) 770–775.

[70] B. van Kollenburg, A.A. Thomas, G. Vermeulen, G.A. Bertrand, C.G. van Berkel, J.C. Pronk, C.G. Proud, M.S. van der Knaap, G.C. Scheper, Regulation of protein synthesis in lymphoblasts from vanishing white matter patients, Neurobiol. Dis. (2005).

[71] G.A. Reynolds, S.K. Basu, T.F. Osborne, D.J. Chin, G. Gil, M.S. Brown, J.L. Goldstein, K.L. Luskey, HMG CoA reductase: a negatively regulated gene with unusual promoter and 5′ untranslated regions, Cell 38 (1984) 275–285.

[72] S. Ueno, L. Foster, G.T. Hifumi, G.I. Tennekoon, A.T. Campagnoni, The simian virus 40 large T antigen does not inhibit translation of the 14-kDa myelin basic protein mRNA in reticulocyte lysates or in transfected cells, J. Neurochem. 64 (1995) 928–931.

[73] E.D. Muse, H. Jurevics, A.D. Toews, G.K. Matsushima, P. Morell, Parameters related to lipid metabolism as markers of myelination in mouse brain, J. Neurochem. 76 (2001) 77–86.

[74] H. Jurevics, C. Largent, J. Hostettler, D.W. Sammond, G.K. Matsushima, A. Kleindienst, A.D. Toews, P. Morell, Alterations in metabolism and gene expression in brain regions during cuprizone-induced demyelination and remyelination, J. Neurochem. 82 (2002) 126–136.

[75] P. Alexandrov, J.G. Cui, Y. Zhao, W.J. Lukiw, 24S-hydroxycholesterol induces inflammatory gene expression in primary human neural cells, Neuroreport 16 (2005) 909–913.

[76] L. Creancier, D. Morello, P. Mercier, A.C. Prats, Fibroblast growth factor 2 internal ribosome entry site (IRES) activity ex vivo and in transgenic mice reveals a stringent tissue-specific regulation, J. Cell Biol. 150 (2000) 275–281.

[77] R. Bansal, S.E. Pfeiffer, FGF-2 converts mature oligodendrocytes to a novel phenotype, J. Neurosci. Res. 50 (1997) 215–228.

[78] M.S. van der Knaap, S.N. Breiter, S. Naidu, A.A. Hart, J. Valk, Defining and categorizing leukoencephalopathies of unknown origin: MR imaging approach, Radiology 213 (1999) 121–133.