ESETISMERTETÉS
CASE REPORT

Vanishing white matter disease, a rare leukodystrophy with mutation in the *EIF2B5* gene

Gabriella SINKÓ[1], Márton TOMPA[2, 3], Zsuzsanna KISS[4], Bernadette KÁLMÁN[2, 5]

[1]Markusovszky University Teaching Hospital, Department of Pediatrics, Szombathely
[2]Markusovszky University Teaching Hospital, Center for Molecular Medicine, Szombathely
[3]Hungarian Centre for Genomics and Bioinformatics, Szentágothai Research Center, Pécs
[4]Markusovszky University Teaching Hospital, Department of Laboratory Medicine, Division of Clinical Genetics, Szombathely
[5]University of Pécs, School of Medicine, Department of Laboratory Medicine, Pécs

PRINT | English | https://doi.org/10.18071/isz.77.0207 | www.elitmed.hu

Correspondent:
Prof. dr. Bernadette KÁLMÁN,
Department of Laboratory Medicine,
Szentágothai Research Center,
University of Pécs,
7624 Pécs, Ifjúság út 20.
E-mail:
Bernadette.kalman@pte.hu
https://orcid.org/0000-0003-2592-3939

Érkezett:
2023. október 25.
Elfogadva:
2023. december 8.

Background – Leukodystrophies, a heterogeneous group of brain and spinal cord disorders, often pose challenges in establishing molecular etiology. Vanishing White Matter Disease (VWMD) is a rare subtype of leukodystrophies presenting with characteristic clinical and MRI features, nevertheless, achieving diagnostic certainty requires genetic studies.
Case presentation – Our patient is a nine year old girl, who developed progressive gait difficulties at around 3-4 years of age. Her brain MRI showed confluent lesions with increased signal intensity in the cerebral and cerebellar white matter on T2/FLAIR sequences, within which hypointense regions appeared with signal intensity resembling that of the cerebrospinal fluid on T1 sequences. Whole exome sequencing identified a homozygous likely pathogenic variant within the *EIF2B5* gene in the proband, which was present in a heterozygous state in both asymptomatic parents. Having the clinical and molecular genetic diagnosis established, we explored therapeutic possibilities for the patient.
Conclusion – VWMD is a severe form of leukodystrophies with little or no disease modifying therapy available until recently. A better understanding of its molecular pathogenesis offers some hope for new inventive therapies.

Keywords: vanishing white matter disease, *EIF2B5*, whole exome sequencing

Eltűnő fehérállomány-betegség, a leukodystrophiák ritka altípusa az *EIF2B5* gén mutációjával

Sinkó G, MD, Tompa M, PhD, Kiss Zs, MD, Kálmán B, MD, DSc, FAAN

Háttér – A leukodystrophiák az agy és a gerincvelő heterogén betegségei, gyakran diagnosztikai kihívásokkal szembesítik a vizsgálót a molekuláris etiológia meghatározásakor. A vanishing white matter disease (VWMD) vagy eltűnő fehérállomány-betegség a leukodystrophiák ritka altípusa, ami jellegzetes klinikai és MRI-sajátosságokkal rendelkezik, de a diagnosztikai bizonyosság érdekében genetikai vizsgálatok szükségesek.
Esetbemutató – Betegünk egy kilencéves kislány, akinek 3-4 éves kora körül kezdődött progresszív járásproblémája. Az MRI-T2/FLAIR-szekvenciákon konfluens fokozott jelintenzitású elváltozások látszottak a hemisphaerialis és a kisagyi fehérállományban, amelyeken belül a T1 szekvenciákon a liquorhoz hasonló jelintenzitású hipointenzív régiók jelentek meg. A teljesexom-szekvenálás homozigóta, valószínű patogén variánst azonosított a proband *EIF2B5* génjében, ami heterozigóta formában volt jelen a tünetmentes szülőkben. A klinikai és molekuláris genetikai diagnózis felállítása után megvizsgáltuk a beteg terápiás lehetőségeit.
Következtetés – A VWMD a leukodystrophiák súlyos formája, amelyhez mostanáig alig vagy egyáltalán nem állt rendelkezésre betegségmódosító terápia. A molekuláris patogenezis jobb megértése reményt ad új innovatív terápiák bevezetéséhez.

Kulcsszavak: vanishing white matter disease, *EIF2B5,* teljesexom-szekvenálás

Leukodystrophies are a heterogeneous group of disorders affecting the white matter in the central nervous system (CNS). Based on magnetic resonance imaging (MRI) characteristics, both hypomyelination and demyelination may be observed in association with various cellular, biochemical and molecular genetic alterations affecting either directly the myelin sheath, the myelin-producing oligodendrocytes or other cellular elements and pathways involved in the maintenance of this axonal insulator[1]. The age of onset, clinical presentation and disease course, distribution and nature of MRI lesions, along with some biochemical laboratory findings often allow us narrowing down the differential diagnosis. Genetic studies, however, have become key elements of the diagnostic work up and may involve targeted single gene studies, panel testing, or broader approaches such as whole exome sequencing (WES). Even when the clinical and MRI findings are suggestive of a specific diagnosis, genetic testing may be necessary for diagnostic validation, therapeutic decision, prognostication and reproductive counselling.

Vanishing white matter disease (VWMD) is a rare form of leukodystrophies inherited in an autosomal recessive manner and caused by homozygous or compound heterozygous mutations in one of the five subunit genes of eIF2B *(EIF2B1-EIF2B5)* (eukaryotic translation initiation factor) (OMIM 603896).

The disease was first described in the 1990s as a malignant encephalopathy with early childhood onset, episodic worsening of neurological symptoms including ataxia, spasticity, cognitive decline, seizures and coma, which lead to early death in a few years[2–4]. The onset and the episodic deterioration of VWMD are typically induced by biological stress such as minor head trauma, infection and fevers. MRI reveals diffuse leukoencephalopathy within which signal alterations resembling those of cerebrospinal fluid may be seen[4]. MRI and spectroscopy as well as autopsy studies suggest that axonal loss underlies the rarefaction of the white matter and myelin[4]. As the underlying genetic abnormality was identified[5] a broader spectrum of disease presentation ranging from early onset, severe phenotype and poor prognosis to late onset, less severe phenotype and varying prognosis have been described[6]. In addition to the step-wise deterioration, patients with progressive disease have also been encountered. In affected females, ovarian dysgenesis has also been reported[7].

Here we describe the case of a nine year old girl who presented with slowly progressive gait and coordination problems without known precipitating factors or family history. The MRI findings were suggestive of VWMD, a suspicion confirmed by genetic studies. When the diagnosis was established, we explored therapeutic options.

Case presentation

The proband is a nine year old girl born from an uncomplicated second pregnancy from unrelated parents. Her perinatal adaptation, early movement and speech development were normal. However, she was noted walking on tiptoe when attending the kindergarten. At eight years of age, her running and walking became increasingly uncoordinated. She was referred to a pediatric neurologist, who detected spastic tone, mild paraparesis, enhanced deep tendon reflexes, appendicular and truncal ataxia prompting her admission to our hospital.

Her neurology exam at admission revealed intact cranial nerves, except for a fine, diminishing horizontal nystagmus in the direction of gaze. Muscle tone, mass and strength were preserved in both upper extremities with 2+ deep tendon reflexes and bilateral Trömner sign. Mild spasticity and 4/5 muscle strength (with some left sided predominance), along with 3+ deep tendon reflexes and ankle clonus, and equivocal plantar responses were found in both lower extremities. Slight uncertainty in the finger-nose test and marked ataxia in the heel-knee test were noted. Her gait was wide-based, paretico-ataxic. Romberg sign was positive. Proprioception, light touch, pain and heat sensation were all preserved. Her speech was intact and cognition appropriate for her age. This charming girl attended primary school with average academic accomplishments, but with some difficulties in mathematics.

Brain MRI in 2022, when the patient was 8 years old, showed bilateral abnormalities affecting almost the entire hemispheric white matter of the cerebrum, but also some areas of the cerebellum, which were hyperintense on T2/FLAIR, and hypointense on T1 sequences. The pathological areas were completely confluent and showed increased diffusion. On both sides of the corona radiata, with left-sided predominance, the reduced T1 signal intensity approached that of the cerebrospinal fluid within areas of T2/FLAIR hyperintensity. The images suggested VWMD, a leukodystrophy subtype **(Figure 1)**. An earlier arylsulfatase test performed on blood cells showed normal enzyme activity, thus excluding metachromatic leukodystrophy[8, 9].

Whole exome sequencing was performed on DNA extracted from EDTA-treated peripheral blood by using the QIAamp® DNA Mini Kit (Qiagen, Hilden, Germany). Applying the DNA Prep with Enrichment (Illumina Inc. San Diego, CA, USA), DNA was first enzymatically tagmented and amplified. Then exome capture was performed by using Illumina Exome Panel probes (Illumina Inc. San Diego, CA, USA). The captured libraries were quality checked by TapeStation 4200 (Agilent Technologies, Santa Clara, CA, USA) and quantified by Qubit 3.0 (Invitrogen, Waltham, MA, USA). The libraries were

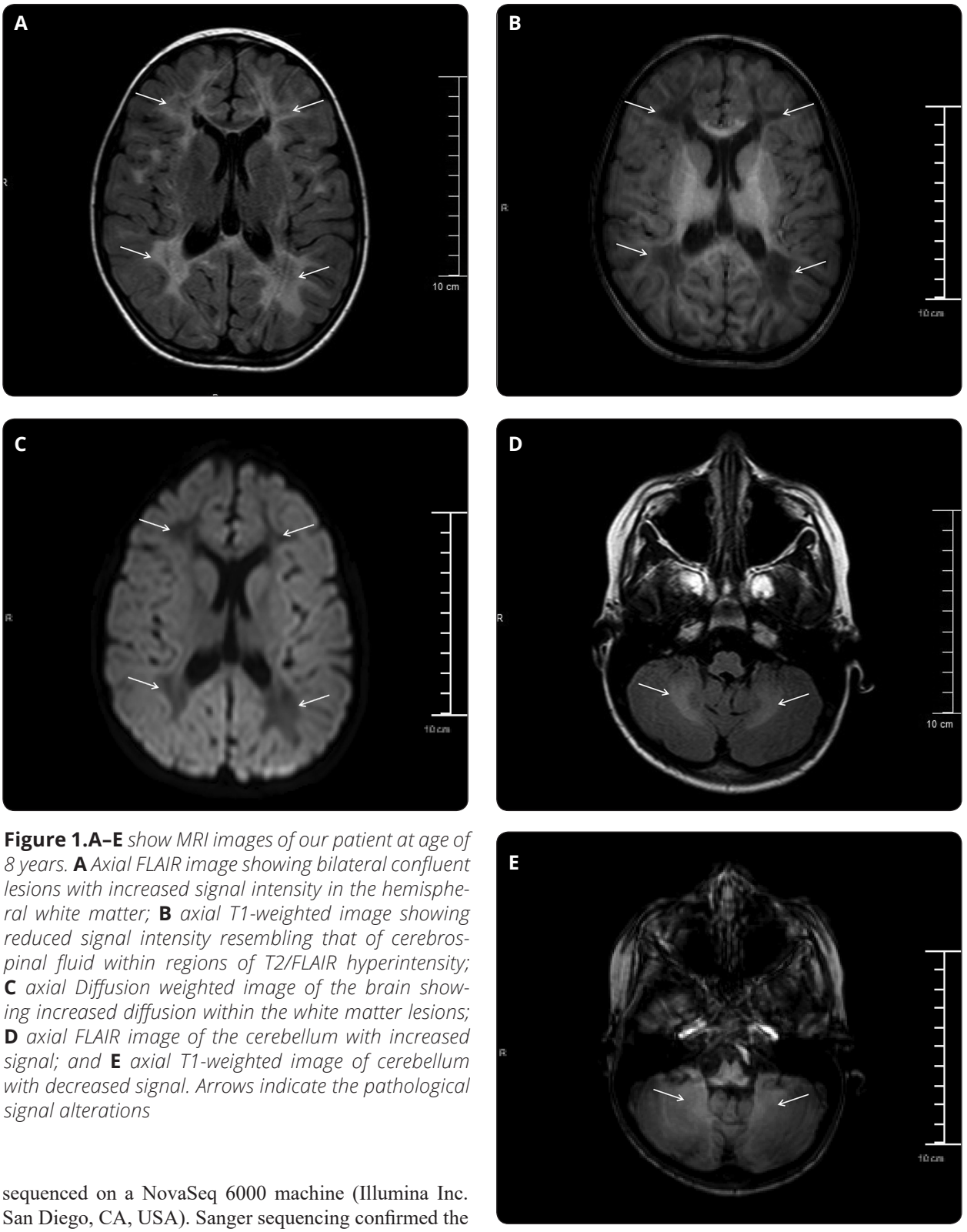

Figure 1.A–E *show MRI images of our patient at age of 8 years.* **A** *Axial FLAIR image showing bilateral confluent lesions with increased signal intensity in the hemispheral white matter;* **B** *axial T1-weighted image showing reduced signal intensity resembling that of cerebrospinal fluid within regions of T2/FLAIR hyperintensity;* **C** *axial Diffusion weighted image of the brain showing increased diffusion within the white matter lesions;* **D** *axial FLAIR image of the cerebellum with increased signal; and* **E** *axial T1-weighted image of cerebellum with decreased signal. Arrows indicate the pathological signal alterations*

sequenced on a NovaSeq 6000 machine (Illumina Inc. San Diego, CA, USA). Sanger sequencing confirmed the presence of variant detected by next generation sequencing (NGS).

Bioinformatic analysis was performed by using the VarSeq software (Golden Helix Inc., Bozeman, MT 59715, USA). A likely pathogenic mutation in homozygous state was detected in the 2/16 exon of the *EIF2B5* gene: NM_003907.3: c.203T>C, NP_003898.2: p.L68S,

on chromosome 3, position 183,854,407 T > C (PM2_Moderate, PP5_Supporting, PM3_Moderate, PP4_Supporting). Both asymptomatic parents were heterozygous carriers of the same mutation and non-consanguineous. Her asymptomatic older sister was not genetically tested at that time.

Discussion

van der Knaap et al[10] subdivide VWMD into six subgroups according to the age of onset (<12 mo, 1–<2y, 2–<4y, 4–<8y, 8–<18y, ≥18y). Our patient presented at 3-4 years of age placing her in the third subgroup. Thus far, she has had a slowly progressive gait disorder without cognitive decline. MRI of the brain showed typical features of VWMD (Figure 1). However, it is important to note that the MRI characteristics depend on the age of onset and disease duration as well as the course, likely explaining, at least in part, the increased diffusion within the confluent white matter regions in our case as opposed to the reported restricted diffusion predominantly found in young patients and short disease duration[10, 11]. The disease entity was confirmed by the detection of a likely pathogenic homozygous mutation within the *EIF2B5* gene using WES. The elF2B protein plays important roles in protein synthesis, and as a component of the integrated stress response pathway (ISR), regulates the rate of mRNA translation under cellular stress. Although no precipitating events were noted in our patient's history, a dysbalance in the IRS response might have contributed to the development of her VWMD[10]. Pathogenic variants can constitutively impair activity of the eIF2B protein and activate the ISR pathway[10].

The detected *EIF2B5* mutation has been reported in four other cases with VWMD[12–15]. The c.203T>C variant changes the hydrophobic leucine to the polar serine in the strongly conserved position of codon 68 in the EIF2B5 molecule. This mutation is located in the ε-subunit of the nucleotidyletransferase-like domain of EIF2B5 and does not affect the catalytic subunit of the guanine exchange activity of the protein. It remains to be explored as to how the p.L68S variant affects the severity of the VWMD phenotype since prior to our case, it has only been found in compound heterozygous state.

Until recently, only symptomatic treatments had been available in VWMD. However, based on the growing information concerning the involvement of ISR in disease pathogenesis, compounds targeting the impaired pathway have been proposed and several direct and indirect eIF2B activators are in preclinical stages of development[10]. One of them is guanabenz, an old α2-adrenergic antihypertensive drug that showed some benefits in the mouse model of VWMD[10, 16]. *In vivo* targeting of the pathogenic variant by CRISPR-CAS technology is also in early stages of development in the mouse model[17].

To allow an efficient collaboration among international VWMD experts, to develop trial designs with more homogeneous clinical subtypes and to use phenotype-adjusted outcome measures when studying cohorts with low numbers of patients, the Vanishing White Matter Consortium (VWM Consortium, www.vwmconsortium.org) was created[18]. This expert team developed a core protocol template for trials in VWMD. When the diagnosis for our patient was established, we contacted the leader of this consortium and registered our patient in the international registry (https://www.vwmconsortium.org/register-as-vanishing-white-matter-patient) with the intention of facilitating her participation in an inventive disease modifying study. After detailed information provided, the parents gave written consent and agreed to visit the Consortium center for participating in the ongoing guanabenz study with inclusion criteria matching to our patient (https://www.vumc.com/departments/center-for-children-with-white-matter-disorders/guanabenz-trial-for-vanishing-white-matter/a-first-trial-of-guanabenz-in-vanishing-white-matter.htm). Our team eagerly follow the disease evolution in this case, and we also hope that either guanabenz or another available study drug will beneficially modify the natural course of this severe condition in our patient.

ACKNOWLEDGEMENTS – Whole exome sequencing and bioinformatic analyses were performed in collaboration with iBioScience Ltd. and the Hungarian Centre for Genomics and Bioinformatics (GenBio) at the Szentagothai Research Centre of the University of Pecs. The authors thankfully acknowledge the support of Dr. Arpad Vadvari, who read the MRI images.

References

1. *Yue NC, Arnold AM, Longstreth WT, Elster AD, Jungreis CA, Leary DHO, et al.* Sulcal, ventricular, and white matter changes at MR imaging in the aging brain: Data from the cardiovascular health study. Radiology 1997;202:33-9. https://doi.org/10.1148/radiology.202.1.8988189
2. *Debette S, Markus HS.* The clinical importance of white matter hyperintensities on brain magnetic resonance imaging: systematic review and meta-analysis. BMJ 2010;341:c3666. https://doi.org/10.1136/bmj.c3666
3. *Wahlund LO, Barkhof F, Fazekas F, Bronge L, Augustin M, Sjögren*

M, et al. A new rating scale for age-related white matter changes-applicable to MRI and CT. Stroke 2001;32:1318-22. https://doi.org/ 10.1161/01.str.32.6.1318
4. *Mäntylä R, Erkinjuntti T, Salonen O, Aronen HJ, Peltonen T, Pohjasvaara T, et al.*Variable agreement between visual rating scales for white matter hyperintensities on MRI. Comparison of 13 rating scales in a poststroke cohort. Stroke 1997;28:1614-23. https://doi.org/10.1161/01.str.28.8.1614
5. *Visser PJ, Verhey FRJ, Hofman PAM, Scheltens P, Jolles J.* Medial temporal lobe atrophy predicts Alzheimer's disease in patients with minor cognitive impairment. J Neurol Neurosurg Psychiatry 2002;72:491-7. https://doi.org/10.1136/jnnp.72.4.491
6. *van der Flier WM, Barkhof F, Scheltens P.* Shifting paradigms in dementia. Toward stratification of diagnosis and treatment using MRI. Ann N Y Acad Sci 2007;1097:215-24. https://doi.org/10.1196/annals.1379.013
7. *Boyle PA, Yu L, Fleischman DA, Leurgans S, Yang J, Wilson RS, et al.* White matter hyperintensities, incident mild cognitive impairment, and cognitive decline in old age. Ann Clin Transl Neurol 2016;3:791-800. https://doi.org/10.1002/acn3.343
8. *Wardlaw JM, Valdés Hernández MC, Muñoz-Maniega S.* What are white matter hyperintensities made of? Relevance to vascular cognitive impairment. J Am Heart Assoc 2015;4:001140. https://doi.org/10.1161/JAHA.114.001140
9. *Silbert LC, Nelson C, Howieson DB, Moore MM, Kaye JA*. Impact of white matter hyperintensity volume progression on rate of cognitive and motor decline. Neurology 2008;71:108-13. https://doi.org/ 10.1212/01.wnl.0000316799.86917.37
10. *Taylor ANW, Kambeitz-Ilankovic L, Gesierich B, Simon-Vermot L, Franzmeier N, Caballero MAA, et al.* Tract-specific white matter hyperintensities disrupt neural network function in Alzheimer's disease. Alzheimers Dement 2017;13:225-35. https://doi.org/10.1016/j.jalz.2016.06.2358
11. *Kandemir Yilmaz M, Yalciner ZB, Cinar S, Unay D.* Evaluating the vascular risk factors using modified visual MRI rating scale in patients with memory decline. *Under-evaluation at Clinical Neuroscience (Ideggyógyászati Szemle)*
12. *Scheltens P, Erkinjunti T, Leys D, Waklund LO, Inzitari D, del Ser T, et al.* White matter changes on CT and MRI: an overview of visual-rating scales. European task force on age-related white matter changes. Eur Neurol 1998;39:80-9. https://doi.org/ 10.1159/000007921
13. *Scheltens P, Barkhof F, Leys D, Pruvo JP, Nauta JJ, Vermersch P, et al.* A semiquantative rating scale for the assessment of signal hyperintensities on magnetic resonance imaging. J Neurol Sci 1993;114:7-12. https://doi.org/10.1016/0022-510x(93)90041-v
14. *Whitwell JL, Peterson RC, Negash S, Weigand SD, Kantarci K, Ivnik RJ, et al.* Patterns of atrophy differ among specific subtypes of mild cognitive impairment. Arch Neurol 2007;64:1130-8. https://doi.org/10.1001/archneur.64.8.1130
15. *Fazekas F, Chawluk JB, Alavi A, Hurtig HI, Zimmerman RA.* MR signal abnormalities at 1.5 T in Alzheimer's dementia and normal aging. AJR Am J Roentgenol 1987;149:351-6. https://doi.org/10.2214/ajr.149.2.351
16. *Yalciner BZ, Kandemir M, Taskale S, Tepe SM, Unay D.* Modified visual magnetic resonance rating scale for evaluation of patients with forgetfulness. Can J Neurol Sci 2019;46:71-8. https://doi.org/10.1017/cjn.2018.333
17. *Koikkalainen JR, Rhodius-Meester HFM, Frederiksen KS, et al; Alzheimer's Disease Neuroimaging Initiative.* Automatically computed rating scales from MRI for patients with cognitive disorders. Eur Radiol 2019;29:4937-47. https://doi.org/10.1007/s00330-019-06067-1