Original Article

Deep White Matter Lesions with Persistent Diffusion Restriction on MRI as a Diagnostic Clue: Neuroimaging of a Turkish Family with Hereditary Diffuse Leukoencephalopathy with Spheroids and Literature Review

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Abstract

Background: Hereditary diffuse leukoencephalopathy with spheroids (HDLS), first described in 1984 is a rare disorder. Generally, it presents at adulthood with dementia, motor impairment, extrapyramidal abnormalities, and epilepsy. Definitive diagnosis is made by brain biopsy. Neuroimaging studies have revealed confluent white matter lesions predominantly in the frontal lobes, corpus callosum, and corticospinal tracts on conventional magnetic resonance imaging. Only a few reports showed diffusion restriction in the cerebral white matter; furthermore, rarer reports emphasized persistent foci of diffusion restriction as a diagnostic imaging marker. **Objective:** Herein, we have aimed to illustrate the first biopsy-proven Turkish HDLS pedigree consisting of 18 persons in 3 generations which contained 4 affected individuals. **Materials and Methods:** Four individuals in the pedigree of HDLS [two affected patients (patient III-1 and patient III-2) and two unaffected individuals (patient II-4 and patient III-5)] were investigated with conventional MRI and Diffusion-weighted imaging (DWI) using 1.5 Tesla (T) scanner. All four individuals were evaluated via neurological examinations and Mini-Mental State Examination. Brain biopsy study was performed on patient III-2. Finally, an extensive literature review involving pathology investigations and neuroimaging studies of HDLS patients was conducted. **Results:** DWIs of two investigated patients showed deep white matter lesions with persistent diffusion restriction. Computed tomography imaging showed punctate mineralization in the lesions. Biopsy specimens of patient III-2 demonstrated axonal spheroids which were typical for HDLS. **Conclusions:** Via the presentation of our pedigree and literature review, we suggest HDSL as a first-line differential diagnosis in patients with undiagnosed adult-onset familial leukoencephalopathy, in particular, those with MRI lesions of frontal white matter and centrum semiovale associated with foci of diffusion restriction and mineralization. Finally, we think that the persistence of the diffusion restriction in deep white matter lesions should be kept in mind as a crucial neuroimaging sign for HDLS.

Keywords: Diagnosis, diffusion-weighted imaging, hereditary diffuse leukoencephalopathy with spheroids, pathophysiology, persistent diffusion restriction, spotty calcification

INTRODUCTION

Leukoencephalopathy with neuroaxonal spheroids is an extremely rare cerebral white matter disease, which can manifest either in a hereditary manner or sporadically. Its manifestations include behavioral or mood changes, dementia, motor impairment, walking impairment, and epilepsy. In contrast to the majority of other leukodystrophies generally manifesting at childhood, the symptoms of hereditary diffuse leukoencephalopathy with spheroids (HDLS) frequently present at adulthood.[1,2] In 1971, Seitelberger first described the term "neuroaxonal spheroid" as neuroaxonal dystrophy that affects cerebellar and cerebral white matter, with resultant secondary demyelination.[2] Thereafter, several pathological investigations have revealed spheroids both in white and grey matter, and have suggested spheroids as a marker of the cerebral degeneration process.[3,4] Since then, a limited number of affected pedigrees and very few sporadic cases have been reported. The disease was demonstrated in a Swedish family (17 cases) in 1984 by Axellson *et al*. In that study, brain biopsy specimens obtained from four patients were also investigated which revealed axonal

dilatations (spheroid) as a characteristic feature.[3] In another large case series, Baba *et al*. identified six cases that were affected within the pedigree of a generation.[4]

Remarkably, the diagnosis of HDLS still constitutes strictly a challenging issue, and definitive diagnosis depends on pathological examination.[5] However, via the aid of the recent studies and genetic characterization of the disease, genetic tests have been proposed as a critical, alternative diagnostic method. Nevertheless, due to the limited awareness among physicians, it can be predicted

that this disease constitutes a crucial underdiagnosed condition. Such that a case series at the Mayo clinic showed that a majority of these patients had received misdiagnoses such as Alzheimer's disease, frontotemporal dementia, atypical parkinsonism, multiple sclerosis and small vessel ischemic disease.[6] However, there are some clinical and neuroimaging clues leading to suspicion of the diagnosis of this entity. Classically, white matter lesions predominantly in the frontal lobes, corpus callosum and pyramidal tracts may suggest HDSL. Nevertheless, these findings are not specific and diagnostic as many other diseases may manifest with these lesions on MRI. However, recently, a limited number of reports have suggested diffusion-weighted imaging (DWI) as an important tool for diagnosis.[7-16]

Herein, a Turkish HDLS pedigree with four affected individuals was investigated with regards to clinical, pathological and brain imaging findings (conventional magnetic resonance imaging [MRI], DWI, proton MR spectroscopy [MRS] and computed tomography [CT]). Particularly, the diagnostic importance of DWI and repeated use of this method at follow-up of these patients are discussed.

MATERIALS AND METHODS

Four individuals in the pedigree of HDLS [two affected patients (patient III-1 and patient III-2) and two unaffected individuals (patient II-4 and patient III-5)] were investigated with conventional MRI and DWI using 1.5 Tesla (T) scanner. MRS was performed for only one of the patients (III-2). Patient III-2 and patient III-8 from this pedigree were investigated using computed tomography (CT) scans of the brain [Other imaging methods could not be applied in case III-5 due to social limitations]. All four individuals were evaluated via neurological examinations and Mini-Mental State Examination (MMSE; Table 1). Brain biopsy study was performed on patient III-2. Biopsy specimens were processed in formalin and stained with hematoxylin-eosin. Also, immunoreactivity to neurofilament protein was investigated. They were stained for CD-68, GFAP, and PAS. Finally, an extensive literature review involving pathology investigations and neuroimaging studies of HDLS patients was conducted.

RESULTS

Sample case (III-2)

A 35-year-old woman consulted our service with the complaints of memory impairment, difficulty in walking and speech impediment with a history of progression within a year. Because of depression, she had been referred to many psychiatric specialists previously. Cranial MRI, performed 7 months before in another clinic, had shown bilateral T2 hyperintense lesions in the corona radiata, centrum semiovale, and pericallosal areas with a hyperintense appearance on DWI sequences which were reported to be suggestive of multiple sclerosis (MS) lesions [Image 1a]. At that time, no treatment was attempted and in the interval period, her walking had worsened, such that 3 months after that admission, she was able to walk without support for only a short distance. On admission to our clinic, she was only able to perform single-stage commands, and her speech was characterized
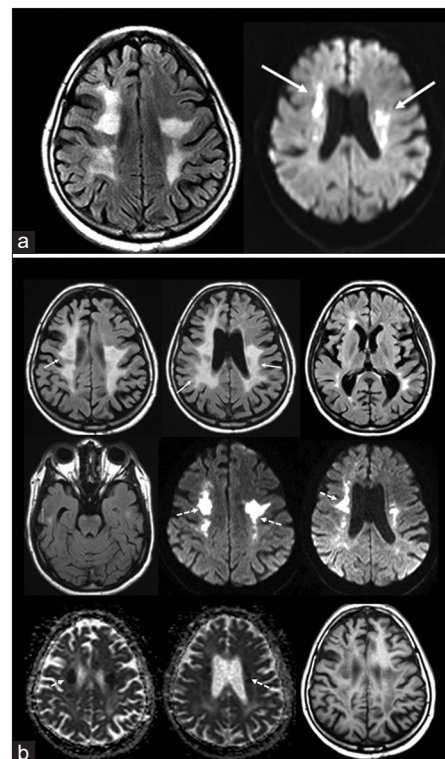

Image 1: (a) Initial MRI images of the patient III-2, performed 7 months prior to admission to our clinic. a, Periventricular T2 hyperintense lesions. b, DWI showing a high signal in the periventricular white matter. (b) Magnetic resonance imaging (MRI) of the patient III-2, performed at admission to our clinic (7 months after the first MRI). FLAIR images show bilateral confluent deep periventricular hyperintense regions (arrows) and U-fibers are relatively spared. DWI showing patchy strong-signal areas (dotted arrows). ADC showing low-signal areas (dotted arrows)

| Table 1: Clinical and laboratory findings of four individuals (two of whom are affected)					
	Age	Symptoms	MMSE	Neurologic Examination	MRI Findings
Sample patient (III-2)	35	Memory impairment, palilalia, gait impairment	6	Severe cognitive impairment, pyramidal, extrapyramidal defects	White matter lesions showing diffusion restriction in bilateral frontal white matter and corona radiata
Brother (III-1)	28	Memory impairment, palilalia	19	Cognitive impairment, palilalia, minimal extrapyramidal defects	Similar lesions showing diffusion restriction
Sister (III-5)	40	No symptoms	22	Normal findings	Chronic white matter lesions
Mother (II-4)	70	No symptoms	21	Normal findings	Normal

by palilalia. Her walking was wide with small steps and on extrapyramidal examination bradykinesia, rigidity and postural instability were observed. She experienced "freezing" episodes during turning or initiation of walking. Cranial MRI revealed T2 hyperintense foci with restricted diffusion in the bilateral deep frontal white matter [Image 1b]. Proton MR Spectroscopy (MRS) was also performed which showed increased choline and lactate with decreased NAA [Image 5]. On history, her father and sister had died after a clinical course due to the progression of similar symptoms those of the patient's. Her father and mother were first-degree relatives which suggested the hereditary leukoencephalopathies in the foreground among differential diagnosis. In laboratory

investigations, routine blood tests were normal (hemogram, biochemistry, folate, B_{12}, thyroid hormones, and lipids). Lysosomal enzymes, peroxisomal enzymes, and blood lactate and pyruvate were within the normal ranges. With the support of radiological findings (persistent diffusion restriction, wide bilateral white matter lesions, and protection of U fibers), we considered the diagnosis of HDLS in the forefront and after informing about the disease and process, a neurosurgical biopsy was recommended to the patient and her relatives. Together with navigational MRI, multiple biopsies were performed from the lesions with diffusion restriction. Biopsy specimens demonstrated axonal spheroids. Additionally, there were scattered GFAP-expressing large reactive astrocytes. CD68-expressing macrophages contained yellow-brown granules that tested positive for PAS staining [Figure 2]. Taken together, the diagnosis of HDLS was made, and the patient
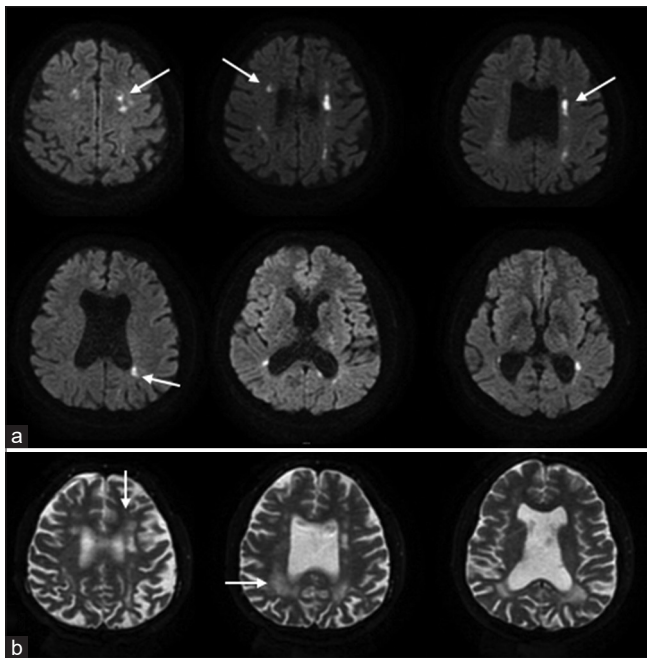

Image 2: Initial MRI findings of case III-1-Six months after the onset of symptoms. (a) DWI showing minute signals (arrows). (b) T2-weighted images display deep periventricular confluent lesions (arrows)
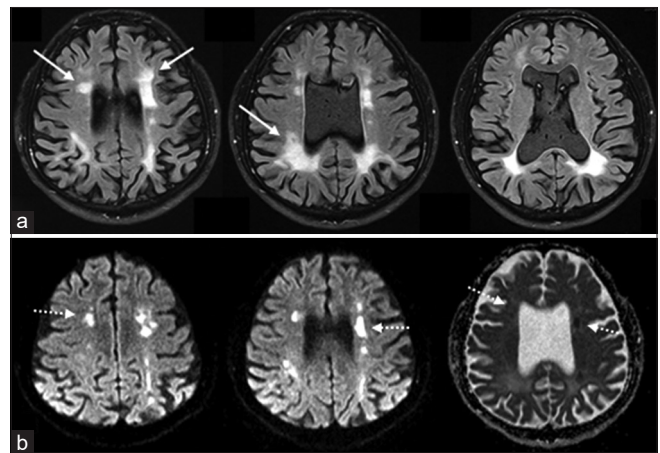
Image 3: Second MRI of case III-1 obtained 2 months after the first MRI. (a) FLAIR images show deep periventricular confluent lesions (arrows). (b) DWI and ADC images display persistent diffusion restriction (dotted arrows)
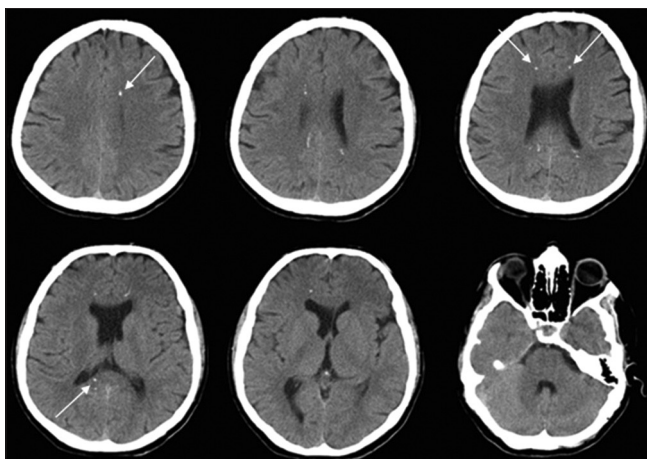
Image 4: Non-enhanced computed tomography of case III-8 reveals multiple tiny calcifications in the frontally predominant periventricular area (arrows), sparing the basal ganglia
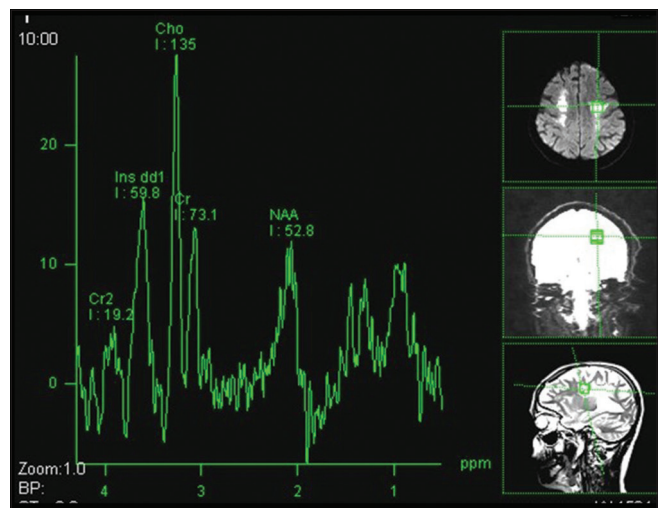
Image 5: Proton MR Spectroscopy (MRS) (TE: 35 msec) of the patient III-2. There is an increase in the choline and lactate levels and a decrease in NAA
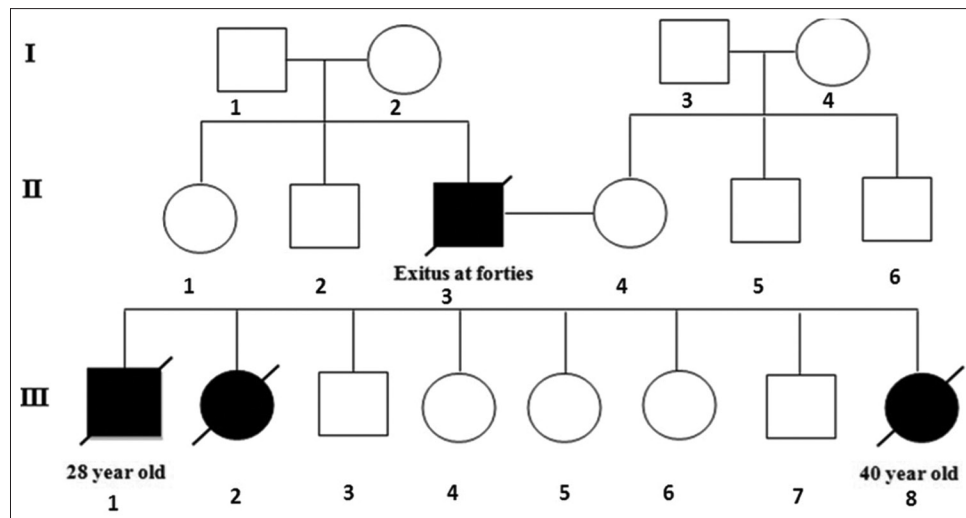
Figure 1: The pedigree of Turkish individuals of hereditary diffuse leukoencephalopathy with spheroids

Figure 2: (a) Axonal spheroids (arrows) in white matter (hematoxylin-eosin stain, ×40 magnification). (b) Axonal spheroids (arrows) expressing neurofilament protein (NFP, ×40)

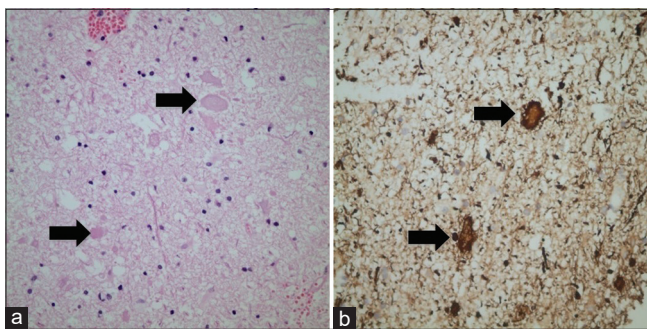and her relatives were informed about the disease and risk for genetic inheritance at follow-up, levodopa was initiated for parkinsonism symptoms which provided only a slight improvement in her gait.

Pathology findings of case III-2

Histologically, cortical laminar architecture was well preserved without an apparent neuronal loss. The axonal spheroids were found in white matter. They were not seen in the areas close to the cortex. They appeared as dark round swollen axons after hematoxylin-eosin staining [Figure 2a]. The spheroids were immunoreactive for neurofilament protein [Figure 2b]. There were scattered GFAP-expressing large reactive astrocytes. CD68-expressing macrophages contained yellow-brown granules that tested positive for PAS staining.

Other patients and healthy individuals of the pedigree

After investigation of the pedigree [Figure 1] of sample patient III-2, several patients with HDLS were identified. Patient II-3 had died following the progression of symptoms of progressive memory impairment and walking disturbance in his 40s. There were no other clinical or radiological data about this patient. Patient III-8, a 40-year-old female, had the symptoms of amnesia, lisping, and walking impediment. Cranial CT was evaluable which showed multiple frontally predominant spotty calcifications.

Patient III-1, a 28-year-old male, had symptoms of progressive walking impediment and memory impairment for the past 6 months. His MMSE score was 19. On neurological examination, his speech was characterized by palilalia, and mild-level parkinsonism signs were present. Motor, sensory, and cerebellar examinations were within normal limits. However, deep tendon reflexes were brisk and muscle tone in the lower extremities were increased compatibly with spasticity. Two cranial MRI scans which were performed between 2-month interval showed T2 hyperintense areas in bilateral corticospinal tracts (cs, cr) and persistent punctate-style diffusion-restricted areas [Images 2 and 3]. Cranial CT again showed multiple periventricular spotty calcification loci [Image 4].

One unaffected individual (III-5, except for a migrainous headache), a 40-year-old female, did not have any neurological symptoms or abnormal findings on neurological examination. Her MMSE score was 22, but according to her sociocultural range, her cognitive status was evaluated to be normal. MRI showed non-specific deep white matter hyperintense lesions with facilitated diffusion. Another unaffected patient (II-4), a 70-year-old female, did not have any symptoms or aberrant neurological signs. Cranial MRI was unremarkable.

Discussion

HDLS is a cerebral white matter disease with autosomal dominant inheritance but with variable penetrance. Latest studies have demonstrated a mutation in the tyrosine kinase domain of the colony stimulating factor 1 receptor (CSFR1) protein as the responsible gene in the pathogenesis.[17-19] Clinical onset often involves symptoms such as dementia, depression, motor deficits, extrapyramidal signs particularly parkinsonism similarly in our case.[20] Arrestingly, due to its frequent association with parkinsonian findings, it has also

Table 2: A literature review of magnetic resonance imaging studies with a diffusion sequence in patients with hereditary diffuse leukoencephalopathy with spheroids

Reference	Cases mentioned in the reports	Sex	Age at onset	MRI interval after symptom	MRI	Follow-up DWI
Freeman *et al*.[10]	Case 2	F	31	-	Bilateral frontally dominant periventricular deep subcortical T2 hyperintense spots, dif r	NA
	Case 3	F	49	6 mo	Bilateral frontal-subcortical confluent hyperintense spots, spotty dif r	NA
	Case 4	F	38	-	Bilateral frontally dominant periventricular deep subcortical T2 hyperintense spots, dif r	NA
Maillart *et al*.[36]	Patient 1	F	33	6 mo	Bilateral frontal periventricular areas, corpus callosum, punctate dif r	NA
	Patient 2	F	33	6 mo	Bilateral frontal periventricular areas, corpus callosum, punctate dif r	NA
Mateen *et al*.[5]	A single case	F	24	7 mo	Bilateral periventricular subcortical T2 hyperintense spots, spotty dif r	Persistent dif r 19 w later
Kleinfeld *et al*.[9]	Patient 1	F	39	2 y	Bilateral corticospinal hyperintense spots	NA
	Patient 4	F	30	11 mo	Bilateral corticospinal hyperintense spots, extensive dif r	NA
	Patient 5	F	47	2 y	Bilateral corticospinal hyperintense spots, spotty dif r	NA
Terasawa *et al*.[17]	A single case	F	51	18 mo	Mild frontal atrophy, spotty dif r	Persistent and increasing dif r 13 and 22 mo later
Battisti *et al*.[50]	Patient 3	M	37	1 y	Brain volume loss, subcortical white matter hyperintensity of both frontal lobes, spotty dif r	NA
Bender *et al*.[37]					Periventricular dif r was seen in three of four patients with DWI	Persistent dif r was seen over a period of 9 months in a patient
Konno *et al*.[14]	Case 2	F	30	-	Periventricular dif r	NA
	Case 3	F	27	-	Periventricular dif r	NA
Codjia *et al*.[15]					Periventricular dif r was seen in six of the eight patients who were investigated by DWI (Details about the patients' data were not included).	NA
Miura *et al*.[16]					Periventricular dif r was reported to be present in eight of the eleven patients who were investigated by DWI (Details about the patients' data were not included).	NA
Current study	Case III-2	F	35	1 y	Bilateral confluent corticospinal hyperintense spots, punctate dif r	Persistent and increasing dif r 7 mo later
	Case III-1	M	28	6 mo	Bilateral confluent corticospinal hyperintense spots, punctate dif r	Persistent and increasing dif r 2 mo later

Abbreviations: Sym on: symptom onset, DWI: Diffusion-weighted imaging, dif r: diffusion restriction, F: female, M: male, mo: months, y: years, w: weeks, NA: not available

been regarded among the differential diagnosis of atypical parkinsonism clinics.[6,21,22] Our sample case (III-2) had initially presented with a depressive mood change, memory impairment, walking impediment and palilalia. At follow-up, her symptoms progressed, and the neurological examination showed severe pyramidal and extrapyramidal signs. Of note, she had also received the misdiagnosis of MS in another center, previously. On the other hand, DWI findings of persistent diffusion restriction suggested HDLS in the forefront and finally, pathological investigations of the brain biopsy specimens (in our index case) provided the definitive diagnosis

of HDLS. Since 1971, numerous pathological studies including brain specimens on patients with HDLS have been published. Data from these studies revealed some characteristic features for pathology examinations. Some reports showed pigment accumulation in CD-68 positive macrophages and to a lesser degree in astrocytes.[10,21,23-30] Although iron-positive cells can be detected, a large amount of this pigment was found to be carried by a lipoprotein termed *seroid*.[4] However, the most characteristic pathological finding in HDLS patients has been defined as the formation of numerous axonal spheroids. Our patient's pathology investigations were also meeting all the

defined criteria of HDLS[29,30] consisting of axonal spheroids and pigmented macrophages [Figure 2].

However, due to its rarity and extremely low awareness, the diagnosis of HDLS generally constitutes a major challenge for physicians. Such that, it has been reported that HDLS patients may frequently receive misdiagnoses such as frontotemporal dementia, MS, cerebrovascular disease, adult-onset metachromatic leukodystrophy, Krabbe disease and X-linked adrenoleukodystrophy.[6] Remarkably, Wrong *et al*. stated that 26.9% of HDLS patients would receive a diagnosis of a probable behavioral variant of frontotemporal dementia if the criteria from the 2011 Frontotemporal Dementia Consensus Statement were used.[31] Ergo, we think that the detailed neuroimaging findings of our HDLS pedigree may give substantial contributions regarding the diagnostic problems of this entity as well as management problems of the patients with clinical suspicion of HDLS.

On this topic, a crucial point of discussion may be the responsible pathophysiology of this HDLS. Although there is an increasing number of research and documentations about this entity, many points of the pathophysiological mechanisms and progression way of this disease still remain to be elucidated. It has been acknowledged that the disease affects equally both axons and myelin sheaths predominantly in the frontal lobe, corpus callosum, and corticospinal descending tracts.[1] Nonetheless, it is not clear if myelin and axonal damage are separate processes or mutually supportive of each other, but some pathological studies support the view of secondary myelin loss due to primary axonal degeneration.[1,4,32] Nonetheless, some other researchers believe that axonal damage is induced by demyelination via an autonomous neurodegeneration process.[33] Besides, the decreased number of oligodendroglia has been reported, particularly in the peripheral portion of the lesion sites. On the basis of these observations, the hypothesis of secondary myelin and axonal damage due to an underlying oligodendroglial defect has formed another explanation. They also supported the view of the reactive nature of axonal spheroids in contrast to the hypothesis describing them as a product of the dystrophic process.[4,34] On the other hand, the most elaborated view is that HDLS is a primary microglial disorder developing as a result of mutations in the tyrosine kinase domain of the CSF1R gene.[19,30]

Overall, the most important aspect of our study was the detailed illustration of the neuroimaging data. Cranial MRIs of our sample case revealed periventricular lesions showing persistent diffusion restriction on DWIs performed within 7-month interval. This finding together with the limited neuroimaging literature data helped to focus on the diagnosis of HDLS. Furthermore, neuroimaging evaluation of her affected brother (case III-1) also disclosed similar findings of persistent diffusion restriction on MRIs within 2 months interval. To date, various neuroradiological methods such as MRI, single-photon emission computerized tomography, positron emission tomography, and BT have been applied to patients with HDLS. Data from the

literature on single-photon emission computerized tomography suggests non-specific findings such as frontotemporal or frontoparietal hypoperfusion; positron emission tomography studies showed generalized hypometabolism and frontotemporal hypometabolism.[4,10,28] Besides, the results of the conventional MRI studies showed that periventricular, callosal, deep white matter patch or confluent-type lesions predominantly in the frontal or frontoparietal areas have been reported to be seen in patients with HDLS.[4,10,28,35] Remarkably, in a crucial MRI study including 20 patients with HDLS, Sundal *et al*. reported that the lesions in the deep and subcortical white matter areas were found to exist at the rate of 93% in these patients. However, remarking that these lesions may be seen in numerous other diseases, it has been several times stated that conventional MRI was a non-specific method and it was unsuitable for diagnostic purposes of HDLS.[5] However, at this point, the use of DWI in clinical practice has begun to be applied in the near future and the results gave promising findings in the diagnosis of various cerebral white matter diseases other than cerebrovascular diseases.[5,9,10,14-17,36-38] Recently, several studies investigating the utility of DWI for the diagnosis of HDLS have been published[5,9,10,14-17,36,39] [Table 2]. In the three of these reports, specific persistent diffusion restriction was particularly noted as in our cases.[5,17,37] The case report of Mateen *et al*. has shown persistent diffusion restriction and low ADC signals on two MRI scans which has been performed intermittently at 19-week interval.[5] Another study of two cases showed frontal white matter dominant involvement with diffusion restriction on DWI scans.[5] Nonetheless, in that study, data regarding a follow-up DWI of these patients has not been mentioned. In another recent report, in a patient with genetically diagnosed HDLS, persistent and increasing diffusion restriction was detected on two DWI scans recorded at 22-week interval.[17] The same study showed increased signals on images corresponding to axonal fibers within sagittal DWI sections which were particularly stated to differ from the localization of U-fibers, frequently reported to be preserved in HDLS.[17,40,41] Restricted diffusion was suggested either by high-grade myelin edema accompanied by leukodystrophy loci or by demyelination spots.[5,42] As a mechanistic explanation, the appearance of persistent diffusion restriction was suggested to be mediated by cytotoxic edema, which can be explained by neurodegenerative processes.[5,42] The same report has suggested that abnormal DWI findings were a marker of the initial phase of the disease, whereas hyperintense loci on T2 MRI sequences were a marker of the late phase of the disease.[5] Our present work suggests that persistent diffusion restriction may be a very crucial and a characteristic finding for HDLS; nonetheless. However, based on our DWI findings of persisting diffusion restriction during the distinct phases of the disease, we do not support the view that restriction on DWI is associated with the early phase of the disease. For example, our cases exhibited T2 hyperintense foci throughout the overall course of the disease [not only in the late phase, (patient III-2)], incompatible with the hypothesis suggested by Mateen *et al*.[5] Of note, U-fibers were spared in MRIs of our patients in accordance with literature data.[4,17,41] Subcortical U-fibers, also known as

short association fibers, represent connections adjacent gyri of the brain, in the very outer parts of the subcortical white matter.[43] These regions are also known to be among the last parts to myelinate of the brain (as late as third and fourth decade) and these localizations have been emphasized to have very slow myelin turnover[44] This feature of the specifically sparring of the white matter regions (U-fibers), having slow myelin turnover, may support the view of an underlying myelin abnormality as a major pathogenetic mechanism of HDLS. This may also indirectly support the hypothesis of intramyelin edema as a probable mechanism of persistent diffusion restriction rather than cytotoxic edema which has been suggested previously.[5] Of note, regions of restricted diffusion were not distributed diffusely rather, they were localized in a punctuate shape. In a unique report investigating the pathological specimens of 5 HDLS patients, it was emphasized that despite the diffuse distribution of white matter degeneration and astrogliosis, activated microglial cells were spatially restricted.[41] We hypothesize that this emphasized distinct distribution of activated microglia cells may correspond to the persistent diffusion restricted punctate areas, which may suggest a possible microglia activation-mediated myelin damage as the mechanism underlying. In addition, reports suggesting the view of microglia dysregulation as the primary cause of the disease, based on the role of CSFR1 in microglia regulations[30,45,46] may support our hypothesis. In another aspect, Konno *et al.*, based on their observation of involvement of brain calcifications nearly similar to the embryonic microglial distributions, have suggested that microglial dysfunction might have a causal relationship with the development of calcifications.[14] In addition to this hypothesis, we also hypothesize that regions of decreased microglial cells, shown in pathological specimens, might be representing the persistent diffusion restricted regions demonstrated in DWIs. Nonetheless, these hypotheses are surely warranted to be clarified in the future studies of multiple targeted stereotactic biopsy specimens from diffusion restricted and unrestricted localizations.

Another important result of our report is that two cranial CT scans of the patients III-2 and III-8 showed multiple spotty calcifications. In the literature, a case of HDLS was reported, with the consistent finding of calcification around the lesion site.[14,17] Multiple calcifications were reported in five of six CT scans from different patients in that study. In the same study, calcifications were reported to be dominantly distributed in white matter adjacent to the anterior horn of the lateral ventricles. Furthermore, in one individual, calcification was confirmed using pathological investigation. In the discussion section, based on the knowledge that CSFR1 pathway is involved in the reorganization of the osteoclast cytoskeletal cycle,[41] they suggested a possible defect in the CSFR1 signaling pathway to be responsible for these foci of calcifications white matter lesions. Neuroimaging finding of spotty calcification has also been emphasized in some recently published other reports as an important sign of HDLS.[16,47,48] A major limitation in our report may be that

genetic investigations were unavailable avoiding to deduce conclusions in this regard. Hence, we plan to perform genetic analyses in the ongoing phase of the study. However, although we cannot evaluate the responsible gene as well as detailed mechanisms of these calcifications in HDLS, we think that re-demonstration of this finding in CT may give substantial perspectives (considering the rarity of the published cases) for diagnostic purposes during the evaluation of patients with pre-diagnosis of HDLS.

MRS data acquired from our sample case showed increased choline and lactate with decreased NAA [Image 5]. In the two published study mentioning MRS in HDLS, an increase in the choline/creatine ratio in the frontal subcortical white matter of a patient with HDLS,[10,24,37,49] whereas a lactate peak was reported in only one study[37] as well as NAA decrease in others.[37,49,50] Taken together, the results of the MRS findings in our case were compatible with the limited literature data. Choline was shown to be a precursor of acetylcholine and cell membrane components. In MRS, choline is a marker of cellular membrane turnover and is therefore known to be upregulated in conjunction with neoplasms, demyelination or gliosis. This increased ratio (choline/creatine) detected in our sample case and in some published studies can be explained as a result of the active demyelination process. However, we believe that MRS findings cannot be a specific marker of HDLS.

In conclusion, herein, we have demonstrated neuroimaging findings of persistent diffusion restriction in two cases in a Turkish pedigree of HDLS. Very few studies have been published reporting diffusion restriction in HDLS patients.[5,9,10,17,36,37] Furthermore, in only two reports, persistent diffusion restriction has been demonstrated on follow-up DWI scans, whereas other studies did not involve any follow-up MRI scans.[5,17] Considering that the literature data illustrating persistent diffusion restriction is extremely rare, we think that the present report provides crucial conclusions remarking persistent diffusion restriction as a useful sign for the diagnosis of HDLS. Therefore, we suggest performing follow-up DWIs as an easy and practical alternative method during the evaluation processes of patients with suspicion of HDLS. This mysterious finding of persistent diffusion restriction may also add crucial perspectives to explain the pathogenesis of HDLS which surely need to be clarified in future studies. We suggest that the primary myelin damage may be the major process underlying pathogenetic mechanisms, we also support the consideration of that microglia-mediated damage may be a critical process. Finally, we think that multiple, spotty calcified lesions on CT should always be kept in mind as a crucial neuroimaging sign of HDLS.

Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form the patient(s) has/have given his/her/their consent for his/her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published

and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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Conflicts of interest

There are no conflicts of interest.

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