


# Taking the Next Step in Double Refractory Disease: Current and Future Treatment Strategies for Chronic Lymphocytic Leukemia

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**Abstract:** Chronic lymphocytic leukemia (CLL) is a monoclonal B-cell lymphoproliferative disease with a high annual incidence in Western countries. As B-cell receptor (BCR) signaling and intrinsic apoptotic resistance play critical roles in the development and survival of CLL cells, therapeutic approaches targeting these pathways have been extensively investigated to tackle this incurable disease. Over the last decade, several Phase 3 trials have confirmed the superior efficacy of covalent Bruton tyrosine kinase inhibitors (cBTKis) and venetoclax, a selective B-cell lymphoma 2 (BCL2) inhibitor, over chemoimmunotherapy. This has been demonstrated in both the treatment-naïve and relapsed/refractory (RR) settings and includes patients with high-risk molecular features. However, these drugs are not curative, with patients continuing to relapse after treatment with both cBTKis and BCL2is, and the optimal treatment strategy for these patients has not been defined. Several novel agents with distinct mechanisms have recently been developed for CLL which have demonstrated efficacy in patients who have previously received cBTKis and BCL2i. In particular, novel BCR-signaling targeting agents have shown promising efficacy in early-phase clinical trials for RR-CLL. Furthermore, cancer immunotherapies such as bispecific antibodies and chimeric antigen receptor T-cells have also shown anti-tumor activity in patients with heavily pretreated RR-CLL. Personalised approaches with these novel agents and combination strategies based on the understanding of resistance mechanisms have the potential to overcome the clinical challenge of what to do next for a patient who has already had a cBTKi and venetoclax.

**Keywords:** CLL, BTK inhibitor, BCL2 inhibitor, bispecific antibody, CAR-T cell therapy

## Introduction

Chronic lymphocytic leukemia (CLL) or small lymphocytic lymphoma (SLL) is an indolent hematological malignancy, characterized by the clonal proliferation and accumulation of functionally incompetent B cells in the blood, bone marrow, lymph nodes and spleen.<sup>1</sup> CLL and SLL are considered to be the same disease with different manifestations. The clinical presentation of CLL is primarily that of a peripheral lymphocytosis, whereas patients with SLL only manifest lymphadenopathy and organomegaly.<sup>2</sup> CLL is the most common adult leukemia in Western countries.<sup>3</sup> Traditional chemotherapeutic agents such as bendamustine, chlorambucil, fludarabine and cyclophosphamide used in combination with anti-CD20 monoclonal antibodies (mAbs) could induce long-term remissions, with these chemoimmunotherapy (CIT) regimens representing the standard treatment options for CLL until the advent of targeted therapies.<sup>4–6</sup>

As B-cell receptor (BCR) signaling mediates the development and differentiation of normal B cells and plays a crucial role in tumorigenesis of B-cell malignancies including CLL, therapeutic approaches targeting key molecules in the BCR-signaling pathways such as Bruton tyrosine kinase (BTK) and phosphatidylinositol-3-kinase (PI3K) were investigated.<sup>7</sup> In 2014, ibrutinib, the first-in-class covalent BTK inhibitor (cBTKi) was approved by the US Food and Drug Administration (FDA) for the treatment of relapsed/refractory (RR) CLL based on its remarkable clinical efficacy.<sup>8</sup> Following this, more selective next-generation cBTKis such as acalabrutinib and zanubrutinib were developed to

improve the efficacy and safety of ibrutinib.<sup>9</sup> Several phase 3 trials confirmed the superior efficacy of these cBTKis as monotherapy or in combination with an anti-CD20 mAb to CITs in the frontline and subsequent treatment settings.<sup>10–13</sup> Another important treatment target is an anti-apoptotic protein B-cell lymphoma 2 (BCL2), which is overexpressed in CLL cells.<sup>14</sup> Venetoclax, an oral selective BCL2 inhibitor (BCL2i), in combination with an anti-CD20 mAb also showed significant improvements in clinical outcomes compared to CITs.<sup>15,16</sup>

These targeted therapies have dramatically transformed the treatment landscape of CLL and long-term survival can be achieved for many patients even those with high-risk features such as *TP53* dysruption and complex karyotypes. However, CLL remains an incurable disease and patients experience disease progression after clinical responses to cBTKis and venetoclax. In this situation, they have limited treatment options with poor clinical outcomes, which highlights a significant unmet medical need.<sup>17</sup> Based on the molecular understanding of clinical resistance to cBTKi and venetoclax, several novel agents that could overcome the resistance mechanisms have entered the clinical investigation. The objectives of this article are to review the biological backgrounds and clinical applications of targeted therapies for CLL, to discuss current clinical challenges in the treatment of CLL, and to explore potential treatment strategies for patients who progressed after both cBTKi- and venetoclax-based therapies.

## The Biological Backgrounds of Targeted Therapies for CLL

The BCR complex with CD79A and CD79B regulates the development and differentiation of B cells by tonic or antigen-dependent BCR-signaling. Tonic BCR-signaling is considered independent of antigen binding and mediated by PI3K/protein kinase B (AKT)/mammalian target of rapamycin (mTOR) pathways.<sup>7</sup> When BCR is engaged by antigen, spleen tyrosine kinase (SYK) and LYN recruit other signaling partner proteins such as BTK, B-cell linker (BLNK), and phospholipase C gamma 2 (PLCG2).<sup>18</sup> BTK and SYK phosphorylate PLCG2, inducing calcium flux and the activation of the protein kinase C beta (PKC $\beta$ ), mitogen-activated protein kinase (MAPK) and nuclear factor kappa light chain enhancer of activated B cells (NF- $\kappa$ B) pathways. Furthermore, the BCR co-receptor CD19 is phosphorylated by LYN to enable the recruitment and activation of PI3K, promoting downstream activation of AKT and mTOR. PI3K generates a second messenger, phosphatidylinositol-3,4,5-triphosphate (PIP3), which activates BTK. The combination of these BTK- and PI3K-dependent signaling pathways regulates the proliferation and survival of B cells (Figure 1).<sup>19</sup>

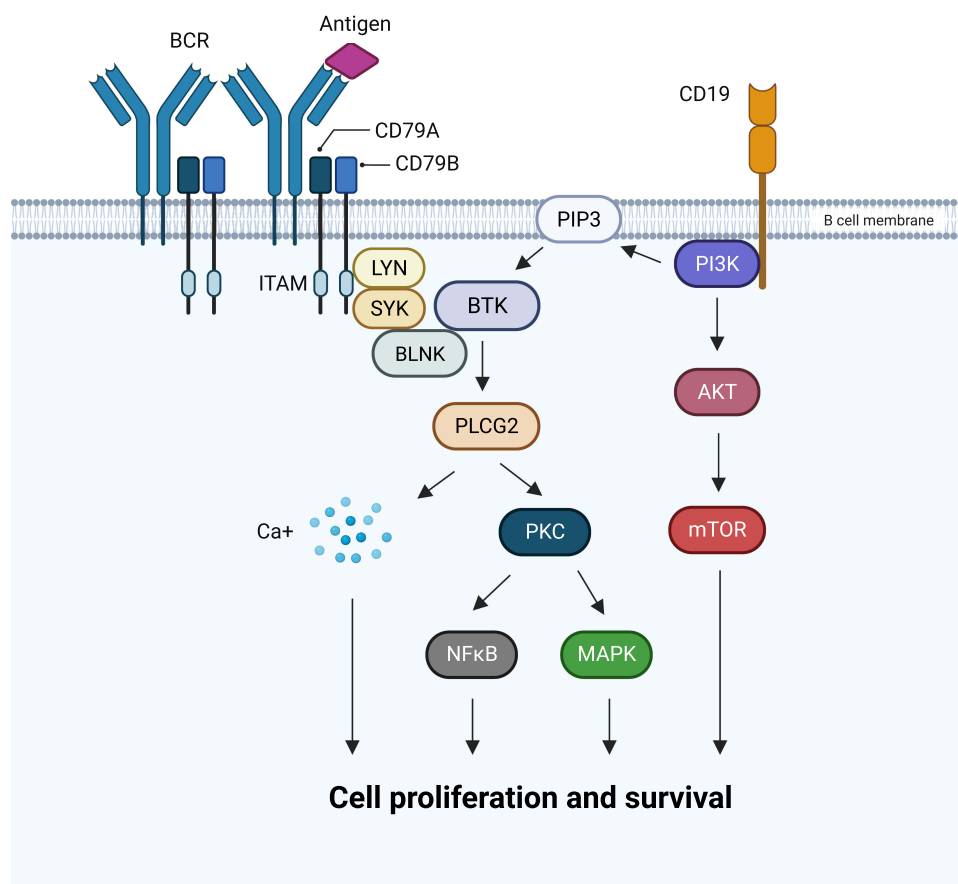
In CLL, BCR-signaling via antigen binding and homotypic interaction plays a crucial role in the development of malignant B cells, consistent with the clinico-pathological features of CLL. Patients with unmutated immunoglobulin heavy chain variable region (*IGHV*) genes have polyreactive low-affinity BCRs that can recognise various foreign antigens and autoantigens, leading to the proliferation and survival of CLL cells. In contrast, CLL cells with mutated *IGHV* genes have high-affinity BCRs with restricted reactivity: therefore, those patients generally show stable disease courses.<sup>20</sup> Furthermore, the fact that one-third of CLL patients have identical or near-identical “stereotyped” BCRs provides evidence for selection pressure by specific antigens.<sup>21</sup>

Apart from the activation of BCR-signaling, dysregulation of intrinsic apoptosis plays another important role in the development and survival of CLL cells.<sup>14</sup> Intrinsic mitochondrial apoptosis is regulated by the balance and interactions between BCL2 family member proteins, including pro-survival proteins such as BCL2, myeloid cell leukemia-1 (MCL1) and B-cell lymphoma-extra large (BCL-XL), pro-apoptotic BCL2 homology domain 3 (BH3)-only proteins such as BCL2 interacting mediator of cell death (BIM) and BCL2 associated agonist of cell death (BAD), and apoptosis effector proteins: BCL2 associated X-protein (BAX) and BCL2 antagonist/killer (BAK).<sup>22</sup> In certain types of tumors, particular pro-survival proteins such as BCL2 in CLL are overexpressed and the balance of these interactions tips towards cell survival. Therefore, small molecules that mimic the action of the BH3-only proteins have been developed, leading to the clinical application of venetoclax, a selective BCL2i, for the treatment of CLL.<sup>23</sup>

## Initial Treatment Options for CLL

### Ibrutinib

The molecular understanding of BCR-signaling pathways and intrinsic apoptosis regulation in B-cell malignancies promoted the development of multiple small molecule drugs that inhibit these pathways. Over the last 10–15 years, significant advances



**Figure 1** Overview of BCR signaling.

**Note:** Created with BioRender.com.

**Abbreviations:** AKT, protein kinase B; BCR, B-cell receptor; BLNK, B-cell linker; BTK, Bruton tyrosine kinase; CD, cluster of differentiation; ITAM, immunoreceptor tyrosine-based activation motif; LYN, LYN proto-oncogene, Src family tyrosine kinase; MAPK, mitogen-activated protein kinase; mTOR, mammalian target of rapamycin; NFκB, nuclear factor kappa light chain enhancer of activated B cells; PI3K, phosphatidylinositol-3-kinase; PIP3, phosphatidylinositol-3,4,5; PKC, protein kinase C; PLCG2, phospholipase C gamma 2; SYK, spleen tyrosine kinase.

have been made in BTK- and BCL2-targeted therapies, which revolutionized treatment strategies for CLL.<sup>24,25</sup> Ibrutinib is the first-in-class, orally bioavailable, highly potent BTK inhibitor, which covalently binds to a cysteine residue (C481) at the ATP binding site of BTK, to inhibit the phosphorylation of BTK and downstream signaling proteins including PLCG2, extracellular signal-regulated kinase (ERK), and NF-κB without affecting T-cell survival.<sup>26,27</sup> Furthermore, ibrutinib inhibits integrin and chemokine-mediated cell adhesion and migration, which is reflected the rapid reduction of lymphadenopathy and transient lymphocytosis in ibrutinib-treated patients.<sup>28,29</sup> In the Phase 1b/2 trial, ibrutinib monotherapy demonstrated full occupancy of BTK and an overall response rate (ORR) of 71% for heavily pretreated RR-CLL patients.<sup>30</sup> In February 2014, ibrutinib was granted accelerated approval from the FDA for the treatment of RR-CLL.<sup>8</sup> The phase 3 RESONATE trial directly compared ibrutinib and ofatumumab, an anti-CD20 mAb, in patients with RR-CLL at risk for a poor outcome. Ibrutinib significantly improved ORR (42.6% vs 4.1%,  $p < 0.001$ ) and median progression-free survival (mPFS) (not reached [NR] vs 8.1 months; hazard ratio [HR] 0.22, 95% confidence interval [CI] 0.15–0.32) compared to ofatumumab. The benefit of ibrutinib was also observed in a subgroup of patients with del(17p).<sup>31</sup>

In the frontline setting, several phase 3 trials have reported remarkable clinical outcomes with ibrutinib as monotherapy and in combination with an anti-CD20 mAb (rituximab or obinutuzumab). In the phase 3 RESONATE-2 trial, 269 treatment-naïve (TN) CLL/SLL patients aged  $\geq 65$  years without del(17p) were randomly assigned to receive ibrutinib or chlorambucil. Ibrutinib showed significantly higher ORR (86% vs 35%,  $p < 0.001$ ), longer PFS (mPFS NR vs 18.9 months; HR 0.16,  $p < 0.001$ ), and longer overall survival (OS) (2-year-OS 98% vs 85%; HR 0.16,  $p = 0.001$ ).<sup>32</sup> Subsequent phase 3 trials showed the superiority of ibrutinib with or without an anti-CD20

mAb to CIT regimens.<sup>33–36</sup> For example, in the phase 3 ECOG-ACRIN E1912 trial, 529 TN-CLL patients aged  $\leq 70$  years without del(17p) were randomly assigned to receive ibrutinib + rituximab (IbR) or fludarabine + cyclophosphamide + rituximab (FCR). IbR prolonged PFS (HR 0.35, 95% CI 0.22–0.56) and OS (HR 0.17, 95% CI 0.05–0.54) compared to FCR.<sup>33</sup> The long-term follow-up results confirmed the PFS improvement in patients with both mutated (HR 0.27,  $p < 0.001$ ) and unmutated *IGHV* (HR 0.27,  $p < 0.001$ ).<sup>37</sup> In the phase 3 iLLUMINATE trial with CLL/SLL patients aged  $\geq 65$  years or  $< 65$  years with co-existing comorbidities, ibrutinib + obinutuzumab (IbObi) showed significantly longer PFS (mPFS NR vs 22 months; HR 0.25, 95% CI 0.16–0.39) and higher undetectable minimal residual disease (uMRD) rate (38% vs 25%) compared to chlorambucil + obinutuzumab (ChlObi). The PFS benefit was also observed in patients with high-risk features of del(17p), *TP53* mutation, del(11q), and unmutated *IGHV*.<sup>34</sup> In the phase 3 Alliance A041202 trial with TN-CLL patients aged  $\geq 65$  years, both ibrutinib and IbR significantly improved PFS compared to bendamustine + rituximab (BR) (HR for ibrutinib vs BR 0.39, 95% CI 0.26–0.58; HR for IbR vs BR 0.38, 95% CI 0.25–0.59). IbR did not improve PFS compared to ibrutinib and there was no significant difference in OS between the three groups.<sup>35</sup>

## Acalabrutinib

Despite the remarkable clinical efficacy, patients may discontinue ibrutinib due to its characteristic adverse events (AEs) such as atrial fibrillation and bleeding, which can be explained by the off-target inhibition of other kinases such as C-terminal Src kinase and Tec kinase.<sup>38,39</sup> Therefore, more selective cBTKis were developed to overcome these challenges. Acalabrutinib is a potent and selective irreversible BTK inhibitor.<sup>40</sup> As the bioavailability of the acalabrutinib capsule is impaired by acid-suppressing therapies, a new tablet form of acalabrutinib, which can be coadministered with proton pump inhibitors, has also been developed.<sup>41</sup> The clinical efficacy of acalabrutinib for CLL has been shown in phase 3 trials. In the phase 3 ASCEND trial for RR-CLL, acalabrutinib significantly prolonged PFS compared to the investigator's choice of IdR (idelalisib + rituximab) or BR (mPFS NR vs 16.8 months; 42-month-PFS 62% vs 19%; HR 0.28, 95% CI 0.20–0.38). Acalabrutinib also showed a favourable trend of OS advantage (median overall survival [mOS] NR vs NR; 42-month-OS 78% vs 65%; HR 0.69, 95% CI 0.46–1.04).<sup>10</sup> In the phase 3 ELEVATE-RR trial, 533 RR-CLL patients with del(17p) or del(11q) were randomised to receive acalabrutinib or ibrutinib. Acalabrutinib was determined to be non-inferior to ibrutinib (mPFS 38.4 months in both arms; HR 1.00, 95% CI 0.79–1.27). Total cardiac events (24.1% vs 30.0%), hypertension (9.4% vs 23.2%), and atrial fibrillation/flutter (9.4% vs 16.0%) were less frequent with acalabrutinib than ibrutinib. Treatment discontinuations due to AEs were less frequent with acalabrutinib (14.7%) than ibrutinib (21.3%).<sup>42</sup>

In the frontline setting, the phase 3 ELEVATE-TN trial evaluated acalabrutinib + obinutuzumab (AcalaObi), acalabrutinib alone, or ChlObi. PFS was significantly prolonged with both AcalaObi (mPFS NR vs 22.6 months; HR 0.1, 95% CI 0.06–0.17) and acalabrutinib (mPFS NR vs 22.6 months; HR 0.20, 95% CI 0.13–0.3) compared to ChlObi. Cytopenia, infusion-related reactions, and grade 3 (G3) or higher infections were observed more frequently with AcalaObi than with acalabrutinib.<sup>11</sup> In the long-term follow-up analysis, AcalaObi prolonged PFS compared to acalabrutinib (HR 0.58, 95% CI 0.39–0.86).<sup>43</sup> Although there has not been direct comparison data of acalabrutinib and other cBTKis in the frontline setting, indirect comparisons have been conducted. A network meta-analysis of the iLLUMINATE, ELEVATE-TN, and CLL14 trials with a total of 1191 patients evaluated the efficacy and safety of frontline IbObi, venetoclax + obinutuzumab (VenObi), AcalaObi and acalabrutinib. No significant differences in PFS were observed between IbObi, VenObi, and acalabrutinib. In contrast, AcalaObi showed significantly improved PFS compared to IbObi (relative risk [RR] 0.43, 95% CI 0.22–0.87) and VenObi (RR 0.29, 95% CI 0.15–0.56). There were no significant differences in the frequency of AEs between these 4 groups.<sup>44</sup> Davids et al conducted a matching-adjusted indirect comparison (MAIC) of acalabrutinib with targeted comparators (ibrutinib, IbObi, and VenObi) using data from the ELEVATE-TN, RESONATE-2, iLLUMINATE and CLL14 trials. After matching baseline characteristics, neither acalabrutinib or AcalaObi significantly improved PFS compared to any other comparators, although both acalabrutinib and AcalaObi showed improved safety outcomes.<sup>45</sup>

## Zanubrutinib

Zanubrutinib is another potent and highly selective cBTKi. In the phase 1 study, BTK occupancy in peripheral blood mononuclear cells (PBMCs) was  $>95\%$  at 4-hour post doses in almost all patients at all doses.<sup>46</sup> In the phase 3 ALPINE

trial for RR-CLL, zanubrutinib showed significantly longer PFS (2-year-PFS 78.4% vs 65.9%; HR 0.65, 95% CI 0.49–0.86), and higher ORR (83.5% vs 74.2%), lower incidence of cardiac disorders (21.3% vs 29.6%) than ibrutinib. Zanubrutinib also showed the PFS benefit in the subgroup of patients with del(17p) or *TP53* mutation.<sup>12</sup> The long-term follow-up analysis confirmed the sustained PFS benefits of zanubrutinib over ibrutinib (3-year-PFS 65.8% and 54.3%; HR 0.67, 95% CI 0.52–0.86).<sup>47</sup> In the frontline setting, the phase 3 SEQUOIA trial evaluated 590 older or frail patients with TN-CLL/SLL. Among them, 479 patients without del(17p) were randomised to receive zanubrutinib or BR, and 111 patients with del(17p) were assigned to receive zanubrutinib. Compared to BR, zanubrutinib significantly prolonged PFS (2-year-PFS 86% vs 70%; HR 0.42, 95% CI 0.28–0.63). The PFS benefit of zanubrutinib was not observed in the subgroup of patients with mutated *IGHV*. In those with del(17p), zanubrutinib showed a 2-year-PFS rate of 89%.<sup>13</sup> The phase 3 clinical trials of cBTKis are summarised in Table 1.

**Table 1** A Summary of the Phase 3 Clinical Trials of Covalent BTK Inhibitors

Trial	Treatments	ORR	mPFS (HR)	mOS (HR)	AE G3 or Higher	Discontinuation Due to AE
RR-CLL						
RESONATE <sup>31</sup>	Ib vs Ofa	43% vs 4%	NR vs 8 mos (0.22)	NR vs NR (0.43)	57% vs 47%	4% vs 4%
ASCEND <sup>10</sup>	Acala vs IdR/BR	83% vs 84%	NR vs 17 mos (0.28)	NR vs NR (0.69)	68% (Acala) vs 92% (IdR) vs 49% (BR)	23% (Acala) vs 56% (IdR/BR)
ELEVATE-RR <sup>42</sup>	Acala vs Ib	81% vs 77%	38 mos vs 38 mos (1.00)	NR vs NR (0.82)	69% vs 75%	15% vs 21%
ALPINE <sup>12</sup>	Zanu vs Ib	84% vs 74%	NR vs 34 mos (0.65)	NR vs NR (0.76)	67% vs 70%	16% vs 23%
TN-CLL						
RESONATE-2 <sup>32</sup>	Ib vs Chl	86% vs 35%	NR vs 19 mos (0.16)	NR vs NR (HR 0.16)	Neutropenia: 10% vs 18%, Hypertension: 4% vs 0%, Diarrhea: 4% vs 0%	9% vs 23%
Alliance A041202 <sup>35</sup>	Ib vs BR	93% vs 81%	NR vs 43 mos (0.39)	NR vs NR	Hematologic: 41% vs 61% Non-hematologic: 74% vs 63%	NA
	IbR vs BR	94% vs 81%	NR vs 43 mos (0.38)	NR vs NR	Hematologic: 39% vs 61% Non-hematologic: 74% vs 63%	NA
ECOG-ACRIN E1912 <sup>33</sup>	IbR vs FCR	96% vs 81%	3y-PFS: 89% vs 73% (0.35)	3y-OS: 99% vs 92% (0.17)	80% vs 80%	NA
FLAIR <sup>36</sup>	IbR vs FCR	(At 9 mos) 91% vs 88%	NR vs 67 mos (0.44)	NR vs NR (1.01)	Febrile neutropenia: 1% vs 4%, Hypertension: 2% vs <1%, Diarrhea: 2% vs 2%	NA
iLLUMINATE <sup>34</sup>	IbObi vs ChlObi	91% vs 81%	NR vs 22 mos (0.25)	NR vs NR (1.08)	Neutropenia: 36% vs 46%, Hypertension: 4% vs 3%, Diarrhea: 3% vs 0%	NA
ELAVATE-TN <sup>11</sup>	AcalaObi vs ChlObi	94% vs 79%	NR vs 23 mos (0.10)	NR vs NR (0.47)	70% vs 70%	11% vs 14%
	Acala vs ChlObi	86% vs 79%	NR vs 23 mos (0.20)	NR vs NR (0.60)	50% vs 70%	9% vs 14%
SEQUOIA <sup>13</sup>	Zanu vs BR	95% vs 85%	NR vs NR (0.42)	NR vs NR (1.07)	53% vs 80%	8% vs 14%

**Abbreviations:** Acala, acalabrutinib; AE, adverse event; BTK, Bruton tyrosine kinase; BR, bendamustine + rituximab; Chl, chlorambucil; CLL, chronic lymphocytic leukemia; FCR, fludarabine + cyclophosphamide + rituximab; G3, grade 3; HR, hazard ratio; Ib, ibrutinib; IbR, ibrutinib + rituximab; IdR, idelalisib + rituximab; mos, months; mOS, median overall survival; mPFS, median progression free survival; NA, not available; NR, not reached; Obi, obinutuzumab; Ofa, ofatumumab; ORR, overall response rate; RR, relapsed/refractory; TN, treatment naïve; Zanu, zanubrutinib.



## Venetoclax-Based Therapies

Small molecules that mimic the action of the BH3-only proteins have been developed to target the intrinsic apoptosis dysregulation in B-cell malignancies. Venetoclax, an oral selective BCL2i, as monotherapy showed high response rates for heavily pretreated CLL patients including those with del(17p).<sup>48,49</sup> As it was found that an anti-CD20 mAb might be able to overcome resistance to venetoclax, the combination of these two agents has been investigated. The phase 3 MURANO trial compared venetoclax + rituximab (VenR) and BR in patients with RR-CLL. The 2-year-PFS rate was significantly higher with VenR than with BR (84.9% vs 36.3%; HR 0.17,  $p < 0.001$ ). The ORRs were 92.3% with VenR and 72.3% with BR. The rate of MRD clearance at the 9-month point in peripheral blood was higher with VenR than with BR (62.4% vs 13.3%). G3 or 4 tumor lysis syndrome was observed in 3.1% of patients with VenR.<sup>50</sup> Long-term follow-up results confirmed the PFS (4-year-PFS 57.3% vs 4.6%; HR 0.19, 95% CI 0.14–0.25) and OS benefits (4-year-OS 85.3% vs 66.8%; HR 0.41, 95% CI 0.26–0.65) with VenR compared to BR.<sup>15</sup>

In the frontline setting, the phase 3 CLL14 trial compared VenObi and ChlObi in 432 TN-CLL patients with coexisting conditions. VenObi significantly prolonged PFS (2-year-PFS 88.2% vs 64.1%; HR 0.35, 95% CI 0.23–0.53) compared to ChlObi. The PFS benefit was also observed in those with *TP53* deletion/mutation and in those with unmutated *IGHV*. The rates of uMRD in peripheral blood and bone marrow were higher with VenObi than with ChlObi (75.5% vs 35.2%, and 56.9% vs 17.1%, respectively).<sup>16</sup> The long-term follow-up analysis reported that the 6-year PFS rates were estimated as 53.1% and 21.7%, respectively.<sup>51</sup> In the phase 3 GAIA-CLL13 trial, VenR and VenObi were evaluated in TN-CLL patients without del(17p) or *TP53* mutation. VenObi significantly improved PFS (3-year-PFS 87.7% vs 75.5%; HR 0.42,  $p < 0.001$ ) and uMRD (86.5% vs 52.0%,  $p < 0.001$ ) compared with FCR/BR, whereas VenR did not.<sup>52</sup>

## Treatment Choices and Sequences of cBTKis and Venetoclax

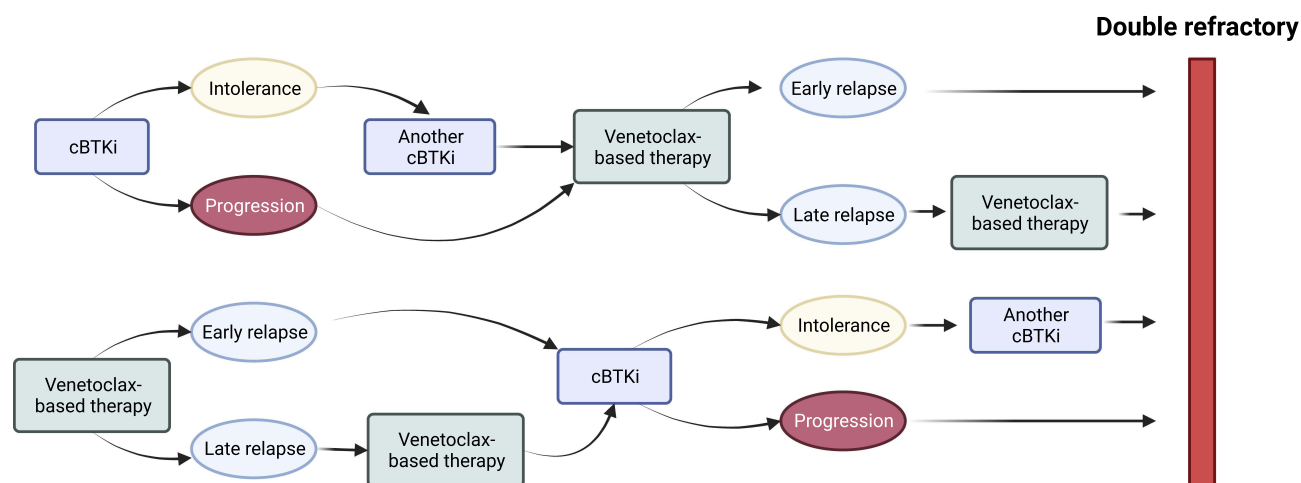
As several phase 3 trials confirmed the superior efficacy of cBTKi- and venetoclax-based therapies to CITs as described above, these two treatments should be used as a first- or second-line treatment for most patients. The patient's symptoms and comorbidities as well as the genetic risk of the disease should be considered for the choice of these agents.<sup>25,53</sup> Previous small studies reported treatment sequences of cBTKi and venetoclax in the first- and second-line setting. In a small Phase 2 trial, venetoclax treatment showed an ORR of 65% and a mPFS of 23.5 months in RR-CLL patients who had previously received ibrutinib.<sup>54</sup> In a small subset of patients who received subsequent ibrutinib therapy after VenR in the MURANO trial, the response rate was 87.5%.<sup>55</sup> Furthermore, a small retrospective study reported that BTKis (ibrutinib or zanubrutinib) showed durable remissions after progression on venetoclax with mPFS of 34 months and mOS of 42 months.<sup>56</sup> Venetoclax retreatment can be considered after a certain period of durable remission by a prior venetoclax-based therapy. An international retrospective study reported the efficacy and safety data of 46 CLL patients treated with venetoclax retreatment. Eighteen patients had received a cBTKi prior to the initial venetoclax therapy. The median duration between the completion of the initial and second venetoclax therapies was 16.1 months. The ORR and mPFS of the second venetoclax therapy were 79.5% and 25 months. In a subgroup of patients with a prior cBTKi, the ORR and mPFS were 56.3% and 15 months.<sup>57</sup>

Based on the results from indirect and direct comparisons between ibrutinib and newer cBTKis, acalabrutinib or zanubrutinib are preferred as the first choice cBTKi for the treatment of TN- and RR-CLL.<sup>12,44,45</sup> Regarding the choice between acalabrutinib and zanubrutinib, there has been no direct comparison. Based on individual patient data of acalabrutinib from the phase 3 ASCEND trial and published aggregated data of zanubrutinib, an unanchored MAIC was performed to compare acalabrutinib and zanubrutinib in the frontline setting. Although the PFS data were similar between acalabrutinib and zanubrutinib (HR 0.90, 95% CI 0.60–1.36), acalabrutinib was found to be associated with lower risks of serious AEs and dose reductions.<sup>58</sup>

## Clinical Challenges in the Era of Targeted Therapies

### Double Refractory Disease

Even though the treatment sequence with cBTKi and venetoclax-based therapies including retreatment approaches are effective, many patients eventually progress after treatment with both classes of targeted therapy (Figure 2).<sup>57,59</sup>



**Figure 2** CLL treatment sequences leading to “double refractory” disease.

**Note:** Created with BioRender.com.

**Abbreviations:** cBTKi, covalent Bruton tyrosine kinase inhibitor; CLL, chronic lymphocytic leukemia.

Prospective clinical data on this “double refractory disease”, are very limited.<sup>60</sup> Most of the clinical evidence derives from retrospective studies and the definitions of clinical resistance to targeted therapies are not consistent, particularly for fixed-duration venetoclax-based therapies. Aronson et al defined “double refractory” disease as a clinical situation where the patient (i) was treated with both a BTKi and venetoclax-based therapy, (ii) experienced disease progression while on a BTKi, and (iii) experienced disease progression while on venetoclax or within 24 months after venetoclax discontinuation, or became resistant to venetoclax retreatment.<sup>61</sup> The prognosis of patients with double refractory disease is poor due to the lack of effective treatments. A retrospective study with 17 double refractory CLL patients reported the mOS was 3.6 months, and the majority of them died from disease progression including 8 patients with Richter’s syndrome (RS).<sup>62</sup> Another retrospective study with 125 patients who had received both a cBTKi and venetoclax (double-exposed) reported the efficacy of subsequent treatments. The ORRs and mPFS were 40.9% and 5 months with PI3K inhibitors, and 31.8% and 3 months with CITs, respectively.<sup>63</sup>

## Resistance Mechanisms

Even though cBTKis can achieve long-term remissions, many patients develop secondary resistance. The mechanisms of secondary resistance have extensively been studied in patients progressing on cBTKis, particularly ibrutinib. Most of those who progressed on ibrutinib develop mutations affecting the C481 residue in the kinase domain of BTK, which hinders the covalent binding of ibrutinib to BTK.<sup>64</sup> Although mutations from cysteine to serine (C481S) are the most common, other mutations such as C481Y/R/F (tyrosine/arginine/phenylalanine) are also observed.<sup>65</sup> The next most common resistance mechanism is gain-of-function mutations of *PLCG2*.<sup>66</sup> Recently, secondary resistance mechanisms to newer cBTKis have been reported. As with the cases of ibrutinib, *BTK* C481 mutations are the most commonly observed in patients who progressed on acalabrutinib.<sup>67</sup> However, *BTK* L528W mutations (from leucine to tryptophan) have been reported as the most common resistance mechanisms to zanubrutinib.<sup>68,69</sup> Recent data with a small number of patients suggest that L528W mutations also show cross-resistance with pirtobrutinib, a novel non-covalent BTK inhibitor.<sup>69,70</sup>

Acquired mutations affecting the BH3-binding domain of *BCL2* have also been reported as the main resistance mechanisms to venetoclax. Blombery et al reported that *BCL2* G101V mutations were identified by next-generation sequencing (NGS) in 7 out of 15 patients who had progressed on continuous venetoclax therapy. The binding of venetoclax to *BCL2* was markedly reduced by this mutation.<sup>71</sup> In addition to G101V mutations, other *BCL2* mutations have been reported (eg D103Y), which, along with overexpression of *BCL-XL* and *MCL1*, have been associated with clinical resistance to venetoclax.<sup>72,73</sup> A recent study with 24 patients who had progressed on BTKi and received continuous venetoclax monotherapy reported that *BCL2* G101V mutations were rare and *BTK* and *PLCG2* mutations

persisted or increased during venetoclax treatment.<sup>74</sup> However, as these data largely derive from continuous venetoclax monotherapy, the clinical implications are not clear for the majority of patients receiving fixed-duration venetoclax-based regimens.

## cBTKi and BCL2i Combinations

The fact that cBTKi and BCL2i have distinct and complementary mechanisms of action, combined with preclinical observations showing that BTK inhibition increases functional dependence on BCL2, paved the way for synergistic combinations of these classes.<sup>75,76</sup> The increase of BCR-pathway mutations during venetoclax therapy after the discontinuation of cBTKi also provides a rationale to delay the emergence of resistance mutations by combination strategies.<sup>74</sup> Furthermore, the cBTKi and venetoclax combination can achieve deep and durable responses with a fixed duration of treatment. As the clearance of MRD is correlated with longer PFS and OS, the uMRD status can predict long-term remissions, highlighting the importance of MRD as a clinical trial endpoint and a biomarker which can guide treatment approaches.<sup>77,78</sup> The phase 2 CAPTIVATE trial investigated different treatment regimens based on the uMRD status after 3 cycles of ibrutinib lead-in followed by 12 cycles of ibrutinib + venetoclax (IbVen) in TN-CLL. In the MRD cohort of this trial, patients who achieved uMRD after the fixed-duration IbVen combination were randomly assigned to ibrutinib or placebo. The 1-year disease-free survival rates were not significantly different between them (100% vs 95%, respectively) ( $p = 0.15$ ).<sup>77</sup> The updated results of this trial showed that no BTK mutations were found at disease progression after the fixed-duration IbVen and the ORR of ibrutinib retreatment was 86%.<sup>78</sup> Another single-arm phase 2 trial investigated the combination treatment with zanubrutinib, obinutuzumab and venetoclax (BOVen), which was discontinued after 8–24 cycles when uMRD was met. Thirty-three patients (89%) reached the pre-specified uMRD status with a median of 10 cycles.<sup>79</sup> Currently, fixed-duration and MRD-guided cBTKi and BCL2i combination with or without an anti-CD20 mAb are being investigated in phase 3 trials (Table 2). In the phase 3 GLOW trial, 211 patients with TN-CLL/SLL were randomly assigned to receive fixed-duration IbVen (3 cycles of ibrutinib lead-in, then 12 cycles of IbVen) or ChlObi (6 cycles). IbVen significantly prolonged PFS compared to ChlObi (HR 0.22, 95% CI 0.13–0.36). The rate of sustained uMRD in the peripheral blood for 3 to 12 months after the end of treatment was 84.5% for IbVen and 29.3% for ChlObi. AEs G3 or higher occurred in 75.5% of those who received IbVen and 69.5% of those who received ChlObi.<sup>80</sup> The 54-month PFS and OS rates were reported as 65.8% and 84.5% for IbVen and 19.1% and 63.1% for ChlObi, respectively.<sup>81</sup> IbVen was approved as the frontline treatment for TN-CLL by the European Commission in

**Table 2** Phase 3 Clinical Trials for BTKi and BCL2i Combinations

Trial	Study Treatments	Clinical Trial Number
GLOW	IbVen vs ChlObi	NCT03462719
FLAIR	IbVen vs FCR	ISRCTN01844152
GAIA-CLL13	VenR vs VenObi vs IbObiVen vs CIT (FCR/BR)	NCT02950051
CLL-16	AcalaVenObi vs VenObi	NCT05197192
CLL-17	Ib vs VenObi vs IbVen	NCT04608318
ECOG-ACRIN EA9161	IbObiVen vs IbObi	NCT03701282
ALLIANCE A041702	IbObi vs IbObiVen	NCT03737981
ACE-CL-311	AcalaVen vs AcalaVenObi vs CIT (FCR/BR)	NCT03836261
MAJIC	MRD-driven AcalaVen vs VenObi	NCT05057494
BGB-11417-301	Zanubrutinib + Sonrotoclax vs VenObi	NCT06073821

**Abbreviations:** AcalaVen, acalabrutinib + venetoclax; AcalaVenObi, acalabrutinib + venetoclax + obinutuzumab; BR, bendamustine + rituximab; ChlObi, chlorambucil + obinutuzumab; CIT, chemoimmunotherapy; IbObi, ibrutinib + obinutuzumab; IbObiVen, ibrutinib + obinutuzumab + venetoclax; IbVen, ibrutinib + venetoclax; MRD, minimal residual disease; VenObi, venetoclax + obinutuzumab; VenR, venetoclax + rituximab.



August 2022.<sup>82</sup> In the phase 3 FLAIR trial, IbVen (with MRD-guided duration) and ibrutinib alone arms were added in 2017. The recent analysis showed that the 4-year-PFS rates were 93.5% for IbVen and 64.8% for FCR (HR 0.13, 95% CI 0.07–0.24) and the 4-year-OS rates were 94.9% for IbVen and 87.3% for FCR (HR 0.31, 95% CI 0.15–0.67).<sup>83</sup> The phase 3 GAIA-CLL13 trial compared VenR, VenObi, ibrutinib + obinutuzumab + venetoclax (IbObiVen) (ibrutinib was discontinued or extended based on the uMRD status), or CIT (FCR or BR) in 926 fit TN-CLL patients without del (17p) or *TP53* mutation. IbObiVen improved 3-year-PFS (90.5% vs 75.5%, HR 0.32,  $p < 0.001$ ) and uMRD at 15 months (92.2% vs 52.0%,  $p < 0.001$ ) compared to CIT. However, the addition of ibrutinib to VenObi did lead to an increase in G3 or 4 infections (21.2% vs 13.2%).<sup>52</sup>

## Novel Targeted Therapies for Double Refractory Disease

### Non-Covalent BTK Inhibitors

As described above, the most common secondary resistance mechanism to cBTKi are *BTK* C481 mutations. Therefore, novel agents that non-covalently bind to BTK at non-C481 sites have been developed with promising clinical efficacy. Pirtobrutinib (LOXO-305) is a novel oral non-covalent BTK inhibitor (ncBTKi), with activity against both wild-type and C481-mutated BTK.<sup>84</sup> Currently, the phase 1/2 BRUIN trial is investigating the safety and efficacy of pirtobrutinib for B-cell malignancies including CLL/SLL. A recent analysis of the ongoing BRUIN trial included 282 CLL/SLL patients who had received a previous BTKi with a median number of prior therapies of 4 (range 1–11). The ORR, ORR including partial response with lymphocytosis (PR-L) and mPFS were 72%, 82% and 19.4 months. In the subgroup of those who had received prior BTKi and BCL2i, the ORR including PR-L and mPFS were 79.7% and 15.9 months.<sup>85</sup> In December 2023, pirtobrutinib achieved accelerated approval from the FDA for the third or later-line treatment of CLL/SLL.<sup>86</sup>

Nemtabrutinib (MK-1026, formerly ARQ-531) is another ncBTKi with high potency against both wild-type and C481S-mutated BTK. In a recent analysis of the phase 1/2 BELLWAVE-001 trial, 57 patients with CLL/SLL were treated at the recommended phase 2 dose of 65 mg. Among them, 54 patients (95%) had a prior BTKi, 24 patients (42%) had both prior BTKi and venetoclax, and 36 patients (63%) had BTK C481S mutation. With the median follow-up of 8.1 months, ORR, median duration of response (mDoR) and mPFS were 56%, 24.4 months and 26.3 months, respectively. In those with prior BTKi and venetoclax, ORR, mDoR and mPFS were 58%, 8.5 months and 10.1 months, respectively.<sup>87</sup>

### BTK Degraders

Another BTK-targeted approach to overcome the resistance to cBTKis and ncBTKis is the degradation of the BTK protein itself rather than the inhibition of the BTK function. BTK degraders induce catalytic ubiquitination of BTK via recruitment of the cereblon E3 ubiquitin ligase complex, leading to BTK degradation by the proteasome.<sup>88</sup> Recent studies reported acquired BTK mutations including T474I gatekeeper mutations on the treatment with ncBTKis, and BTK degraders can bind to BTK proteins with T474 mutations.<sup>89</sup> NX-2127 is a novel oral BTK degrader that also degrades IKAROS family zinc finger 1 (IKZF1) and IKZF3, inducing immunomodulatory activity.<sup>90</sup> Recently, preliminary data of the phase 1 NX-2127-001 trial have been reported. Among 17 RR-CLL patients, all had received a prior BTKi and 13 patients (76.5%) had also received venetoclax. In 12 response-evaluable CLL patients, although the best ORR was 33%, ORR increased with longer follow-up (16.7% at 2 months, 42.9% at 4 months, and 50% at 6 months). Responses were also observed in double-refractory patients and those who progressed on a ncBTKi.<sup>91</sup>

NX-5948 is an oral small molecule that selectively degrades BTK. In a preclinical study, NX-5948 catalysed the rapid degradation of BTK and potently inhibited B-cell activation in human PBMCs. Oral administration of NX-5948 demonstrated the degradation of BTK in mice and cynomolgus monkeys.<sup>92</sup> Preliminary data of the ongoing phase 1 NX-5948-301 trial showed rapid, robust and sustained BTK degradation with NX-5948 at 50 mg and 100 mg dose levels in patients with RR B-cell malignancies.<sup>88</sup> In addition to NX-2127 and NX-5948, other BTK degraders: BGB-16673 (NCT05006716 and NCT05294731), ABBV-101 (NCT05753501), and AC676 (NCT05780034) are also currently under clinical investigation for B-cell malignancies including CLL.

## Novel BCL2 Targeted Therapies

Novel BCL2is are currently under development and there are some preliminary data for RR-CLL, although the clinical data among patients who progressed on venetoclax are very limited. Lisoftoclax is a highly selective and potent BCL2i. Preliminary data with 141 RR-CLL/SLL patients in the phase 2 trial have recently been reported. Seventeen patients (12%) had progressed on a BTKi (n = 15) and/or venetoclax (n = 3). The ORRs with lisoftoclax monotherapy, lisoftoclax + acalabrutinib, and lisoftoclax + rituximab were 65%, 98%, and 87%, respectively.<sup>93</sup> Sonrotoclax (BGB-11417) is a novel highly selective and potent BCL2i with a favourable pharmacokinetics profile and a broad therapeutic index.<sup>94</sup> According to the preliminary data for CLL/SLL in the phase 1 BGB-11417-101 trial, the ORRs with sonrotoclax monotherapy and in combination with zanubrutinib were 67% (6/8) and 95% (19/25), respectively.<sup>95</sup> LOXO-338 is a novel BCL2i developed to achieve selectivity over BCL-XL.<sup>96</sup> The phase 1 LOXO-BCL-20001 trial is currently evaluating LOXO-338 monotherapy and in combination with pirtobrutinib in RR B-cell malignancies including CLL/SLL (NCT05024045).

## Immunotherapeutic Approaches

Recent advancements in cancer immunotherapy have dramatically changed the treatment landscape in hematological malignancies as well as solid tumors.<sup>97</sup> In CLL, observations regarding the immunological effect of allogeneic hematopoietic stem cell transplantation (graft-versus-leukemia effect) provide a rationale to develop immunotherapeutic approaches to boost anti-tumor immune responses. Currently, several novel antibody-based and adoptive immunotherapies have shown promising efficacy as a monotherapy or in combination with targeted therapies in heavily pre-treated RR-CLL.<sup>98</sup>

## Monoclonal Antibodies (mAbs)

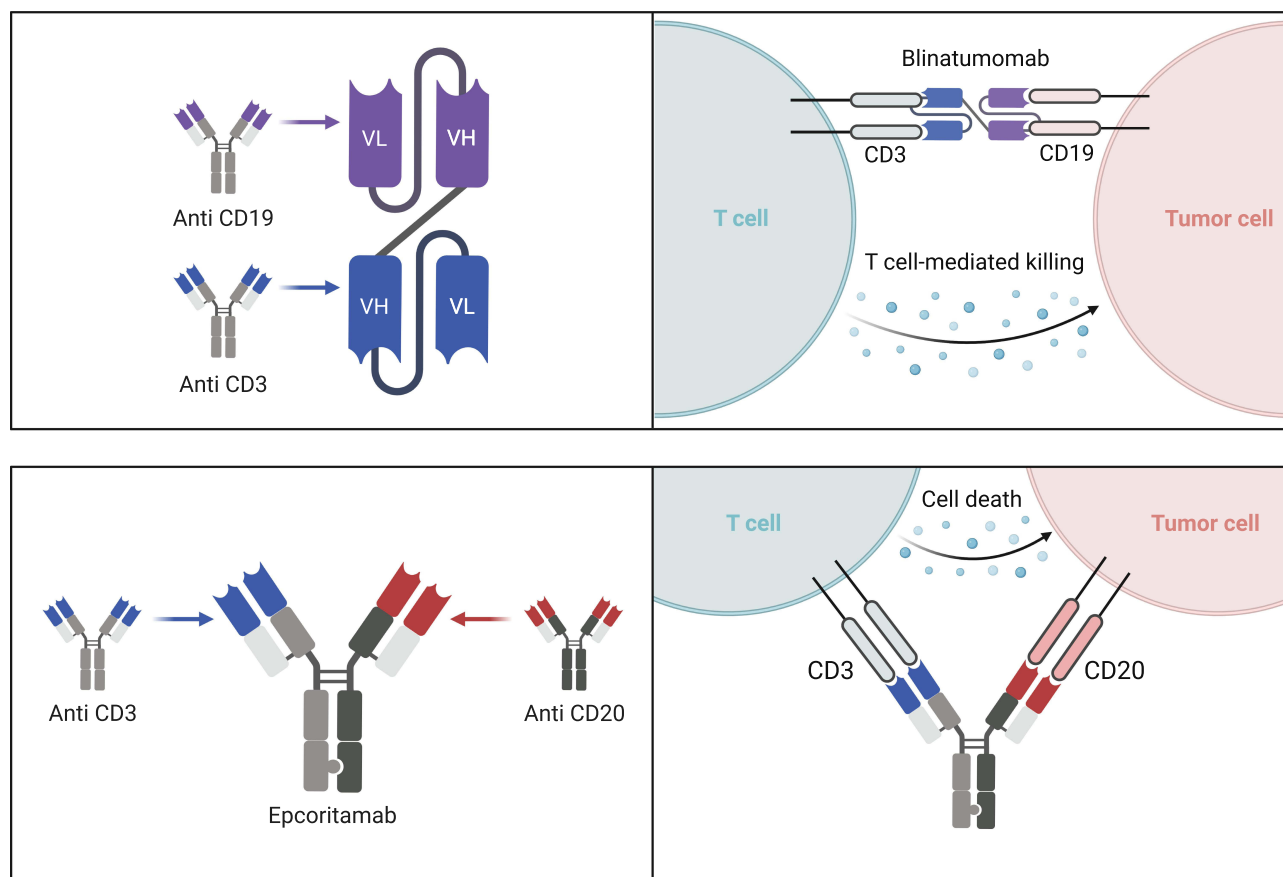
CD20 has been considered a safe and effective target for B-cell malignancies including CLL.<sup>99</sup> Rituximab and obinutuzumab have been widely used for the treatment of CLL in combination with chemotherapies and targeted therapies.<sup>17</sup> Ofatumumab is a fully humanised anti-CD20 mAb, which has shown efficacy in combination with chemotherapies and PI3K inhibitors.<sup>100–102</sup> Although these anti-CD20 mAbs can be used as monotherapy in later lines, the clinical efficacy is very limited for patients who progressed on venetoclax treatment, which is usually combined with an anti-CD20 mAb.<sup>59</sup> Another commonly targeted antigen is CD19, which is continuously expressed on the surface of all stages of B cells.<sup>103</sup> Tafasitamab, a novel anti-CD19 mAb with enhanced CD16 affinity, has shown promising efficacy in RR-CLL. In a cohort of the phase 2 COSMOS trial, 11 patients who had received a prior BTKi were treated with tafasitamab in combination with idelalisib. One patient also had received previous treatment with venetoclax. The best ORR was 90.9% and uMRD was achieved in 2 of 8 patients.<sup>104</sup> Currently, tafasitamab in combination with zanubrutinib for TN-CLL (NCT05718869), in combination with piasclisib, a next-generation PI3K $\delta$  inhibitor, for RR-NHL or CLL (NCT04809467), and in combination with acalabrutinib and obinutuzumab for TN-CLL (NCT05943496) are under clinical investigation.

Immune checkpoint inhibitors (ICIs) block inhibitory receptors on immune cells or their ligands on tumor cells such as programmed cell death protein 1 (PD1) and programmed death-ligand 1 (PD-L1) to overcome tumor immune escape mechanisms, leading to remarkable long-term tumor responses in many types of solid cancers.<sup>98</sup> Anti-PD1/PD-L1 antibodies showed significant efficacy results in multiple hematological malignancies such as Hodgkin's lymphomas and diffuse large B-cell lymphoma (DLBCL).<sup>105,106</sup> However, the clinical efficacy of ICI monotherapy for CLL is disappointing which is likely to reflect the pseudo-exhausted state of T-cells in CLL.<sup>107</sup> In a small phase 2 trial with pembrolizumab, an anti-PD1 antibody, the ORRs in 16 RR-CLL and 9 RS patients were 0% and 44%, respectively.<sup>108</sup> For the treatment of RS, combination strategies with targeted therapies have shown promising results. Nivolumab, an anti-PD1 antibody, in combination with ibrutinib reported an ORR of 46%.<sup>109</sup> Furthermore, atezolizumab, an anti-PD-L1 antibody, in combination with venetoclax and obinutuzumab showed a 100% response rate for previously untreated RS.<sup>110</sup>

## Bispecific Antibodies

A novel promising immunotherapeutic approach has been the development of bispecific antibodies (bsAbs), which redirect T cells to tumor cells, leading to potent tumor cell lysis (Figure 3). Currently, many bsAbs based on a variety of platforms, are under clinical development in hematological malignancies as well as solid cancers.<sup>111</sup> Blinatumomab, a CD3/CD19 bispecific T cell engager (BiTE), was the first bsAb evaluated for the treatment of CLL. In a small phase 2 trial with 9 heavily pre-treated patients with RS, blinatumomab treatment was well tolerated with no G3 or higher cytokine release syndrome (CRS) and 1 case of G3 neurotoxicity. However, the ORR and mPFS were only 22% and 1.9 months.<sup>112</sup> Currently, phase 1 studies are evaluating blinatumomab and lenalidomide combination in non-Hodgkin lymphoma (NHL) including SLL (NCT02568553) and blinatumomab-expanded T cells in indolent NHL and CLL (NCT03823365).

CD3/CD20-targeting bsAbs have also shown promising efficacy for RS. Epcoritamab is a subcutaneously administered CD3/CD20 bsAb, redirecting and activating T cells to kill CD20-expressing tumor cells. The phase 1b/2 EPCORE CLL-1 trial is investigating epcoritamab monotherapy and in combination with venetoclax for RR-CLL and epcoritamab monotherapy and in combination with lenalidomide or R-CHOP for RS.<sup>113</sup> Initial data included 10 patients with RS who were treated with epcoritamab monotherapy. No cases of G3 or more CRS and no case of immune effector cell-associated neurotoxicity syndrome (ICANS) were observed. The ORR was 60% with a complete response rate of 50%.<sup>114</sup> Epcoritamab was granted accelerated approval from the FDA for the treatment of adult RR-DLBCL including DLBCL arising from indolent lymphoma in May 2023.<sup>115</sup> Glofitamab, a CD3/CD20 bsAb with two anti-CD20 binding domains, also showed promising efficacy for RS. In the phase 1/2 trial with 11 patients with RS, the ORR and complete response



**Figure 3** Mechanism of action of bispecific antibodies. Blinatumomab is a scFv-based bispecific antibody (bsAb) (upper left). Epcoritamab is an IgG-like bsAb (lower left). These bsAbs redirect T cells to tumor cells (lower left and right).

**Note:** Created with BioRender.com.

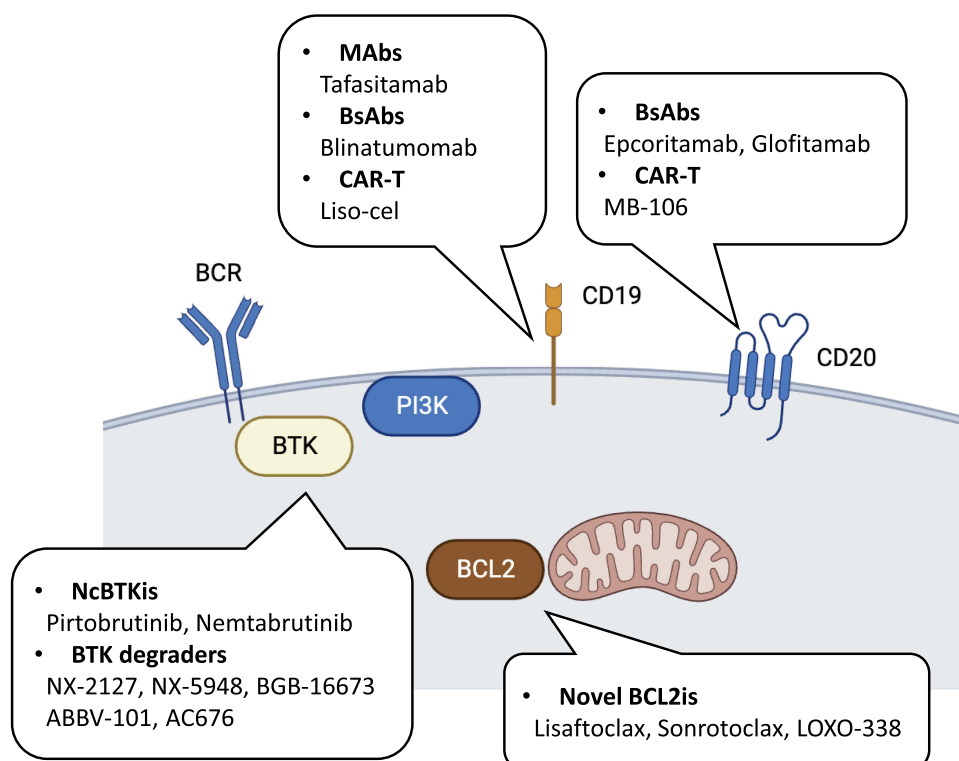
**Abbreviations:** CD, cluster of differentiation; Ig, immunoglobulin; scFv, single chain variable fragment; VH, variable fragment heavy chain; VL, variable fragment light chain.

rate were 63.6% and 45.5%, respectively. Glofitamab showed a manageable safety profile with 2 cases of G3 or 4 CRS and 1 case of G3 ICANS.<sup>116</sup> Glofitamab was also granted accelerated approval from the FDA for the treatment of adult RR-DLBCL in June 2023.<sup>117</sup>

## Chimeric Antigen Receptor T Cell Therapies

In addition to antibody-based therapies, chimeric antigen receptor (CAR)-T cell therapies have shown remarkable results with long-term complete remissions in hematological malignancies such as DLBCL and B-cell acute lymphoblastic leukemia (ALL).<sup>118–120</sup> In autologous CAR-T cell therapies, T cells derived from the patient are genetically engineered to express CARs on the cell surface. A CAR is composed of a single chain variable fragment (scFV) that recognises a specific tumor antigen and intracellular T-cell activation domains. The CAR-T cells are infused back into the patient and recognise the target antigens on the tumor cells, inducing downstream signals to kill the tumor cells. Although the first experimental use of CAR-T cells was reported as the treatment of CLL in 2011,<sup>121</sup> further development was outpaced by other hematological malignancies and CAR-T therapies are still in the investigational stage for CLL.<sup>122</sup>

The phase 1/2 TRANSCEND CLL004 trial has recently reported the preliminary data on lisocabtagene maraleucel (liso-cel), an autologous CD19-targeting CAR-T cell product, with 118 RR-CLL/SLL patients. All patients had received a previous BTKi. In the primary efficacy analysis set, 50 patients received liso-cel at a target dose of  $100 \times 10^6$  CAR T cells after lymphodepleting chemotherapy. Among them, the rate of complete response or remission (CR) and CR with incomplete marrow recovery was 20%.<sup>123</sup> In order to improve the clinical outcomes of CAR-T cell therapies for CLL, there are some challenges to overcome. As the loss of CD-19 represents one of the main causes of relapse, other target molecules are being investigated.<sup>124</sup> MB-106 is a third-generation CD20-targeted CAR-T cell product. In the initial report from the ongoing phase 1/2 trial for high-risk B-NHL and CLL, 16 patients including 1 CLL patient were treated



**Figure 4** Novel targeted agents for double refractory disease.

**Note:** Created with BioRender.com.

**Abbreviations:** BCL2, B-cell lymphoma 2; BCL2is, B-cell lymphoma 2 inhibitors; BTK, Bruton tyrosine kinase; BCR, B-cell receptor; BsAbs, bispecific antibodies; CAR-T, chimeric antigen receptor T-cells; Liso-cel, lisocabtagene maraleucel; MAbs, monoclonal antibodies; NcBTKis, non-covalent Bruton tyrosine kinase inhibitors; PI3K, phosphatidylinositol-3-kinase.

with MB-106. The ORR was 94% and the CLL patient had a complete response and uMRD in peripheral blood and bone marrow.<sup>125</sup> Other potential target antigens that are highly expressed on CLL cells include receptor tyrosine kinase-like orphan receptor 1 (ROR1) and Fc receptor for immunoglobulin M (FcμR).<sup>126,127</sup> Furthermore, as ibrutinib has beneficial effects on the tumor microenvironment and can expand CAR-T cells, a combination strategy may overcome the resistance to CD19-targeted CAR-T therapies.<sup>128</sup> In the phase 1/2 TRANSCEND CLL004 trial, ibrutinib and liso-cel combination showed an ORR of 95% in 19 RR-CLL patients who had previously received ibrutinib.<sup>129</sup>

## Conclusion

In the last 10–15 years, small molecule agents targeting key proteins in BCR-signaling and intrinsic apoptotic pathways have significantly improved clinical outcomes of CLL. Current treatment algorithms with cBTKi- and venetoclax-based therapies based on patient characteristics and treatment responses can bring long-term survival to patients with this incurable disease. Extensive biological research has revealed the mechanisms that underlie resistance to cBTKis and venetoclax, enabling the clinical development of novel targeted agents. The promising preliminary data of ncBTKis and BTK degraders suggest that the use of these BTK-targeted agents may be the next treatment approach for double refractory disease. In addition, novel cancer immunotherapies such as bsAbs and CAR-T therapies can expand the treatment armamentarium for RR-CLL including double-refractory disease. Considering the distinct mechanisms of action, combination strategies with immunotherapies and targeted therapies are also being extensively investigated. As described in this article, there are a wide array of potential new treatment strategies for double-refractory disease (Figure 4). In order to accelerate the clinical application of these treatments, further research on reliable biomarkers for appropriate patient selection and treatment sequence is necessary. Furthermore, effective collaboration among academia, industry, regulatory agents, and patients in multiple regions is increasingly important to efficiently conduct patient-centered clinical trials of novel treatments for CLL.

## Abbreviations

AcalaObi, acalabrutinib + obinutuzumab; AE, adverse event; AKT, protein kinase B; ALL, acute lymphoblastic leukemia; BAD, BCL2 associated agonist of cell death; BAK, BCL2 antagonist/killer; BAX, BCL2 associated X-protein; BCL2, B-cell lymphoma 2; BCL2i, BCL2 inhibitor; BCL-XL, B-cell lymphoma-extra large; BCR, B-cell receptor; BH3, BCL2 homology domain 3; BIM, BCL2 interacting mediator of cell death; BiTE, bispecific T cell engager; BLNK, B-cell linker; BOVen, zanubrutinib + obinutuzumab + venetoclax; BR, bendamustine + rituximab; bsAb, bispecific antibody; BTK, Bruton tyrosine kinase; cBTKi, covalent BTK inhibitor; CAR, chimeric antigen receptor; ChlObi, chlorambucil + obinutuzumab; CI, confidence interval; CIT, chemoimmunotherapy; CLL, chronic lymphocytic leukemia; CR, complete response or remission; CRS, cytokine release syndrome; ERK, extracellular signal-regulated kinase; FCR, fludarabine + cyclophosphamide + rituximab; FcμR, Fc receptor for immunoglobulin M; FDA, US Food and Drug Administration; G, grade; HR, hazard ratio; IbObi, ibrutinib + obinutuzumab; IbR, ibrutinib + rituximab; IbVen, ibrutinib + venetoclax; ICI, immune checkpoint inhibitor; ICANS, immune effector cell-associated neurotoxicity syndrome; IdR, idelalisib + rituximab; IGHV, immunoglobulin heavy chain variable region; IKZF, IKAROS family zinc finger; mAb, monoclonal antibody; MAIC, matching-adjusted indirect comparison; MAPK, mitogen-activated protein kinase; MCL1, myeloid cell leukemia-1; mDoR, median duration of response; mOS, median overall survival; mPFS, median progression free survival; mTOR, mammalian target of rapamycin; ncBTKi, non-covalent BTK inhibitor; NF-κB, nuclear factor kappa light chain enhancer of activated B cells; NGS, next-generation sequencing; NHL, non-Hodgkin's lymphoma; NR, not reached; ORR, overall response rate; OS, overall survival; PBMC, peripheral blood mononuclear cell; PD1, programmed cell death protein 1; PD-L1, programmed death-ligand 1; PI3K, phosphatidylinositol-3-kinase; PIP3, phosphatidylinositol-3,4,5-triphosphate; PKCβ, protein kinase C beta; PLCG2, phospholipase C gamma 2; PR-L, partial response with lymphocytosis; ROR1, receptor tyrosine kinase-like orphan receptor 1; RR, relapsed or refractory; RR, relative risk; RS, Richter's syndrome; scFV, single chain variable fragment; SLL, small lymphocytic lymphoma; SYK, spleen tyrosine kinase; TN, treatment naïve; uMRD, undetectable minimal residual disease; VenObi, venetoclax + obinutuzumab; VenR, venetoclax + rituximab.



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## Disclosure

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