Bendamustine: A review of pharmacology, clinical use and immunological effects (Review)

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Abstract. Bendamustine is an alkylating agent classified into the group of nitrogen mustard analogues, synthesized almost sixty years ago. It was registered in former East Germany in 1971 and approved by the US Food and Drug Administration in 2008 for treatment of chronic lymphocytic leukemia and indolent B-cell non-Hodgkin lymphoma. Considering its beneficial properties in the therapy of relapsed or refractory hematological malignancies, synergistic effects with other antineoplastic agents and increasing recent reports on its immunomodulatory effects, bendamustine has once again gained its justified attention. The uniqueness of bendamustine-mediated effects should be observed keeping in mind its distinctive structure with structural similarities to both alkylating agents and purine analogs. In the present review, the current knowledge on the use of bendamustine in oncology, its pharmacokinetics, mechanism of action and toxicity was summarized. In addition, its immune-modulating effects that have not been fully elucidated so far are emphasized, hoping to encourage further investigations of this unique drug.

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1. Introduction

According to the Anatomical Therapeutic Chemical (ATC) classification system (1), bendamustine belongs to the group of antineoplastic and immunomodulating agents (L), antineoplastic agents (L01), alkylating agents (L01A), nitrogen mustard analogues (L01AA) and is given an ATC code L01AA09. Chemically, bendamustine is 4-{5-[bis(2-chloroethyl)amino]-1-methyl-2-bezimidazolyl} butyric acid hydrochloride. It was first synthesized in the early 1960s at the Institute for Microbiology and Experimental therapy in Jena, in the former East German Democratic Republic (2). Nitrogen mustard, an alkylating drug after which the L01AA ATC group was named, was used in the World War I as chemical weapon causing skin lesions, blindness, lung damage, nausea, and vomiting. After learning about mutagenic properties of nitrogen mustard and its effects on lowering the white blood cells count, many other alkylating agents had been developed with a goal to treat malignant tumors (3). The synthesis of bendamustine was based on the idea to produce a nitrogen mustard compound less toxic and at least as effective as other alkylating agents. As observed in Fig. 1, the specific mechanism of action of bendamustine is related to its unique structure with similarities to both alkylating agents and purine analogs.

Bendamustine was first registered in 1971 as a treatment for both hematological malignancies and solid tumors. Although not widely prescribed in East Germany, the interest for this drug increased after the reunification of Germany in the 1990s, mostly as a treatment for lymphoid malignancies. The drug gained US Food and Drug Administration (FDA) approval in 2008 and European Medicines Agency approval in 2010. Bendamustine is also on the World Health Organization list of essential medicines (4).

There are several excellent reviews regarding the use of bendamustine in hematological and solid malignancies (2,5-8). Since the understanding of its role in hematology has changed considerably in the past ten years, the present review focused on more recent clinical trials investigating the efficacy and safety of bendamustine in comparison or in combination with novel, targeted agents. Additionally, more recent studies were reviewed regarding its pharmacology, and finally, some light was shed on bendamustine-mediated immunological effects, proving to be of great importance in the current COVID-19 pandemic.

2. Bendamustine-mechanism of action, cell death and cell cycle

Cytotoxic effects of bendamustine primarily result from alkylation-mediated DNA damage and possibly to a lesser extent from antimetabolite properties of its benzimidazole ring. Bendamustine is a bifunctional alkylating agent containing two reactive groups that can bond to separate DNA sites, a feature characteristic of other nitrogen mustards-related agents such as cyclophosphamide, chlorambucil and melphalan (9). Monofunctional alkylating agents, on the other hand, contain a single active chemical moiety that is able to modify only a single DNA site (9). In comparison with other alkylating drugs, bendamustine shows a unique cytotoxicity profile such as improved penetration and localization within DNA, and more DNA double-strand breaks that persist longer (3). Upon alkylation, DNA damage induces DNA damage repair signaling pathways (9). In contrast to other alkylating agents that induce a repair of DNA by O-6-methylguanine-DNA methyltransferase and base excision repair (BER), bendamustine appears to preferably induce BER DNA repair (10). This mechanism could additionally be an asset to its unique effects since this type of repair is more complex, takes longer to perform and therefore further diminishes the capacity of cells to repair the damage (3). Except for the mere addition of the alkyl group, another mechanism of bendamustine cytotoxic properties is cross-linking of DNA, creating links within strands (intrastrand cross-linking) and between strands (interstrand cross-linking), the latter caused by formation of covalent bonds of the electrophilic alkyl group of bendamustine with electron-rich nucleophilic moieties (3,8,9,11). If the cell is unable to repair DNA damage, cell cycle progression is inhibited and cell death via apoptotic mechanism occurs.

It has still not been fully elucidated to what extent may the uniqueness of bendamustine-mediated effects be related to its benzimidazole ring. Leoni hypothesized that two mechanisms of action could be responsible for it (12). First, a direct antimetabolite activity of bendamustine could be exerted by its incorporation into newly synthesized DNA molecules or by inhibiting ribonucleotide reductase or other enzymes involved in the generation of deoxynucleoside triphosphates. Second, the benzimidazole ring may enhance the alkylating activity of bendamustine, possibly by facilitating nuclear transport and allowing the drug to reach higher concentrations in the nucleus or by inhibiting DNA repair (12). In mantle-cell lymphoma (MCL), diffuse large B-cell lymphoma (DLBCL), Burkitt lymphoma and multiple myeloma (MM) cell lines, bendamustine showed synergistic effects with pyrimidine

analogues and elicited DNA damage response and subsequent apoptosis faster and with a shorter exposure time than other alkylating agents examined (13). Although bendamustine cellular uptake is debatable, Hiraoka *et al* (13) reported that it is at least partly mediated through nucleoside transporters, suggesting its purine analogue-like properties. On the other hand, Arimany-Nardi *et al* (14) found no interaction of bendamustine with hCNT and hENT proteins, known to mediate the uptake of purine and pyrimidine drug analogs, suggesting a lack of their role in cellular uptake of the drug and emphasizing the importance of human organic cation transporter 1. The role of organic transporters was further corroborated by the finding that renal human organic anion transporter 3 increases the susceptibility of lymphoma cells to bendamustine uptake (15).

Schwänen *et al* (16) first reported *in vitro* efficacy of bendamustine alone or in combination with fludarabine in inducing apoptosis in B-cell chronic lymphocytic leukemia (CLL) cells (16). Notably, apoptosis is not the only mechanism of bendamustine-mediated cytotoxic effects, since it causes an alternative mechanism called mitotic catastrophe that bypasses apoptosis which is often impaired in tumor cells (11). Normally, when DNA damage or DNA replication stress occurs, these changes are detected by check points that arrest the cell cycle at either the G1-S (G1 check point) or the G2-M (G2 check point) transition to prevent the accumulation and propagation of genetic errors during cell division and to allow DNA repair to take place (9,17). Occurrence of damaged DNA in form of double-strand breaks (DSBs) triggers ataxia telangiectasia-mutated (ATM) check point protein kinase and downstream targets, protein kinase called checkpoint kinase (Chk)2 and transcription factor p53, most important for prevention of cells to enter S phase (9,17). Due to the repair mechanism of DSBs or due to replication stress which arises during S phase, single-strand breaks are generated and ataxia telangiectasia and Rad3-related protein and Chk1 signaling pathways are activated. If no DNA repair is achieved, cells do not enter mitosis but undergo apoptotic cell death or senescence, often by TP53-dependent mechanisms (9,17). In the case of mitotic catastrophe, if for example, TP53 is mutated, there is an insufficient G2 check point regulation and cells enter mitosis with significant DNA damage followed by apoptosis, necrosis and senescence (17,18). Gene expression profiling studies conducted by Leoni *et al* (10) demonstrated bendamustine-mediated inhibition of expression of genes involved in DNA repair and mitotic checkpoints indicating that the assumed intercalation of the drug into the DNA and downregulation of check point inhibitors could be the mechanism behind mitotic catastrophe.

Different experimental models have shown different effects of bendamustine on the cell cycle. The drug causes significantly more T-cell lymphoma cells to be arrested in the S-phase than chlorambucil or phosphoramide (10), and similar effect was observed in both MM (19,20) and MCL cell lines (21). However, in different experimental models, bendamustine induced ATM-Chk2-Cdc2-mediated arrest in G2 phase of the cell cycle of MM cells and p53-mediated apoptosis, the latter augmented by inhibition of p38 MAPK (22). In human DLBCL cell lines, the drug increased the proportion of cells in G2-M and bendamustine-induced activation of the
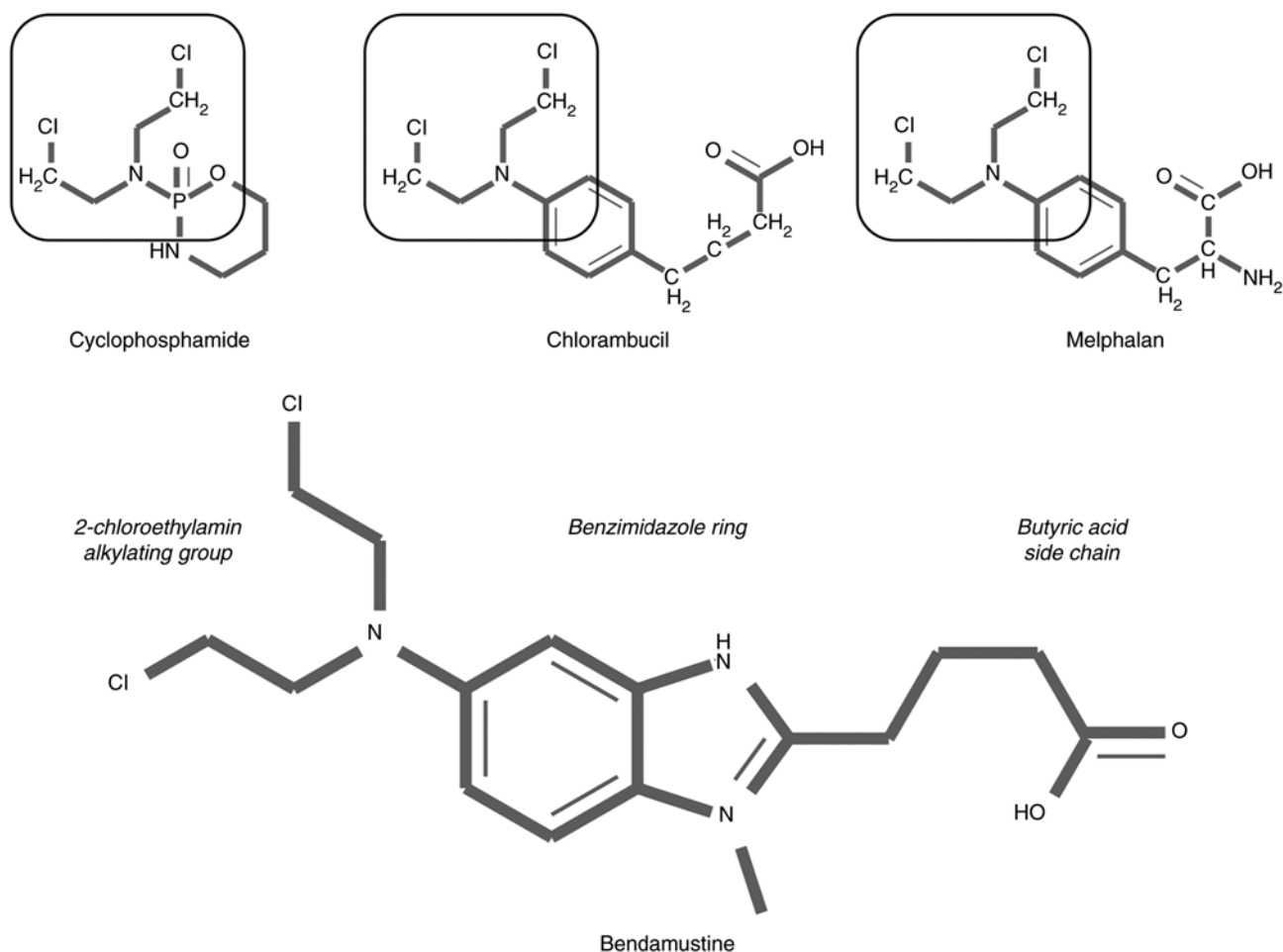
Figure 1. Chemical structure of bendamustine and similarities with other alkylating drugs.

ATM pathway and accumulation of surviving cells at G2-M phase was inhibited by surviving suppressant (23). An explanation of these findings may come from studies on HeLa cells suggesting a dose-dependent effect on cell cycle checkpoints and DNA repair (24). Low concentrations of bendamustine transiently arrested cells in G2, which then entered mitosis and divided normally, while a 4-fold higher concentration arrested cells in S phase resulting in aberrant mitosis and cell death (24). Proposed mechanism of action of bendamustine is shown in Fig. 2.

3. Bendamustine-pharmacokinetics

Pharmacokinetics of bendamustine has been studied, in addition to humans, in mice, rats and dogs. The extent of binding and formation of metabolites is different among species, but while some authors considered this to be clinically relevant (5), others suggested that the few new metabolic products detected in the human mass balance study that had not been observed in rats, largely represent adducts that are formed by reaction of bendamustine with endogenous compounds in the urine and conclude that the metabolic elimination of bendamustine is qualitatively the same in humans and rats (25,26). Although the pharmacokinetics of multiple-dose administration of the drug have not been investigated, there is a significant correlation

between nausea and maximum drug concentration (Cmax) observed in population pharmacokinetic (6,8,25). Cmax of bendamustine depends on the dose. When administered at doses of 30-200 mg/m^2, Cmax varies between 0.1-30 μg/ml and is reached after the mean time of 29.6 min (5,8). Central volume of distribution following intravenous (IV) administration is 8.6-19.3 l and steady-state volume of distribution is 15.8-20.5 l (5,8). In a previous study in which bendamustine, at a dose of 75 mg/m^2 for two days, was a component of IV administered salvage R-B(O)AD (rituximab, bendamustine, vincristine, cytarabine, dexamethasone) regimen for the treatment of primary central nervous system lymphoma, the Cmax mean for plasma and cerebrospinal fluid were 2,669 and 0.397 ng/ml, respectively, and patients with response at deep tumor sites displayed higher trends in peak exposure (27).

After IV administration, high percentage (>95%) of the drug is bound to proteins, mainly albumin, and is unlikely to displace or be displaced by other highly protein-binding drugs (5). Bendamustine is mainly nonenzymatically hydrolyzed to the markedly less active metabolites HP1 and HP2, and metabolized in the liver to active M3 and M4 via CYP1A2 (25). Since their concentrations in the plasma are significantly lower than those of the parent drug, the main therapeutic effect is due to bendamustine itself (25). Opinions on the importance of renal excretion of the drug,
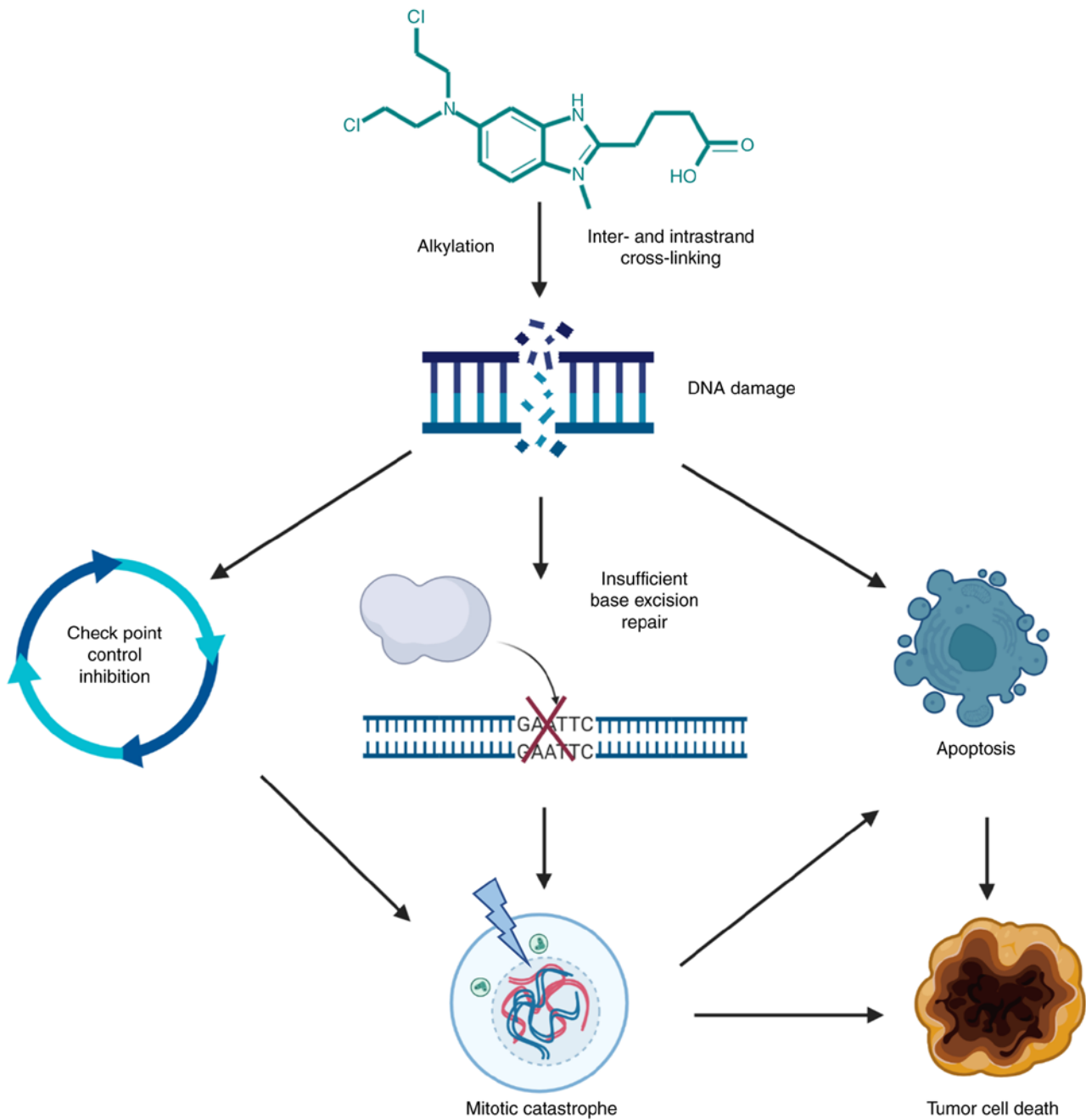
Figure 2. Potential mechanism of action of bendamustine. The figure was created with BioRender.com.

and therefore its safety in patients with renal failure, vary. Bendamustine is eliminated by the kidneys with mean elimination half-life of 28.2 min and mean total clearance of 639.4 ml/min (8). Peak metabolite concentrations are found in the urine 1 h after administration, but only 3% of the drug is excreted through kidneys unmetabolized (25,26). Preclinical and the human mass balance study demonstrated 76-90% recovery of radiolabeled bendamustine in the excreta, with varying proportions being recovered in urine (20-50%) and in feces (25-70%) (8,25,26). Based on these data, Dubbelman *et al* (26) suggested that, in contrast to the position of the majority of authors and the official FDA prescribing information (28), renal or hepatic impairment would not be expected to have an important effect on the systemic exposure to bendamustine (25,26).

4. Bendamustine in hematological and solid malignancies

The use of bendamustine in hematological malignancies frequently deviates from its official labels, both in terms of indications as well as dosage. Regarding the latter, doses higher than 90 mg/m^2 for 2 consecutive days are rarely used, and are frequently reduced to 70 mg/m^2 in pretreated or unfit patients. A detailed list of all potential clinical uses is beyond the scope of the present review. Main indications for bendamustine are CLL, indolent non-Hodgkin lymphomas (iNHL) and MCL. Other potential indications include MM, DLBCL and Hodgkin lymphoma (HL) as well as lymphodepletion prior to chimeric antigen receptor T-cell (CAR-T) infusion. Results of major studies are presented in Table I. The use of bendamustine has lately dwindled, due to the appearance of more effective and

Table I. Bendamustine in hematological malignancies.

Clinical trial	Disease (status)	Objective	Phase	N	PFS	OS	(Refs.)
	CLL (TN)	CLB vs. B	III	319	Median PFS 8.8 months vs. 21.2 months	Median OS 78.8 months vs. NR	Knauf et al (29)
NCT00769522 CLL10	CLL (TN)	FCR vs. BR	III	561	Median PFS 55.2 months vs. 41.7 months	91 vs. 92% (3 years)	Eichhorst et al (30)
NCT01886872 Alliance	CLL (TN)	BR vs. ibrutinib vs. R + ibrutinib	III	547	74 vs. 87 vs. 88% (2 years)	95 vs. 90 vs. 94% (2 years)	Woyach et al (31)
NCT03336333 SEQUOIA	CLL/SLL (TN)	Zanubrutinib vs. BR	III	479	85.5 vs. 69.5% (2 years)	94.3 vs. 94.6% (2 years)	Tam CS et al: Blood 138: 396, 2021
NCT02970318 ASCEND	CLL (r/r)	BR/IdR vs. acalabrutinib	III	310	Median PFS 16.5 months vs. NR	Median OS NR 91% vs. 94% (1 year)	Ghia et al (32)
NCT01611090 HELIOS	CLL (r/r)	Ibrutinib + BR vs. BR	III	578	Median PFS 65.1 months vs. 14.3 months	Median OS NR 75.7 vs. 61.2 % (5 years)	Fraser et al (33)
NCT02005471 MURANO	CLL (r/r)	VenR vs. BR	III	389	Median PFS NR vs. 17 months	Median OS NR 91.9 vs. 86.6% (2 years)	Seymour et al (34)
NCT01332968 GALLIUM	FL (TN)	G + CHOP/CVP/B vs. R + CHOP/CVP/B	III	1202	80.0 vs. 73.3% (3 years) 82 vs. 75% (3 years) 70.5 vs. 63.2% (5 years)	94 vs. 92.1% (3 years) 94 vs. 92.1% (3 years) 90.2 vs. 89.4% (5 years)	Marcus et al (35) Hiddemann et al (36) Townsend W et al: J Clin Oncol 38: 8023, 2020
NCT00991211 StiL NHL 1-2003	iNHL, MCL (TN)	BR vs. R-CHOP	III	514	Median PFS 69.5 months vs. 31.2 months	Median OS NR 84 vs. 82%	Rummel et al (37)
NCT00877006 BRIGHT	iNHL, MCL (TN)	BR vs. R-CHOP/R-CVP	III	447	65.5 vs. 55.8% (5 years)	81.7 vs. 85% (5 years)	Flinn et al (38)
NCT01059630 GADOLIN	iNHL (R-r)	BG vs. B	III	396	Median PFS NR vs. 14.9 months	Median OS NE events 18 vs. 20%	Sehn et al (39)
				413	Median PFS 25.8 months vs. 14.1 months	Median OS NE events 25.5 vs. 34.9%	Cheson et al (40)
NCT01456351 StiL NHL 2-2003	iNHL, MCL (relapsed)	BR vs. FR	III	230	Median PFS 34.2 months vs. 11.7 months	Median OS 109.7 months vs. 49.1 months	Rummel et al (41)
NCT01662050	MCL (TN)	R-BAC	retrospective cohort study	57	76% (35 months)	86% (2 years)	Visco et al (42)
	MCL (r/r)	R-BAC	II	36	Median PFS 10.1 months	Median OS 12.5 months	McCulloch et al (43)
NCT01412879 S1106	MCL (TN)	RH vs. BR	II	52	62 vs. 66% (5 years)	74 vs. 80% (5 years)	Kamdar et al (44)
NCT01234467	DLBCL (TN)	BR	II	23	Median PFS 5.4 months	Median OS 10.2 months	Park et al (45)
NCT01626352	DLBCL (TN)	OB	II	21	Median PFS 8.6 months	Median OS 12. months	Flinn et al (46)

Table I. Continued.

Clinical trial	Disease (status)	Objective	Phase	N	PFS	OS	(Refs.)
NCT02257567	DLBCL (r/r)	Pola-BR vs. BR	Ib/II	192	Median PFS 9.2 months vs. 3.7 months	Median OS 12.4 months vs. 4.7 months	Sehn et al (47)
NCT01657331	HL (r/r)	BV + B	I/II	65	Median PFS 7.5 months (phase II)	Median OS 43.3 months (phase I) and NR (phase II)	O'Connor et al (48)
NCT02499627	HL (r/r)	BV + B	II	40	67.3% (3 years)	88.1% (3 years)	Broccoli et al (49)
NCT01874054	HL (r/r)	BV + B	I/II	55	60.3% (3 years)	92% (3 years)	LaCasce et al (50)
LYSA	MCL (TN)	BeEAM vs. BEAM (Conditioning prior to ASCT)	Multicenter retrospective	168	84 vs. 63% (3 years)	93 vs. 84% (3 years)	Hueso et al (51)
	LBCL	FC vs. B (Lympho-depletion)	Multicenter retrospective	113	22 vs. 27% (1 year)	41 vs. 49% (2 years)	Ghilardi G et al: Blood 138: 1438-1440, 2021
NCT00916058	MM (TN+r/r)	B + melphalan	II	35	Median PFS 47 months	Median OS NR	Gomez-Arteaga et al (52)
NCT02095834	MM (r/r)	B + Car + D	I	17	Median PFS 11.1 months	Median OS 56.3 months	Lee et al (53)

PFS, progression-free survival; OS, overall survival; ASCT, autologous stem-cell transplantation; B, bendamustine; BEAM, carmustine, etoposide, cytarabine, and melphalan; BeEAM, bendamustine, etoposide, cytarabine, and melphalan; BG, bendamustine plus obinutuzumab; BR, bendamustine plus rituximab; BV, brentuximab vedotin; Car, carfilzomib; cHL, classical Hodgkin lymphoma; CHOP, cyclophosphamide, doxorubicin, vincristine, and prednisone; CLB, chlorambucil; CLL, chronic lymphocytic leukemia; CVP, cyclophosphamide, vincristine, and prednisone; D, dexamethasone; DLBCL, diffuse large B-cell lymphoma; FC, fludarabine plus cyclophosphamide; FCR, fludarabine, cyclophosphamide and rituximab; FL, follicular lymphoma; FR, fludarabine plus rituximab; G, obinutuzumab; HL, Hodgkin lymphoma; IdR, idelalisib plus rituximab; iNHL, indolent non-Hodgkin lymphoma; LBCL, large B-cell lymphoma; MCL, mantle-cell lymphoma; MM, multiple myeloma; NE, not estimated; NR, not reached; OB, ofatumumab plus bendamustine; Pola-BR, polatuzumab vedotin plus bendamustine and rituximab; R-BAC, rituximab plus bendamustine, and low dose cytarabine; R, rituximab; RH, R-hyper-CVAD (rituximab plus hyperfractionated cyclophosphamide, vincristine, doxorubicin and dexamethasone, alternating with high dose cytarabine and methotrexate); R-r, rituximab-refractory; r/r, relapsed/refractory; SLL, small lymphocytic lymphoma; TN, treatment-naïve; VenR, venetoclax plus rituximab.

less toxic agents, but also due to an apparent increased risk of lethal outcome of COVID-19 in bendamustine-treated patients (as seen below).

Bendamustine was approved for front-line treatment of CLL at a dose of 100 mg/m^2 on days 1 and 2 every 4 weeks for six cycles, after showing superiority to chlorambucil monotherapy (29). A study of the German CLL Group, comparing bendamustine plus rituximab (BR) to fludarabine, cyclophosphamide and rituximab (FCR) showed that the former resulted in decreased progression-free survival (PFS), while overall survival (OS) remained similar (30). However, in elderly patients PFS was not different between treatment groups, and BR caused significantly fewer infectious complications (30). This rendered BR the treatment of choice in elderly patients with CLL until the appearance of targeted agents. All three approved Bruton's tyrosine kinase inhibitors (BTKi), ibrutinib, acalabrutinib and zanubrutinib, have been shown in randomized trials to improve PFS, but notably not OS in comparison with BR (31,32) (Table I). Ibrutinib + BR demonstrated that both PFS and OS benefit over BR alone (33,54,55). In patients with relapsed or refractory (r/r) CLL, the combination of rituximab with venetoclax, a BCL2 inhibitor, was shown to be superior to BR (34). Thus at present, bendamustine is only rarely used in CLL; most patients receive targeted agents, BTKi and venetoclax, and frail elderly chlorambucil with or without anti-CD20 monoclonal antibodies (MoAbs).

Bendamustine, first alone and later in combination with anti-CD20 MoAbs has been widely investigated for treatment of iNHL, including follicular lymphoma (FL), marginal-zone lymphoma (MZL), and lymphoplasmacytic lymphoma/Waldenstrom macroglobulinemia (LPL/WM) (2,56). StiL was the most important randomized trial proving the efficacy of this drug in front-line treatment of iNHL (37). Patients with FL, MZL, MCL and LPL treated with BR had significantly longer PFS compared with those treated with rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP) (median PFS 69.5 months vs. 31.2 months) (37). These findings were corroborated by the BRIGHT study (38,57). In patients with relapsed iNHL, BR was revealed to improve PFS and OS in comparison with fludarabine plus rituximab (41). In patients refractory to rituximab, the combination of bendamustine and obinutuzumab, an alternative anti-CD20 MoAb, was identified to be superior to bendamustine monotherapy (39,40). Thus, bendamustine became the treatment of choice for front-line treatment of iNHL as well as for patients who relapsed after R-CHOP or rituximab plus cyclophosphamide, vincristine, and prednisone (R-CVP). This position was recently challenged when some real-life series and the GALLIUM suggested an increase in late infection-related mortality in vulnerable patients treated with bendamustine (35,36,58-60). Thus currently, numerous centers that used to treat iNHL patients with bendamustine only 2 or 3 years ago are switching back to CHOP/CVP or even, particularly in the USA, to lenalidomide.

MCL is a type of B-NHL occurring preferentially in elderly males and combining the aggressive clinical behavior of large B-cell lymphoma with the continuous tendency for relapses of iNHL. Young fit patients, typically treated with rituximab, a high-dose cytarabine containing regimen (for instance dexamethasone, high-dose cytarabine and cisplatin; DHAP),

alone or alternating with R-CHOP (R-CHOP/R-DHAP), are autografted in 1st remission and then continue with rituximab maintenance. Bendamustine is one of the most effective cytotoxic agents for MCL. Randomized studies have shown BR to be superior to R-CHOP and R-CVP (37,38,57). The American S1106 study compared BR induction with R-hyper-CVAD (RH, rituximab plus hyperfractionated cyclophosphamide, vincristine, doxorubicin and dexamethasone, alternating with high dose cytarabine and methotrexate) in patients with previously untreated MCL and who were candidates for autologous stem-cell transplantation (ASCT) (44,61). Both regimens showed excellent antitumor activity, but BR was far less toxic and had less stem cell mobilization failure rates (44). Italian studies with the combination of rituximab, bendamustine and high-dose cytarabine produced excellent results with long-term PFS even without rituximab maintenance (42,43). Real-life series have confirmed the superiority of bendamustine-based treatment in MCL over CHOP-like regimens (62,63). BTKi, while very active in MCL, are only approved for relapsed disease. Therefore, even in the current situation, bendamustine-based regimens remain the mainstay of MCL treatment, at least for patients unfit for aggressive chemotherapy, such as DHAP. Notably, there are previous studies suggesting that bendamustine-based regimens are less sensitive to p53 mutations (3,64,65), which are a negative prognostic factor in patients treated with R-CHOP/R-DHAP (66). A possible explanation for this observation may be the propensity of bendamustine to eliminate cells via the mitotic catastrophe mechanism.

BR is active in DLBCL (45), but the duration of remission is short and is mainly used for palliation. Additionally, modest efficacy was shown when bendamustine was combined with ofatumumab, another anti-CD20 MoAb (46). Recently, the combination of polatuzumab vedotin, a conjugated anti-CD79b MoAb and BR was shown to significantly improve outcomes in patients with r/r DLBCL unsuitable for intensive chemotherapy and ASCT (47). This combination, Pola-BR, has been registered and approved for this indication in both USA (67), and EU (68) and is the current treatment of choice for this group of patients. Bendamustine is active in HL with median PFS 5.2 months, but again the duration of remission is short (69). The combination of bendamustine with brentuximab vedotin, an anti-CD30 conjugated MoAb has been shown to be an effective salvage and is occasionally used instead of more aggressive chemotherapy regimens prior to stem cell transplantation (48-50). Bendamustine is also used instead of carmustine in combination with etoposide, cytarabine and melphalan for conditioning prior to ASCT (51), and as an alternative lymphodepleting therapy prior to the infusion of tisagenlecleucel, one of the registered CAR-T cell products (Ghilardi G et al: Blood 138: 1438-1440, 2021). Although active in MM (52,53), its use has all but disappeared even before the current COVID-19 pandemic, due to appearance of other, more effective and less toxic agents: anti-CD38 monoclonal antibodies, proteasome inhibitors and immunomodulators. Still, an occasional patient with MM, failing these options or unsuitable for them, may benefit from bendamustine.

There are several recent studies of bendamustine effects in treating certain solid tumors (70-73). In pretreated women with HER2-negative metastatic breast cancer in combination with

capecitabine OR was 46% and median PFS was 7.5 months (70). In a phase II study of relapsed chemotherapy sensitive or resistant small-cell lung cancer, median time to progression was 4.0 months, median OS was 4.8 months, and the response rate was 26% (71). In patients with refractory soft tissue sarcoma who were treated with bendamustine, the estimated 3-month and 6-month PFS for all histological subtypes was 35.3 and 23.5%, respectively (72). In an open trial including women with advanced ovarian cancer last updates posted in 2018 reported on median OS of 393 days (73).

5. Bendamustine related side-effects in clinical trials

Patients with cancer frequently experience problems which can be caused by their disease, antineoplastic treatment, or other, unrelated causes. This should be considered when interpreting published data. Detailed data on frequency of most important side-effects are presented in Table II.

In patients treated with bendamustine monotherapy, most frequently reported treatment-emergent adverse events (AE) were fever, skin reactions, nausea, vomiting and hematological AEs: granulocytopenia and thrombocytopenia (74-76) Fatigue, mucositis and infections were very common, but were rarely higher than grade 2, at least during and immediately after treatment (74-76).

Toxicity reports on studies comparing bendamustine-MoAb combinations with CHOP-MoAb combinations are not completely consistent. Universally, bendamustine was associated with a very low rate of alopecia and peripheral sensory neuropathy and higher incidence of skin reactions and gastrointestinal problems (6,8,37,57). Granulocytopenia and thrombocytopenia as well as infections during treatment and neutropenic fever were usually more frequent and severe with CHOP (37). Lymphopenia, a frequently disregarded side-effect, universally occurs more frequently with bendamustine (57).

Toxicities of BR and fludarabine + rituximab were similar, most commonly myelosuppression and infections (41). As already mentioned, FCR was more toxic than BR. There were more cases of severe neutropenia and infections (84 vs. 59% and 39 vs. 25%, respectively) (30). The increased frequency of infectious complications with FCR was, as aforementioned, more pronounced in patients older than 65 years (30).

The combination of venetoclax plus rituximab was associated with more AEs of grade 3 or 4 in comparison with BR in the MURANO study (82.0 vs. 70.2%) (34). Particularly grade 3-4 neutropenia was more frequent in the former arm (57.7 vs. 38.8%) (34). On the other hand, febrile neutropenia and grade 3 or 4 infections or infestations were more common in the BR group (34).

In the GALLIUM study, treatment with bendamustine was associated with marked and prolonged reductions in T-cell counts and higher rates of grade 3 to 5 infection and second neoplasm during the maintenance and follow-up phases, whereas CHOP was associated with higher rates of grade 3 to 5 neutropenia during the induction phase (35,36). Non-relapse-related fatal AEs, although with small absolute numbers, were more common among patients who received bendamustine (5.6% of patients in the obinutuzumab group and 4.4% of those in the rituximab group) than among those treated with CHOP (1.6 and 2.0%, respectively) or CVP (1.6 and

1.8%), and pose a concern in this population of patients (35). The frequency of deaths was higher in patients treated with bendamustine (10%) than in patients treated with CHOP (5%) or CVP (8%) (36). Furthermore, patients who had not previously started new anticancer treatment had higher proportion of fatal AEs when treated with bendamustine (4%) than with CHOP (2%) or CVP (2%) (36).

These data suggested that, while the acute toxicities of bendamustine are less prominent than that of CHOP, the drug seems to have a prolonged effect, probably immunological, leading to an increased risk of late infections, particularly in patients receiving prolonged anti-CD20 MoAb maintenance.

6. Immunological effects of bendamustine

A recent detailed review by Stokes *et al* (77) reported bendamustine effects in both murine models and clinical setting as pre-transplant conditioning drug and its immunomodulatory properties in graft-versus-host disease (GVHD). In the present review, general immunological effects of bendamustine not necessarily restricted to GVHD are discussed.

Hematological malignancies are known to have a direct effect on the immunological system; in patients with CLL there is an impaired production of polyclonal immunoglobulins due to anomalous CD40-CD40 ligand relations and reduction of CD40 ligand, suppression of $CD95^+$ plasma cells in the bone marrow, and impaired inhibition by T cells (78). Furthermore, in those patients the numbers of T helper cells are reduced with augmented number of T suppressor cells; a CD200 marker expressed on CLL cells stimulates differentiation of $CD4^+$ T cells into regulatory T cells (Tregs), which express CTLA-4, CD270 and PD-L1. NK-cell activity, phagocytosis and complement amounts have been also reported to be impaired and all these facts greatly contribute to insufficient immunological response to infectious stimuli in patients with CLL (78). Although no differences on the rate of infections between bendamustine and other alkylating agents or fludarabine were reported in a meta-analysis of randomized controlled trials by Gafter-Gvili *et al* (79), bendamustine is associated with an increased risk of bacterial infections and opportunistic infections, including cytomegalovirus, varicella zoster virus, histoplasmosis and *Pneumocystis jirovecii*-caused pneumonia (59). Furthermore, a recent retrospective multicenter cohort study by Lamure *et al* (80) on the determinants of outcome in COVID-19-hospitalized patients with lymphoma, reported on bendamustine-associated death in patients with relapsed/refractory lymphoma. A similar effect has been observed in our patients (Aurer I *et al*: Blood 138: 3553, 2021).

Bendamustine-induced lymphopenia, whether as monotherapy or in combination, has been widely reported in both hematological and non-hematological malignancies (37,45,57,70,75,81-90). Lymphopenia ranged from 5% in rituximab-refractory patients with iNHL (85) to 75% of patients with grade 3-4 hematological toxicity receiving BR (37,90) or even to 91% in patients treated for triple negative breast cancer (89). The latter group was characterized with pronounced decline in $CD4^+$ cells, with 86% having grade 4 depressed $CD4^+$ counts ($<50/\mu l$) (89). In FL patients treated with bendamustine, marked reductions in $CD3^+$ and $CD3^+CD4^+$ T cells were seen during induction

Table II. Bendamustine-related side-effects in clinical trials.

Clinical trial	Disease (status)	Objective	Phase	N	Bendamustine dose	Side-effects (% of patients)	(Refs.)
	iNHL (R-r)	B	II	76	120 mg/m² IV on days 1 and 2 of six 3-week cycles	Grade 3-4 neutropenia (54%), thrombocytopenia (25%), and anemia (12%). All grade nausea (72%), vomiting (41%), fatigue (49%), constipation (26%), anorexia (34%), fever (25%), cough (29%) and diarrhea (30%).	Friedberg et al (74)
	CLL (TN)	CLB vs. B	III	319	100 mg/m² on days 1 to 2 of six 4-week cycles	Grade 3-4 neutropenia (10.6 vs. 23%), thrombocytopenia (7.9 vs. 11.8%), and anemia (0 vs. 2.5%). All grade nausea (19.3 vs. 13.9%), vomiting (15.5 vs. 6.6%), and diarrhea (9.9 vs. 4%). Grade 3-4 infections (3 vs. 8%).	Knauf et al (75)
NCT00769522 CLL10	CLL (TN)	FCR vs. BR	III	561	90 mg/m² IV on days 1 and 2 of six 4-week cycles	Grade 3-4 hematologic AEs (90 vs. 67%)-neutropenia (85 vs.59%), leukopenia (81 vs. 48%), thrombocytopenia (21 vs. 14%), anemia (14 vs. 11%). Grade 3-4 infections (39 vs. 25%).	Eichhorst et al (30)
NCT02005471 MURANO	CLL (r/r)	VenR vs. BR	III	389	70 mg/m² IV on days 1 and 2 of six 4-week cycles	Grade 3-4 AEs (82 vs. 70.2%). Grade 3-4 neutropenia (57.7 vs. 38.8%), anemia (10.8 vs. 13.8%), thrombocytopenia (5.7 vs. 10.1%), and febrile neutropenia (3.6 vs. 9.6%). Grade 3-4 infections and infestations (17.5 vs. 21.8%)	Seymour et al (34)
NCT01332968 GALLIUM	FL (TN)	G + CHOP/CVP/B vs. R + CHOP/ CVP/B	III	1202	90 mg/m² IV on days 1 and 2 of six 4-week cycles	G-group (CHOP/CVP/B)-grade 3-5 AEs (89%/69%/69%), grade 3-5 neutropenia (71%/46%/30%), grade 3-5 infections (12%/13%/20%), fatal AEs (2%/2%/6%) R-group (CHOP/CVP/B)-grade 3-5 AEs (74%/54%/67%), grade 3-5 neutropenia (55%/23%/30%), grade 3-5 infections (12%/13%/26%), fatal AEs (2%/2%/5%)	Hiddemann et al (36)
NCT00991211 StiL NHL 1-2003	iNHL, MCL (TN)	BR vs. R-CHOP	III	514	90 mg/m² IV over 30-60 min on days 1 and 2 of six 4-week cycles	Hematological AEs (30 vs. 68%). Grade 3-4 leukopenia (37 vs. 72%) and neutropenia (29 vs. 69%). All grade alopecia (0 vs. 100%), infections (37 vs. 50%), peripheral neuropathy (7 vs. 29%), stomatitis (6 vs. 19%), erythematous skin reactions (16 vs. 9%).	Rummel et al (37)

Table II. Continued.

Clinical trial	Disease (status)	Objective	Phase	N	Bendamustine dose	Side-effects (% of patients)	(Refs.)
NCT00877006 BRIGHT	iNHL, MCL (TN)	BR *vs.* R-CHOP/R-CVP	III	447	90 mg/m^2 IV on days 1 and 2 of six 4-week cycles	Grade 3-4 neutropenia (39-49% vs. 87%/56%) and lymphopenia (61-63% vs. 33%/28%). All grade vomiting (25-29% vs. 13%/13%), drug-hypersensitivity (13-17% vs. 6%/3%), peripheral neuropathy/paresthesia (9-14% vs. 44%/47%), and alopecia (3-4% vs. 51%/21%). Grade 3-4 infections (7-12% vs. 5%/7%)	Flinn *et al* (38)
NCT01456351 StiL NHL 2-2003	iNHL, MCL (re-lapsed)	BR *vs.* FR	III	230	90 mg/m^2 IV on days 1 and 2 of six 4-week cycles	Grade 3-4 leukopenia (13 vs. 12%), neutropenia. (9 vs. 9%), anemia (1 vs. 1%), and thrombocytopenia (2 vs. 2%). All grade infections (30 vs. 27%)	Rummel *et al* (41)
NCT01412879 S1106	MCL (TN)	RH *vs.* BR	II	52	90 mg/m^2 IV on days 1 and 2 of six 4-week cycles	Grade 3-4 anemia (59 vs. 8.6%), neutropenia (65 vs. 34%), febrile neutropenia (29 vs. 14%), and thrombocytopenia (71 vs. 17%).	Chen *et al* (61)

AEs, adverse events; B, bendamustine; BR, bendamustine plus rituximab; CHOP, cyclophosphamide, doxorubicin, vincristine, and prednisone; CLB, chlorambucil; CLL, chronic lymphocytic leukemia; CVP, cyclophosphamide, vincristine, and prednisone; FCR, fludarabine, cyclophosphamide and rituximab; FL, follicular lymphoma; FR, fludarabine plus rituximab; G, obinutuzumab; iNHL, indolent non-Hodgkin lymphoma; MCL, mantle-cell lymphoma; R, rituximab; RH, R-hyper-CVAD (rituximab plus hyperfractionated cyclophosphamide, vincristine, doxorubicin and dexamethasone, alternating with high dose cytarabine and methotrexate); R-r, rituximab-refractory; r/r, relapsed/refractory; TN, treatment-naive; VenR, venetoclax plus rituximab.
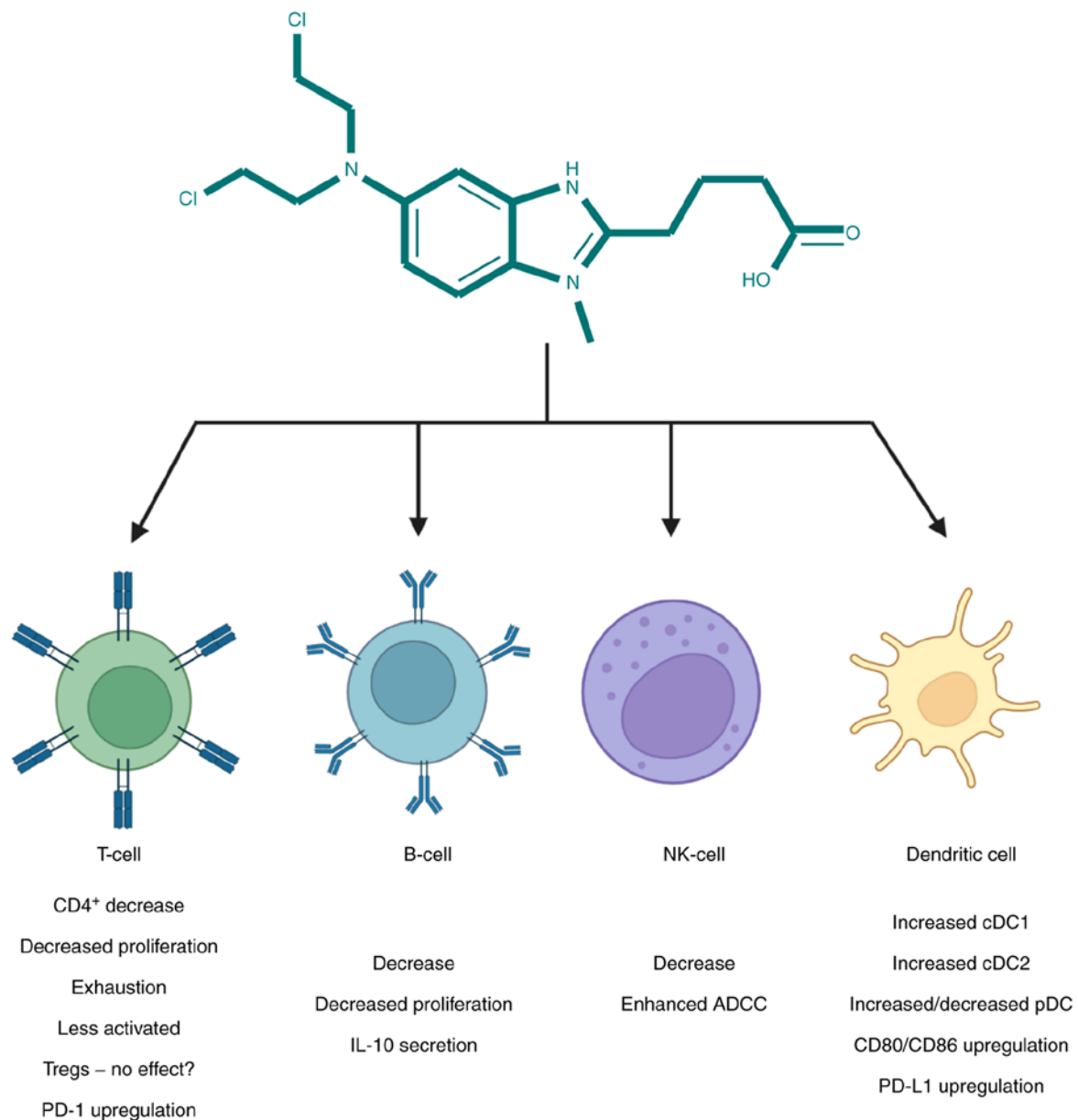
Figure 3. Bendamustine-mediated effects on immune cells. The figure was created with BioRender.com. Tregs, regulatory T cells; cDCs, conventional dendritic cells; ADCC, antibody-dependent cell cytotoxicity; pDC, plasmacytoid dendritic cells.

with prolonged recovery during and after maintenance (36). Prolonged lymphopenia and low CD4^{+} T-cell counts, for at least 7-9 months were also observed in relapsed or refractory patients with iNHL and MCL (83). Recent population-based analysis by Martínez-Calle *et al* (91) following BR treatment in patients with low grade lymphoproliferative disease revealed that median times to lymphocyte count recovery ($\geq 1 \times 10^9$/l) and CD4^{+} recovery ($\geq 0.2 \times 10^9$/l) were 26 and 24 months, respectively, and late recovery was associated with risk of serious infection.

T-cells showed less proliferative properties when incubated with bendamustine (92). In a case report of systemic CMV infection following BR treatment, low γ/δ T-cell frequency and hyperactivated/exhausted CD4^{+} and CD8^{+} T-cell phenotypes unable to face CMV challenge was reported (93). In a major histocompatibility complex mismatched murine transplant model, combination of bendamustine with total body irradiation (BEN-TBI) showed no difference in donor Tregs, defined as CD4+CD25^{+}FoxP3^{+} and measured in peripheral blood, when compared with cyclophosphamide plus TBI, and proliferative properties of splenic Tregs did not differ between groups either (94). In the same study, *in vitro* generation of Tregs was not affected by the increasing concentrations of bendamustine, but mice treated with bendamustine had less activated donor T-cells measured by CD25 expression (94). In a study of FOXP3^{+} Tregs in patients with gastric malt lymphoma, immunohistochemistry revealed depletion of Tregs at the end of treatment, that was slightly deeper in cases treated with bendamustine or fludarabine than in those treated with antibiotics, with a continuous decline in FOXP3^{+} cells up to one year (95). Stokes *et al* (94) observed that in peripheral blood of BEN-TBI conditioned mice, treatment induced an increase in absolute numbers of Th2 cells on day +7 and decrease in numbers of Th17 on day +14 but the effects were

not detected at any other time points (94). The same group exposed murine bone marrow-derived dendritic cells (BMDC) to bendamustine and incubated them with allogenic T-cells. Upon incubation, T-cells exhibited an increase in markers of T-cell exhaustion, as well as markers of activated T-cells, ICOS and CD69, and an increase in PD-1, negative regulator of immune response, followed by allogenic CD4$^+$ cell death (96). Since PD-1 is so far considered to be crucial in T-cell exhaustion, combining bendamustine with anti-PD-1 antibody could be beneficial in both anticancer and anti-microbe setting (97). Notably, CD69 expression on CLL cells is considered to be a predictor of response to bendamustine since tumor cells derived from lymphoid tumor niches harbored higher CD69 expression and were less sensitive to bendamustine than their peripheral blood counterparts (98). The majority of the studies support findings that it is mostly CD4$^+$ T cell count that is decreased with a concomitant decrease in CD4/CD8 ratio and an insufficient T cell recovery in patients treated with bendamustine, independent of the type of malignancy involved (99), a fact most recently corroborated in a study by Yamasaki *et al* (100).

Bendamustine-mediated lymphopenia is also extended to B cells, with a previous study reporting predominant B-cell cytotoxicity (86), resulting in secondary hypogammaglobulinemia and susceptibility to infections. Nonetheless, incidence of hypogammaglobulinemia after BR is not very different from that reported in association with rituximab, suggesting rituximab- or disease-mediated causes of reduced immunoglobulin levels (75,79,86,87,91,99). Suggestive of not only diminished numbers but also altered function of B-cells are reports whereupon incubation of murine B cells with bendamustine showed less proliferation in response to LPS (92). In addition, IL-10 production by B cells among peripheral blood mononuclear cell was significantly enhanced by addition of bendamustine (101).

NK-cells are known to be impaired in both numbers and activity in hematological malignancies (78). In a study by Bremer *et al* (87), NK-cells dropped by ~60% within the first 3 weeks after bendamustine therapy. However, data of a recent study presented on ASH in 2020 showed that in a model with obinutuzumab-induced antibody-dependent cell cytotoxicity (ADCC) resistant clones, pretreatment of effector NK cells with bendamustine enhanced ADCC induction of obinutuzumab, which was followed by the increased expression of CD107, a NK-cell degranulation marker (Yamashita-Kashima Y *et al*: Blood 136: 11-12, 2020).

Due to the crucial role of dendritic cells (DC) in both pathogenesis of GVHD and graft-vs. leukemia (102), their function and phenotype have been studied in response to bendamustine. In a murine model, bendamustine + TBI increased the proportion of plasmacytoid DC, type 1 conventional DC (cDC1s), and type 2 conventional DCs (cDC2s), whereas in human monocyte-derived DCs, bendamustine treatment decreased the number of plasmacytoid DCs and increased those of cDC1 and cDC2s (96). The same study demonstrated that bendamustine-treated murine BMDC showed concentration-dependent increase in CD80, CD86, and PD-L1 expression and dampened response to lipopolysaccharide (LPS). Bendamustine immunomodulatory properties were additionally confirmed by a decrease in secretion of pro-inflammatory cytokines IL-6, TNFα, CCL5, and CCL2 by BMDC in response to LPS (96). Bendamustine effects on T, B, NK and DC are shown in Fig. 3.

Various and often insufficiently explained immunological effects induced by bendamustine can be found in a study where two cases demonstrated hypersensitivity to bendamustine but with different mechanisms: one with type I hypersensitivity reaction and another with type IVb or type IVc hypersensitivity reaction. Additionally, one patient exhibited bendamustine-induced drug fever in whom neither a type I nor a type IV hypersensitivity mechanism to this drug was demonstrated (103). Furthermore, in a recent study by Chan *et al* a case of severe bendamustine-induced autoimmune hemolytic anemia in a patient with splenic marginal zone lymphoma was reported (104).

7. Conclusion

The approval by FDA of bendamustine in 2008 was referred to as the revival of an old unjustly ignored drug, an alkylator that, by virtue of its structure, also has antimetabolite properties and therefore improved antitumor efficacy than classical alkylators without increased toxicity. At one time, it seemed to be the best cytotoxic agent for treatment of indolent lymphoproliferative disorders. Bendamustine is acutely relatively well tolerated, easier to handle than CHOP or similar regimens. It does not cause alopecia, a side-effect which physicians frequently ignore, but patients, particularly female, not infrequently find difficult to bear, is not cardiotoxic and not excreted by kidneys in a meaningful amount, thus increasing its target patient population. However, bendamustine causes deep and prolonged lymphopenia, affecting both T and B-cell lineages. The latter in the current COVID-19 pandemics, together with the appearance of more effective and less toxic targeted agents for treatment of CLL resulted in a sharp decline in its use, which remains more or less unaffected only in mantle-cell lymphoma, at least for the time being.

Experimental data suggested that bendamustine causes cytotoxicity through multiple mechanisms, some of which seem independent of p53. This, coupled with its profound effect on various lymphocyte subsets and lack of major hematotoxicity presents an opportunity to develop it further, as in the Pola-BR combination or CAR-T setting. Physicians using bendamustine must be aware of its prolonged effects and to continue to carefully and regularly monitor treated patients, in order to be able to recognize and treat late complications and thus derive the greatest benefit from this versatile and interesting drug.

Availability of data and materials

Not applicable.

Authors' contributions

HL and IA conceived the manuscript and wrote the article. HL, TS and AB performed the literature research and prepared the figures. DV and DB revised and edited the manuscript. All authors read and approved the final manuscript. Data authentication is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

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References

1. WHO collaborating centre for drug statistics methodology, ATC classification index with DDDs, Oslo, Norway, 2021.
2. Cheson BD and Rummel MJ: Bendamustine: Rebirth of an Old Drug. J Clin Oncol 27: 1492-1501, 2009.
3. Cheson BD and Leoni L: Bendamustine: Mechanism of action and clinical data. Clin Adv Hematol Oncol 9 (8 Suppl 19): S1-S11, 2011.
4. World Health Organization model list of essential medicines, 21st list, 2019. Geneva: World Health Organization, 2019.
5. Gandhi V and Burger JA: Bendamustine in B-cell malignancies: The new 46-year-old kid on the block. Clin Cancer Res 15: 7456-7461, 2009.
6. Garnock-Jones KP: Bendamustine: A review of its use in the management of indolent non-Hodgkin's lymphoma and mantle cell lymphoma. Drugs 70: 1703-1718, 2010.
7. Tageja N: Bendamustine: Safety and efficacy in the management of indolent non-hodgkins lymphoma. Clin Med Insights Oncol 5: 145-156, 2011.
8. Hoy SM: Bendamustine: A review of its use in the management of chronic lymphocytic leukaemia, rituximab-refractory indolent non-Hodgkin's lymphoma and multiple myeloma. Drugs 72: 1929-1950, 2012.
9. Fu D, Calvo JA and Samson LD: Balancing repair and tolerance of DNA damage caused by alkylating agents. Nat Rev Cancer 12: 104-120, 2012.
10. Leoni LM, Bailey B, Reifert J, Bendall HH, Zeller RW, Corbeil J, Elliott G and Niemeyer CC: Bendamustine (Treanda) displays a distinct pattern of cytotoxicity and unique mechanistic features compared with other alkylating agents. Clin Cancer Res 14: 309-317, 2008.
11. Leoni LM and Hartley JA: Mechanism of action: The unique pattern of bendamustine-induced cytotoxicity. Semin Hematol 48 (Suppl 1): S12-S23, 2011.
12. Leoni LM: Bendamustine: Rescue of an effective antineoplastic agent from the mid-twentieth century. Semin Hematol 48 (Suppl 1): S4-S11, 2011.
13. Hiraoka N, Kikuchi J, Yamauchi T, Koyama D, Wada T, Uesawa M, Akutsu M, Mori S, Nakamura Y, Ueda T, et al: Purine analog-like properties of bendamustine underlie rapid activation of DNA damage response and synergistic effects with pyrimidine analogues in lymphoid malignancies. PLoS One 9: e90675, 2014.
14. Arimany-Nardi C, Montraveta A, Lee-Vergés E, Puente XS, Koepsell H, Campo E, Colomer D and Pastor-Anglada M: Human organic cation transporter 1 (hOCT1) as a mediator of bendamustine uptake and cytotoxicity in chronic lymphocytic leukemia (CLL) cells. Pharmacogenomics J 15: 363-371, 2015.
15. Hagos Y, Hundertmark P, Shnitsar V, Marada VV, Wulf G and Burckhardt G: Renal human organic anion transporter 3 increases the susceptibility of lymphoma cells to bendamustine uptake. Am J Physiol Physiol 308: F330-F338, 2015.
16. Schwänen C, Hecker T, Hübinger G, Wölfle M, Rittgen W, Bergmann L and Karakas T: In vitro evaluation of bendamustine induced apoptosis in B-chronic lymphocytic leukemia. Leukemia 16: 2096-2105, 2002.
17. Matthews HK, Bertoli C and de Bruin RAM: Cell cycle control in cancer. Nat Rev Mol Cell Biol 23: 74-88, 2022.
18. Vitale I, Galluzzi L, Castedo M and Kroemer G: Mitotic catastrophe: A mechanism for avoiding genomic instability. Nat Rev Mol Cell Biol 12: 385-392, 2011.
19. Cai B, Lyu H, Huang J, Wang S, Lee CK, Gao C and Liu B: Combination of bendamustine and entinostat synergistically inhibits proliferation of multiple myeloma cells via induction of apoptosis and DNA damage response. Cancer Lett 335: 343-350, 2013.
20. Cai B, Wang S, Huang J, Lee CK, Gao C and Liu B: Cladribine and bendamustine exhibit inhibitory activity in dexamethasone-sensitive and -resistant multiple myeloma cells. Am J Transl Res 5: 36-46, 2013.
21. Visco C, Castegnaro S, Chieregato K, Bernardi M, Albiero E, Zanon C, Madeo D and Rodeghiero F: The cytotoxic effects of bendamustine in combination with cytarabine in mantle cell lymphoma cell lines. Blood Cells Mol Dis 48: 68-75, 2012.
22. Gaul L, Mandl-Weber S, Baumann P, Emmerich B and Schmidmaier R: Bendamustine induces G2 cell cycle arrest and apoptosis in myeloma cells: The role of ATM-Chk2-Cdc25A and ATM-p53-p21-pathways. J Cancer Res Clin Oncol 134: 245-253, 2008.
23. Kaneko N, Mitsuoka K, Amino N, Yamanaka K, Kita A, Mori M, Miyoshi S and Kuromitsu S: Combination of YM155, a survivin suppressant, with bendamustine and rituximab: A new combination therapy to treat relapsed/refractory diffuse large B-cell lymphoma. Clin Cancer Res 20: 1814-1822, 2014.
24. Beeharry N, Rattner JB, Bellacosa A, Smith MR and Yen TJ: Dose dependent effects on cell cycle checkpoints and DNA repair by bendamustine. PLoS One 7: e40342, 2012.
25. Darwish M, Bond M, Hellriegel E, Robertson P Jr and Chovan JP: Pharmacokinetic and pharmacodynamic profile of bendamustine and its metabolites. Cancer Chemother Pharmacol 75: 1143-1154, 2015.
26. Dubbelman AC, Rosing H, Darwish M, D'Andrea D, Bond M, Hellriegel E, Robertson P Jr, Beijnen JH and Schellens JH: Pharmacokinetics and excretion of 14C-bendamustine in patients with relapsed or refractory malignancy. Drugs R D 13: 17-28, 2013.
27. Kim T, Choi HY, Lee HS, Jung SH, Ahn JS, Kim HJ, Lee JJ, Yoo HD and Yang DH: Clinical response and pharmacokinetics of bendamustine as a component of salvage R-B(O)AD therapy for the treatment of primary central nervous system lymphoma (PCNSL). BMC Cancer 18: 729, 2018.
28. Cephalon Inc. Treanda (bendamustine hydrochloride for injection) for intravenous infusion: US prescribing information, 2021.
29. Knauf WU, Lissitchkov T, Aldaoud A, Liberati AM, Loscertales J, Herbrecht R, Juliusson G, Postner G, Gercheva L, Goranov S, et al: Bendamustine compared with chlorambucil in previously untreated patients with chronic lymphocytic leukaemia: Updated results of a randomized phase III trial. Br J Haematol 159: 67-77, 2012.
30. Eichhorst B, Fink AM, Bahlo J, Busch R, Kovacs G, Maurer C, Lange E, Köppler H, Kiehl M, Sökler M, et al: First-line chemoimmunotherapy with bendamustine and rituximab versus fludarabine, cyclophosphamide, and rituximab in patients with advanced chronic lymphocytic leukaemia (CLL10): An international, open-label, randomised, phase 3, non-inferiority trial. Lancet Oncol 17: 928-942, 2016.
31. Woyach JA, Ruppert AS, Heerema NA, Zhao W, Booth AM, Ding W, Bartlett NL, Brander DM, Barr PM, Rogers KA, et al: Ibrutinib regimens versus chemoimmunotherapy in older patients with untreated CLL. N Engl J Med 379: 2517-2528, 2018.
32. Ghia P, Pluta A, Wach M, Lysak D, Kozak T, Simkovic M, Kaplan P, Kraychok I, Illes A, de la Serna J, et al: ASCEND: Phase III, randomized trial of acalabrutinib versus idelalisib plus rituximab or bendamustine plus rituximab in relapsed or refractory chronic lymphocytic leukemia. J Clin Oncol 38: 2849-2861, 2020.
33. Fraser GAM, Chanan-Khan A, Demirkan F, Santucci Silva R, Grosicki S, Janssens A, Mayer J, Bartlett NL, Dilhuydy MS, Loscertales J, et al: Final 5-year findings from the phase 3 HELIOS study of ibrutinib plus bendamustine and rituximab in patients with relapsed/refractory chronic lymphocytic leukemia/small lymphocytic lymphoma. Leuk Lymphoma 61: 3188-3197, 2020.

34. Seymour JF, Kipps TJ, Eichhorst B, Hillmen P, D'Rozario J, Assouline S, Owen C, Gerecitano J, Robak T, De la Serna J, *et al*: Venetoclax-rituximab in relapsed or refractory chronic lymphocytic leukemia. N Engl J Med 378: 1107-1120, 2018.

35. Marcus R, Davies A, Ando K, Klapper W, Opat S, Owen C, Phillips E, Sangha R, Schlag R, Seymour JF, *et al*: Obinutuzumab for the first-line treatment of follicular lymphoma. N Engl J Med 377: 1331-1344, 2017.

36. Hiddemann W, Barbui AM, Canales MA, Cannell PK, Collins GP, Dürig J, Forstpointner R, Herold M, Hertzberg M, Klanova M, *et al*: Immunochemotherapy with obinutuzumab or rituximab for previously untreated follicular lymphoma in the GALLIUM study: Influence of chemotherapy on efficacy and safety. J Clin Oncol 36: 2395-2404, 2018.

37. Rummel MJ, Niederle N, Maschmeyer G, Banat GA, von Grünhagen U, Losem C, Kofahl-Krause D, Heil G, Welslau M, Balser C, *et al*: Bendamustine plus rituximab versus CHOP plus rituximab as first-line treatment for patients with indolent and mantle-cell lymphomas: An open-label, multicentre, randomised, phase 3 non-inferiority trial. Lancet 381: 1203-1210, 2013.

38. Flinn IW, van der Jagt R, Kahl B, Wood P, Hawkins T, MacDonald D, Simpson D, Kolibaba K, Issa S, Chang J, *et al*: First-line treatment of patients with indolent non-Hodgkin lymphoma or mantle-cell lymphoma with bendamustine plus rituximab versus R-CHOP or R-CVP: Results of the BRIGHT 5-year follow-up study. J Clin Oncol 37: 984-991, 2019.

39. Sehn LH, Chua N, Mayer J, Dueck G, Trněný M, Bouabdallah K, Fowler N, Delwail V, Press O, Salles G, *et al*: Obinutuzumab plus bendamustine versus bendamustine monotherapy in patients with rituximab-refractory indolent non-Hodgkin lymphoma (GADOLIN): A randomised, controlled, open-label, multicentre, phase 3 trial. Lancet Oncol 17: 1081-1093, 2016.

40. Cheson BD, Chua N, Mayer J, Dueck G, Trněný M, Bouabdallah K, Fowler N, Delwail V, Press O, Salles G, *et al*: Overall survival benefit in patients with rituximab-refractory indolent non-Hodgkin lymphoma who received obinutuzumab plus bendamustine induction and obinutuzumab maintenance in the GADOLIN study. J Clin Oncol 36: 2259-2266, 2018.

41. Rummel M, Kaiser U, Balser C, Stauch M, Brugger W, Welslau M, Niederle N, Losem C, Boeck HP, Weidmann E, *et al*: Bendamustine plus rituximab versus fludarabine plus rituximab for patients with relapsed indolent and mantle-cell lymphomas: A multicentre, randomised, open-label, non-inferiority phase 3 trial. Lancet Oncol 17: 57-66, 2016.

42. Visco C, Chiappella A, Nassi L, Patti C, Ferrero S, Barbero D, Evangelista A, Spina M, Molinari A, Rigacci L, *et al*: Rituximab, bendamustine, and low-dose cytarabine as induction therapy in elderly patients with mantle cell lymphoma: A multicentre, phase 2 trial from Fondazione Italiana Linfomi. Lancet Haematol 4: e15-e23, 2017.

43. McCulloch R, Visco C, Eyre TA, Frewin R, Phillips N, Tucker DL, Quaglia FM, McMillan A, Lambert J, Crosbie N and Rule S: Efficacy of R-BAC in relapsed, refractory mantle cell lymphoma post BTK inhibitor therapy. Br J Haematol 189: 684-688, 2020.

44. Kamdar M, Li H, Chen RW, Rimsza LM, Leblanc ML, Fenske TS, Shea TC, Barr PM, Phillips TJ, Leonard JP, *et al*: Five-year outcomes of the S1106 study of R-hyper-CVAD vs R-bendamustine in transplant-eligible patients with mantle cell lymphoma. Blood Adv 3: 3132-3135, 2019.

45. Park SI, Grover NS, Olajide O, Asch AS, Wall JG, Richards KL, Sobol AL, Deal AM, Ivanova A, Foster MC, *et al*: A phase II trial of bendamustine in combination with rituximab in older patients with previously untreated diffuse large B-cell lymphoma. Br J Haematol 175: 281-289, 2016.

46. Flinn IW, Erter J, Daniel DB, Mace JR and Berdeja JG: Phase II study of bendamustine and ofatumumab in elderly patients with newly diagnosed diffuse large B-cell lymphoma who are poor candidates for R-CHOP chemotherapy. Oncologist 24: 1035-e623, 2019.

47. Sehn LH, Hertzberg M, Opat S, Herrera AF, Assouline S, Flowers CR, Kim TM, McMillan A, Ozcan M, Safar V, *et al*: Polatuzumab vedotin plus bendamustine and rituximab in relapsed/refractory DLBCL: Survival update and new extension cohort data. Blood Adv 6: 533-543, 2022.

48. O'Connor OA, Lue JK, Sawas A, Amengual JE, Deng C, Kalac M, Falchi L, Marchi E, Turenne I, Lichtenstein R, *et al*: Brentuximab vedotin plus bendamustine in relapsed or refractory Hodgkin's lymphoma: An international, multicentre, single-arm, phase 1-2 trial. Lancet Oncol 19: 257-266, 2018.

49. Broccoli A, Argnani L, Botto B, Corradini P, Pinto A, Re A, Vitolo U, Fanti S, Stefoni V and Zinzani PL; Fondazione Italiana Linfomi ONLUS: First salvage treatment with bendamustine and brentuximab vedotin in Hodgkin lymphoma: A phase 2 study of the Fondazione Italiana Linfomi. Blood Cancer J 9: 100, 2019.

50. LaCasce AS, Bociek RG, Sawas A, Caimi P, Agura E, Matous J, Ansell SM, Crosswell HE, Islas-Ohlmayer M, Behler C, *et al*: Three-year outcomes with brentuximab vedotin plus bendamustine as first salvage therapy in relapsed or refractory Hodgkin lymphoma. Br J Haematol 189: e86-e90, 2020.

51. Hueso T, Gastinne T, Garciaz S, Tchernonog E, Delette C, Casasnovas RO, Durot E, Houot R, Tessoulin B, Tournilhac O, *et al*: Bendamustine-EAM versus BEAM regimen in patients with mantle cell lymphoma undergoing autologous stem cell transplantation in the frontline setting: A multicenter retrospective study from lymphoma study association (LYSA) centers. Bone Marrow Transplant 55: 1076-1084, 2020.

52. Gomez-Arteaga A, Mark TM, Guarneri D, Christos PJ, Gergis U, Greenberg JD, Hsu J, Mayer SA, Niesvizky R, Pearse RN, *et al*: High-dose bendamustine and melphalan conditioning for autologous stem cell transplantation for patients with multiple myeloma. Bone Marrow Transplant 54: 2027-2038, 2019.

53. Lee HC, Feng L, Oriabure O, Graham V, Chen W, Badillo M, Lu R, Lee HJ, Jain P, Manasanch EE, *et al*: A phase one trial of carfilzomib, bendamustine, and dexamethasone in relapsed and/or refractory multiple myeloma. Am J Hematol 96: E243-E246, 2021.

54. Chanan-Khan A, Cramer P, Demirkan F, Fraser G, Silva RS, Grosicki S, Pristupa A, Janssens A, Mayer J, Bartlett NL, *et al*: Ibrutinib combined with bendamustine and rituximab compared with placebo, bendamustine, and rituximab for previously treated chronic lymphocytic leukaemia or small lymphocytic lymphoma (HELIOS): A randomised, double-blind, phase 3 study. Lancet Oncol 17: 200-211, 2016.

55. Fraser G, Cramer P, Demirkan F, Silva RS, Grosicki S, Pristupa A, Janssens A, Mayer J, Bartlett NL, Dilhuydy MS, *et al*: Updated results from the phase 3 HELIOS study of ibrutinib, bendamustine, and rituximab in relapsed chronic lymphocytic leukemia/small lymphocytic lymphoma. Leukemia 33: 969-980, 2019.

56. Cheson BD, Brugger W, Damaj G, Dreyling M, Kahl B, Kimby E, Ogura M, Weidmann E, Wendtner CM and Zinzani PL: Optimal use of bendamustine in hematologic disorders: Treatment recommendations from an international consensus panel-an update. Leuk Lymphoma 57: 766-782, 2016.

57. Flinn IW, van der Jagt R, Kahl BS, Wood P, Hawkins TE, Macdonald D, Hertzberg M, Kwan YL, Simpson D, Craig M, *et al*: Randomized trial of bendamustine-rituximab or R-CHOP/R-CVP in first-line treatment of indolent NHL or MCL: The BRIGHT study. Blood 123: 2944-2952, 2014.

58. Martin P, Chen Z, Cheson BD, Robinson KS, Williams M, Rajguru SA, Friedberg JW, van der Jagt RH, LaCasce AS, Joyce R, *et al*: Long-term outcomes, secondary malignancies and stem cell collection following bendamustine in patients with previously treated non-Hodgkin lymphoma. Br J Haematol 178: 250-256, 2017.

59. Fung M, Jacobsen E, Freedman A, Prestes D, Farmakiotis D, Gu X, Nguyen PL and Koo S: Increased risk of infectious complications in older patients with indolent non-Hodgkin lymphoma exposed to bendamustine. Clin Infect Dis 68: 247-255, 2019.

60. Pezzullo L, Giudice V, Serio B, Fontana R, Guariglia R, Martorelli MC, Ferrara I, Mettivier L, Bruno A, Bianco R, *et al*: Real-world evidence of cytomegalovirus reactivation in non-Hodgkin lymphomas treated with bendamustine-containing regimens. Open Med (Wars) 16: 672-682, 2021.

61. Chen RW, Li H, Bernstein SH, Kahwash S, Rimsza LM, Forman SJ, Constine L, Shea TC, Cashen AF, Blum KA, *et al*: RB but not R-HCVAD is a feasible induction regimen prior to auto-HCT in frontline MCL: Results of SWOG study S1106. Br J Haematol 176: 759-769, 2017.

62. Smith A, Roman E, Appleton S, Howell D, Johnson R, Burton C and Patmore R: Impact of novel therapies for mantle cell lymphoma in the real world setting: A report from the UK's haematological malignancy research network (HMRN). Br J Haematol 181: 215-228, 2018.

63. Bašić-Kinda S, Mišura Jakobac K, Sinčić-Petričević J, Deak D, Vodanović M, Jakić-Bubalo M, Mitrović Z, Grubešić A, Dreta B, Županić Krmek D, *et al*: Improvement in the outcomes of mantle cell lymphoma in the last decade: A real-life non interventional study of the croatian cooperative group for hematologic diseases. Croat Med J 62: 455-463, 2021.

64. Roué G, López-Guerra M, Milpied P, Pérez-Galán P, Villamor N, Montserrat E, Campo E and Colomer D: Bendamustine is effective in p53-deficient B-cell neoplasms and requires oxidative stress and caspase-independent signaling. Clin Cancer Res 14: 6907-6915, 2008.
65. Aurer I: Mantle cell lymphoma in patients not eligible for autologous stem cell transplantation. Curr Opin Oncol 31: 374-379, 2019.
66. Aukema SM, Hoster E, Rosenwald A, Canoni D, Delfau-Larue MH, Rymkiewicz G, Thorns C, Hartmann S, Kluin-Nelemans H, Hermine O, et al: Expression of TP53 is associated with the outcome of MCL independent of MIPI and Ki-67 in trials of the European MCL network. Blood 131: 417-420, 2018.
67. FDA approves polatuzumab vedotin-piiq for diffuse large B-cell lymphoma, 2019.
68. Nikitorowicz-Buniak J: CHMP recommends EMA approval of polatuzumab vedotin for the treatment of adult patients with R/R DLBCL, 2019.
69. Moskowitz AJ, Hamlin PA Jr, Perales MA, Gerecitano J, Horwitz SM, Matasar MJ, Noy A, Palomba ML, Portlock CS, Straus DJ, et al: Phase II study of bendamustine in relapsed and refractory Hodgkin lymphoma. J Clin Oncol 31: 456-460, 2013.
70. Rinnerthaler G, Gampenrieder SP, Petzer A, Hubalek M, Petru E, Sandholzer M, Andel J, Balic M, Melchardt T, Hauser-Kronberger C, et al: Capecitabine in combination with bendamustine in pretreated women with HER2-negative metastatic breast cancer: Results of a phase II trial (AGMT MBC-6). Ther Adv Med Oncol 13: 17588359211042301, 2021.
71. Lammers PE, Shyr Y, Li CI, Hutchison AS, Sandler A, Carbone DP, Johnson DH, Keedy VL and Horn L: Phase II study of bendamustine in relapsed chemotherapy sensitive or resistant small-cell lung cancer. J Thorac Oncol 9: 559-562, 2014.
72. Hartmann JT, Mayer F, Schleicher J, Horger M, Huober J, Meisinger I, Pintoffl J, Käfer G, Kanz L and Grünwald V; German sarcoma group: Bendamustine hydrochloride in patients with refractory soft tissue sarcoma: A noncomparative multicenter phase 2 study of the German sarcoma group (AIO-001). Cancer 110: 861-866, 2007.
73. Chambers SK: Open trial of bendamustine hydrochloride in women with advanced ovarian cancer. NCT00867503, 2012.
74. Friedberg JW, Cohen P, Chen L, Robinson KS, Forero-Torres A, La Casce AS, Fayad LE, Bessudo A, Camacho ES, Williams ME, et al: Bendamustine in patients with rituximab-refractory indolent and transformed non-Hodgkin's lymphoma: Results from a phase II multicenter, single-agent study. J Clin Oncol 26: 204-210, 2008.
75. Knauf WU, Lissichkov T, Aldaoud A, Liberati A, Loscertales J, Herbrecht R, Juliusson G, Postner G, Gercheva L, Goranov S, et al: Phase III randomized study of bendamustine compared with chlorambucil in previously untreated patients with chronic lymphocytic leukemia. J Clin Oncol 27: 4378-4384, 2009.
76. Cheson BD, Friedberg JW, Kahl BS, Van der Jagt RH and Tremmel L: Bendamustine produces durable responses with an acceptable safety profile in patients with rituximab-refractory indolent non-Hodgkin lymphoma. Clin Lymphoma Myeloma Leuk 10: 452-457, 2010.
77. Stokes J, Molina MS, Hoffman EA, Simpson RJ and Katsanis E: Immunomodulatory effects of bendamustine in hematopoietic cell transplantation. Cancers (Basel) 13: 1702, 2021.
78. Allegra A, Tonacci A, Musolino C, Pioggia G and Gangemi S: Secondary immunodeficiency in hematological malignancies: Focus on multiple myeloma and chronic lymphocytic leukemia. Front Immunol 12: 738915, 2021.
79. Gafter-Gvili A, Gurion R, Raanani P, Shpilberg O and Vidal L: Bendamustine-associated infections-systematic review and meta-analysis of randomized controlled trials. Hematol Oncol 35: 424-431, 2017.
80. Lamure S, Duléry R, Di Blasi R, Chauchet A, Laureana C, Deau-Fischer B, Drenou B, Soussain C, Rossi C, Noël N, et al: Determinants of outcome in Covid-19 hospitalized patients with lymphoma: A retrospective multicentric cohort study. EClinicalMedicine 27: 100549, 2020.
81. Fraser C, Brown P, Megason G, Ahn HS, Cho B, Kirov I, Frankel L, Aplenc R, Bensen-Kennedy D, Munteanu M, et al: Open-label bendamustine monotherapy for pediatric patients with relapsed or refractory acute leukemia: Efficacy and tolerability. J Pediatr Hematol Oncol 36: e212-e218, 2014.
82. García Muñoz R, Izquierdo-Gil A, Muñoz A, Roldan-Galiacho V, Rabasa P and Panizo C: Lymphocyte recovery is impaired in patients with chronic lymphocytic leukemia and indolent non-Hodgkin lymphomas treated with bendamustine plus rituximab. Ann Hematol 93: 1879-1887, 2014.
83. Saito H, Maruyama D, Maeshima AM, Makita S, Kitahara H, Miyamoto K, Fukuhara S, Munakata W, Suzuki T, Kobayashi Y, et al: Prolonged lymphocytopenia after bendamustine therapy in patients with relapsed or refractory indolent B-cell and mantle cell lymphoma. Blood Cancer J 5: e362, 2015.
84. Coutre SE, Flinn IW, de Vos S, Barrientos JC, Schreeder MT, Wagner-Johnson ND, Sharman JP, Boyd TE, Fowler N, Dreiling L, et al: Idelalisib in combination with rituximab or bendamustine or both in patients with relapsed/refractory chronic lymphocytic leukemia. Hemasphere 2: e39, 2018.
85. Rummel MJ, Janssens A, MacDonald D, Keating MM, Zaucha JM, Davis J, Lasher J, Babanrao Pisal C, Izquierdo M and Friedberg JW: A phase 3, randomized study of ofatumumab combined with bendamustine in rituximab-refractory iNHL (COMPLEMENT A + B study). Br J Haematol 193: 1123-1133, 2021.
86. Schöffski P, Seeland G, Engel H, Grünwald V, Paul H, Merkle K, Kowalski R and Ganser A: Weekly administration of bendamustine: A phase I study in patients with advanced progressive solid tumours. Ann Oncol 11: 729-734, 2000.
87. Bremer K: High rates of long-lasting remissions after 5-day bendamustine chemotherapy cycles in pre-treated low-grade non-Hodgkin's-lymphomas. J Cancer Res Clin Oncol 128: 603-609, 2002.
88. Klippstein A, Schneider CP, Sayer HG and Höffken K: Pneumocystis carinii pneumonia as a complication of bendamustine monotherapy in a patient with advanced progressive breast cancer. J Cancer Res Clin Oncol 129: 316-319, 2003.
89. Layman RM, Ruppert AS, Lynn M, Mrozek E, Ramaswamy B, Lustberg MB, Wesolowski R, Ottman S, Carothers S, Bingman A, et al: Severe and prolonged lymphopenia observed in patients treated with bendamustine and erlotinib for metastatic triple negative breast cancer. Cancer Chemother Pharmacol 71: 1183-1190, 2013.
90. Burotto M, Stetler-Stevenson M, Arons E, Zhou H, Wilson W and Kreitman RJ: Bendamustine and rituximab in relapsed and refractory hairy cell leukemia. Clin Cancer Res 19: 6313-6321, 2013.
91. Martínez-Calle N, Hartley S, Ahearne M, Kasenda B, Beech A, Knight H, Balotis C, Kennedy B, Wagner S, Dyer MJS, et al: Kinetics of T-cell subset reconstitution following treatment with bendamustine and rituximab for low-grade lymphoproliferative disease: A population-based analysis. Br J Haematol 184: 957-968, 2019.
92. Stokes J, Hoffman EA, Zeng Y, Larmonier N and Katsanis E: Post-transplant bendamustine reduces GvHD while preserving GvL in experimental haploidentical bone marrow transplantation. Br J Haematol 174: 102-116, 2016.
93. Cona A, Tesoro D, Chiamenti M, Merlini E, Ferrari D, Marti A, Codecà C, Ancona G, Tincati C, d'Arminio Monforte A and Marchetti G: Disseminated cytomegalovirus disease after bendamustine: A case report and analysis of circulating B- and T-cell subsets. BMC Infect Dis 19: 881, 2019.
94. Stokes J, Hoffman EA, Molina MS, Kummet N, Simpson RJ, Zeng Y and Katsanis E: Bendamustine with total body irradiation conditioning yields tolerant T-cells while preserving T-cell-dependent graft-versus-leukemia. Oncoimmunology 9: 1758011, 2020.
95. García M, Bellosillo B, Sánchez-González B, García-Payarols F, Seoane A, Ferrer AM, Gimeno E, Barranco LE, Torner A, Solé F, et al: Study of regulatory T-cells in patients with gastric malt lymphoma: Influence on treatment response and outcome. PLoS One 7: e51681, 2012.
96. Molina MS, Hoffman EA, Stokes J, Kummet N, Smith KA, Baker F, Zúñiga TM, Simpson RJ and Katsanis E: Regulatory dendritic cells induced by bendamustine are associated with enhanced Flt3 expression and alloreactive T-cell death. Front Immunol 12: 699128, 2021.
97. Lepik KV, Mikhailova NB, Kondakova EV, Zalyalov YR, Fedorova LV, Tsvetkova LA, Kotselyabina PV, Borzenkova ES, Babenko EV, Popova MO, et al: A study of safety and efficacy of nivolumab and bendamustine (NB) in patients with relapsed/refractory Hodgkin lymphoma after nivolumab monotherapy failure. Hemasphere 4: e401, 2020.

98. Montraveta A, Lee-Vergés E, Roldán J, Jiménez L, Cabezas S, Clot G, Pinyol M, Xargay-Torrent S, Rosich L, Arimany-Nardí C, *et al*: CD69 expression potentially predicts response to bendamustine and its modulation by ibrutinib or idelalisib enhances cytotoxic effect in chronic lymphocytic leukemia. Oncotarget 7: 5507-5520, 2016.

99. Gafter-Gvili A and Polliack A: Bendamustine associated immune suppression and infections during therapy of hematological malignancies. Leuk Lymphoma 57: 512-519, 2016.

100. Yamasaki S, Matsushima T, Minami M, Kadowaki M, Takase K and Iwasaki H: Clinical impact of bendamustine exposure on lymphopenia risk after bendamustine and rituximab combination therapy for follicular lymphoma: A single-institute retrospective study. Ann Hematol 101: 209-211, 2022.

101. Lu L, Yoshimoto K, Morita A, Kameda H and Takeuchi T: Bendamustine increases interleukin-10 secretion from B cells via p38 MAP kinase activation. Int Immunopharmacol 39: 273-279, 2016.

102. Stenger EO, Turnquist HR, Mapara MY and Thomson AW: Dendritic cells and regulation of graft-versus-host disease and graft-versus-leukemia activity. Blood 119: 5088-5103, 2012.

103. Barbarroja-Escudero J, Sanchez-Gonzalez MJ, Antolin-Amerigo D, Rodriguez-Rodriguez M and Alvarez-Mon M: Hypersensitivity reactions and drug fever by bendamustine: A case report of three patients. Allergol Int 64: 109-111, 2015.

104. Chan M, Silverstein WK, Nikonova A, Pavenski K and Hicks LK: Bendamustine-induced immune hemolytic anemia: A case report and systematic review of the literature. Blood Adv 4: 1756-1759, 2020.