

Case Study

Diagnosis of Richter Transformation in CNS Using CSF cell-free DNA and RNA Sequencing by NGS



- Liquid biopsy using CSF is less invasive and lower risk than tissue biopsy in brain tumors and cranial metastases
- CSF is useful for molecularly characterizing brain lesions
- NGS on CSF can identify disease transformation that are challenging to identify by other modalities

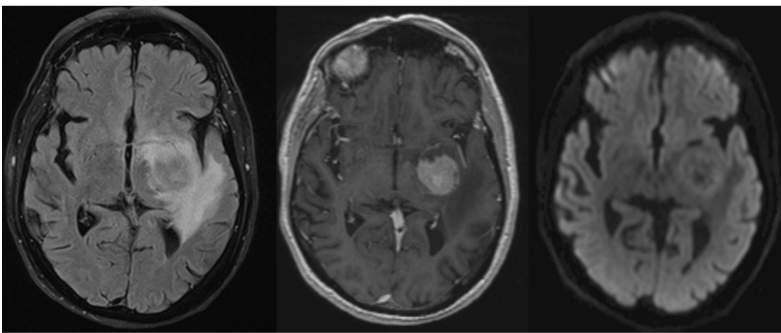
Background

We present a case of an 80-year-old patient with a history of chronic lymphocytic leukemia (CLL) in remission, who developed progressive neurologic symptoms. Imaging revealed a solitary brain lesion concerning high-grade tumor. CSF cytology and flow cytometry were inconclusive, but cerebrospinal fluid (CSF) NGS revealed a complex mutational profile consistent with aggressive diffuse large B-cell lymphoma (DLBCL) with biallelic TP53 abnormalities, consistent with Richter transformation of CLL involving the central nervous system (CNS). The case highlights the diagnostic utility of CSF-based cell-free DNA and RNA NGS in the setting of high-risk biopsy and ambiguous radiologic differentials.

T2 FLAIR

T1-Post

Diffusion



CSF Cytological and Chemical Analysis

- Elevated Protein and glucose
- Cytology and Flow Cytometry: No monoclonal B cells detected; negative for malignant cells

Clinical History

The patient has a history of chronic lymphocytic leukemia (CLL), diagnosed years ago and treated with six cycles of R-CHOP chemotherapy, presented with few weeks of progressive headache, short-term memory impairment, and worsening word-finding difficulties. The patient had been in remission and followed annually.

Neurologic Imaging

MRI brain showed an irregular, peripherally enhancing lesion with vasogenic edema in the left temporoparietal region.

- T2-FLAIR: Extensive surrounding hyperintensity with mass effect
- T1 Post-contrast: Heterogeneous enhancement
- Diffusion-weighted imaging: Peripheral diffusion restriction

Differential Diagnosis

- Primary CNS tumor (e.g., high-grade glioma)
- CNS lymphoma (possibly recurrent DLBCL)
- Brain metastasis from unknown primary
- Less likely: Langerhans cell histiocytosis (LCH), atypical infection, inflammatory lesion

Laboratory Findings

- Elevated LDH and Beta-2 Microglobulin

CSF NGS Findings

- Somatic mutations: XRCC2, CHEK2, ETV6, TP53, PIK3R1, IDH2, SF3B1, MYD88, PIM1 (7 mutations), SETBP1, KMT2D, IRF4 (2), KMT2C, DNMT3A (2), KAT6A
- Chromosomal abnormalities: 1p-, proximal 1q+, distal 1q-, -6, +12, 17p- (TP53 deletion)
- Clonality:
 - B-cell: Detected (IGHV3-33 / IGKV3-20)
 - T-cell: Not detected
- Blast markers: Low-level expression (CD1A, CD34, CD117, TdT)
- B-cell markers: Low-level with abnormal pattern
- T-cell markers: Increased but normal pattern
- mRNA Expression:
 - MYC and BCL2: Increased (suggestive of promoter hijacking)
 - Low CCND1 and SOX11 expression
- Viral RNA: EBV, HPV, TTV — Not detected
- HLA Genotyping: A*24:02-A*02:01; B*27:05-B*18:01; C*07:01-C*02:02

Findings were most consistent with Richter transformation of CLL to aggressive diffuse large B-cell lymphoma involving the CNS, with biallelic TP53 alterations. The detection of XRCC2 and CHEK2 mutations at high levels raises the possibility of underlying germline predisposition.

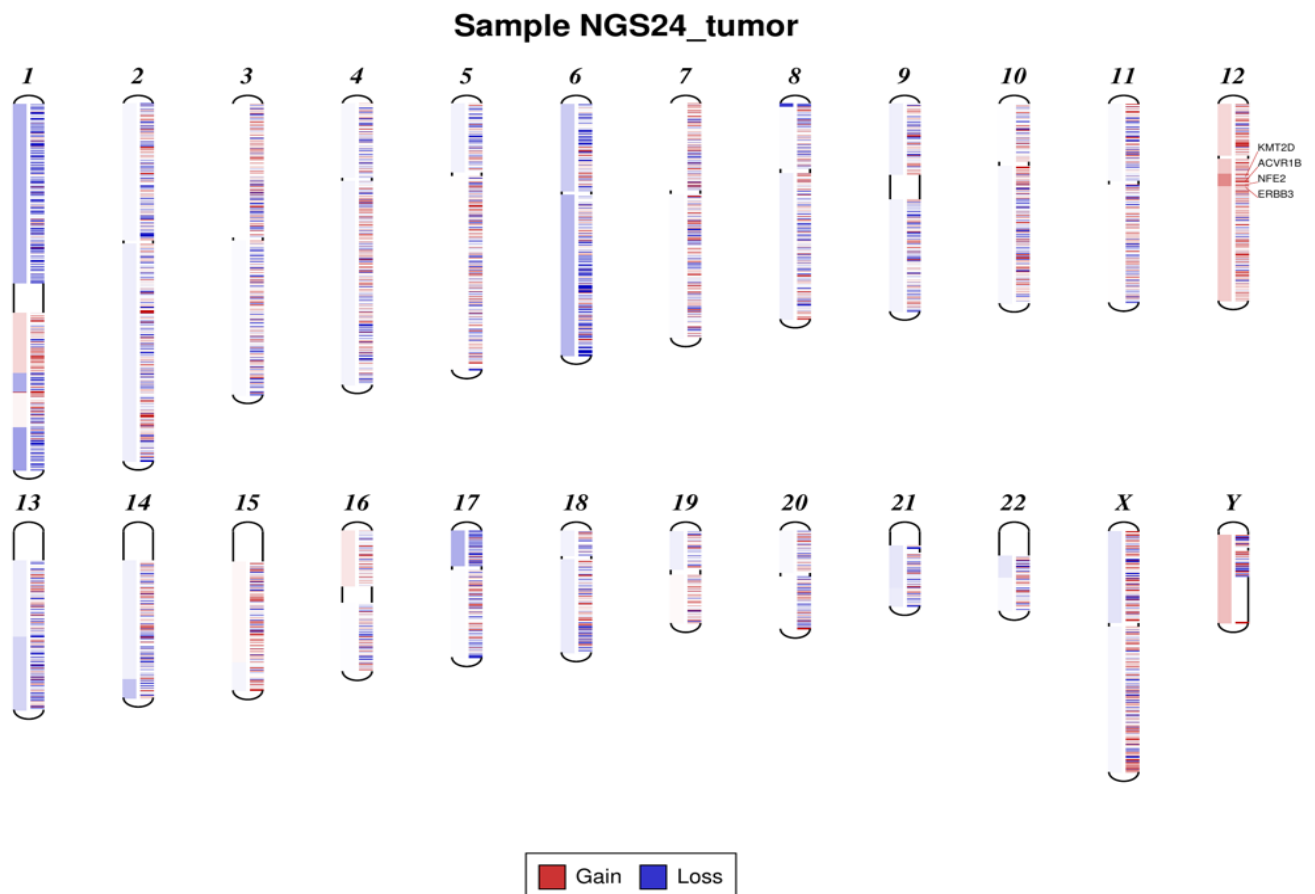
Discussion

Richter transformation to DLBCL is a known complication of CLL, occurring in 2–10% of cases, and is rarely associated with isolated CNS involvement. CNS involvement is more common with Richter transformation, but is difficult to diagnose due to nonspecific imaging findings, frequent absence of malignant cells in CSF cytology, and high-risk of biopsy in eloquent brain areas. This case underscores the pivotal role of CSF-based NGS cell-free DNA and RNA sequencing, which provided a molecular diagnosis and guided treatment in a case where conventional diagnostics were limited.

Conclusion

This case illustrates the importance of advanced molecular diagnostics, particularly CSF-based next-generation sequencing of cell-free DNA and RNA, in evaluating CNS lesions and leptomeningeal involvement in patients with a history of malignancy. When traditional diagnostics—including CSF cytology and flow cytometry—were inconclusive, CSF NGS enabled a successful diagnosis with low morbidity. Notably, the diagnosis of CNS lymphoma with MYD88 alteration was achieved despite the patient being on high-dose dexamethasone, which can suppress cellular yield and confound conventional testing.

The molecular results facilitated a minimal time to next treatment without the delay and complications of post-operative recovery. Furthermore, this approach avoided empiric treatment missteps, which carry risk in cases with overlapping radiographic differentials (e.g., high-grade glioma, infection, metastasis). In summary, this case emphasizes that CSF NGS may represent a frontline diagnostic strategy in patients with high-risk CNS lesions and prior hematologic malignancy when traditional methods are limited by feasibility or sensitivity.



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