

Evaluation of a New Criterion for Detecting Prion Disease With Diffusion Magnetic Resonance Imaging

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 Supplemental content

IMPORTANCE Early diagnosis is a requirement for future treatment of prion diseases. Magnetic resonance imaging (MRI) with diffusion-weighted images and improved real-time quaking-induced conversion (RT-QuIC) in cerebrospinal fluid (CSF) have emerged as reliable tests.

OBJECTIVES To assess the sensitivity and specificity of diffusion MRI for the diagnosis of sporadic Creutzfeldt-Jakob disease (sCJD) with a new criterion (index test) of at least 1 positive brain region among the cortex of the frontal, parietal, temporal, and occipital lobes; the caudate; the putamen; and the thalamus.

DESIGN, SETTING, AND PARTICIPANTS This diagnostic study with a prospective and a retrospective arm was performed from January 1, 2003, to October 31, 2018. MRIs were collected from 1387 patients with suspected sCJD consecutively referred to the National Prion Disease Pathology Surveillance Center as part of a consultation service.

INTERVENTION Magnetic resonance imaging. Four neuroradiologists blinded to the diagnosis scored the MRIs of 200 randomly selected patients. One neuroradiologist scored the MRIs of all patients.

MAIN OUTCOMES AND MEASURES Sensitivity and specificity of the index test compared with currently used criteria and CSF diagnostic (improved RT-QuIC, 14-3-3, and tau CSF tests).

RESULTS A total of 872 patients matched the inclusion criteria (diffusion MRI and autopsy-confirmed diagnosis), with 619 having sCJD, 102 having other prion diseases, and 151 having nonprion disease. The primary analysis included 200 patients (mean [SD] age, 63.6 [12.9] years; 100 [50.0%] male). Sensitivity of the index test of 4 neuroradiologists was 90% to 95% and superior to sensitivity of current MRI criteria (69%-76%), whereas specificity was 90% to 100% and unchanged. Interrater reliability of the 4 neuroradiologists was high ($\kappa = 0.81$), and individual intrarater reliability was excellent ($\kappa \geq 0.87$). The sensitivity of the index test of 1 neuroradiologist for 770 patients was 92.1% (95% CI, 89.7%-94.1%) and the specificity was 97.4% (95% CI, 93.4%-99.3%) compared with a sensitivity of 69.8% (95% CI, 66.0%-73.4%; $P < .001$) and a specificity of 98.0% (95% CI, 94.3%-99.6%; $P > .99$) according to the current criteria. For 88 patients, index test sensitivity (94.9%; 95% CI, 87.5%-98.6%) and specificity (100%; 95% CI, 66.4%-100%) were similar to those of improved RT-QuIC (86.1% [95% CI, 76.5%-92.8%] and 100% [95% CI, 66.4%-100%], respectively). Lower specificities were found for 14-3-3 and tau CSF tests in 452 patients.

CONCLUSIONS AND RELEVANCE In this study, the diagnostic performance of diffusion MRI with the new criterion was superior to that of current standard criteria and similar to that of improved RT-QuIC. These results may have important clinical implications because MRI is noninvasive and typically prescribed at disease presentation.

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Considerable progress has been made in the diagnosis of sporadic Creutzfeldt-Jakob disease (sCJD)¹⁻⁵ using magnetic resonance imaging (MRI).⁶⁻¹³ Initially, CJD diagnostic criteria comprised only the presence of abnormal signal in the caudate and putamen because of the low sensitivity of conventional MRI sequences in detecting cortical abnormalities.^{14,15} Subsequently, Zerr et al¹⁶ updated sCJD diagnostic criteria, adding MRI signal hyperintensities in the cortex of at least 2 lobes, excluding the frontal lobes on diffusion-weighted imaging (DWI) and fluid-attenuated inversion recovery (FLAIR). These criteria were adopted for national surveillance purposes by many countries, including the US; they are referred to in this article as the current standard.¹⁷ However, these criteria might be too conservative, missing the diagnosis of patients with frontal cortex and/or only 1 lobe involvement or with only the caudate or putamen affected.

Progress has also been made with cerebrospinal fluid (CSF) tests. Satisfactory sensitivity (70%-85%) but low specificities (40%-74%) have been reported for 14-3-3 and tau.^{18,19} Recently, an improved version of real-time quaking-induced conversion^{20,21} (RT-QuIC, also known as IQ-CSF or second-generation RT-QuIC) showed a remarkable increase in sensitivity with no loss of specificity, reaching a sensitivity of 95% and a specificity of 100%.²²⁻²⁴ This study assessed the sensitivity and specificity of diffusion MRI for the diagnosis of sCJD with a new criterion (index test) of at least 1 positive brain region among the cortex of the frontal, parietal, temporal, and occipital lobes; the caudate; the putamen; and the thalamus and compared the index test's performance with that of currently used criteria. We also compared diffusion MRI diagnostic performance with CSF test diagnostic performance.

Methods

This diagnostic study used MRIs from 1387 patients with suspected sCJD that were consecutively collected by the National Prion Disease Pathology Surveillance Center (NPDPS) in Cleveland, Ohio, from January 1, 2003, to October 31, 2018, following prospective and retrospective approaches. The prospective approach included living patients whose MRIs were collected through an MRI consultation service starting from 2009. The retrospective approach included the MRI retrieval of autopsied individuals referred to the NPDPS. Inclusion criteria were MRI study with DWI and apparent diffusion coefficient (ADC) maps available and autopsy diagnosis made at the NPDPS. Clinical records of patients without autopsy performed at the NPDPS were not available. Written informed consent was provided by the patient or legal next of kin for autopsy through the Centers for Disease Control and Prevention-funded NPDPS Autopsy Program. All imaging and laboratory data were anonymized. This study was approved by the University Hospitals Institutional Review Board, and informed consent was waived by the institutional review board on deceased individuals for the purpose of this study.

Neuropathologic Analysis

Brain examination was performed by histopathologic analysis, including prion protein immunohistochemistry and West-

Key Points

Question Is diffusion magnetic resonance imaging (MRI) using a new criterion of at least 1 positive brain region for diagnosis of sporadic Creutzfeldt-Jakob disease (sCJD) more accurate than currently used criteria and 3 commonly used cerebrospinal fluid tests?

Findings In this diagnostic study of 1387 patients with suspected sCJD, the new criterion increased the sensitivity of MRI for autopsy-confirmed sCJD compared with current applied criteria. The diagnostic performance of diffusion MRI was comparable with the improved version of the real-time quaking-induced conversion test in cerebrospinal fluid.

Meaning The findings suggest that diffusion MRI is an accurate test for establishing a diagnosis of prion diseases in the appropriate clinical context.

ern blot, and subtype diagnosis of prion disease was established or excluded.^{2,3,25} Details on the procedures for the identification of pure and mixed sCJD subtypes are reported in the eMethods in the [Supplement](#).

CSF tests of IQ-CSF, 14-3-3, and total tau were performed and reported by the NPDPS as previously described.^{18,26} Diagnostic reliability analysis of 14-3-3 did not include patients with indeterminate results, and the total tau test result was considered positive for values greater than 1150 pg/mL.

Diffusion MRI Analysis

Diffusion MRI lesion profiles were generated for each patient by grading the signal hyperintensities of 12 brain regions on a 4-point ordinal scale: 0 for no hyperintensity, 1 for questionable, 2 for sCJD-related hyperintensity associated with low diffusivity on ADC maps, and 3 for presence of extensive sCJD-related hyperintensity with low diffusivity in that specific region. FLAIR images were systematically inspected before DWI in all patients. Both hemispheres were inspected, and the highest score was recorded for each brain region. Symmetry or asymmetry of the signal hyperintensities also was recorded. The score was assigned to 5 cortical regions (frontal and parietal, including the precuneus, temporal, and occipital lobes), the caudate, the putamen, the thalamus, limbic structures (cingulate, insula, and hippocampus), and the cerebellum. MRI results were considered positive when at least 1 of 8 regions, excluding 3 limbic structures and cerebellum, was scored 2 or higher. The cingulate, insula, hippocampus, and cerebellum were excluded because they can be spontaneously hyperintense on DWI of healthy individuals.^{27,28} Diagnostic values of the proposed criterion (index test) were computed considering neuropathologic findings as the reference standard and were compared with current standard criteria: involvement of both the caudate and the putamen or at least 2 cortical regions among parietal, temporal, and occipital lobes.^{16,17}

Three neuroradiologists each with 15 years of experience (A.B., M.G., and R.L.) and 1 neuroradiologist with 1 year of experience (M.E.M.M.) blinded to clinical data and diagnosis scored the diffusion MRIs of 200 individuals with autopsy-confirmed diagnosis. The interrater reliability (IRR) among the

Table 1. Demographics of 200 Patients With Autopsy-Confirmed Diagnosis of sCJD by Subtype and Nonprion Diseases

Subtype	Total, No. (%) ^a	Male/female, No.	Median (IQR)		
			Age at onset, y	Disease duration, mo	Time from onset to MRI, d
sCJD (n = 150)					
MM1	52 (34.7)	26/26	66 (61-74)	2.4 (1.8-3.4)	44 (25-69)
MV1	10 (6.7)	6/4	68 (59-74)	3.7 (2.1-10.4)	73 (42-104)
MM2	10 (6.7)	5/5	68 (59-75)	11.0 (7.1-12.6)	60 (29-163)
MV2C	7 (4.7)	3/4	69 (65-75)	9.1 (6.6-26.9)	70 (3-147)
MV2K	9 (6.0)	6/3	69 (65-73)	11.5 (8.5-12.4)	164 (128-245)
VV1	5 (3.3)	2/3	58 (57-87)	12.3 (8.1-13.0)	144 (103-246)
VV2	23 (15.3)	11/12	66 (59-74)	5.2 (4.6-7.0)	112 (89-129)
MM1 + 2	18 (12.0)	8/10	63 (56-66)	3.4 (2.7-8.0)	58 (18-85)
MV1 + 2C	3 (2.0)	2/1	67 (65-70)	27.5 (20.5-27.5)	245 (242-256)
MV1 + 2K	3 (2.0)	2/1	69 (65-72)	10.5 (8.8-10.6)	75 (70-106)
MV1 + 2K+C	1 (0.7)	0/1	64	8.5 ^b	133 ^b
MV2K+C	6 (4.0)	2/4	65 (60-75)	6.8 (5.9-11.7)	183 (107-188)
VV1 + 2	1 (0.7)	1/0	62	12.8 ^b	253 ^b
NOS	2 (1.3)	1/1	63 (61-65)	20.1 (15.1-25.1)	398 (351-446)
Nonprion diseases (n = 50)					
Alzheimer disease	6 (12.0)	2/4	65 (55-73)	NA	NA
Inflammatory (encephalitis, vasculitis)	5 (10.0)	3/2	57 (29-75)	NA	NA
Vascular (stroke, anoxic encephalopathy)	4 (8.0)	3/1	64 (50-65)	NA	NA
NOS	35 (70.0)	17/18	56 (49-66)	NA	NA

Abbreviations: IQR, interquartile range; MRI, magnetic resonance imaging; NA, not available; NOS, not otherwise specified; sCJD, sporadic Creutzfeldt-Jakob disease.

^a For the sCJD group, percentages do not add to 100% because of rounding.

^b The entries without IQRs correspond to sCJD subtypes with only 1 patient (MV1 + 2K + C and VV1 + 2). In these cases, the IQRs would be the same as the median value reported in the entry.

neuroradiologists was measured. The intrarater reliability for each neuroradiologist was measured in a subset of MRI studies from 60 individuals that each neuroradiologist read with an interval of at least 1 year. One neuroradiologist (A.B.) read and scored all MRIs in electronic format.

Statistical Analysis

The IRR analysis was performed for 200 individuals (150 with sCJD and 50 with nonprion diseases) randomly selected from the 619 individuals with sCJD by stratified sampling (strata were disease subtypes) and from 151 individuals with nonprion disease by simple random sampling. The Fleiss κ and intraclass correlation coefficients (ICCs) were used to measure interrater agreement in the diagnosis and in scoring each region, respectively. Intrarater analysis was assessed with the Cohen κ coefficient and percentage agreement in a subsample of 60 individuals (30 with sCJD and 30 with nonprion diseases) selected from the 200 individuals through the same sampling procedure described above. Further details on these statistical analyses are reported in the eMethods in the Supplement.

The McNemar test was used to compare diagnostic parameters (ie, accuracy, sensitivity, and specificity) of the proposed and current diffusion MRI criteria for each neuroradiologist from the subsample of 200 individuals and for 1 neuroradiologist (A.B.) from all 619 individuals with sCJD and 151 individuals with nonprion disease. Sensitivities of these criteria were also compared in 102 individuals with other prion diseases (A.B.). Diagnostic parameters of diffusion MRI scored

by this neuroradiologist (A.B.) were compared with those of IQ-CSF, 14-3-3, and total tau tests performed on the same individuals using the McNemar test.

The Fisher exact test was used to compare sensitivity values of the same diagnostic test between sCJD subtypes. Statistical significance level was set at a 2-sided $P < .05$. Statistical analyses were performed using R software, version 3.6.0 (R Foundation for Statistical Computing).

Results

Of the 1387 patients, 872 had a DWI sequence available and autopsy diagnosis performed at the NPDPC. The following diagnoses were made: sCJD (619 cases), other prion diseases (102 cases), and nonprion diseases (151 cases). Patients with other prion diseases included familial CJD, variably protease-sensitive prionopathy, sporadic fatal insomnia, Gerstmann-Sträussler-Scheinker disease, fatal familial insomnia, and variant CJD. The diagnostic performance of 4 neuroradiologists applying the proposed and current MRI criteria was computed in a subsample of 200 individuals (150 with sCJD and 50 with nonprion diseases) (mean [SD] age, 63.6 [12.9] years; 100 [50.0%] male). The flowchart of case selection and patient demographics is given in Table 1 and the eFigure and eTable 1 in the Supplement. Comparison between demographic data of the 200 individuals with those of the larger cohort is given in eTable 2 in the Supplement.

Table 2. Comparison of Sensitivity and Specificity Achieved by 4 Neuroradiologists Using Proposed and Current Magnetic Resonance Imaging Criteria

Diagnostic parameter	No. of correctly identified patients/ No. of all patients examined (%) [95% CI]		P value
	New criterion ^a	Current criteria ^b	
Neuroradiologist 1 (M.G.) (n = 200)			
Sensitivity	142/150 (94.7) [89.8-97.7]	114/150 (76.0) [68.4-82.6]	<.001
Specificity	45/50 (90.0) [78.2-96.7]	45/50 (90.0) [78.2-96.7]	ND
Neuroradiologist 2 (R.L.) (n = 200)			
Sensitivity	141/150 (94.0) [88.9-97.2]	110/150 (73.3) [65.5-80.2]	<.001
Specificity	48/50 (96.0) [86.3-99.5]	49/50 (98.0) [89.4-99.9]	>.99
Neuroradiologist 3 (M.E.M.M.) (n = 200)			
Sensitivity	135/150 (90.0) [84.0-94.3]	104/150 (69.3) [61.3-76.6]	<.001
Specificity	48/50 (96.0) [86.3-99.5]	49/50 (98.0) [89.4-99.9]	>.99
Neuroradiologist 4 (A.B.) (n = 200)			
Sensitivity	138/150 (92.0) [86.4-95.8]	111/150 (74.0) [66.2-80.8]	<.001
Specificity	50/50 (100.0) [92.9-100]	50/50 (100.0) [92.9-100]	ND

Abbreviations: ND, not determined (P value cannot be computed in the McNemar test when diagnostic tests are in perfect agreement).

^a At least 1 positive result in the following key brain regions: cortex of the frontal, parietal, temporal and occipital lobes, caudate, putamen, and thalamus, excluding limbic structures and cerebellum.

^b Both caudate and putamen or the cortex of at least 2 lobes excluding the frontal lobes.^{16,17}

Comparative Diagnostic Reliability Study of Proposed vs Current Standard MRI Criteria

Sensitivity values of 3 of the neuroradiologists (A.B., M.G., and R.L.) in 200 individuals using the proposed criteria were 92%, 95%, and 94%, and corresponding specificities were 100%, 90%, and 96%; sensitivity and specificity of the other neuroradiologist (M.E.M.M.) were 90% and 96%, respectively (Table 2). Sensitivities were significantly greater than those obtained using current criteria (new criterion vs current criteria: 94.7% vs 76.0% for neuroradiologist 1, 94.0% vs 73.3% for neuroradiologist 2, 90.0% vs 69.3% for neuroradiologist 3, and 92.0% vs 74.0% for neuroradiologist 4; all $P < .001$), whereas specificities were unchanged. Sensitivity and specificity were not significantly different between the neuroradiologists (sensitivities: 94.7% for neuroradiologist 1, 94.0% for neuroradiologist 2, 90.0% for neuroradiologist 3, and 92.0% for neuroradiologist 4; specificities: 90.0% for neuroradiologist 1, 96.0% for neuroradiologist 2, 96.0% for neuroradiologist 3, and 100.0% for neuroradiologist 4). Thus, neuroradiologist experience did not affect use of new diffusion MRI criteria.

Interrater agreement in the diagnosis was good (mean [SE] $\kappa = 0.814$ [0.041]) with the proposed criteria, and it was significantly higher ($P < .001$) when compared with that obtained by using the current criteria (mean [SE] $\kappa = 0.646$ [0.041]). The agreement in the scores was excellent, with ICCs between 0.86 and 0.93 in 6 of 12 brain regions. It was good for temporal cortex (ICC, 0.83), cingulate and insula (ICC, 0.81), occipital cortex (ICC, 0.77), and cerebellum (ICC, 0.70) and fair for the hippocampus (ICC, 0.58) (eTable 3 in the Supplement). Intrarater reliability in 60 individuals was excellent for all neuroradiologists; for each of the neuroradiologists, κ was 1.0, 0.90, 0.93, and 0.87, with percentage agreements of 100%, 95%, 97%, and 93%.

Diagnostic Reliability of Diffusion MRI

The performance of the index test in 770 individuals (619 with sCJD and 151 with nonprion disease) scored by 1 neuroradi-

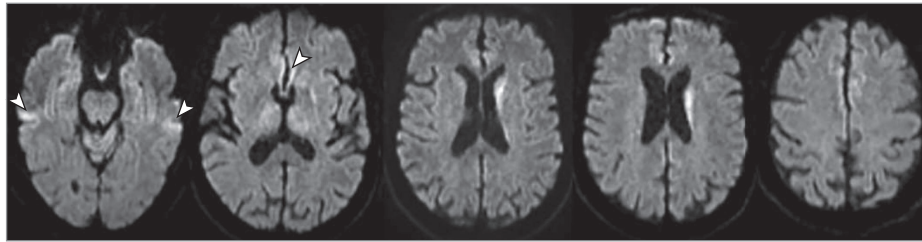
ologist (A.B.) is reported in eTable 4 in the Supplement. Accuracy of diffusion MRI applying the new criterion was 93.1% (95% CI, 91.1%-94.8%) and sensitivity was 92.1% (95% CI, 89.7%-94.1%), and these values were significantly higher than applying the currently used criteria¹⁶ (75.3% [95% CI, 72.1%-78.3%] and 69.8% [432 of 619; 95% CI, 66.0%-73.4%]; $P < .001$). Specificity values were not significantly different between criteria (97.4% [95% CI, 93.4%-99.3%] and 98.0% [95% CI, 94.3%-99.6%]; $P > .99$). These results are equivalent to those obtained by the same neuroradiologist (A.B.) in the subsample of 200 individuals. The new criterion correctly identified a significantly higher number of patients with sCJD with respect to current criteria in VV1, VV2, MM1, and MM2 pure subtypes, as well as for MM1 + 2 mixed form (eTable 5 in the Supplement). Other pure and mixed subtypes did not show any significant difference.

An accurate positive MRI diagnosis was made in 138 patients who would have had a false-negative MRI result applying the current criteria: 53 with only 1 positive region, 61 with 2 positive regions, and 24 patients with 3 positive regions. In particular, patients with the following combinations of diffusion MRI signal abnormalities would have missed the diagnosis with the current criteria: 27 patients with abnormality only in the parietal lobes, 12 patients with abnormality only in the caudate, 42 patients with abnormalities in the parietal and frontal cortices, 9 patients with abnormalities in the caudate and thalami, and 19 patients with abnormalities in the parietal and frontal cortices and caudate. Five demonstrative cases are shown in Figure 1 and Figure 2.

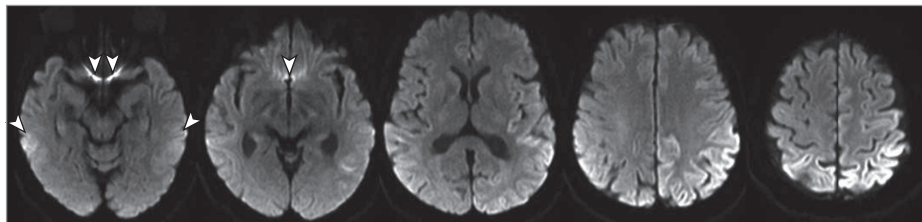
A follow-up diffusion MRI was available in 24 of 49 patients with initially false-negative results, and the result was positive in 16 of 24 (66.7%). Accuracy and sensitivity increased to 95% when all MRI studies were considered. Diagnostic sensitivity for pure and mixed sCJD subtypes ranged from 83% to 100% for the new criterion with no statistically significant differences between subtypes (93% for MM1, 95% for MM2, 93% for MV1, 100% for MV2C, 92% for MV2K, 90%

Figure 1. Magnetic Resonance Imaging Comparative Study of Patients With Sporadic Creutzfeldt-Jakob Disease (sCJD) Using the Newly Proposed and the Currently Used Criteria

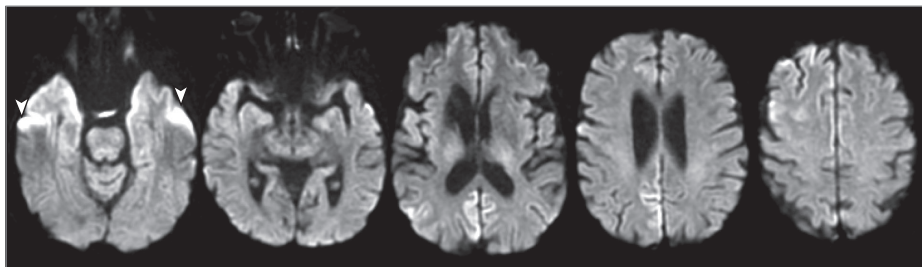
A Abnormality in the caudate only



B Abnormalities in both parietal lobes



C Abnormalities in the parietal and frontal lobes only



Three patients with autopsy-confirmed diagnoses of sCJD who were rated positive with our new criterion and negative with the current criteria¹⁶ were selected. Five axial diffusion-weighted images ($b = 1000$) of sections at the level of the temporal lobes, striatum, thalamus, insula, occipital, temporal, frontal, and parietal lobes are shown. Arrowheads indicate susceptibility artifacts at the base of the skull. A, A man in his 70s with sCJD VV2 with signal hyperintensity in the body of the left caudate, without evidence of signal abnormality in the putamina and neocortical ribbon. Subtle signal hyperintensity in the thalami was considered insufficient to rate these structures abnormal (score of 1). Subtle hyperintensity in cingulate and insula is within normal limits. B, A woman in her 60s with sCJD MM1 with signal hyperintensity in the cortical ribbon of both parietal lobes, left greater than right, and left precuneus without evidence of abnormality in the striatum, thalami, and cortical ribbon of the temporal and occipital lobes. C, A woman in her 70s with sCJD MM1 with signal hyperintensity in the precuneus (right greater than left) and right parietal cortex. There is signal hyperintensity in the dorsal gyri of the right frontal lobe and bilateral cingulate and no evidence of signal abnormality in the striatum, thalami, and cortical ribbon of the temporal and occipital lobes.

for VV1, 84% for VV2, 96% for MM1 + 2, 92% for MV1 + 2C, 83% for MV1 + 2K, 100% for MV1 + 2K + C, 92% for MV2K + C, 67% for VV1 + 2, and 100% for not otherwise specified; $P = .09$) (eTable 5 in the Supplement). Sensitivity of MRI in 102 patients with other prion diseases was 63.7% (65 of 102) (eTable 6 in the Supplement). Diffusion-weighted imaging signal hyperintensities were rated more prominent than FLAIR in 490 of 536 patients (91.4%), equal in 43 of 536 patients (8.0%), and less prominent in 3 of 536 patients (0.6%).

Comparative Diagnostic Reliability Study of Diffusion MRI vs CSF Tests

Diagnostic parameters of diffusion MRI provided by the neuroradiologist who scored all 770 images were compared with the results of CSF tests obtained from the same patients during the same visit. Data on both diffusion MRI and IQ-CSF were available in 79 patients with sCJD and 9 with nonprion disease. Diagnostic accuracy was 95%, sensitivity was 95%, and specificity was 100% for MRI, and diagnostic accuracy was 88%, sensitivity was 86%, and specificity was 100% for IQ-CSF, with no significant differences in sensitivity ($P = .12$) and perfect agreement for specificity (Table 3). No statistically significant differences were observed among sCJD subtypes

(eTable 7 in the Supplement). Sensitivity was 100% when both MRI and IQ-CSF results were considered.

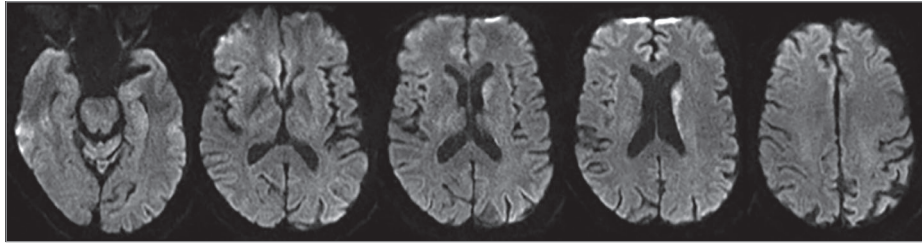
New MRI criterion and CSF total tau and 14-3-3 proteins were compared in 452 patients (391 patients with sCJD and 61 with nonprion disease) who had undergone all 3 tests. In this cohort, diffusion MRI achieved 93% accuracy, 92% sensitivity, and 100% specificity, surpassing both total tau (83% accuracy, 87% sensitivity, and 61% specificity) and 14-3-3 (84% accuracy, 92% sensitivity, and 23% specificity) (Table 3). Diffusion MRI sensitivity (92% vs 87%, $P = .03$) and specificity (100% vs 61%, $P < .001$) significantly exceeded those of total tau, whereas only specificity was significantly improved (100% vs 23%, $P < .001$) compared with 14-3-3. However, the 14-3-3 test was nondiagnostic in 92 of 452 patients (20.4%).

Discussion

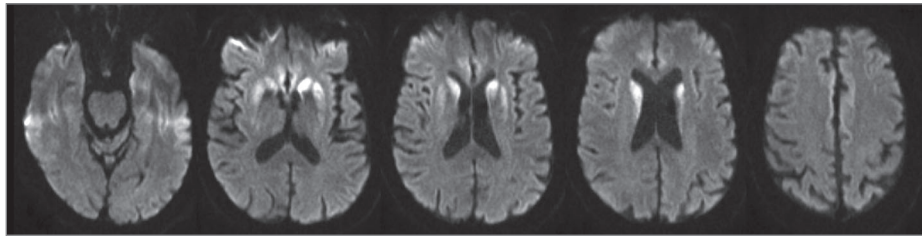
To our knowledge, this study was the largest diffusion MRI study in individuals with definitive prion disease.^{1,16,29-31} With the development of new potential therapies to alter the natural history of prion disease, early diagnosis is essential to maximize the chance of salvaging brain tissue.³² It is important that

Figure 2. Clinical Implication of the Magnetic Resonance Imaging (MRI) Newly Proposed Criteria for Early Diagnosis

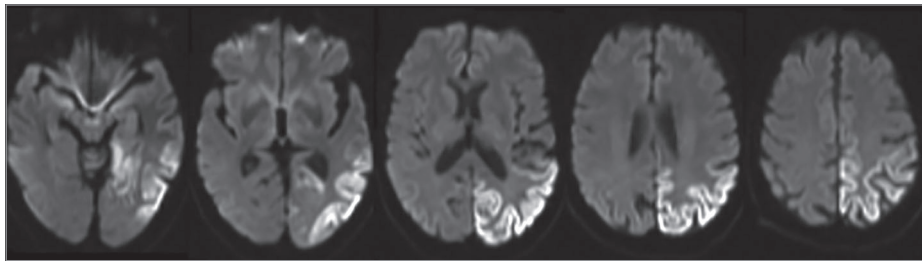
A Abnormalities in the head and body of the left caudate



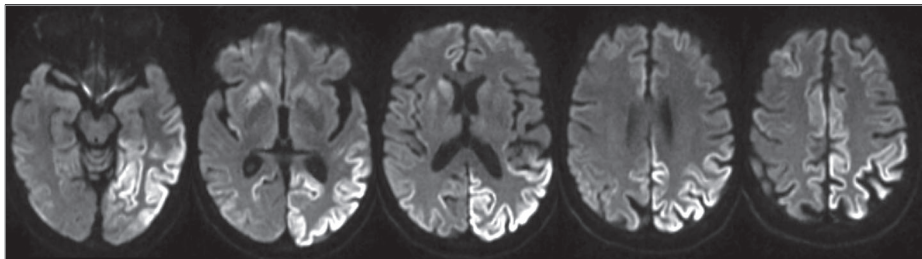
B Abnormalities in the right caudate and both putamina



C Abnormality in the cortical ribbon of the left parietal lobe



D Abnormalities in the right caudate and putamen



The MRI study was performed on 2 patients with autopsy-confirmed diagnoses of sporadic Creutzfeldt-Jakob disease (sCJD) at disease onset and follow-up to assess the clinical implication of our new criterion for early diagnosis. Five axial diffusion-weighted images ($b = 1000$) sections at the level of the temporal lobes, striatum, thalamus, insula, occipital, temporal, frontal, and parietal lobes are shown. A, MRI of a man in his 60s with sCJD VV2 showed signal hyperintensity in the head and body of the left caudate without evidence of signal abnormality in the putamina and cortical ribbon. The subtle signal hyperintensity in the cingulate was considered insufficient to define this structure as abnormal (score of 1). B, Two months later, diffusion-weighted imaging of the same patient showed the appearance of signal hyperintensity in the right caudate and both putamina but not in the thalami. Both study results were positive with the new proposed criterion, whereas diagnosis would have

been delayed for 2 months with the criteria currently used. C, Diffusion-weighted imaging of a man in his 60s with sCJD MV1 + 2K showed a single large lesion with signal hyperintensity in the cortical ribbon of the left parietal lobe, including the left precuneus and extending barely to the posterior cortex of the left temporal lobe and ventral cortex of the occipital lobes as well as no evidence of signal abnormality in the striatum and cortical ribbon of the other lobes. A second MRI 2 months later (not shown) was unchanged. D, Six months later, diffusion-weighted imaging of the same patient showed new signal hyperintensities in the right caudate and putamen, cortical ribbon of the right cingulate, right parietal lobe, and bilateral frontal lobes in addition to more extensive signal hyperintensities in the left parietal and lingular cortices. The use of the current criteria delayed the diagnosis for 6 months.

practitioners are aware of the diagnostic potential of diffusion MRI because it is the most widely available diagnostic test.

In a cohort of 200 patients with autopsy-confirmed diagnoses scored independently by 4 neuroradiologists, we found that

Table 3. Comparative Diagnostic Reliability Study of Diffusion MRI vs CSF Tests in sCJD

Variable	Diffusion MRI and improved RT-QulC (IQ-CSF), No. of correctly identified patients/No. of all examined patients (%) [95% CI] ^{a,b}			Diffusion MRI, total tau, and 14-3-3, No. of correctly identified patients/No. of all examined patients (%) [95% CI] ^{c,d}				
	Diffusion MRI (n = 88)	Improved RT-QulC (n = 88)	P value	Diffusion MRI (n = 452)	Total tau (n = 452)	14-3-3 (n = 452)	Diffusion MRI vs total tau P value	Diffusion MRI vs 14-3-3 P value
Accuracy	84/88 (95.5) [88.8-98.8]	77/88 (87.5) [78.7-93.6]	.12	422/452 (93.4) [90.7-95.5]	377/452 (83.4) [79.7-86.7]	304/360 (84.4) [80.3-88.0]	<.001	.007
Sensitivity	75/79 (94.9) [87.5-98.6]	68/79 (86.1) [76.5-92.8]	.12	361/391 (92.3) [89.2-94.8]	340/391 (87.0) [83.2-90.1]	295/321 (91.9) [88.4-94.6]	.03	.44
Specificity	9/9 (100) [66.4-100]	9/9 (100) [66.4-100]	ND	61/61 (100) [94.1-100]	37/61 (60.7) [47.3-72.9]	9/39 (23.1) [11.1-39.3]	<.001	<.001

Abbreviations: CSF, cerebrospinal fluid; RT-QulC, real-time quaking-induced conversion assay performed on cerebrospinal fluid (also known as IQ-CSF); MRI, magnetic resonance imaging; ND, not determined (P value cannot be computed in the McNemar test when diagnostic tests are in perfect agreement); sCJD, sporadic Creutzfeldt-Jakob disease.

^a Diffusion MRI examination results were read by 1 neuroradiologist, and diagnostic performance was measured according to the new criterion; for details on MRI criteria, see Table 2.

^b Includes 79 patients with sCJD and 9 with nonprion disease, all of whom underwent both diffusion MRI and improved RT-QulC.

^c Includes 391 patients with sCJD and 61 with nonprion disease, all of whom underwent diffusion MRI, total tau, and 14-3-3 tests.

^d Nondiagnostic reports of 14-3-3 test were recorded in 92 patients who were excluded from the analyses.

the results of newly proposed diffusion MRI criterion matched or exceeded values previously reported for currently used MRI sCJD diagnostic criteria.^{1,8,9} Moreover, sensitivity values of the 4 neuroradiologists applying the current criteria were similar to those obtained by Zerr et al¹⁶ in 233 individuals DWI. We found that the use of our at least 1 region criterion significantly improved the sensitivity, especially for patients with sCJD with MM and VV genotypes, whereas specificity remained unchanged. Of note, similar diagnostic performances were achieved by the 4 neuroradiologists regardless of their years of experience. Diagnostic sensitivity reached 100% when both MRI and IQ-CSF were considered, suggesting that the 2 tests are complementary, and they should be used together in the diagnostic workup of patients with suspected sCJD.

The new criterion is simple, robust, and practical to use. On the contrary, the requirement of 2 or more positive brain regions by current standard criteria is likely to limit the detection of positive cases more so at early stages of the disease. This study showed that in the appropriate clinical context the probable diagnosis of sCJD should be raised with MRI in patients presenting with a single positive region showing DWI abnormality in the parietal cortex or in the caudate and in patients with 2 positive regions in the cortical ribbon of the parietal and frontal lobes. The sensitivity of MRI with inclusion also of limbic structures and cerebellum was even higher than with 8 brain regions. Notwithstanding, we excluded limbic structures and cerebellum to avoid false-positive readings by less experienced neuroradiologists. Furthermore, our study supported the superiority of the DWI over FLAIR, and the findings suggest that DWI should be acquired for all patients with rapidly progressive encephalopathy.^{27,33,34}

Few MRI studies^{16,35,36} have analyzed individual sCJD subtypes; moreover, they examined only the main subtypes because of the small sample size of the autopsy-confirmed subtypes. Our study found high diagnostic performance of diffusion MRI in all subtypes, including mixed subtypes and, for the first time to our knowledge, the recently distinguished MV2C and MV2K. The excellent sensitivity of our criterion for almost all sCJD subtypes may inform in vivo iden-

tification of sCJD subtypes by MRI, which is the focus of a forthcoming study.

MRI sensitivity in other prion diseases characterized by spongiform changes (familial CJD, sporadic fatal insomnia, and variant CJD) was above 75%. As expected, sensitivity was lower in Gerstmann-Sträussler-Scheinker disease, variably protease-sensitive prionopathy, and fatal familial insomnia, which rarely have vacuoles on histopathologic analysis. These results suggest that DWI signal hyperintensity may be associated with the spongiform change rather than other histopathologic hallmarks of prion diseases and confirm the results of previous studies.^{13,34}

The topography of the diffusion signal abnormality is heterogeneous in sCJD because it reflects the multiple configurations in which key brain regions are affected.³⁶⁻³⁸ Furthermore, the involvement of the cerebral cortex is often asymmetrical. This heterogeneity of signal abnormalities at presentation, which is mostly associated with the distinct subtypes, may be a confounder in the interpretation of the findings by the neuroradiologists.³⁶ Despite the good diagnostic values reported in the literature, clinical use of MRI may be hampered by the rarity of the disease and variations among sCJD subtypes. Diffusion MRI signal hyperintensity was often not correctly interpreted on the initial MRI at referring hospitals in a study of 103 patients with CJD.³⁹ Retrospective reading of the original MRI reports found that the diffusion MRI signal abnormalities were rarely missed. On the contrary, often the neuroradiologist failed to raise the possibility of sCJD diagnosis because these findings were usually considered as an improbable presentation of a common disease rather than a typical presentation of a rare one. The most frequent sCJD misdiagnoses on MRI were acute cortical ischemic infarct, venous thrombosis, encephalitis, or metabolic disorders.³⁹ All the above diseases are typically associated with diffusion MRI signal abnormalities and clinical signs that are different from those found in sCJD. Nevertheless, any diagnostic test must be interpreted in the appropriate clinical context, which includes ruling out treatable illnesses and potential mimickers. When a patient presents with signs of rapidly progressive encephal-

lopathy and sCJD is suspected, even if the initial MRI findings do not suggest the diagnosis, review of the study by an experienced neuroradiologist should be encouraged to facilitate early diagnosis. The MRI consultation program was started at the NPDPS to address this need.

A remarkable finding of our study was that diffusion MRI diagnostic parameters were comparable to those of the IQ-CSF. As expected, diffusion MRI exceeded the diagnostic values of older CSF tests, such as total tau and 14-3-3. These findings were not affected by patient population, including whether the procedures were performed on the same patients, as we did in this study, or by comparing data from separate cohorts, as reported previously.^{18,26} The IQ-CSF sensitivity for all sCJD cases (86%) was lower than the sensitivity values reported in the literature (92%-95%).^{24,26} A possible contributing factor to this discrepancy was the different representation of individual subtypes in this sCJD cohort compared with previous studies.^{24,26,40}

IQ-CSF sensitivities differed by sCJD subtype. Although sCJD MM1 and VV2, the 2 most common subtypes, have similar sensitivities, MM2 sensitivity was 100% for diffusion MRI and 67% in this study and 80% in previous studies for IQ-CSF.^{24,26,40} Although more of the rare sCJD subtypes need to be analyzed, these results suggest that the 2 tests are complementary and should both be performed.

Limitations

There are a few limitations in this study. The diffusion MRI lesion profile score was assigned semiquantitatively by neuro-

radiologists from 2 different medical centers with expertise in evaluating patients with prion disease. The high prevalence of prion disease (3:1) in the cohort of 200 individuals may overestimate test performance. Despite the high mean IRR values of the neuroradiologists, the reliability of the scoring system may be lower when used by less expert radiologists or neurologists. In the future, a strategy to extract the MRI signal from diffusion MRI and automatically generate a lesion profile score may improve the accuracy and consistency of CJD diagnosis.

Conclusions

The diagnostic performance of diffusion MRI with the new criterion was superior to that of current standard criteria and equal to that of IQ-CSF in this study performed in, to our knowledge, the largest autopsied cohort with suspected CJD to date. When both diagnostic tests were used, the sensitivity reached 100%. MRI appear to offer several advantages over IQ-CSF in terms of an early diagnostic marker because it is noninvasive and a more accessible test. Moreover, positive MRI findings may suggest sCJD diagnosis even when it is not suspected yet by the physician. Future studies should examine the capability of MRI to identify sCJD subtypes using the brain regions scoring procedure proposed in this study. The effective use of DWI and ADC maps in the diagnosis of sCJD requires that general radiologists become aware of the importance of correctly interpreting the diffusion abnormalities that are characteristic of this rare disease.

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REFERENCES

- Puoti G, Bizzi A, Forloni G, Safar JG, Tagliavini F, Gambetti P. Sporadic human prion diseases: molecular insights and diagnosis. *Lancet Neurol*. 2012;11(7):618-628. doi:10.1016/S1474-4422(12)70063-7
- Gambetti P, Cali I, Notari S, Kong Q, Zou WQ, Surewicz WK. Molecular biology and pathology of prion strains in sporadic human prion diseases. *Acta Neuropathol*. 2011;121(1):79-90. doi:10.1007/s00401-010-0761-3
- Parchi P, de Boni L, Saverioni D, et al. Consensus classification of human prion disease histotypes allows reliable identification of molecular subtypes: an inter-rater study among surveillance centres in Europe and USA. *Acta Neuropathol*. 2012;124(4):517-529. doi:10.1007/s00401-012-1002-8
- Collins SJ, Sanchez-Juan P, Masters CL, et al. Determinants of diagnostic investigation sensitivities across the clinical spectrum of sporadic Creutzfeldt-Jakob disease. *Brain*. 2006;129(Pt 9):2278-2287. doi:10.1093/brain/awl159
- Zerr I, Parchi P. Sporadic Creutzfeldt-Jakob disease. In: Pocchiari M, Manson J, eds. *Handbook of Clinical Neurology*. Vol 153. Elsevier; 2018:155-174.

6. Bizzi A, Peoc'h K. Amended diagnostic protocol increases the early diagnosis of sporadic Creutzfeldt-Jakob disease. *Neurology*. 2018;91(4):155-156. doi:10.1212/WNL.0000000000005871
7. Gaudino S, Gangemi E, Colantonio R, et al. Neuroimaging of human prion diseases, diagnosis and differential diagnosis. *Radiol Med*. 2017;122(5):369-385. doi:10.1007/s11547-017-0725-y
8. Vitali P, Maccagnano E, Caverzasi E, et al. Diffusion-weighted MRI hyperintensity patterns differentiate CJD from other rapid dementias. *Neurology*. 2011;76(20):1711-1719. doi:10.1212/WNL.0b013e31821a4439
9. Manix M, Kalakoti P, Henry M, et al. Creutzfeldt-Jakob disease: updated diagnostic criteria, treatment algorithm, and the utility of brain biopsy. *Neurosurg Focus*. 2015;39(5):E2. doi:10.3171/2015.8.FOCUS15328
10. Terasawa Y, Fujita K, Izumi Y, Kaji R. Early detection of familial Creutzfeldt-Jakob disease on diffusion-weighted imaging before symptom onset. *J Neurol Sci*. 2012;319(1-2):130-132. doi:10.1016/j.jns.2012.04.004
11. Baiardi S, Magherini A, Capellari S, et al. Towards an early clinical diagnosis of sporadic CJD VV2 (ataxic type). *J Neurol Neurosurg Psychiatry*. 2017;88(9):764-772. doi:10.1136/jnnp-2017-315942
12. Fragoso DC, Gonçalves Filho AL, Pacheco FT, et al. Imaging of Creutzfeldt-Jakob disease: imaging patterns and their differential diagnosis. *Radiographics*. 2017;37(1):234-257. doi:10.1148/rg.2017160075
13. Figini M, Alexander DC, Redaelli V, et al. Mathematical models for the diffusion magnetic resonance signal abnormality in patients with prion diseases. *Neuroimage Clin*. 2014;7:142-154. doi:10.1016/j.nicl.2014.11.017
14. Meissner B, Körtner K, Bartl M, et al. Sporadic Creutzfeldt-Jakob disease: magnetic resonance imaging and clinical findings. *Neurology*. 2004;63(3):450-456. doi:10.1212/01.WNL.0000136225.80445.C9
15. Tschampa HJ, Kallenberg K, Urbach H, et al. MRI in the diagnosis of sporadic Creutzfeldt-Jakob disease: a study on inter-observer agreement. *Brain*. 2005;128(Pt 9):2026-2033. doi:10.1093/brain/awh575
16. Zerr I, Kallenberg K, Summers DM, et al. Updated clinical diagnostic criteria for sporadic Creutzfeldt-Jakob disease. *Brain*. 2009;132(Pt 10):2659-2668. doi:10.1093/brain/awp191
17. Centers for Disease Control and Prevention. CDC's diagnostic criteria for Creutzfeldt-Jakob disease. CDC website. 2018. Accessed September 30, 2019. <https://www.cdc.gov/prions/cjd/diagnostic-criteria.html>
18. Hamlin C, Puoti G, Berri S, et al. A comparison of tau and 14-3-3 protein in the diagnosis of Creutzfeldt-Jakob disease. *Neurology*. 2012;79(6):547-552. doi:10.1212/WNL.0b013e318263565f
19. Lattanzio F, Abu-Rumeileh S, Franceschini A, et al. Prion-specific and surrogate CSF biomarkers in Creutzfeldt-Jakob disease: diagnostic accuracy in relation to molecular subtypes and analysis of neuropathological correlates of p-tau and Aβ42 levels. *Acta Neuropathol*. 2017;133(4):559-578. doi:10.1007/s00401-017-1683-0
20. Atarashi R, Satoh K, Sano K, et al. Ultrasensitive human prion detection in cerebrospinal fluid by real-time quaking-induced conversion. *Nat Med*. 2011;17(2):175-178. doi:10.1038/nm.2294
21. Wilham JM, Orrú CD, Bessen RA, et al. Rapid end-point quantitation of prion seeding activity with sensitivity comparable to bioassays. *PLoS Pathog*. 2010;6(12):e1001217. doi:10.1371/journal.ppat.1001217
22. Orrú CD, Bongianini M, Tonoli G, et al. A test for Creutzfeldt-Jakob disease using nasal brushings. *N Engl J Med*. 2014;371(6):519-529. doi:10.1056/NEJMoa1315200
23. Orrú CD, Groveman BR, Hughson AG, Zanusso G, Coulthart MB, Caughey B. Rapid and sensitive RT-QuIC detection of human Creutzfeldt-Jakob disease using cerebrospinal fluid. *mBio*. 2015;6(1):e02451-e14. doi:10.1128/mBio.02451-14
24. Franceschini A, Baiardi S, Hughson AG, et al. High diagnostic value of second generation CSF RT-QuIC across the wide spectrum of CJD prions. *Sci Rep*. 2017;7(1):10655. doi:10.1038/s41598-017-10922-w
25. Parchi P, Strammiello R, Notari S, et al. Incidence and spectrum of sporadic Creutzfeldt-Jakob disease variants with mixed phenotype and co-occurrence of PrPSc types: an updated classification. *Acta Neuropathol*. 2009;118(5):659-671. doi:10.1007/s00401-009-0585-1
26. Foutz A, Appleby BS, Hamlin C, et al. Diagnostic and prognostic value of human prion detection in cerebrospinal fluid. *Ann Neurol*. 2017;81(1):79-92. doi:10.1002/ana.24833
27. Young GS, Geschwind MD, Fischbein NJ, et al. Diffusion-weighted and fluid-attenuated inversion recovery imaging in Creutzfeldt-Jakob disease: high sensitivity and specificity for diagnosis. *AJNR Am J Neuroradiol*. 2005;26(6):1551-1562.
28. Hirai T, Korogi Y, Yoshizumi K, Shigematsu Y, Sugahara T, Takahashi M. Limbic lobe of the human brain: evaluation with turbo fluid-attenuated inversion-recovery MR imaging. *Radiology*. 2000;215(2):470-475. doi:10.1148/radiology.215.2.r00ma06470
29. Wang LH, Bucelli RC, Patrick E, et al. Role of magnetic resonance imaging, cerebrospinal fluid, and electroencephalogram in diagnosis of sporadic Creutzfeldt-Jakob disease. *J Neurol*. 2013;260(2):498-506. doi:10.1007/s00415-012-6664-6
30. Forner SA, Takada LT, Bettcher BM, et al. Comparing CSF biomarkers and brain MRI in the diagnosis of sporadic Creutzfeldt-Jakob disease. *Neuro Clin Pract*. 2015;5(2):116-125. doi:10.1212/CPJ.0000000000000111
31. Rudge P, Hyare H, Green A, Collinge J, Mead S. Imaging and CSF analyses effectively distinguish CJD from its mimics. *J Neurol Neurosurg Psychiatry*. 2018;89(5):461-466. doi:10.1136/jnnp-2017-316853
32. Collinge J, Gorham M, Hudson F, et al. Safety and efficacy of quinacrine in human prion disease (PRION-1 study): a patient-preference trial. *Lancet Neurol*. 2009;8(4):334-344. doi:10.1016/S1474-4422(09)70049-3
33. Shiga Y, Miyazawa K, Sato S, et al. Diffusion-weighted MRI abnormalities as an early diagnostic marker for Creutzfeldt-Jakob disease. *Neurology*. 2004;63(3):443-449. doi:10.1212/01.WNL.0000134555.59460.5D
34. Lodi R, Parchi P, Tonon C, et al. Magnetic resonance diagnostic markers in clinically sporadic prion disease: a combined brain magnetic resonance imaging and spectroscopy study. *Brain*. 2009;132(Pt 10):2669-2679. doi:10.1093/brain/awp210
35. Krasnianski A, Kallenberg K, Collie DA, et al. MRI in the classical MM1 and the atypical MV2 subtypes of sporadic CJD: an inter-observer agreement study. *Eur J Neurol*. 2008;15(8):762-771. doi:10.1111/j.1468-1331.2008.02209.x
36. Meissner B, Kallenberg K, Sanchez-Juan P, et al. MRI lesion profiles in sporadic Creutzfeldt-Jakob disease. *Neurology*. 2009;72(23):1994-2001. doi:10.1212/WNL.0b013e3181a96e5d
37. Meissner B, Kallenberg K, Sanchez-Juan P, et al. Isolated cortical signal increase on MR imaging as a frequent lesion pattern in sporadic Creutzfeldt-Jakob disease. *AJNR Am J Neuroradiol*. 2008;29(8):1519-1524. doi:10.3174/ajnr.A1122
38. Tschampa HJ, Kallenberg K, Kretschmar HA, et al. Pattern of cortical changes in sporadic Creutzfeldt-Jakob disease. *AJNR Am J Neuroradiol*. 2007;28(6):1114-1118. doi:10.3174/ajnr.A0496
39. Carswell C, Thompson A, Lukic A, et al. MRI findings are often missed in the diagnosis of Creutzfeldt-Jakob disease. *BMC Neurol*. 2012;12:153. doi:10.1186/1471-2377-12-153
40. Green AJE. RT-QuIC: a new test for sporadic CJD. *Pract Neurol*. 2019;19(1):49-55. doi:10.1136/practneurol-2018-001935