THERAPEUTIC ADVANCES in
Medical Oncology

Review

Novel insights into the biomarkers and therapies for primary central nervous system lymphoma

Ther Adv Med Oncol

2022, Vol. 14: 1–22

DOI: 10.1177/17588359221093745

Yujia Zhai, Xiangxiang Zhou and Xin Wang

***Abstract*:** Primary central nervous system lymphoma (PCNSL) is a rare and highly aggressive extranodal type of non-Hodgkin lymphoma. After the introduction and widespread use of high-dose-methotrexate (HD-MTX)-based polychemotherapy, treatment responses of PCNSL have been improved. However, long-term prognosis for patients who have failed first-line therapy and relapsed remains poor. Less invasive diagnostic markers, including the circulating tumor DNAs (ctDNAs), microRNAs, metabolomic markers, and other novel biomarkers, such as a proliferation inducing ligand (APRIL) and B-cell activating factor of the TNF family (BAFF), have shown potential to distinguish PCNSL at an early stage, and some of them are related with prognosis to a certain extent. Recent insights into novel therapies, including Bruton tyrosine kinase (BTK) inhibitors, immunomodulatory drugs, immune checkpoint inhibitors, PI3K/mTOR inhibitors, and chimeric antigen receptor (CAR) T cells, have revealed encouraging efficacy in treatment response, whereas the duration of response and long-term survival of patients with relapsed or refractory PCNSL (r/r PCNSL) need further improvement. In addition, the diagnostic efficiency of novel markers and the antitumor efficacy of novel therapies are needed to be assessed further in larger clinical trials. This review provides an overview of recent research on novel diagnostic markers and therapeutic strategies for PCNSL.

Keywords: CAR T cells, diagnostic markers, novel agents, precision therapies, primary central nervous system lymphoma

Received: 20 February 2021; revised manuscript accepted: 24 March 2022.

Correspondence to:
Xiangxiang Zhou
Department of Hematology, Shandong Provincial Hospital Affiliated to Shandong First Medical University, Jinan 250021, Shandong, China

School of Medicine, Shandong University, Jinan, China

Shandong Provincial Engineering Research Center of Lymphoma, Jinan, China

Branch of National Clinical Research Center for Hematologic Diseases, Jinan, China

National Clinical Research Center for Hematologic Diseases, The First Affiliated Hospital of Soochow University, Suzhou, China
xiangxiangzhou@sdu.edu.cn

Xin Wang
Department of Hematology, Shandong Provincial Hospital, Cheeloo College of Medicine, Shandong University, No. 324, Jingwu Road, Jinan 250021, Shandong, China

Department of Hematology, Shandong Provincial Hospital Affiliated to Shandong First Medical University, Jinan, China

School of Medicine, Shandong University, Jinan, China

Introduction

Primary central nervous system lymphoma (PCNSL) is a highly aggressive extranodal type of non-Hodgkin lymphoma (NHL), with an overall incidence of approximately 0.5/100,000 per year.[1] However, there are still numerous challenges in terms of its early diagnosis and effective treatment. As reported in previous retrospective studies, the proper diagnosis of PCNSL patients is often delayed due to nonspecific initial clinical symptoms and nonspecific findings obtained from magnetic resonance imaging (MRI) – the commonly used diagnostic technique. A study by the French oculo-cerebral lymphoma network (LOC) of 1002 immunocompromised PCNSL patients showed a median diagnosis delay of 35 days (range 0–6.7 years),[2] similar to the 47-day delay of 327 immunocompetent patients with PCNSL in Spain.[3] Notably, early diagnosis followed by timely treatment of patients with PCNSL with lower tumor burden and better performance conditions could lead to better prognosis.[4] Thus, novel diagnostic markers of PCNSL are urgently required.

The majority (90%) of patients with PCNSL present with diffuse large B-cell lymphomas (DLBCL), which express pan B-cell marker (CD20). The classical first-line treatment regimen R-CHOP (rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone) showed poorer efficacy for PCNSL than for systemic DLBCL, partly due to its inefficacy in crossing the blood-brain barrier (BBB).[5] Comprehensive

Shandong Provincial Engineering Research Center of Lymphoma, Jinan, China

Branch of National Clinical Research Center for Hematologic Diseases, Jinan, China

National Clinical Research Center for Hematologic Diseases, The First Affiliated Hospital of Soochow University, Suzhou, China
xinw007@126.com

Yujia Zhai
Department of Hematology, Shandong Provincial Hospital, Cheeloo College of Medicine, Shandong University, Jinan, China

chemotherapy based on high-dose methotrexate (HD-MTX) has become a cornerstone treatment for PCNSL.[6] Whole-brain radiotherapy (WBRT) and high-dose chemotherapy (HDC) supported by autologous stem cell transplantation (ASCT) are recent options for consolidation treatment.[7] Children with PCNSL, who are below 19 years without immunodeficiency, could achieve an exceptional event-free survival (EFS) of 74% ± 5% on receiving treatment followed by a histological subtype-driven (including HD-MTX and HD cytarabine) and radiation-free protocol.[8] Regrettably, PCNSL remains a disease with high refractory and relapse rate, with more than 20% of PCNSL patients showing resistance to first-line therapies and nearly half of patients eventually suffering a relapse.[9] Although the median overall survival (OS) has improved significantly from 2.5 months to 26 months over the past few decades,[10] the 5-year survival rate of patients with PCNSL is still very low, especially in elderly and fragile patients who cannot tolerate chemotherapy. Overall, the limited prognosis of PCNSL highlights the need for novel therapeutic agents.

Based on recent molecular insights into the underlying PCNSL-related genomic alterations in patients, many clinical trials have been established to assess the efficacy of novel therapies against PCNSL. This review focuses on the recently discovered novel diagnostic biomarkers and therapeutic strategies for PCNSL in immunocompetent patients.

Novel diagnostic markers for PCNSL

Liquid biopsy biomarkers

Circulating tumor DNAs. Circulating tumor DNAs (ctDNAs) are DNA fragments derived from apoptosis, necrosis, or secretion of tumor cells. Pretreatment ctDNA levels have been demonstrated to be independent prognostic predictors of aggressive lymphomas.[11] In an investigation on six patients with CNSL (including five PCNSL), the digital polymerase chain reaction (PCR) analyses of cerebrospinal fluid (CSF)-derived ctDNAs proved effective for detecting somatic mutations in accordance with next generation sequencing (NGS) analysis of biopsy tissues.[12] Another study evaluating the CSF and plasma from 6 patients with restricted CNSL, 1 with systemic and CNS lymphoma, and 12 with systemic lymphoma, indicated that CSF ctDNAs might detect CNS relapse earlier and the presence of residual disease better than conventional methods, such as flow cytometry (FC).[13] NGS analysis was performed on 11 newly diagnosed PCNSL patients who received ibrutinib-based therapy. CtDNAs in CSF were detected before and during treatment, and the results showed sensitive surveillance of tumor burden and assessment of treatment responses.[14]

Although more than 70 aberrant genes have been reported in PCNSL, a half of which are related to the nuclear factor-κB (NF-κB) pathway, MYD88 and CD79B are the only two that have been reported to have the diagnostic abilities in liquid biopsy.[15] The potential application of MYD88 L265P mutation in noninvasive diagnosis of PCNSL has been implied in clinical cases. The successful detection of the MYD88 L265P mutation in CSF using droplet digital PCR (ddPCR) has been documented in secondary CNSL (SCNSL).[16] The detection rate of MYD88 L265P in the CSF of 14 patients with PCNSL was 86% in a retrospective study, and in three cases of which wherein no lymphoma cells were detected using cytology and FC, ddPCR could detect ctMYD88 L265P at the time of diagnosis.[17] In a recent retrospective study of 17 patients who were newly diagnosed with PCNSL, the concordance of MYD88 L265P mutation between CSF and tissue samples was found to be 82%.[18] In a phase Ib trial of an ibrutinib-based therapy for patients with r/r CNSL, high ctDNA mutations (60%) in B-cell antigen receptor (BCR) pathway genes, including MYD88 L265P and CD79B Y196, were detected in the CSF using MSK-HemPACT (an NGS-based tumor-sequencing assay), and the changes of these mutations were consistent with treatment responses.[19] In addition, concomitant assessment of CSF MYD88 L265P mutation and IL-10 could significantly improve the diagnostic sensitivity and specificity of distinguishing PCNSL from other CNS disorders by 94% and 98%, respectively.[18]

Notably, a recent study that analyzed MYD88 and CD79B mutations in 27 patients with PCNSL at diagnosis showed significantly lower detection levels of these two hotspot mutations in blood-derived ctDNAs than in stereotactic biopsies, suggesting that MYD88 and CD79B mutations in blood-derived ctDNAs did not have reliable diagnostic value for PCNSL.[20]

MicroRNAs. MicroRNAs (miRNAs) are small non-coding RNAs, 20–24 nucleotide long, and some of them have been indicated to

have diagnosis efficacy for lymphomas. Several miRNAs, such as miR-107, have shown diagnostic and prognostic indicator potential for DLBCL.[21] On one hand, a few members of CSF-derived miRNAs have demonstrated encouraging diagnostic accuracy for PCNSL from other brain diseases. A meta-analysis indicated that CSF-based miRNA assays seem to be more accurate and sensitive for the diagnosis of PCNSL than other widely used techniques, including neuroimaging MRI, cytopathologic, and immunophenotypic analyses of CSF cells.[22] Several studies have demonstrated that miR-19b, miR-21, and miR-92a quantified by qRT-PCR were significantly increased in CSF collected from patients with PCNSL,[23] compared with those from the control group with other neurologic disorders.[24] Combined analyses of these three miRNAs in CSF have shown diagnostic accuracy of PCNSL with a sensitivity of 95.7% and specificity of 96.7%. In addition, significantly lower miR-30c levels in CSF detected by miRNA arrays could distinguish patients with PCNSL from those with SCNSL with 90.9% sensitivity and 85.5% specificity.[25]

The diagnostic efficiency and prognostic value of blood-derived miRNAs in PCNSL have also been investigated. A study based on available blood samples from patients entering the G-PCNSL-SG1 trial compared the peripheral blood miRNA levels in a short-term survival group (median OS of 3 months) of 20 patients with PCNSL with those in a long-term survival group (median OS of 55 months) using NGS. According to the results, a total of 12 miRNAs were significantly deregulated between these two groups. Among them, miR-151a-5p and miR-151b differed prominently and exhibited the largest differential expression.[26] While considering the limited number of available patients, verifications in larger population cohorts are required. Higher serum miR-21 levels were also associated with poorer prognosis in another survival analysis that included 56 patients with PCNSL. Multivariate Cox proportional hazard analysis demonstrated that serum miR-21 expression level could be an independent prognostic biomarker for PCNSL.[27]

For formalin-fixed paraffin-embedded (FFPE) biopsy tissues from PCNSL, there was also a distinction in the miRNA expression panel between PCNSL and nodal DLBCL, with 13 significantly upregulated and 5 significantly downregulated miRNAs.[28] Among them, the upregulated miRNAs in PCNSL included miR-17-5p, miR-20a, miR-9 (related to the Myc pathway), miR-30b/c, and miR-155. The downregulated miRNAs consisted of miR-199a, miR-214, miR-193b, and miR-145, which are putative tumor-suppressor miRNAs. Furthermore, PCNSL showed a significantly increased expression level of miR-17-5p and a significantly decreased level of miR-127 compared to those in primary testicular lymphoma (PTL).[23] Nevertheless, a recent study indicated that the miRNA levels in CSF and FFPE biopsy tissues from PCNSL patients did not match with non-overlapped miRNAs differentiated compared with tissues from nonmalignant brain lesions.[29] One explanation for such discrepancies is that CSF miRNAs may be derived not only from lymphoma cells but also from other cells, including those from the subarachnoid space, ventricular system, ventricular choroid plexus, and spinal cord.[30] Differentially expressed miRNAs between PCNSL and other diseases are shown in Table 1.

Metabolomic biomarkers

Metabolomics can identify and quantify small molecular metabolic substrates and products of various metabolic pathways. Analytical tools for metabolomics mainly include the integration of mass spectrometers (MSs), ion mobility systems (IMSs), gas chromatography (GC), capillary electrophoresis (CE), liquid chromatography (LC), and nuclear magnetic resonance (NMR) spectrometers.[31] Cancer cells have metabolomic pathways distinct from those of normal cells regarding the absorption of nutrients for immortal proliferation. Excessive glycolysis *via* PI3 K/AKT/mTOR and RAS/MAPK signaling has been reported in MTX-resistant PCNSL-derived cells.[32] A metabolomics analysis of 163 CSF samples collected from clinical patients (including 59 with PCNSL) showed that 81, 17, and 188 metabolic features were significantly altered in PCNSL compared with those in non-tumorous brain lesions, SCNSL, and patients with lung adenocarcinoma with brain metastases (MBT), respectively. Three independent panels based on 5 (L-glutamine, 5-aminoimidazole, L-cystine, butyrylcarnitine 9, phytosphingosine), 4 (valyl-methionine, inositol phosphate, homocysteine, 5-aminoimidazole), and 13 different metabolites could effectively diagnose PCNSL *versus* non-tumorous brain lesions, SCNSL, and MBT, respectively.[33] A phase I study (NCT01542918) of lenalidomide also revealed the metabolomic profile of CSF obtained from 14 relapsed PCNSL patients

Table 1. MiRNAs differentially expressed between PCNSL and other diseases at diagnosis.

Samples	Comparation	Methods	Upregulated	Downregulated
FFPE biopsy samples	Nodal DLBCL	qRT-PCR	miR-9 miR-20b miR-155 miR-340 miR-17-5p miR-148a miR-30b miR-27b miR-26b miR-146b miR-20a miR-30c miR-let-7g	miR-199a miR-214 miR-432 miR-193b miR-145
	PTL	qRT-PCR	miR-17-5p	miR-127
	n-ML[a]	qRT-PCR	miR-155 miR-196b	miR-9 miR-125b, miR-let-7b
Blood samples	LTS: STS[b]	NGS	miR-151a-5p miR-151b miR-106a-5p miR-17-5p miR-183-3p miR-30d-5p miR-96-5p miR-194-5p miR-503-5p	miR-219-5p miR-6130 miR-181c-3p
	Glioblastomas, CNS inflammation, metastases, healthy controls, respectively	qRT-PCR	miR-21	
CSF samples	Various neurologic disorders[c]	qRT-PCR	miR-21 miR-19b miR-92a	
	SCNSL	microRNA arrays	miR-222	miR-16 miR-30b miR-30c miR-191 miR-204
	n-ML[a]	qRT-PCR	miR-21 miR-19b miR-92a	

CSF, cerebrospinal fluid; FFPE, formalin-fixed paraffin-embedded; MiRNA, MicroRNAs; NGS, next generation sequencing; OS, overall survival; PCNSL, primary central nervous system lymphoma; PCR, polymerase chain reaction; PTL, primary testicular lymphoma; qRT-PCR, quantitative reverse transcription-PCR; SCNSL, secondary central nervous system lymphoma.
[a]n-ML, nonmalignant brain lesions (including gliosis, cavernous malformation, cyst, abscess, hematoma, ischemic necrosis, lymphocytic inflammation, toxoplasmosis, focal cortical dysplasia).
[b]LTS: STS, long-term survival group (median OS of 55 months) with those in short-term survival group (median OS of 3 months) of PCNSL patients.
[c]Various neurologic disorders (including CNS inflammation, neurocardiogenic syncope, tension headache, brain infarction, polyneuropathy, migraine, epilepsy, and transient ischemic attack).

compared with that obtained from 14 controls without malignant diseases. Among all 145 enrolled metabolites, 36 that were mainly involved in energy metabolism pathways were significantly elevated in patients with PCNSL compared with those in controls. Significantly higher levels of lactate, 5-methylthioadenosine, and kynurenine in the CSF helped to differentiate PCNSL from non-malignant diseases. Elevated levels of both lactate and kynurenine in the CSF were associated with shorter survival.[34] In addition, another quantitative metabolomic analysis demonstrated that elevated CSF lactate and reduced GABA (γ-aminobutyric acid) were significantly related to the lesion size of lymphoma as well as neurocognitive deficits in both PCNSL and SCNSL.[35]

Functional imaging

Compared with traditional anatomical imaging, positron emission tomography (PET) can detect abnormal functional areas and is valuable in cancer diagnosis. However, the most routinely used tracer [^{18}F] fluorodeoxyglucose (FDG) has its limitations in diagnosing PCNSL due to the high physiologic cerebral glucose metabolism[36] and it is not a specific radiotracer that could increase in several benign conditions. Several novel probes have shown potential specificity in the diagnosis of PCNSL and exert research value in further clinical trials.

[^{18}F] fludarabine (FDB) can be trapped after being transported into cells by equilibrative nucleoside transporters, ENT1 and ENT2, and initial phosphorylation by deoxycytidine kinase (dCK).[37] As increased levels of dCK expression are observed in lymphoid malignancies, [^{18}F] FDB was evaluated to target lymphoid cells. In a human CNSL xenograft rat, [^{18}F] FDB exhibited a tumor-to-background ratio (TBR) that was two to threefold higher than that of [^{18}F] FDG. Therefore, compared to using [^{18}F] FDG ($p = 0.04$), [^{18}F] FDB-PET shows encouraging potential to distinguish PCNSL from glioblastoma ($p < 0.01$).[38]

CXCR4 is highly expressed on PCNSL cells and binds to CXCL12 to establish the CXCL12/CXCR4 signaling pathway, which is important in the homing of tumor cells.[39] PET imaging using ^{68}Ga-Pentixafor, a CXCR4-directed tracer, could exhibit PCNSL in excellent contrast to the surrounding brain parenchyma, and might be a novel diagnostic biomarker for PCNSL.[40] Considering the high CXCR4 expression levels in glioblastoma,[39] the efficacy of CXCR4-directed imaging in distinguishing PCNSL from glioma should be investigated further in prospective clinical trials.

Other novel biomarkers

A proliferation inducing ligand, B-cell activating factor of the tumor necrosis factor family, and their receptors. Two significantly homologous members of the tumor necrosis factor (TNF) superfamily, B-cell activating factor of the TNF family (BAFF) and a proliferation inducing ligand (APRIL), play important roles in the pathogenesis of B-cell malignancies. Serum BAFF levels have been observed to be higher in patients with aggressive NHL and are related to treatment response and OS.[41] Immunohistochemical staining has shown that BAFF and APRIL are both expressed in PCNSL tissue specimens.[42] APRIL and BAFF have the potential to be reliable diagnostic biomarkers for PCNSL. A prospective analysis of the CSF collected from 116 patients with focal brain lesions containing 53 CNSLs (30 of which were PCNSL) demonstrated that CSF levels of APRIL and BAFF in patients with CNSL were significantly higher than those in patients with any other focal lesions, such as primary brain tumors (PBTs), MBT, and autoimmune-inflammatory diseases.[43] The elevated APRIL levels in CSF revealed promising potential application as diagnostic marker for CNSL, with a specificity of 93.7% and sensitivity of 62.3%. The integration of BAFF levels with APRIL levels further improved the diagnostic accuracy to a specificity of 96.1% and a sensitivity of 77.3%. In the subgroup analysis, patients with PCNSL who achieved complete response (CR) showed remarkable decrease in both APRIL and BAFF CSF levels, compared with those in untreated patients.

APRIL and BAFF, as well as their two receptors, B-cell maturation antigen (BCMA) and transmembrane activator and CAML interactor (TACI), could serve as potential diagnostic biomarkers for PCNSL. Soluble TACI and BCMA were found to be elevated in the CSF of patients with PCNSL, compared with those in patients with other neurological diseases.[44] The TACI level in the CSF could differentiate PCNSL with high specificity (88.3%) and sensitivity (87.9%). The diagnostic specificity increased to 96.7% when CSF BCMA was combined with TACI levels. Notably, serum TACI and BCMA levels were not elevated in patients with PCNSL and do not

exhibit diagnostic value. In a small sample study of nine patients with PCNSL, the combination of elevated levels of CSF BAFF and its receptor TACI revealed 100% sensitivity and 100% specificity for the diagnosis of PCNSL.[45]

Currently, no single diagnostic modality is either sensitive enough or specific enough for clinical application. Integration of multivariable diagnostic modalities is necessary for the early diagnosis and treatment selection in PCNSL. When a patient presents with initial clinical symptoms of a brain tumor, MRI is commonly the first important diagnostic test. Functional imaging such as PET can be a more specific complement to this step. If PCNSL is suspected based on imaging findings, tissue-based brain pathology biopsy, which is still considered as the gold standard, is necessary for the definite diagnosis of PCNSL, unless fluid-based analysis is diagnostic. Unfortunately, the clinical use of tissue-based biopsy is limited mainly due to the risk of postoperative complications, such as intracranial hemorrhage and functional damage. In some cases, the tumor location is difficult or unsuitable to access for tissue-based biopsy, so fluid-based diagnostic techniques could be less invasive and more helpful. Besides, the use of corticosteroids hampers histopathological diagnosis, as they can reduce diagnostic sensitivity.[15] Fluid-based analysis of cytomorphology, such as FC should be performed. Compared with traditional liquid analysis, novel biomarkers including ctDNAs, miRNAs, and metabolomic markers might have advantages in terms of sensitivity and specificity. These fluid-based diagnostic markers facilitate more accurate stratification, prognosis evaluation, and choice of targeted treatment at an early stage, which could be optional choices. An overview of diagnostic value of novel biomarkers is summarized in Table 2. Liquid biopsy can overcome tumor heterogeneity to detect minimal residual disease (MRD) during early relapse. However, in current clinical practice, only a subset of patients can be diagnosed definitively based on typical liquid biopsy. Also, concerns regarding the waiting time for fluid-based diagnostic results and prioritization of different techniques remain to be resolved. Therefore, tissue-based brain biopsy should be carried out in a timely manner if there are no contraindications. In addition, to distinguish PCNSL from SCNSL, whole-body PET-CT and bone marrow biopsy should be performed. Furthermore, ocular examinations, such as vitrectomy, are required for the detection of ocular involvement.

Novel therapies for PCNSL

Bruton tyrosine kinase inhibitors

Ibrutinib. Bruton tyrosine kinase (BTK) links BCR and Toll-like receptor (TLR) signaling, thus preventing the activation of NF-κB pathway.[47] Ibrutinib is a primary inhibitor of BTK, and has single-drug activity in chronic lymphocytic leukemia (CLL)[48] and other B-cell malignant tumors,[49] including mantle cell lymphoma (MCL), Waldendström macroglobulinemia (WM), marginal zone lymphoma, and follicular lymphoma (FL). Systematic sequencing of PCNSL revealed that CD79B of the BCR signaling pathway accompanied with MYD88 is more frequently mutated in PCNSL than in systemic DLBCL,[50] suggesting promising antitumor efficacy of ibrutinib.

Ibrutinib monotherapy has been observed to reveal an encouraging overall response rate (ORR) of 50% in 14 r/r PCNSL patients. Among them, two patients achieved long-lasting CR of 8 months without severe toxicities in a multicenter retrospective case series.[51] The median duration of response and progression-free survival (PFS) in all patients were 4 and 6 months, respectively. The final outcomes of a phase II clinical trial for r/r PCNSL and primary vitreoretinal lymphoma (PVRL) conducted by LYSA and LOC also demonstrated that ibrutinib monotherapy (560 mg/day) was highly clinically effective (70% of evaluable patients achieved disease control) and tolerated (only one patient experienced fatal pulmonary aspergillosis), with a median PFS of 4.8 months in all.[52] In addition, another single-center study (NCT02315326) including 13 r/r PCNSL and 7 r/r SCNSL patients also showed good single-agent activity of ibrutinib. In total, 10 (77%) patients with PCNSL [5 CR and 5 partial response (PR)] and 5 with SCNSL (including 4 CR) responded. However, the duration of response was short, with the median PFS of 4.6 months (95% CI = [2.4, 7.5] months) in PCNSL and 7.43 months in SCNSL. Genomic analysis revealed that the mutation within the coiled-coil domain of CARD11 seemed to be associated with ibrutinib resistance.[53] The latest study by Lewis also reported similar results.[54] Even with an encouraging ORR of 58% and CR rate of 55% for ibrutinib across the entire

Table 2. Overview of diagnostic value of novel biomarkers for PCNSL.

Biomarker	Number of patients	Sample	Methods	Sensitivity	Specificity	Reference
Combination of MYD88 and IL-10	36 PCNSL and 106 neurological controls	CSF	TaqMan-based PCR assay for MYD88	94%	98%	Ferreri *et al.*[18]
Combination of miR-21, miR-19b, and miR-92a	23 PCNSL and 30 controls	CSF	qRT-PCR	95.7%	96.7%	Baraniskin *et al.*[24]
	30 PCNSL and 23 n-ML	CSF	qRT-PCR	63.33%	80.77%	Zajdel *et al.*[29]
MiR-30c	55 PCNSL and 11 SCNSL	CSF	qRT-PCR	90.9%	85.5%	Baraniskin *et al.*[25]
Combination of miR-155 and miR-let-7b	35 PCNSL and 23 n-ML	FFPET	qRT-PCR	96%	98%	Zajdel *et al.*[29]
MiR-21 and RNU2-1f	72 PCNSL and 47 controls	CSF	qRT-PCR	91.7%	95.7%	Baraniskin *et al.*[46]
APRIL	53 CNSL (30 PCNSL) and 63 controls	CSF	ELISA	62.3%	93.7%	Mulazzani *et al.*[43]
BAFF		CSF	ELISA	47.1%	93.7%	
Elevation of APRIL and/or BAFF				77.3%	96.1%	
TACI	33 PCNSL and 143 controls	CSF	ELISA	87.9%	88.3%	Thaler *et al.*[44]
BCMA			ELISA	72.7%	71.8%	
Combination of TACI and BCMA				63.9%	96.7%	
Combination of TACI and BAFF	9 PCNSL and 73 controls	CSF	ELISA	100%	100%	Mizutani *et al.*[45]

APRIL, a proliferation inducing ligand; BAFF, B-cell activating factor of the tumor necrosis factor family; BCMA, B-cell maturation antigen; CSF, cerebrospinal fluid; ELISA, enzyme linked immunosorbent assay; FFPET, formalin-fixed paraffin-embedded tissue; n-ML, nonmalignant brain lesions; PCNSL, primary central nervous system lymphoma; PCR, polymerase chain reaction; qRT-PCR, quantitative reverse transcription-PCR; RNU2-1f, circulating U2 small nuclear RNA fragments; SCNSL, secondary central nervous system lymphoma; TACI, transmembrane activator and CAML interactor.

cohort of 33 patients with CNSL (9 PCNSL and 24 SCNSL), the median PFS was 10.2 months for SCNSL and short, at 3.1 months, for PCNSL. Thus, considering the relatively short duration of responses in PCNSL, as several studies have reported, combination therapies of ibrutinib for the treatment of PCNSL are needed. A phase Ib study of 18 patients with PCNSL (five untreated, 13 r/r) investigating the potential of ibrutinib monotherapy and together with chemotherapy (DA-TEDDi-R), showed significant response improvement with ibrutinib achieving significant CNS penetration and high free-drug concentrations.[55] During ibrutinib monotherapy (maximum dose of 840 mg/day) treatment, 94% of patients (17/18) showed tumor reductions and 83% (15/18) achieved PR. Out of the 14 evaluable patients who carried on with DA-TEDDi-R, 86% achieved CR, suggesting ibrutinib could strengthen the efficacy of chemotherapy.

Considering the high incidence of aspergillosis (39%) in this trial, two patients developed aspergillosis during ibrutinib monotherapy and five

patients developed aspergillosis during DA-TEDDi-R treatment, and other clinical trials of ibrutinib-based combination therapies are needed to reduce the risk of adverse events (AEs). Another phase Ib clinical trial (NCT02315326) explored ibrutinib (560 or 840 mg/d) /HD-MTX/rituximab combination in 15 patients with CNSL (9 PCNSL and 6 SCNSL).[19] This three-drug combination was well tolerated without dose-limiting toxicity (DLT) and a high response rate (89% in PCNSL and 67% in SCNSL) with a median duration of response of 12.8 months. A phase Ib clinical trial (NCT03703167) of ibrutinib combined with lenalidomide and rituximab for r/r CNSL is ongoing. In addition, ibrutinib as a maintenance treatment for elderly patients with primary CNS DLBCL is being investigated in a phase II clinical trial (NCT02623010). Patients who achieved MRI-documented CR or PR after rituximab and HD-MTX induction treatment would receive maintenance treatment including ibrutinib 560 mg/day until disease progression or occurrence of limiting toxicities.

Other BTK inhibitors

Other novel BTK inhibitors with better selectivity toward BTK than ibrutinib are investigated in PCNSL patients, including tirabrutinib, zanubrutinib, and acalabrutinib, which intend to improve antitumor effect and reduce AEs attributed to the inhibition of off-target enzymes.[56] Tirabrutinib (ONO/GS-4059) has exhibited positive antitumor effects and well safety profiles in r/r B-cell malignancies.[57] It was first approved at the recommended dosage of 480 mg/day in 2020 for the treatment of r/r PCNSL in Japan.[58] A phase I/II study was carried out for tirabrutinib in 44 enrolled patients with r/r PCNSL, including 20 on 320 mg/day, 7 on 480 mg/day, and 17 on 480 mg/day under fasted conditions. The ORR was 63.6% (60.0%, 100.0%, and 52.9%, respectively, for each treatment group), and the overall median PFS was 2.9 months (2.1, 11.1, and 5.8 months, respectively, for each treatment group). Common AEs of grade $\geqslant$ 3 were leukopenia, lymphopenia, neutropenia, and erythema multiforme. Only one patient in the 480 mg treatment group had Grade 5 AEs.[59] Zanubrutinib was developed in China and approved by the US Food and Drug Administration (FDA) for patients with MCL who have received $\geqslant$ 1 prior therapy. In addition, zanubrutinib has revealed promising antitumor activity and good tolerance in the treatment of WM and CLL/small lymphocytic lymphoma (SLL).[60] A successful case of refractory PCNSL treated with zanubrutinib has been reported.[61] The 53-year-old patient who developed progressive disease (PD) after two courses of HD-MTX-based regimen achieved CR after subsequent three courses of zanubrutinib plus chemotherapy. The patient received ASCT after four courses of zanubrutinib with chemotherapy, and his MRI revealed stable CR. Based on the case above, it would be interesting to test whether zanubrutinib could be added into first-line regimens for PCNSL to improve curative effect. The phase II clinical trial (NCT04938297) that evaluates the efficacy and toxicity of zanubrutinib in combination with rituximab and lenalidomide, followed by zanubrutinib or lenalidomide maintenance, is recruiting. Acalabrutinib (ACP-196) is another more selective second-generation BTK inhibitor with less off-target effects than ibrutinib. It has been approved for the treatment of relapsed MCL and CLL. Acalabrutinib has recently been registered in a phase II clinical trial (NCT04548648) and in combination with durvalumab (a PD-1 inhibitor) in another phase I clinical trial for PCNSL and SCNSL (NCT04462328).

Immunomodulatory drugs

Lenalidomide. Lenalidomide is an oral second-generation immunomodulatory drug (IMiD) that has exhibited direct effects against B-NHL cells[62] and indirect antitumor effects mediated by various immune cells in the tumor microenvironment (TME), including T, natural killer (NK), and dendritic cells in B-NHL.[63]

Lenalidomide monotherapy as a salvage therapy has shown promising efficiency and good tolerance for the treatment of PCNSL in several clinical studies. A retrospective case series reported that two out of six patients with recurrent PCNSL who received lenalidomide monotherapy as salvage treatment achieved CR, including one patient with a remission duration of 24 months.[64] One MTX-resistant acquired immune deficiency syndrome (AIDS)-related PCNSL patient was also reported to maintain remission for up to 4 years with lenalidomide monotherapy (10 mg/day) plus anti-retroviral therapy.[65] A phase I study (NCT01542918) including 14 relapsed CNSL patients (six with PCNSL and eight with SCNSL) demonstrated the tolerability, antitumor efficiency, and CSF penetration of lenalidomide as the ORR was 64% (9/14), and the response

duration to this monotherapy in four patients prolonged for 18 months with all manageable toxicities. Considering the effectiveness of lenalidomide through the BBB and the absence of DLT, this study recommended an initial dose of lenalidomide monotherapy at 15 mg/day to treat relapsed CNSL.[34]

Meanwhile, lenalidomide monotherapy has been revealed to be an optional maintenance therapy following the response to genotoxic therapy for relapsed PCNSL, as an alternative to WBRT or HDC. The same group conducted a retrospective analysis of 10 patients with relapsed PCNSL with a median age of 61.5 years. These patients received salvage therapies of either HD-MTX or focal irradiation to demonstrate the outcomes of low-dose (5–10 mg/day) lenalidomide monotherapy as maintenance treatment, with an encouraging median OS of 45 months, and only one patient progressed to receive WBRT.[34] Among them, the response duration of patients in CR 2 through 5 after salvage treatment followed by lenalidomide maintenance was more than 6 times longer than that of patients who relapsed in CR1 after receiving standard induction therapies of MT-R (HD-MTX, temozolomide, and rituximab). As the poorer outcomes in elderly patients with PCNSL remain a significant problem,[10] low-dose lenalidomide was again used as maintenance in 13 patients, all of whom were ⩾70 years old and achieved PR or CR after MT-R with or without EA (etoposide, cytarabine) induction therapy.[66] Lenalidomide maintenance (median time of 18.9 months on lenalidomide) also revealed good tolerance in these elderly patients and median PFS was not reached, with only one death occurring due to complications from myocardial infarction in a median follow up of 31.6 months.

In addition to lenalidomide monotherapy, due to a synergistic effect between lenalidomide and rituximab in a mouse lymphoma model,[67] investigations of the combination treatment are underway. LOC and LYSA conducted a phase II study of lenalidomide combined with intravenous rituximab (R^2) for r/r PCNSL and PVRL. The ORR of 45 assessable patients (34 PCNSL and 11 PVRL) after a median number of 7 R^2 treatment cycles was 35.6%, including 13 CR/unconfirmed CR (uCR; 29%) and 3 PR (7%) cases. In total, 18 patients continued to the lenalidomide maintenance phase, and 5 completed 12 cycles. Without unexpected toxicities, the median PFS and OS reached 7.8 and 17.7 months at a median follow up of 19.2 months. The peripheral baseline CD4/CD8 ratio ⩾1.6 was significantly related to a longer PFS of 9.5 months.[68]

Pomalidomide. A novel third-generation IMiD, pomalidomide, has shown significant CNS penetration and effective antitumor effect against CNSL in murine models.[69] A phase I clinical trial (NCT 01722305) investigated pomalidomide (5 mg/day orally on Days 1 to 4 of each 28-day cycle) and weekly dexamethasone for r/r PCNSL and PVRL.[70] This combination showed a tolerable toxicity profile and significant curative activity, with an overall ORR of 48% (12 of 25), including 6 CR, 2 uCR, and 4 PR, as well as a median PFS of 9 months for responders.

Immune checkpoint inhibitors

Nivolumab. Programmed cell death protein 1 (PD-1) is an important immune checkpoint receptor expressed on activated T cells. The expression of PD-1 and its ligands (PD-L1 and PD-L2) have been extensively investigated for the treatment of lymphoma. High expression of transcript variants of PD-1 showed an ability to predict poor prognosis in PCNSL.[71] A recent study revealed that based on tumor mutational burden (TMB), high PD-L1 expression was observed in 37.5% of the 48 PCNSL subjects and more than 90% expressed intermediate-to-high TMB, indicating that immune checkpoint inhibitors may be beneficial to PCNSL.[72] Furthermore, larger clinical studies on PCNSL are warranted to investigate the checkpoint inhibitor therapies. A retrospective study of TME using high-plex immunohistochemistry in 36 CNSLs, 22 of which were PCNSLs, reported that PD-L1 expression level in lymphoma cells of all cases was 28%, which also supported immune checkpoint inhibitor treatment in PCNSL.[73]

Nivolumab is a humanized anti-PD-1 antibody approved by the FDA as second-line therapy for r/r classical Hodgkin lymphoma (cHL).[74] Recent evidence of frequent increases in the copy numbers of 9p24.1/PD-L1 and PD-L2, accompanied by increased expression of PD-L1 and PD-L2 in PCNSL,[75] has provided a rationale for immune checkpoint inhibitors treatment regimens. A case series of five patients (four with r/r PCNSL) demonstrated that nivolumab was effective and well tolerated, with all achieving objective radiographic responses (four CR and one PR) and symptomatic responses to nivolumab.[76] Three

out of all involved patients remained progression-free up to 13–17 months after the initiation of nivolumab. Another case report showed that a PCNSL patient with a very poor performance status (ECOG 4) achieved MRI almost CR and regained consciousness after six cycles of 3 mg/kg single-agent nivolumab administration.[77] Nivolumab combined with lenalidomide exhibited good tolerance and antitumor activity as salvage therapy in a patient with sanctuary site CNS DLBCL,[78] whose MRI showed near CR after 2 months of the combination treatment. A multicenter phase II trial of nivolumab for the treatment of r/r PCNSL and PTL has recently been completed (NCT02857426). Regrettably, with only one of 47 enrolled patients with PCNSL completing nivolumab treatment for 2 years, ORR was 6.4%, and PFS and OS were 1.41 and 8.64 months, respectively. Clinical trials in larger populations to investigate the effect of nivolumab are urgently required.

Nivolumab was also investigated as maintenance therapy after the patient achieved CR with MTX-based therapy followed by HDC/ASCT. In another case report of a patient with PCNSL and with two late relapses, a prolonged CR of more than 24 months without nivolumab-related toxicities indicated that nivolumab might be the drug of choice for the maintenance of remission.[79] In addition to being used as salvage and maintenance therapy, nivolumab monotherapy in the front-line treatment of PCNSL has shown promising effects in one PCNSL patient with poor tolerance for induction chemotherapy.[77]

Pembrolizumab. Pembrolizumab, another humanized anti-PD-1 antibody that has different epitope binding sites compared with nivolumab, has been reported to have a promising antitumor effect in r/r cHL and was granted FDA approval in June 2018 for r/r primary mediastinal B-cell lymphoma (PMBCL).[80] Pembrolizumab has also demonstrated antitumor responses for CNSL in a case series.[81] A patient with r/r PCNSL after treatment including lenalidomide, achieved MRI CR following two doses of 200 mg pembrolizumab with 1 mg dexamethasone. The CR was maintained for more than 22 months and to 24 months after discontinuing pembrolizumab. Registered phase II trials evaluating the role of pembrolizumab in the treatment of r/r PCNSL are now recruiting (NCT02779101, NCT03255018, NCT04421560).

PI3K/mTOR inhibitors

Temsirolimus. The PI3K/mTOR pathway is a critical pro-survival pathway that adjusts the synthesis of proteins to modulate cell growth and proliferation.[82] Temsirolimus, an inhibitor of mTOR, has shown encouraging efficiency in the treatment of r/r MCL and DLBCL. A phase II trial of 37 r/r PCNSL patients with r/r PCNSL demonstrated that temsirolimus monotherapy at a dose of 75 mg/week had a positive effect with 54% ORR, including 13.5% CR.[83] However, median PFS and OS were short at 2.1 and 3.7 months, respectively, with 13.5% treatment-associated mortality mainly from severe infections. In addition, 14 blood–CSF pairs were collected from nine patients, whereas only one patient showed a 2 ng/mL concentration of temsirolimus in the CSF. No temsirolimus or its main metabolite, sirolimus, was found in any other patient. The lack of a detectable concentration of temsirolimus and its main metabolite in CSF was inconsistent with its activity.

Other PI3K/mTOR inhibitors. In CD79B-mutant PCNSL, the PI3K/mTOR signaling axis is highly activated to promote tumor survival.[53] The PI3K inhibitor BKM120, buparlisib, could result in cell death in slice cultures from xenograft CD79B-mutant PCNSL.[84] This preclinical study demonstrated that combining ibrutinib with inhibitors of p110α/p110δ PI3K (BAY80-6946) or mTOR (INK128) could deepen and extend its responses in CD79B-mutant PCNSL. However, this needs to be investigated in advanced clinical trials. A phase Ib/II clinical trial (NCT03581942) on BAY80-6946 (copanlisib) for r/r PCNSL is ongoing. The recently completed phase II clinical trial (NCT02301364) of buparlisib in four patients with r/r CNSL showed three patients with PD and one PR. Another phase II trial (NCT02669511) investigating the pan PI3K/mTOR inhibitor, PQR309, in 21 relapsed PCNSL patients has been completed; however, no results had been posted at the time of writing this review.

Chimeric antigen receptor T-cell therapy

Chimeric antigen receptor (CAR) T-cells have been investigated by many groups for the treatment of lymphoma and have shown promising efficacy. Four CD19 targeted CAR T-cell therapies for r/r B-cell lymphoma have achieved FDA approval.[85] However, considering the risk of neurotoxicity from CAR T-cell therapy, initial trials

on patients with PCNSL have not met the current medical needs.[86] Abramson and colleagues studied a refractory DLBCL patient with CNS involvement who was treated with CD19 CAR T-cells. The patient's brain MRI showed CR after 1 month, and the CNS response lasted for 12 months of follow up.[87] A recent retrospective study on eight patients with SCNSL suggested that CD19 CAR T-cell therapies could be feasible for use in CNSL as two patients achieved CR and two PR at D + 28 after infiltration, without high rates of cytokine release syndrome (CRS).[88] Several studies have reported the clinical use of CD19 targeted CAR T-cells for PCNSL. A phase I clinical trial (NCT02153580) in a small cohort (five patients) demonstrated that CD19 CAR T-cells generated from autologous T-cells is safe and feasible for the treatment of PCNSL. Three patients achieved CR, and the other two remained stable disease (SD). Even all five patients occurred grade $\geqslant 1$ CRS and neurotoxicity, and these toxicities were tolerable and manageable.[89] The French LOC reported their first clinical experience of commercial CD19 CAR T-cell therapy for r/r PCNSL, which supported the clinical application of CD19 CAR T-cells. The best response in nine patients was five CR and one PR, with median PFS 122 days (210 days for responders).[90] A phase I/II clinical trial of tisagenlecleucel (the second approved anti-CD19 CAR T-cell therapy) in patients with r/r PCNSL demonstrated its safety and efficacy (NCT04134117).[91] Overall, 58.3% of 12 patients enrolled revealed response, including 6 CR and 1 PR. And, 58.3% of patients experienced Grade 1 CRS, and 41.6% experienced low grade immune cell associated neurotoxicity syndrome (ICANS). Seven patients remained alive, and three sustained remissions at a median time of 12.2 months after the infusion of tisagenlecleucel.

Bispecific CAR T-cell therapies, including CAR19/CAR70 T-cells and CAR19/CAR22 T-cells, have been reported to be used in PCNSL. A 67-year-old male patient with a second relapse of PCNSL who received CAR19/CAR70 T-cell therapy achieved CR in 1 month and disease-free survival for over 17 months of follow up without CRS. This case highlights the safety and antitumor efficacy of dual-target CAR19/CAR70 T-cells in PCNSL. CAR19 and CAR70 T-cells were detected after 10 months of CAR T-cell infusion.[92] In a study of five patients with chemorefractory B-cell CNSL who received CAR19/CAR22 T-cell therapy intravenously, one of the two patients who had achieved CR relapsed at the eighth month, and three patients who had achieved PR had PD by the third month, and all five patients had occurred CRS. As remission did not last long, this study suggested CAR T-cell therapy should be combined with other treatments.[93]

Mulazzan and colleagues developed a PCNSL mouse model to demonstrate the *in vivo* potential of CAR T-cells targeting human CD19 for the treatment of PCNSL. Although intravenous injection (IV) of anti-CD19 CAR T-cells could not effectively control tumor growth due to poor tumor infiltration of CAR T-cells, intracerebral injection (ICV) of CAR T-cells showed substantial antitumor effects with deep infiltration into the tumor, leading to long-term survival. Anti-CD19 CAR T-cells remained detectable up to 159 days after the complete eradication of tumors.[94] Another CNSL mouse model also showed that compared with IV, ICV of the anti-CD19 CAR T-cell therapy with superior proliferative potential resulted in significantly lower lymphoma burden and complete tumor regression. ICV-administered CAR T-cells can also migrate to peripheral blood and exhibit memory T-cell characteristics under undetectable tumor burden.[95] Studies in mouse models have inspired clinical trials of ICV of CD19 CAR T-cell therapies on PCNSL patients seem to be needed.

Different clinical trials investigating approved anti-CD19 CAR T-cell therapies for PCNSL are currently recruiting patients. The first approved anti-CD19 CAR T-cell therapy is axicabtagene ciloleucel (axi-cel) for the treatment of r/r large B-cell lymphoma, based on the ZUMA-1 trial.[96] The safety and efficacy of axi-cel for r/r PCNSL and SCNSL are being tested in a recruiting phase I trial (NCT04608487). The enrolled participants will be divided into two groups, one including patients with r/r PCNSL and SCNSL without lymphoma outside the CNS, and the other including those with r/r lymphoma outside CNS, with active or previously treated involvement of CNS by the lymphoma. Fludarabine and cyclophosphamide will be administered to minimize the interference from immune system cells, thus to help axi-cel work more effectively. This trial expects to enroll 18 participants and be followed for up to 15 years. The most recently approved anti-CD19 CAR T-cell therapy lisocabtagene maraleucel (liso-cel) is for patients with r/r large B-cell lymphoma.[85] One goal of the recruiting phase II

TRANSCENDWORLD trial (NCT03484702) is to evaluate the efficacy of liso-cel in PCNSL and SCNSL. The CAROUSEL trial (NCT04443829) is a phase I trial of autologous anti-CD19 CAR T-cell therapy following preconditioning lymphodepleting (LD) chemotherapy with cyclophosphamide, FDB, and pembrolizumab in r/r PCNSL, which will recruit soon. All patients will be treated on Theme 1 of the study with 250×10^6 CD19 CAR T-cells IV following LD chemotherapy as described above. Patients achieving SD or PD without severe toxicity on Day 28 after 250×10^6 CD19 CAR T-cells will receive Dose 2 of 25×10^6 CD19 CAR T-cells IV following LD chemotherapy.

However, data on CAR T-cell therapy for PCNSL remain scarce. CAR T-cell-related toxicities, including neurotoxicity and CRS, prevent its successful application in the treatment of patients with PCNSL. Even recent studies investigating the efficacy and safety supported the clinical application of CAR T-cell therapy for PCNSL, and further clinical trials in larger cohorts and longer follow-up time of CAR T-cell therapies for PCNSL are still recruiting patients. There are only single-targeted anti-CD19 CAR T-cell therapy-based trials and clinical trials in larger population cohorts of different dual-targeted CAR T-cell therapies, such as CAR19/CAR70 T-cells and CAR19/CAR22 T-cells are expected. Table 3 shows results of clinical trials on novel therapies for patients with PCNSL.

Other novel therapies

Selinexor, a selective inhibitor of nuclear export (SINE), binds to the nuclear export protein XPO1 (CRM1) and inhibits its activity, which is important for the maintenance of cellular homeostasis in mediating the transport of many tumor suppressor proteins from the nucleus to the cytoplasm.[97] Selinexor was approved as a third-line therapy for DLBCL based on data from the phase IIb SADAL study.[98] A case report revealed that a patient with SCNSL achieved CR based on brain MRI after oral administration of selinexor for 5 months. Regrettably, Grade 3 fatigue and Grade 2 anorexia during selinexor treatment led to dose reduction and its stoppage eventually in the patient.[99] Selinexor was capable of inhibiting the growth of tumors to extend survival in PCNSL mouse models, and mouse survival could be further prolonged after combination with ibrutinib. This finding might provide a preclinical rationale for this two-novel-drug strategy for PCNSL.[100] Chidamide, an oral subtype-selective histone deacetylase inhibitor (HDACI), has been approved by China FDA for the treatment of r/r peripheral T-cell lymphoma (PTCL).[101] A recent study reported that chidamide could significantly upregulate the expression of CD20 on the surface of DLBCL cell lines; thus, it could synergize with rituximab to inhibit tumor growth of DLBCL.[102] Chidamide combined with rituximab and HD-MTX is being studied in an ongoing phase II trial (NCT04516655), for previously untreated patients with PCNSL. Obinutuzumab, a new generation anti-CD20 antibody, has greater antibody-dependent cellular cytotoxicity (ADCC) and lower complement-dependent cytotoxicity than rituximab.[103] The GALLIUM trial reported that the replacement of rituximab with obinutuzumab in first-line treatment for patients with previously untreated FL resulted in significantly longer PFS.[104] The effect of obinutuzumab maintenance on the duration of response in patients with CD20+ PCNSL who achieved CR or PR after first-line treatment with HD-MTX-based chemotherapy is being investigated in a phase II trial (NCT02498951). Venetoclax, an orally bioavailable inhibitor of BCL-2, has revealed to be a safe and effective novel agent for the treatment of NHL.[105] Venetoclax has shown good penetration in CSF in CLL with CNS involvement.[106] Overexpression of BCL-2 also occurs in PCNSL patients and increases risk stratification.[107] A phase Ib clinical trial (NCT04073147) of venetoclax in combination with obinutuzumab is being conducted for r/r PCNSL.

In addition to novel drugs, traditional chemotherapeutic drugs, which were not commonly used in the treatment of PCNSL, such as temozolomide, bendamustine, and pemetrexed, were evaluated in retrospective case series and prospective clinical trials for PCNSL. Adding TNF-α coupled with NGR (NGR-hTNF) to the classical R-CHOP regimen could increase BBB permeabilization to increase CNS bioavailability of anticancer drugs and was reported to be a safe and highly effective combination therapy in PCNSL.[5] Table 4 summarizes the ongoing clinical trials of novel therapies for patients with PCNSL.

Conclusion and future directions

Although the treatment of PCNSL has achieved significant advances over the past decades, it

Table 3. Results of clinical trials on novel therapies for PCNSL patients.

Medication	Study type	Trial number	Patients' number	Setting	Primary objective	Response	PFS	OS	Toxicity	Reference
Ibrutinib monotherapy	phase II	NCT02542514	52 recruited (44 evaluable)	R/R DLBCL-PCNSL or PVRL	DC rate after 2 months of treatment	70% DC rate; 59% ORR	4.8 months	19.2 months	Two pulmonary aspergillosis (one fatal)	Soussain *et al.*[52]
	phase I/II	NCT02315326	20 (13 PCNSL)	R/R PCNSL and SCNSL	Safety and ORR	10 with PCNSL respond (5 CR), 5 with SCNSL respond (4 CR)	4.6 months in PCNSL, 7.43 months in SCNSL	15 months in PCNSL, not reached in SCNSL	No DLT occurred	Grommes *et al.*[53]
	phase Ib	NCT02203526	18	PCNSL	A tolerated dose of ibrutinib	94% showed tumor reductions (83% PR)			Two Grade 5 pulmonary/CNS aspergillosis	Lionakis *et al.*[55]
Ibrutinib and DA-TEDDi-R			16 (14 evaluable)			12 CR or CRu, 1 PR, 1 PD	15.3 months	Not reached at 1 year	53% cycles of Grade 4 neutropenia, five pulmonary aspergillus, two CNS aspergillus	
Ibrutinib with HD-MTX and rituximab	phase Ib	NCT02315326	15 (9 PCNSL)	R/R PCNSL and SCNSL	MTD of ibrutinib	80% ORR (89% in PCNSL and 67% in SCNSL)	9.2 months (not reached in PCNSL)	71.1% 1-year OS	Three non-DLT Grade 4 AEs	Grommes *et al.*[19]
Tirabrutinib	phase I/II	JapicCTI-173646	44	R/R PCNSL	ORR	63.6% ORR	2.9 months	Not reached with median follow up of 9.1 months	13 serious AEs, one Grade 5 AE	Narita *et al.*[59]
Lenalidomide monotherapy	phase I	NCT01542918	14 (6 PCNSL)	Relapsed CNSL	Safety, MTD, CSF penetration of lenalidomide	64% ORR	Six patients' response durations ⩾9 months	15.5 months	Three ⩾Grade 3 infections	Rubenstein *et al.*[34]
Lenalidomide + intravenous rituximab	phase II	NCT01956695	45 assessable (34 PCNSL)	R/R PCNSL and PVRL	ORR	35.6% ORR (CR/uCR 29%)	7.8 months	17.7 months	44% Grade 3 to 4 neutropenia	Ghesquieres *et al.*[68]
Pomalidomide and dexamethasone	phase I	NCT01722305	25 eligible (23 PCNSL)	R/R PCNSL and PVRL	MTD of pomalidomide with weekly dexamethasone	48% ORR (six CR, two uCR, and four PR)	9 months for responders, and 5.3 for PCNSL		62.5% Grade 3 or 4 toxicities	Tun *et al.*[70]
Nivolumab	phase II	NCT02857426	47 PCNSL (1 finished treatment)	R/R PCNSL or PTL	ORR	6.4% ORR	1.41 months for PCNSL	8.64 months for PCNSL	72.34% serious AEs for PCNSL	
Temsirolimus	phase II	NCT00942747	37	R/R PCNSL	ORR	54% ORR (13.5% CR)	2.1 months	3.7 months	13.5% treatment-associated mortality	Korfel *et al.*[83]
Buparlisib (BKM120)	phase II	NCT02301364	4	R/R PCNSL and SCNSL	PFS	One PR, three PD	39 days	196 days	All with serious AEs	

(Continued)

Table 3. (Continued)

Medication	Study type	Trial number	Patients' number	Setting	Primary objective	Response	PFS	OS	Toxicity	Reference
CD19 CAR T-cells	phase I	NCT02153580	5	R/R PCNSL	Safety	Three CR, two SD	–	–	All with grade ⩾ 1 CRS and neurotoxicity	Siddiqi *et al.*[89]
Tisagenlecleucel	phase I/II	NCT04134117	12	R/R PCNSL	Tolerability and toxicity	Six CR, one PR	–	–	58.3% Grade 1 CRS, 41.6% low grade ICANS (One Grade 3)	Frigault *et al.*[91]

AEs, adverse events; CNS, central nervous system; CR, complete response; CRS, cytokine release syndrome; CSF, cerebrospinal fluid; DA-TEDDi-R, dose-adjusted temozolomide, etoposide, liposomal doxorubicin, dexamethasone, and rituximab; DC, disease control; DLT, dose-limiting toxicity; ICANS, immune cell associated neurotoxicity syndrome; MTD, the maximal tolerated dose; ORR, overall response rate; OS, overall survival; PCNSL, primary central nervous system lymphoma; PD, progressive disease; PFS, progression-free survival; PTL, primary testicular lymphoma; PR, partial response; PVRL, primary vitreoretinal lymphoma; R/R, relapsed and refractory; SCNSL: secondary central nervous system lymphoma; SD, stable disease.

remains a highly recurrent disease with poor prognosis. A schematic summary of the development of therapies for PCNSL is illustrated in Figure 1. Novel diagnostic markers, such as ctDNAs, miRNAs, metabolomic markers, and others, facilitate early diagnostic accuracy, and they are consistent with prognosis. Nevertheless, the actual results of clinical use of novel biomarkers are limited to date and are required for broad clinical application. For newly diagnosed PCNSL, especially in young patients, induction therapy of HD-MTX-based chemotherapy and consolidation treatment of ASCT or WBRT are recent mainstream choices. However, there is no consensus on the optimal regimens for r/r and elderly PCNSL patients. The prognostic outcomes of r/r PCNSL patients remain nonideal. Salvage therapy followed by consolidation of HDC/ASCT could relatively prolong survival. While, as clinical studies are limited, the optimal treatment for r/r PCNSL has not been fully defined. With the development of immunotherapy, novel agents, including BTK inhibitors, immunomodulatory drugs, immune checkpoint inhibitors, PI3K/mTOR inhibitors, and CAR T-cell therapy, have shown encouraging efficacy based on response rates. Regrettably, the durations of response for r/r patients (ranging from 4 to 24 months) need further improvement, and might depend on a more appropriate schedule for the combination of different novel and classical therapies. Besides, most of the studies that have clinical results are relatively small; therefore, further clinical studies in larger populations need to be conducted to achieve a reliable conclusion. Other novel therapies, such as antibody-drug conjugates, which have emerged as promising treatment options for both lymphomas[108] and brain glioblastomas, might be valuable in the treatment of PCNSL and require further investigations in clinical trials. Targeting the TME components which play important roles in various processes of B-cell lymphoma could also provide novel insights into the precise treatment of PCNSL.[109] Considering that it is unlikely for a novel agent to cure such a PD as PCNSL, different polychemotherapy combinations of several novel and traditional therapies should be investigated further.

Author contributions

Yujia Zhai: Data curation; Investigation; Methodology; Writing – original draft; Writing – review & editing.

Table 4. Ongoing clinical trials on novel therapies for patients with PCNSL.

Study type	Date opened	Clinical trial registration no.	Patient population	Trial agents
phase I	August 2014	NCT02203526	PCNSL	Ibrutinib and TEDDI-R
phase I/II	December 2014	NCT02315326	R/R PCNSL and R/R SCNSL	Ibrutinib; HD-MTX; Rituximab + HD-MTX
phase II	June 2016	NCT02779101	R/R PCNSL	Pembrolizumab
phase II	July 2016	NCT02498951	Patients with CD20 + PCNSL who achieved CR or PR to first-line treatment with HD-MTX-based chemotherapy	Obinutuzumab maintenance
phase II	October 2016	NCT02623010	Elderly patients with PCNSL	Ibrutinib as maintenance treatment
phase II	February 2018	NCT03255018	R/R GZL, PCNSL and other extranodal DLBCL	Pembrolizumab
phase II	April 2018	NCT03484702	DLBCL, NOS; HGBL; FL3B; PCNSL and SCNSL	Lisocabtagene maraleucel
phase Ib/II	August 2018	NCT03581942	R/R PCNSL	Copanlisib in combination with ibrutinib
phase Ib	January 2019	NCT03703167	R/R PCNSL and R/R SCNSL	Ibrutinib in combination with R^2
phase I	January 2019	NCT03798314	R/R PCNSL and R/R PVRL	Nivolumab and pomalidomide combination
phase II	February 2019	NCT03770416	R/R CNSL	Nivolumab and ibrutinib
phase II	May 2019	NCT04052659	R/R PMBCL, PT/NKCL and PCNSL	Sintilimab (IBI308)
phase II	June 2019	NCT03495960	Newly diagnosed PCNSL > 70	Lenalidomide maintenance following MTX/Rituximab-based induction
phase II	June 2019	NCT04438044	R/R PCNSL and R/R SCNSL	ICP-022
phase I	July 2019	NCT04022980	Older (⩾65 years) patients with PCNSL	Nivolumab consolidation following HD-MTX
phase I/II	August 2019	NCT04120350	Newly diagnosed PCNSL	R^2-MTX regimen combined with lenalidomide maintenance
phase II	January 2020	NCT04129710	R/R PCNSL	Ibrutinib in combination with MRE *versus* lenalidomide in combination with MRE
phase II	February 2020	NCT04070040	Recurrent PCNSL	Camrelizumab
phase Ib	May 2020	NCT04073147	R/R PCNSL	Venetoclax and obinutuzumab
phase II	May 2020	NCT04401774	PCNSL patients completed HD-MTX but have persistent cfDNA in CSF after treatment despite imaging response.	Nivolumab maintenance

(Continued)

Table 4. (Continued)

Study type	Date opened	Clinical trial registration no.	Patient population	Trial agents
phase Ib/II	August 2020	NCT04421560	Recurrent PCNSL	Pembrolizumab in combination with ibrutinib and rituximab
phase II	September 2020	NCT04516655	Untreated PCNSL	Chidamide in combination with rituximab and HD-MTX
phase II	September 2020	NCT04548648	R/R PCNSL and R/R SCNSL	Acalabrutinib
phase Ib/II	October 2020	NCT04446962	Newly diagnosed PCNSL patients aged 18–60 years	Lenalidomide and ibrutinib in combination with R-MPV
phase II	October 2020	NCT04481815	Newly diagnosed PCNSL	R^2-MTX *versus* R-MTX
phase II	November 2020	NCT04457869	R/R PCNSL and R/R SCNSL	F520
phase II	November 2020	NCT04627753	Transplantation ineligible patients with PCNSL	Lenalidomide and rituximab maintenance
phase II	December 2020	NCT04514393	Newly diagnosed PCNSL	MTX, ibrutinib, and temozolomide
phase I	December 2020	NCT04608487	R/R PCNSL and R/R SCNSL	Axicabtagene ciloleucel
phase I	January 2021	NCT04609046	PCNSL	R^2-MTX and nivolumab induction followed by lenalidomide and nivolumab maintenance
phase I	February 2021	NCT04462328	PCNSL and SCNSL	Acalabrutinib and durvalumab
phase I	February 2021	NCT04443829	R/R PCNSL	CD19 CAR T-cells
phase Ib	February 2021	NCT04688151	PCNSL	Combining rituximab, acalabrutinib, and durvalumab
phase II	March 2021	NCT04899427	R/R PCNSL	Orelabrutinib combined with PD-1 inhibitor
phase II	May 2021	NCT04938297	PCNSL and SCNSL	Rituximab, zanubrutinib in combination with lenalidomide, followed by zanubrutinib or lenalidomide maintenance
phase II	July 2021	NCT04947319	R/R PCNSL	Tirabrutinib

CR, complete response; DLBCL, diffuse large B-cell lymphoma; FL3B, follicular lymphoma Grade 3B; GZL, Gray-Zone lymphoma; HD-MTX, high-dose-methotrexate; HGBL, high-grade B-cell lymphoma; MRE, methotrexate, rituximab, etoposide; PCNSL, primary central nervous system lymphoma; PD, progressive disease; PMBCL, primary mediastinal large B-cell lymphoma; PR, partial response; PTL, primary testicular lymphoma; PT/NKCL, peripheral T/NK-cell lymphoma; PVRL, primary vitreoretinal lymphoma; R^2, lenalidomide in combination with rituximab; R/R, relapsed and refractory; R-MPV, rituximab-methotrexate procarbazine vincristine; R-MTX, rituximab-methotrexate; SCNSL, secondary central nervous system lymphoma; TEDDI-R, temozolomide, etoposide, doxil, dexamethasone, ibrutinib, rituximab.

Xiangxiang Zhou: Conceptualization; Formal analysis; Funding acquisition; Project administration; Resources; Supervision; Validation; Writing – review & editing.

Xin Wang: Conceptualization; Funding acquisition; Project administration; Resources; Supervision; Validation; Writing – review & editing.

ORCID iD
Xiangxiang Zhou https://orcid.org/0000-0003-0725-716X

Funding
The authors disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This study was

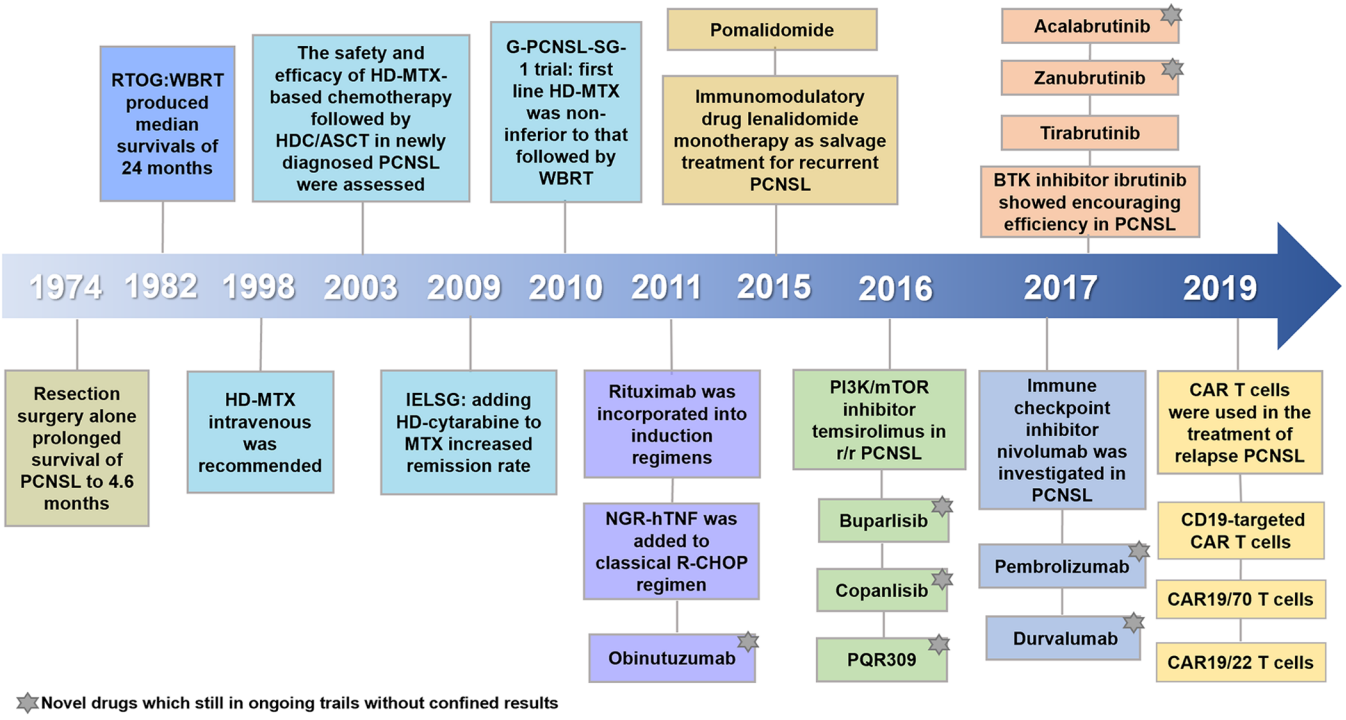

Figure 1. A schematic summary of the development of therapies for PCNSL.
PCNSL, primary central nervous system lymphoma. RTOG, radiation therapy oncology group; IELSG, international extranodal lymphoma study group.

supported by National Natural Science Foundation (No. 82170189, No. 82070203, No. 81800194, No. 81770210, No. 81473486, and No. 81270598); Key Research and Development Program of Shandong Province (No. 2018CXGC1213); Development Project of Youth Innovation Teams in Colleges and Universities of Shandong Province (No. 2020KJL006); China Postdoctoral Science Foundation (No. 2021T140422, No. 2020M672103); Technology Development Projects of Shandong Province (No. 2017GSF18189); Translational Research Grant of NCRCH (No. 2021WWB02, No. 2020ZKMB01); Shandong Provincial Natural Science Foundation (ZR2021YQ51); Technology Development Project of Jinan City (No. 202134034); Taishan Scholars Program of Shandong Province; Shandong Provincial Engineering Research Center of Lymphoma; Academic Promotion Programme of Shandong First Medical University (No. 2019QL018, No. 2020RC006).

Conflict of interest statement

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

References

1. Makranz C, Arkadir D, Nachmias B, *et al.* Neurological misdiagnoses of lymphoma. *Neurol Sci* 2021; 42: 1933–1940.
2. Houillier C, Soussain C, Ghesquières H, *et al.* Management and outcome of primary CNS lymphoma in the modern era: an LOC network study. *Neurology* 2020; 94: e1027–e1039.
3. Velasco R, Mercadal S, Vidal N, *et al.* Diagnostic delay and outcome in immunocompetent patients with primary central nervous system lymphoma in Spain: a multicentric study. *J Neurooncol* 2020; 148: 545–554.
4. Haldorsen IS, Espeland A, Larsen JL, *et al.* Diagnostic delay in primary central nervous system lymphoma. *Acta Oncol* 2005; 44: 728–734.
5. Ferreri AJM, Calimeri T, Conte GM, *et al.* R-CHOP preceded by blood-brain barrier permeabilization with engineered tumor necrosis factor-α in primary CNS lymphoma. *Blood* 2019; 134: 252–262.
6. Zhang Y and Zhou DB. Primary central nervous system lymphoma: status and advances in diagnosis, molecular pathogenesis, and treatment. *Chinese Med J* 2020; 133: 1462–1469.

7. Ferreri AJM. Therapy of primary CNS lymphoma: role of intensity, radiation, and novel agents. *Hematology* 2017; 2017: 565–577.

8. Attarbaschi A, Abla O, Ronceray L, *et al.* Primary central nervous system lymphoma: initial features, outcome, and late effects in 75 children and adolescents. *Blood Adv* 2019; 3: 4291–4297.

9. Langner-Lemercier S, Houillier C, Soussain C, *et al.* Primary CNS lymphoma at first relapse/progression: characteristics, management, and outcome of 256 patients from the French LOC network. *Neuro Oncol* 2016; 18: 1297–1303.

10. Mendez JS, Ostrom QT, Gittleman H, *et al.* The elderly left behind-changes in survival trends of primary central nervous system lymphoma over the past 4 decades. *Neuro Oncol* 2018; 20: 687–694.

11. Kurtz DM, Scherer F, Jin MC, *et al.* Circulating tumor DNA measurements as early outcome predictors in diffuse large B-cell lymphoma. *J Clin Oncol* 2018; 36: 2845–2853.

12. Hickmann AK, Frick M, Hadaschik D, *et al.* Molecular tumor analysis and liquid biopsy: a feasibility investigation analyzing circulating tumor DNA in patients with central nervous system lymphomas. *BMC Cancer* 2019; 19: 192.

13. Bobillo S, Crespo M, Escudero L, *et al.* Cell free circulating tumor DNA in cerebrospinal fluid detects and monitors central nervous system involvement of B-cell lymphomas. *Haematologica* 2021; 106: 513–521.

14. Chen F, Pang D, Guo H, *et al.* Clinical outcomes of newly diagnosed primary CNS lymphoma treated with ibrutinib-based combination therapy: a real-world experience of off-label ibrutinib use. *Cancer Med* 2020; 9: 8676–8684.

15. Hiemcke-Jiwa LS, Leguit RJ, Snijders TJ, *et al.* Molecular analysis in liquid biopsies for diagnostics of primary central nervous system lymphoma: review of literature and future opportunities. *Crit Rev Oncol Hematol* 2018; 127: 56–65.

16. Zorofchian S, Lu G, Zhu JJ, *et al.* Detection of the MYD88 p.L265P mutation in the CSF of a patient with secondary central nervous system lymphoma. *Front Oncol* 2018; 8: 382.

17. Rimelen V, Ahle G, Pencreach E, *et al.* Tumor cell-free DNA detection in CSF for primary CNS lymphoma diagnosis. *Acta Neuropathol Commun* 2019; 7: 43.

18. Ferreri AJM, Calimeri T, Lopedote P, *et al.* MYD88 L265P mutation and interleukin-10 detection in cerebrospinal fluid are highly specific discriminating markers in patients with primary central nervous system lymphoma: results from a prospective study. *Br J Haematol* 2021; 193: 497–505.

19. Grommes C, Tang SS, Wolfe J, *et al.* Phase 1b trial of an ibrutinib-based combination therapy in recurrent/refractory CNS lymphoma. *Blood* 2019; 133: 436–445.

20. Montesinos-Rongen M, Brunn A, Tuchscherer A, *et al.* Analysis of driver mutational hot spots in blood-derived cell-free DNA of patients with primary central nervous system lymphoma obtained before intracerebral biopsy. *J Mol Diagn* 2020; 22: 1300–1307.

21. Liu J, Han Y, Hu S, *et al.* Circulating exosomal MiR-107 restrains tumorigenesis in diffuse large B-cell lymphoma by targeting 14-3-3η. *Front Cell Dev Biol* 2021; 9: 667800.

22. Wei D, Wan Q, Li L, *et al.* MicroRNAs as potential biomarkers for diagnosing cancers of central nervous system: a meta-analysis. *Mol Neurobiol* 2015; 51: 1452–1461.

23. Robertus JL, Harms G, Blokzijl T, *et al.* Specific expression of miR-17-5p and miR-127 in testicular and central nervous system diffuse large B-cell lymphoma. *Mod Pathol* 2009; 22: 547–555.

24. Baraniskin A, Kuhnhenn J, Schlegel U, *et al.* Identification of microRNAs in the cerebrospinal fluid as marker for primary diffuse large B-cell lymphoma of the central nervous system. *Blood* 2011; 117: 3140–3146.

25. Baraniskin A, Chomiak M, Ahle G, *et al.* MicroRNA-30c as a novel diagnostic biomarker for primary and secondary B-cell lymphoma of the CNS. *J Neurooncol* 2018; 137: 463–468.

26. Roth P, Keller A, Hoheisel JD, *et al.* Differentially regulated miRNAs as prognostic biomarkers in the blood of primary CNS lymphoma patients. *Eur J Cancer* 2015; 51: 382–390.

27. Mao X, Sun Y and Tang J. Serum miR-21 is a diagnostic and prognostic marker of primary central nervous system lymphoma. *Neurol Sci* 2014; 35: 233–238.

28. Fischer L, Hummel M, Korfel A, *et al.* Differential micro-RNA expression in primary CNS and nodal diffuse large B-cell lymphomas. *Neuro Oncol* 2011; 13: 1090–1098.

29. Zajdel M, Rymkiewicz G, Sromek M, *et al.* Tumor and cerebrospinal fluid microRNAs in primary central nervous system lymphomas. *Cancers* 2019; 11: 1647.

30. Shalaby T and Grotzer MA. Tumor-associated CSF microRNAs for the prediction and

evaluation of CNS malignancies. *Int J Mol Sci* 2015; 16: 29103–29119.

31. Wishart DS. Metabolomics for investigating physiological and pathophysiological processes. *Physiol Rev* 2019; 99: 1819–1875.
32. Takashima Y, Hayano A and Yamanaka R. Metabolome analysis reveals excessive glycolysis via PI3K/AKT/mTOR and RAS/MAPK signaling in methotrexate-resistant primary CNS lymphoma-derived cells. *Clin Cancer Res* 2020; 26: 2754–2766.
33. Wang FX, Chen K, Huang FQ, *et al.* Cerebrospinal fluid-based metabolomics to characterize different types of brain tumors. *J Neurol* 2020; 267: 984–993.
34. Rubenstein JL, Geng H, Fraser EJ, *et al.* Phase 1 investigation of lenalidomide/rituximab plus outcomes of lenalidomide maintenance in relapsed CNS lymphoma. *Blood Adv* 2018; 2: 1595–1607.
35. Geng H, Tsang M, Subbaraj L, *et al.* Tumor metabolism and neurocognition in CNS lymphoma. *Neuro-Oncology* 2021; 23: 1668–1679.
36. Mayerhoefer ME, Umutlu L and Schöder H. Functional imaging using radiomic features in assessment of lymphoma. *Methods* 2021; 188: 105–111.
37. Barré L, Hovhannisyan N, Bodet-Milin C, *et al.* [(18)F]-fludarabine for hematological malignancies. *Front Med* 2019; 6: 77.
38. Hovhannisyan N, Fillesoye F, Guillouet S, *et al.* [(18)F]fludarabine-PET as a promising tool for differentiating CNS lymphoma and glioblastoma: comparative analysis with [(18)F]FDG in human xenograft models. *Theranostics* 2018; 8: 4563–4573.
39. Zhang H, Maeda M, Shindo M, *et al.* Imaging CXCR4 expression with iodinated and brominated cyclam derivatives. *Mol Imaging Biol* 2020; 22: 1184–1196.
40. Herhaus P, Lipkova J, Lammer F, *et al.* CXCR4-targeted PET imaging of central nervous system B-cell lymphoma. *J Nucl Med* 2020; 61: 1765–1771.
41. Novak AJ, Grote DM, Stenson M, *et al.* Expression of BLyS and its receptors in B-cell non-Hodgkin lymphoma: correlation with disease activity and patient outcome. *Blood* 2004; 104: 2247–2253.
42. Birnbaum T, Langer S, Roeber S, *et al.* Expression of B-cell activating factor, a proliferating inducing ligand and its receptors in primary central nervous system lymphoma. *Neurol Int* 2013; 5: e4.
43. Mulazzani M, Huber M, Borchard S, *et al.* APRIL and BAFF: novel biomarkers for central nervous system lymphoma. *J Hematol Oncol* 2019; 12: 102.
44. Thaler FS, Laurent SA, Huber M, *et al.* Soluble TACI and soluble BCMA as biomarkers in primary central nervous system lymphoma. *Neuro-Oncology* 2017; 19: 1618–1627.
45. Mizutani H, Nakane S, Ikeda T, *et al.* CSF TACI and BAFF levels in patients with primary CNS lymphoma as novel diagnostic biomarkers. *Ann Clin Transl Neurol* 2018; 5: 1611–1616.
46. Baraniskin A, Zaslavska E, Nöpel-Dünnebacke S, *et al.* Circulating U2 small nuclear RNA fragments as a novel diagnostic biomarker for primary central nervous system lymphoma. *Neuro-Oncology* 2015; 18: 361–367.
47. Hendriks RW, Yuvaraj S and Kil LP. Targeting Bruton's tyrosine kinase in B cell malignancies. *Nat Rev Cancer* 2014; 14: 219–232.
48. Byrd JC, Brown JR, O'Brien S, *et al.* Ibrutinib versus ofatumumab in previously treated chronic lymphoid leukemia. *N Engl J Med* 2014; 371: 213–223.
49. Advani RH, Buggy JJ, Sharman JP, *et al.* Bruton tyrosine kinase inhibitor ibrutinib (PCI-32765) has significant activity in patients with relapsed/refractory B-cell malignancies. *J Clin Oncol* 2013; 31: 88–94.
50. Nakamura T, Tateishi K, Niwa T, *et al.* Recurrent mutations of CD79B and MYD88 are the hallmark of primary central nervous system lymphomas. *Neuropathol Appl Neurobiol* 2016; 42: 279–290.
51. Chamoun K, Choquet S, Boyle E, *et al.* Ibrutinib monotherapy in relapsed/refractory CNS lymphoma: a retrospective case series. *Neurology* 2017; 88: 101–102.
52. Soussain C, Choquet S, Blonski M, *et al.* Ibrutinib monotherapy for relapse or refractory primary CNS lymphoma and primary vitreoretinal lymphoma: final analysis of the phase II 'proof-of-concept' iLOC study by the Lymphoma study association (LYSA) and the French oculo-cerebral lymphoma (LOC) network. *Eur J Cancer* 2019; 117: 121–130.
53. Grommes C, Pastore A, Palaskas N, *et al.* Ibrutinib unmasks critical role of bruton tyrosine kinase in primary CNS lymphoma. *Cancer Discov* 2017; 7: 1018–1029.

54. Lewis KL, Chin CK, Manos K, *et al*. Ibrutinib for central nervous system lymphoma: the Australasian Lymphoma Alliance/MD Anderson Cancer Center experience. *Br J Haematol* 2021; 192: 1049–1053.

55. Lionakis MS, Dunleavy K, Roschewski M, *et al*. Inhibition of B cell receptor signaling by ibrutinib in primary CNS lymphoma. *Cancer Cell* 2017; 31: 833–843.e835.

56. Liclican A, Serafini L, Xing W, *et al*. Biochemical characterization of tirabrutinib and other irreversible inhibitors of Bruton's tyrosine kinase reveals differences in on – and off – target inhibition. *Biochim Biophys Acta Gen Subj* 2020; 1864: 129531.

57. Walter HS, Rule SA, Dyer MJ, *et al*. A phase 1 clinical trial of the selective BTK inhibitor ONO/GS-4059 in relapsed and refractory mature B-cell malignancies. *Blood* 2016; 127: 411–419.

58. Dhillon S. Tirabrutinib: first Approval. *Drugs* 2020; 80: 835–840.

59. Narita Y, Nagane M, Mishima K, *et al*. Phase I/II study of tirabrutinib, a second-generation Bruton's tyrosine kinase inhibitor, in relapsed/refractory primary central nervous system lymphoma. *Neuro-Oncology* 2021; 23: 122–133.

60. Tam CS, Opat S, Simpson D, *et al*. Zanubrutinib for the treatment of relapsed or refractory mantle cell lymphoma. *Blood Adv* 2021; 5: 2577–2585.

61. Cheng Q, Wang J, Lv C, *et al*. Successful management of a patient with refractory primary central nervous system lymphoma by zanubrutinib. *Onco Targets Ther* 2021; 14: 3367–3372.

62. Verhelle D, Corral LG, Wong K, *et al*. Lenalidomide and CC-4047 inhibit the proliferation of malignant B cells while expanding normal CD34+ progenitor cells. *Cancer Res* 2007; 67: 746–755.

63. Gribben JG, Fowler N and Morschhauser F. Mechanisms of action of lenalidomide in B-cell non-hodgkin Lymphoma. *J Clin Oncol* 2015; 33: 2803–2811.

64. Houillier C, Choquet S, Touitou V, *et al*. Lenalidomide monotherapy as salvage treatment for recurrent primary CNS lymphoma. *Neurology* 2015; 84: 325–326.

65. Gupta NK, Wang CC, Mannis GN, *et al*. Regression of methotrexate-resistant AIDS-related primary central nervous system lymphoma with lenalidomide plus combination anti-retroviral therapy. *Leuk Lymphoma* 2017; 58: 2748–2751.

66. Vu K, Mannis G, Hwang J, *et al*. Low-dose lenalidomide maintenance after induction therapy in older patients with primary central nervous system lymphoma. *Br J Haematol* 2019; 186: 180–183.

67. Hernandez-Ilizaliturri FJ, Reddy N, Holkova B, *et al*. Immunomodulatory drug CC-5013 or CC-4047 and rituximab enhance antitumor activity in a severe combined immunodeficient mouse lymphoma model. *Clin Cancer Res* 2005; 15: 5984–5992.

68. Ghesquieres H, Chevrier M, Laadhari M, *et al*. Lenalidomide in combination with intravenous rituximab (REVRI) in relapsed/refractory primary CNS lymphoma or primary intraocular lymphoma: a multicenter prospective 'proof of concept' phase II study of the French Oculo-Cerebral lymphoma (LOC) Network and the Lymphoma Study Association (LYSA). *Ann Oncol* 2019; 30: 621–628.

69. Li Z, Qiu Y, Personett D, *et al*. Pomalidomide shows significant therapeutic activity against CNS lymphoma with a major impact on the tumor microenvironment in murine models. *PLoS ONE* 2013; 8: e71754.

70. Tun HW, Johnston PB, DeAngelis LM, *et al*. Phase 1 study of pomalidomide and dexamethasone for relapsed/refractory primary CNS or vitreoretinal lymphoma. *Blood* 2018; 132: 2240–2248.

71. Takashima Y, Kawaguchi A, Sato R, *et al*. Differential expression of individual transcript variants of PD-1 and PD-L2 genes on Th-1/Th-2 status is guaranteed for prognosis prediction in PCNSL. *Sci Rep* 2019; 9: 10004.

72. Ou A, Sumrall A, Phuphanich S, *et al*. Primary CNS lymphoma commonly expresses immune response biomarkers. *Neurooncol Adv* 2020; 2: vdaa018.

73. Marcelis L, Antoranz A, Delsupehe A-M, *et al*. In-depth characterization of the tumor microenvironment in central nervous system lymphoma reveals implications for immune-checkpoint therapy. *Cancer Immunol Immun* 2020; 69: 1751–1766.

74. Davis KL, Fox E, Merchant MS, *et al*. Nivolumab in children and young adults with relapsed or refractory solid tumours or lymphoma (ADVL1412): a multicentre, open-label, single-arm, phase 1-2 trial. *Lancet Oncol* 2020; 21: 541–550.

75. Chapuy B, Roemer MG, Stewart C, *et al*. Targetable genetic features of primary testicular and primary central nervous system lymphomas. *Blood* 2016; 127: 869–881.

76. Nayak L, Iwamoto FM, LaCasce A, *et al*. PD-1 blockade with nivolumab in relapsed/refractory primary central nervous system and testicular lymphoma. *Blood* 2017; 129: 3071–3073.

77. El-Tawab R, Hamada A, Elhagracy R, *et al*. Promising effect of PDL1 inhibitors in the front-line management of primary aggressive central nervous system lymphoma: a case report. *Hematol Oncol Stem Cell Ther*. Epub ahead of print 30 June 2020. DOI: 10.1016/j.hemonc.2020.06.003.

78. Madabhavi IV, Revannasiddaiah S, Sarkar MS, *et al*. Sanctuary site central nervous system relapse-refractory DLBCL responding to nivolumab and lenalidomide. *Oral Oncol* 2019; 93: 122–124.

79. Terziev D, Hutter B, Klink B, *et al*. Nivolumab maintenance after salvage autologous stem cell transplantation results in long-term remission in multiple relapsed primary CNS lymphoma. *Eur J Haematol* 2018; 101: 115–118.

80. Sheikh S and Kuruvilla J. Pembrolizumab for the treatment of diffuse large B-cell lymphoma. *Expert Opin Biol Ther* 2019; 19: 1119–1126.

81. Graber JJ, Plato B, Mawad R, *et al*. Pembrolizumab immunotherapy for relapsed CNS lymphoma. *Leuk Lymphoma* 2020; 61: 1766–1768.

82. Fingar DC and Blenis J. Target of rapamycin (TOR): an integrator of nutrient and growth factor signals and coordinator of cell growth and cell cycle progression. *Oncogene* 2004; 23: 3151–3171.

83. Korfel A, Schlegel U, Herrlinger U, *et al*. Phase II trial of temsirolimus for relapsed/refractory primary CNS lymphoma. *J Clin Oncol* 2016; 34: 1757–1763.

84. Maira SM, Pecchi S, Huang A, *et al*. Identification and characterization of NVP-BKM120, an orally available pan-class I PI3-kinase inhibitor. *Mol Cancer Ther* 2012; 11: 317–328.

85. Hou J-Z, Ye JC, Pu JJ, *et al*. Novel agents and regimens for hematological malignancies: recent updates from 2020 ASH annual meeting. *J Hematol Oncol* 2021; 14: 66.

86. Holstein SA and Lunning MA. CAR T-cell therapy in hematologic malignancies: a voyage in progress. *Clin Pharmacol Ther* 2020; 107: 112–122.

87. Abramson JS, McGree B, Noyes S, *et al*. Anti-CD19 CAR T cells in CNS diffuse large-B-cell lymphoma. *N Engl J Med* 2017; 377: 783–784.

88. Frigault MJ, Dietrich J, Martinez-Lage M, *et al*. Tisagenlecleucel CAR T-cell therapy in secondary CNS lymphoma. *Blood* 2019; 134: 860–866.

89. Siddiqi T, Wang X, Blanchard MS, *et al*. CD19-directed CAR T-cell therapy for treatment of primary CNS lymphoma. *Blood Adv* 2021; 5: 4059–4063.

90. Alcantara M, Houillier C, Blonski M, *et al*. CAR T-cell therapy in primary central nervous system lymphoma: the clinical experience of the French LOC network. *Blood* 2022; 139: 792–796.

91. Frigault MJ, Dietrich J, Gallagher KME, *et al*. Safety and efficacy of tisagenlecleucel in primary CNS Lymphoma: a phase I/II clinical trial. *Blood*. Epub ahead of print 15 February 2022. DOI: 10.1182/blood.2021014738.

92. Tu S, Zhou X, Guo Z, *et al*. CD19 and CD70 dual-target chimeric antigen receptor T-cell therapy for the treatment of relapsed and refractory primary central nervous system diffuse large B-cell lymphoma. *Front Oncol* 2019; 9: 1350.

93. Li T, Zhao L, Zhang Y, *et al*. CAR T-cell therapy is effective but not long-lasting in B-cell lymphoma of the brain. *Front Oncol* 2020; 10: 1306.

94. Mulazzani M, Fräßle SP, von Mücke-Heim I, *et al*. Long-term in vivo microscopy of CAR T cell dynamics during eradication of CNS lymphoma in mice. *P Natl Acad Sci* 2019; 116: 24275–24284.

95. Wang X, Huynh C, Urak R, *et al*. The cerebroventricular environment modifies CAR T cells for potent activity against both central nervous system and systemic lymphoma. *Cancer Immunol Res* 2021; 9: 75–88.

96. Abramson JS, Palomba ML, Gordon LI, *et al*. Lisocabtagene maraleucel for patients with relapsed or refractory large B-cell lymphomas (TRANSCEND NHL 001): a multicentre seamless design study. *Lancet* 2020; 396: 839–852.

97. Azmi AS, Uddin MH and Mohammad RM. The nuclear export protein XPO1 – from biology to targeted therapy. *Nat Rev Clin Oncol* 2021; 18: 152–169.

98. Kalakonda N, Maerevoet M, Cavallo F, *et al*. Selinexor in patients with relapsed or refractory diffuse large B-cell lymphoma (SADAL): a single-arm, multinational, multicentre, open-label, phase 2 trial. *Lancet Haematol* 2020; 7: e511–e522.

99. Bobillo S, Abrisqueta P, Carpio C, *et al.* Promising activity of selinexor in the treatment of a patient with refractory diffuse large B-cell lymphoma and central nervous system involvement. *Haematologica* 2018; 103: e92–e93.

100. Jiménez I, Carabia J, Bobillo S, *et al.* Repolarization of tumor infiltrating macrophages and increased survival in mouse primary CNS lymphomas after XPO1 and BTK inhibition. *J Neurooncol* 2020; 149: 13–25.

101. Shi Y, Jia B, Xu W, *et al.* Chidamide in relapsed or refractory peripheral T cell lymphoma: a multicenter real-world study in China. *J Hematol Oncol* 2017; 10: 69.

102. Guan XW, Wang HQ, Ban WW, *et al.* Novel HDAC inhibitor Chidamide synergizes with Rituximab to inhibit diffuse large B-cell lymphoma tumour growth by upregulating CD20. *Cell Death Dis* 2020; 11: 20.

103. Herter S, Herting F, Mundigl O, *et al.* Preclinical activity of the type II CD20 antibody GA101 (obinutuzumab) compared with rituximab and ofatumumab in vitro and in xenograft models. *Mol Cancer Ther* 2013; 12: 2031–2042.

104. Marcus R, Davies A, Ando K, *et al.* Obinutuzumab for the first-line treatment of follicular lymphoma. *N Engl J Med* 2017; 377: 1331–1344.

105. Das M. Venetoclax active and safe in non-Hodgkin lymphoma. *Lancet Oncol* 2017; 18: e136.

106. Reda G, Cassin R, Dovrtelova G, *et al.* Venetoclax penetrates in cerebrospinal fluid and may be effective in chronic lymphocytic leukemia with central nervous system involvement. *Haematologica* 2019; 104: e222–e223.

107. Hatzl S, Posch F, Deutsch A, *et al.* Immunohistochemistry for c-myc and bcl-2 overexpression improves risk stratification in primary central nervous system lymphoma. *Hematol Oncol* 2020; 38: 277–283.

108. Chu Y, Zhou X and Wang X. Antibody-drug conjugates for the treatment of lymphoma: clinical advances and latest progress. *J Hematol Oncol* 2021; 14: 88.

109. Liu Y, Zhou X and Wang X. Targeting the tumor microenvironment in B-cell lymphoma: challenges and opportunities. *J Hematol Oncol* 2021; 14: 125.