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Case report

Early diagnosis of progressive multifocal leukoencephalopathy in untreated HIV infection via ultrasensitive PCR testing for JC virus: A case report☆

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ABSTRACT

Progressive multifocal leukoencephalopathy (PML) is a fatal demyelinating disease triggered by the reactivation of JC virus (JCV) in individuals with immunodeficiency, particularly those with untreated human immunodeficiency virus (HIV) infection. This case report describes a 46-year-old HIV-positive man who initially presented with neurological symptoms and was incorrectly diagnosed as cerebral infarction. Although standard real-time polymerase chain reaction (PCR) testing for JCV in cerebrospinal fluid (CSF) at a commercial laboratory was negative, neuroimaging and clinical suspicion prompted ultrasensitive PCR testing at a national laboratory. This test detected a low viral load of JCV (28 copies/mL), confirming the diagnosis of PML. The patient underwent treatment with antiretroviral therapy and corticosteroids to prevent immune reconstitution inflammatory syndrome; however, his neurological symptoms persisted. This case highlights the importance of ultrasensitive CSF JCV testing for early PML diagnosis when standard PCR tests are inconclusive, particularly in HIV patients with atypically low JCV levels. It also highlights the diagnostic challenges of PML and emphasizes the clinical value of advanced PCR techniques for timely and accurate diagnosis in similar cases.

Introduction

Progressive multifocal leukoencephalopathy (PML) is a demyelinating disease caused by JC virus (JCV) reactivation in immunocompromised individuals [1,2]. PML diagnosis is based on clinical presentation, imaging findings, and polymerase chain reaction (PCR) testing for JCV in the cerebrospinal fluid (CSF) [3]. To date, no specific therapy has been established for PML. Treatment strategies include antiretroviral therapy (ART) to address the underlying immunosuppression that impairs the host immune response to the virus [4].

Historically, human immunodeficiency virus (HIV)-associated PML was thought to be associated with a high JCV copy number [3]. However, with development of antiretroviral therapy (ART), PML cases with lower JCV copy number have been documented [5]. The 1-year survival rate of PML patients with low CD4 + counts (< 100 cells/μL) is 39 % [6], underscoring the importance of early diagnosis and highlighting the need for ultrasensitive PCR testing when standard results are negative.

We present the case of a patient with HIV-associated PML for whom ultrasensitive PCR for PML was helpful in diagnosis.

Case report

A 46-year-old man who has sex with men was admitted to a local hospital, initially diagnosed with cerebral infarction based on the presence of right facial nerve palsy, dysarthria, and right-hand sensory impairment. The patient was alert and oriented, with vital signs within normal limits and afebrile. Cranial nerves VII, IX, and X were impaired, with drooping of the right corner of the mouth and dysarthria. The patient demonstrated a positive Barré sign in the upper extremities with evidence of right-side drift. Sensory examination revealed right-hand

☆ All authors met the ICMJE authorship criteria and were involved in the clinical care and management of the patient, collection of data, and drafting of the manuscript.

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reduction. Brain magnetic resonance imaging (MRI) showed increased diffusion-weighted imaging and fluid-attenuated inversion recovery (FLAIR) sequences in the left frontoparietal lobe subcortical white matter. Positive HIV screening led to referral to our outpatient clinic, where confirmatory testing verified a diagnosis of HIV infection.

His initial CD4 + lymphocyte count was 119 per μL, and 7.06×10^5 copies per mL of HIV-1 RNA were detected. Plasma tests for Toxoplasma IgG antibodies were negative (< 1.6 IU/mL); PCR for cytomegalovirus and JCV were also negative (< 200 copies/mL). Lumbar puncture revealed no abnormalities, and CSF JCV was undetectable with standard PCR (< 200 copies/mL). Other CSF studies were unremarkable, including PCR for Epstein-Barr virus and Toxoplasma, and the FilmArray® Meningitis/Encephalitis multiplex PCR panel (bioMérieux Japan Ltd., Tokyo, Japan). MRI on admission showed an asymmetric patchy hyperintense lesion on T2/FLAIR, called the Milky Way sign (Fig. 1A, B), and a hypointense lesion on T1 (Fig. 1C) in the subcortical white matter of the left frontoparietal lobe.

The patient was admitted to the hospital with a presumptive diagnosis of PML.

The patient could not be definitively diagnosed with PML because the commercially available PCR test yielded a negative result However, because of negative commercial real-time CSF PCR for JCV (< 200 copies/mL), the patient could not be definitively diagnosed with PML. Consequently, an ultrasensitive CSF JCV PCR test was performed and detected 28 copies/mL of CSF JCV, and the patient was diagnosed with PML. To confirm these results, the same test was repeated 2 weeks and approximately 1 month later, yielding positive results with increasing JCV loads of 117 and 1644 copies/mL, respectively. Given that blood JCV PCR testing has low sensitivity, it was not performed in this case.

The patient began ART (bictegravir, emtricitabine, and tenofovir alafenamide) and prednisolone (60 mg) to prevent the onset of PML-related immune reconstitution inflammatory syndrome (IRIS). The patient's neurological symptoms progressively worsened, and MRI performed 28 and 53 days after admission showed expansion of the FLAIR high-signal area (Fig. 1D, E). No evidence of contrast enhancement or edema was observed, which is inconsistent with IRIS, therefore, the prednisolone was tapered. At the patients' and families' request, treatment was continued at a hospital closer to home. After a 49-day tapering of prednisolone, the patient was discharged 83 days after admission.

Discussion

PML develops from reactivation of latent JCV infection acquired during childhood. This reactivation, often due to immunodeficiency as seen in untreated HIV with a CD4 + count of 119 cells/μL, leads to viral proliferation in oligodendrocytes and cerebral white matter demyelination [1,7].

The American Academy of Neurology defines two diagnostic criteria for PML: clinical features and pathological findings [3]. Diagnoses often rely on clinical criteria, (definite, probable, possible, and not PML) based on neurological symptoms, imaging, and CSF JCV PCR results [3, 8].

In HIV-associated PML, CSF JCV copy numbers are typically higher than those in non-HIV cases [3]. However, ART has lowered these numbers, complicating detection with standard PCR methods [9]. Comparing the pre-ART (1992–1995) and ART (1996–2002) era PML cases, the positive detection rate for CSF JCV dropped from 89.5 % to 57.5 % [10]. A Danish study observed a decline in PML incidence from 3.3 cases per 1000 PYR in 1995–1996 (pre-ART) to 1.3 cases in 2000–2006 (ART) [11]. Similarly, Japanese surveillance data show a trend toward lower JCV loads [12]. Standard PCR testing for CSF JCV has 74 % sensitivity (detecting 200 copies/mL), whereas ultrasensitive PCR exceeds 95 % sensitivity (detecting 10 copies/mL) [3]. A study comparing ultrasensitive PCR to brain tissue examination found 85 % sensitivity, increased to 95 % with follow-up testing [13]. HIV-positive patients with low JCV viral loads or high CD4 + counts often experience diagnostic delays [10]. A report exists of HIV-associated PML diagnosed by ultrasensitive PCR at 19 copies/mL despite negative standard tests [5].

Ultrasensitive CSF JCV PCR test that extracts highly pure and enriched nucleic acids from large volumes of CSF by manual handling [5,13] is conducted at the National Institute of Infectious Diseases (NIID) in Japan [13]. Real-time PCR, which can detect JCV by concentrating viral particles in CSF samples with a detection threshold as low as 10 copies/mL [5], is performed using primers and probes targeting the conserved region of the JCV T gene [14] and the Roche LightCycler 96 System (Roche Diagnostics, Mannheim, Germany) [13].

In therapeutic cases for autoimmune and hematologic diseases, JCV levels often fall below 200 copies/mL, making detection challenging without ultrasensitive PCR [3,11,15]. However, in Japan, in cases of

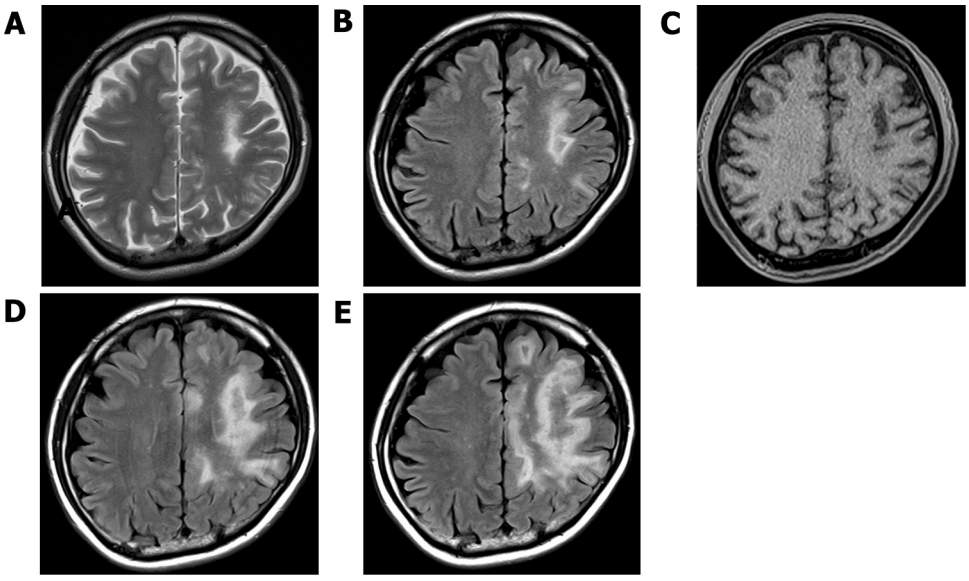

Fig. 1. Initial magnetic resonance imaging (MRI) of the brain demonstrated high T2 signals (A) and high FLAIR signals (B) with low T1 signals (C) in the subcortical white matter of the left frontal parietal lobe. No mass lesions with contrast enhancement are present. High FLAIR signals 28 days after admission (D) and 53 days after admission (E). Increases in high-signal intensity are observed over time.

suspected HIV-associated PML, standard CSF JCV testing in commercial laboratories is typically preferred, and the NIID's ultrasensitive detection of CSF JCV is not well understood. Compared with an epidemiologic study analyzing admission data of patients with PML in the Diagnosis Procedure Combination (DPC) database in Japan, the number of HIV-associated PML cases identified by the laboratory surveillance based on CSF JCV testing at the NIID during a similar period was only approximately 60 %, however, for PML cases associated with the treatment of autoimmune and hematological diseases, NIID data was comparable to the DPC database study [11,16]. Therefore, HIV-associated PML cases with very low CSF JCV levels may be missed during early diagnosis due to infrequent ultrasensitive PCR testing.

Here, standard commercial PCR testing failed to detect CSF JCV despite strong clinical and imaging evidence of PML, yet ultrasensitive PCR detected 28 copies/mL of CSF JCV; the patient was diagnosed with "definite PML." Though a few cases of HIV-associated PML with CSF JCV (approximately 100 copies/mL) were identified at the NIID, the clinical presentation, course, and prognosis of PML in HIV-positive patients with very low levels of CSF JCV are unclear [11]. This report significantly demonstrates the importance of ultrasensitive PCR testing for CSF JCV to resolve diagnostic uncertainty and enable timely treatment in HIV-associated PML cases where standard PCR tests are inconclusive. We recommend initial CSF JCV testing for the diagnosis of PML be performed in commercial and national laboratories. However, potential limitations with any PCR-based assay include human error, analytical failure, or mechanical failure of the equipment. Although laboratory standards aim to minimize such errors, repeating PCR tests in cases of high clinical suspicion may be beneficial. In this case, to address these potential limitations, a follow-up ultrasensitive PCR test was performed 2 weeks later, confirming a positive result with an increased viral load. This sequential testing further supported the reliability of the initial diagnosis.

There is no specific treatment for PML, and therapeutic approaches mainly focus on ART to counteract the underlying immunosuppression that weakens the host immune response to JCV [4]. Early initiation of ART is important to build an intact cellular immune response, and immune reconstitution through ART may halt disease progression in the central nervous system. However, rapid or excessive immune reconstitution can result in extensive inflammation and damage to the virus-infected cells, a phenomenon known as PML with immune reconstitution inflammatory syndrome (PML-IRIS) [1,17]. PML-IRIS is associated with high morbidity and mortality rates. A systematic review and meta-analysis found that 16.7 % of patients with AIDS and prior PML developed PML-IRIS [18]. Currently, no specific treatment has been established for PML-IRIS. ART remains the cornerstone of therapy; however, the role of corticosteroids remains controversial [19,20]. In the present case, corticosteroids were used to manage the risk of IRIS; however, the neurological symptoms worsened and required dose adjustments. This highlights the challenge of balancing immune recovery and inflammatory control during PML treatment.

In conclusion, this case highlights the value of ultrasensitive PCR for diagnosing HIV-associated PML when standard PCR is inconclusive. As ART reduces JCV levels in patients with PML, ultrasensitive diagnostics are critical for timely and accurate diagnosis and management of PML and PML-IRIS in the ART era.

Authorship statement

A.K. and T.N. supervised the study. All authors approved the final version of the manuscript. All authors met the ICMJE authorship criteria. K. Nakano, A.K., T.N., R.K., S.A., E.I., N.A., H.U., D.M., T.A., K. T., and H.G. were involved in the clinical care and management of the patient, collection of data, and drafting of the manuscript. K. Nakamichi performed the PCR testing for CSF JCV and edited the manuscript.

CRediT authorship contribution statement

Teruya Katsuji: Writing – review & editing, Supervision, Funding acquisition, Conceptualization. **Nakano Kenji:** Writing – original draft, Data curation, Conceptualization. **Gatanaga Hiroyuki:** Writing – review & editing, Supervision. **Kawashima Akira:** Writing – review & editing, Supervision, Conceptualization. **Nakamoto Takato:** Writing – review & editing, Supervision, Conceptualization. **Nakamichi Kazuo:** Writing – review & editing, Funding acquisition, Data curation. **Mizushima Daisuke:** Writing – review & editing, Supervision. **Aoki Takahiro:** Writing – review & editing, Supervision. **Uemura Haruka:** Writing – review & editing, Supervision. **Kuwata Ryo:** Writing – review & editing, Supervision. **Abe Seitaro:** Writing – review & editing, Supervision. **Inoue Eri:** Writing – review & editing, Supervision. **Ando Naokatsu:** Writing – review & editing, Supervision.

Patient consent

Written informed consent for publication was obtained from the patient. Ethical committee approval was not required for this study.

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Declaration of Competing Interest

We declare no competing interests.

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