CD30 is a member of the tumor necrosis factor receptor superfamily. It is characteristically expressed in certain hematopoietic malignancies, including anaplastic large cell lymphoma and Hodgkin lymphoma, among others. The variable expression of CD30 on both normal and malignant lymphoid cells has focused research efforts on understanding the pathogenesis of CD30 upregulation, its contribution to lymphomagenesis through anti-apoptotic mechanisms, and its effect on cell survival. Given the restriction of CD30 to certain tumor types, the logical extension of this has been to attempt to exploit it as a therapeutic target. The efficacy of naked anti-CD30 antibodies in practice was, however, modest. Moreover, combinations with bacterial toxins and radioimmunoconjugates have also had limited success. The development of the antibody-drug compound brentuximab vedotin (BV), however, has rejuvenated interest in CD30 as a tumor target. Phase I and II clinical trials in Hodgkin lymphoma, peripheral T-cell lymphoma, cutaneous T-cell lymphoma, and even CD30-expressing B-cell lymphomas, have shown the compound is well tolerated, but more importantly, able to deliver meaningful disease control even in patients with multiply relapsed or refractory disease. FDA approval has been granted for its use in relapsed Hodgkin lymphoma and systemic anaplastic large cell lymphoma. A recent phase III trial of BV in cutaneous T-cell lymphoma has confirmed its superiority to standard of care therapies. In this manuscript, we explore the history of CD30 as a tumor marker and as a therapeutic target, both in the laboratory and in the clinic, with a view to understanding future avenues for further study.

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CD30 MOLECULE—CLONING AND CHARACTERIZATION OF TISSUE EXPRESSION

CD30, also known as Ki-1 or TNFRSF8, was first identified in 1982 using a monoclonal antibody (mAb) derived from a Hodgkin lymphoma (HL) cell line. The CD30 molecule was subsequently cloned and characterized as a 120 kD transmembrane glycoprotein receptor belonging to the tumor necrosis factor receptor (TNFR) superfamily, with intracellular, trans-membrane and extracellular domains. Sequence similarity between CD30 and other TNFR molecules is limited to the extracellular components; in CD30, these are comprised of six cysteine-rich repeats. Further delineation of CD30 epitopes suggested the extracellular components adopt a flower-like configuration. Later structural studies on other TNFR-related molecules—alone and in complex with ligands—suggested the extracellular cysteine-rich repeats most likely adopt an extended conformation. This may have implications for antibody binding.

CD30 ligand (CD30L, also known as TNFSF8 or CD153) is a membrane-bound cytokine with sequence homology to other members of the tumor necrosis factor (TNF) family. CD30L can be detected in vitro on a subset of activated lymphocytes, histiocytes and granulocytes, and has been demonstrated on Reed–Sternberg cells and some T-cell lymphomas, although not consistently on anaplastic large cell lymphomas (ALCL). Additionally, an 88 kD form of soluble CD30 (sCD30) can be detected in vivo in inflammatory states and CD30-positive hematologic malignancies, and is presumed to represent a cleavage by-product of the extracellular portion of CD30.

CD30 is expressed on a small subset of activated T and B lymphocytes, and a variety of lymphoid neoplasms, with the highest expression in classical HL and ALCL. It has been demonstrated with variable expression and intensity in some cases of peripheral T-cell lymphoma, not otherwise specified (PTCL-NOS); adult T-cell leukemia/lymphoma; cutaneous T-cell lymphoma (CTCL); extra-nodal NK-T-cell lymphoma; and a variety of B-cell non-HLs, including diffuse large B-cell lymphoma, particularly EBV-positive diffuse large B-cell lymphoma. Neoplastic mast cells in advanced systemic mastocytosis have also been shown to be CD30-positive. Less commonly, CD30 expression is seen in certain non-hematopoietic malignancies, including germ cell tumors and testicular embryonal carcinomas.

The BerH2 antibody is used for routine assessment of CD30 expression in tissue specimens, with good correlation between immunohistochemical protein expression and specific mRNA levels. Kim et al. reported the use of multi-spectral imaging in their study of brentuximab vedotin (BV) in CTCL, quantifying CD30 expression in biopsies otherwise designated CD30-negative by immunohistochemistry (IHC), thus defining a ‘low-level positive’ tumor group. Double antibody-conjugate techniques were employed to assess whether CD30 expression was
representative of tumor, or of tumor-infiltrating inflammatory cells such as cytotoxic T-lymphocytes or macrophages. While these techniques are not widely available, these results raise interesting questions about the contribution of the tumor microenvironment to lymphomagenesis, the specificity of CD30 as a treatment target, and how to best assess patient suitability for anti-CD30 therapies.

While CD30-positivity in ALCL is defined as tumor cell expression of 75% or higher, diagnostic cut-offs for other tumor types have not been universally agreed upon. In mycosis fungoides (MF), for example, CD30 positivity is much less than in ALCL, with one group reporting median epidermal staining of 14% in non-transformed cases, with higher expression levels in more advanced stages and large-cell transformation. It is a matter of debate whether CD30 expression levels permit stratification of expected responses to anti-CD30 therapies.

ROLE OF CD30 IN HEALTH AND DISEASE

Given the relative restriction of CD30 expression, early studies sought to characterize the driving factors behind CD30 upregulation in normal tissue. Initial studies showed that CD30 expression on lymphocytes could be induced by in vitro antigenic stimulation by mitogens or viruses, especially HIV, EBV and HTLV-1. Subsequent studies demonstrated that CD30 expression was not uniform across all activated lymphocytes, instead being limited to subpopulations of CD4+/CD45RO+ and CD8+ T cells in lymph nodes and the thymic medulla. CD30 expression appears higher in CD4+ and CD8+ cells producing a Th2-type cytokine response, although subsequent studies have also demonstrated CD30 expression on Th0 and Th1-specific cells. Additionally, building on the work of Stein et al., Catoretti et al. demonstrated that CD30 expression in B-lymphocytes is restricted to a minority population of stimulated B immunoblasts located at the edge of the germinal center and the extrafollicular region, with co-expression of markers of acute activation.

CD30 knockout mice have been studied in an attempt to understand the role of CD30 expression, albeit with somewhat conflicting results. Amakawa et al. demonstrated that the development and maintenance of memory T cells, as well as T-helper cell-dependent B-cell class-switching, were not impaired in response to mitogenic stimulation in their CD30 mutant mice, which the researchers felt argued against CD30 being involved in maintenance of the immune response. The researchers additionally found that CD30 mutant mice had increased thymic volume and increased numbers of circulating double-antigen positive T cells, suggesting a role for CD30 in negative selection in the thymus, although this has not been confirmed by later researchers. Subsequent studies have, however, reported contrasting results to these earlier conclusions. Gaspal et al. demonstrated that secondary antibody production—a process dependent on follicular T cells—was impaired in mice deficient in CD30, with loss of a sustained germinal center response, even though the primary response to antigen stimulation was normal, a result recapitulated in CD30-/-CD40-/- mice. These results were replicated by Kennedy et al. Overall, these findings underscore a role for CD30 in immune surveillance and cross-talk between B and T cells.

Subsequent studies have assessed the effects of CD30 stimulation in order to understand the cell signaling pathways linked to CD30. Epitope stimulation of CD30 results in receptor trimerization and signal transduction via the recruitment of TNFR-associated factor (TRAF) and TRAF-binding proteins, generating a signaling complex. TRAF2, as well as TRAF1 and TRAF5, have all been implicated in the signaling process. Downstream effects of CD30 stimulation are mediated in part by nuclear factor kappa B (NFkB), as well as by mitogen-activated protein kinase/extracellular signal-regulated kinase pathways, as outlined in Figure 1.

The restricted expression of CD30 suggests the possibility that CD30 plays a role in the development and propagation of HL and ALCL. However, defining if and how CD30 expression relates specifically to lymphomagenesis—rather than being merely a marker of cell of origin—has proven challenging. Certainly the finding that the NFkB and mitogen-activated protein kinase/extracellular signal-regulated kinase pathways are integral to CD30-mediated signaling suggests that CD30 expression may confer a proliferative and anti-apoptotic benefit in neoplastic cells. Horie et al. proposed a link between CD30 overexpression and ligand-independent stimulation of the NFkB pathways in HL cells, underscoring a possible link between CD30 expression and tumor perpetuation. These findings were not replicated by Hirsch et al., who instead suggested that NFkB activation in HL is constitutive and unrelated to CD30. In contrast, Watanabe et al. showed that CD30 upregulation in HL and ALCL cell lines might be linked by a self-perpetuating loop through the mitogen-activated protein kinase/extracellular signal-regulated kinase pathway to the expression of JunB, a member of the activator protein (AP)-1 transcription factor family, with diverse effects including a possible link to malignant transformation.

Leading on from this, additional studies sought to define the role of CD30 stimulation in lymphoma pathogenesis; however, interpretation of the results is hampered somewhat by the use of differing ligands between studies. Early studies assessed the effects of the CD30 cognate, CD30L, in different cell lines. CD30L binding showed pleiotropic effects in vitro, with activation and enhanced cytokine secretion in HL cells, but a pro-apoptotic effect in ALCL cells. Subsequent assessment of CD30 stimulation in
CD30 as a tumor target

Given the overexpression of CD30 in certain lymphoma subtypes and some non-lymphoid neoplasms, it seems logical to exploit it as a therapeutic target.

Monoclonal antibody monotherapy

Preclinical studies. The development of humanized anti-CD30 mAbs paved the way for clinical trials of immunotherapy. Initial preclinical studies in the mid- to late 1990s used murine anti-CD30 mAbs, with reported improved disease-free survival rates in xenograft mice treated with M44 or HeFi-1, and, in a later study using HeFi-1, tumor growth arrest or regression in an ALCL xenograft model. Leading on from these studies, focus was directed to the creation of humanized mAbs, resulting in the development of SGN-30, a chimeric mouse-human antibody, and the fully humanized mAb 5F11 (MDX-060, or iratumumab).

Studies reported between 2002 and 2010 examined the effects of SGN-30 in HL and adult T-cell leukemia/lymphoma cell lines and mouse models. In contrast to earlier studies using non-humanized mAbs, investigators were able to demonstrate cellular growth arrest and DNA fragmentation in vitro in HL cell lines treated with SGN-30. Moreover, tumor regression and improved survival was observed in a mouse xenograft cohort. One explanation advanced for this unexpected response in HL was that SGN-30 itself promotes receptor cross-linking and multimerization, potentially promoting growth arrest and pro-apoptotic signaling. Similarly, 5F11 was able to promote cell growth arrest in CD30-positive cell lines when receptor cross-linking occurred, enhancing antibody-dependent cell cytotoxicity. Tumor regression was seen in HL xenograft mouse models.

Clinical studies. Based on promising results in preclinical studies, phase I and II trials of both SGN-30 and 5F11 were undertaken. However, the results of early phase studies did not seem to offer significant promise for the use of ‘naked’ anti-CD30 mAbs in practice.

The phase I trial of SGN-30 was conducted in 2002 and 2003, including 24 patients with relapsed or refractory HL or CD30-positive non-HL. SGN-30 was administered weekly for six doses, and was well tolerated. Clinical benefits were modest, with six patients achieving stable disease, and one patient with primary cutaneous ALCL achieving a complete response (CR).

A subsequent phase II trial of SGN-30 in patients with HL and ALCL used the same dosing schedule with doses of either 6 mg/kg or 12 mg/kg; the regimen was again well tolerated. Clinical outcomes were, however, disappointing. Of the 38 patients with relapsed/refractory HL, 11 patients achieved stable disease, while none achieved CR or partial responses (PR). In the ALCL cohort of 41 patients, two CR and five PR were attained.

Duvcic et al. reported phase II Results for the use of SGN-30 in relapsed/refractory cutaneous ALCL and other CD30-positive lymphoproliferative disorders. In this study, objective responses were seen in 16 of 23 patients (70%), with 10 patients achieving CR and six achieving PR. Overall clinical benefit rate was 87%, and median duration of objective response was 84 days. There appeared to be a dose-response effect, with some patients achieving clinical responses at 12 mg/kg despite having minimal benefit at 4 mg/kg.

5F11 (MDX-060), a fully humanized anti-CD30 mAb, was the focus of a phase I/II trial in HL and systemic ALCL. Twenty-one patients were treated in the phase I cohort and 51 in the phase II cohort; of these patients, 25 achieved stable disease, two achieved PR and four achieved CR, with a median duration of response of less than 6 months in all groups. The investigators noted high rates of corticosteroid use in this study population, with 31 patients receiving steroids during treatment, including four patients with objective responses. Further clinical trials have not been pursued.

Imunoconjugates

Immunotoxins. Early studies of anti-CD30 immunoconjugates focused on immunotoxins, albeit with limited success. The BerH2-saporin anti-CD30 immunotoxin used a ribosome-inactivating protein type 1 (RIP-1), with demonstrable efficacy in vitro and in animal studies. Later combinations included conjugates of BerH2 and other RIP-1s, the anti CD30-Pseudomonas exotoxin A conjugate Ki-4 (scFv-ETA), and the anti-CD30 ricin A-chain immunotoxin. However, the efficacy of these immunotoxins in humans was hampered by high rates of the development of anti-therapeutic antibodies, CD30 down-regulation, or non-specific binding of the immunotoxin to soluble CD30.

Radioimmunoconjugates. Anti-CD30 radioimmunoconjugates have shown mixed results in preclinical studies, with limited scope for clinical development. A conjugate of Ki-4 and radiolabeled iodine (I-131) was tested in 22 patients with HL, with one CR and five PR, but had significant side effects, with seven patients experiencing grade 4 hematologic toxicity. Later combinations using Ki-4 and 5F11 antibodies with variable iodination showed high in vitro tumor cell specificity, but minimal efficacy in vivo in mouse models. The combination of HeFi-1 with either radiolabeled astatine (At-211) or yttrium (Y-90) showed preliminary efficacy in murine models; however, no clinical trials eventuated.

Brentuximab vedotin. The development of a novel antibody-drug conjugate (ADC), BV (also referred to as SGN-35 or cAC10-vcMMAE), seemed to overcome some of the problems seen with SGN-30. Brentuximab vedotin consists of the humanized IgG1 mAb SGN-30, in combination with the antimitotic agent monomethylauristatin E (MMAE), joined by a cathepsin cleavable linker (valine-citrulline). Brentuximab vedotin acts through binding of the ADC to CD30-positive cells, followed by receptor endocytosis and release of MMAE upon exposure to intracellular lysozymes. This results in inhibition of tubulin formation and cell apoptosis.
CR. While the median progression-free survival (PFS) was 1.8 mg/kg.82 Fatigue, nausea, diarrhea, neutropenia, and peripheral neuropathy, including 11 CR, with a median duration of response of 9.7 months. Tumor regression was seen in 36 of 42 patients (86%) responses, including 11 CR, with a median duration of response of 16.6 months. Median PFS was four months (0.6+ months). 87

Following on from this, Maeda et al. demonstrated a growth inhibitory effect of both SGN-30 and SGN-35 in adult T-cell leukemia/lymphoma cell lines. More importantly, mouse models treated with SGN-35 showed tumor regression.60

Blatt et al. examined the effects of BV in CD30-positive systemic mastocytosis. The researchers confirmed CD30 expression in systemic mastocytosis cell lines, and demonstrated cell apoptosis and death in response to BV. Interestingly, BV exposure did not appear to increase the risk of histamine release, and, moreover, appeared to downregulate IgE-mediated manifestations of histamine release.23

Brentuximab vedotin—preclinical studies. Preclinical studies confirmed efficacy of BV in vitro and in mouse models. Francisco et al. used BV in HL and ALCL cell lines, demonstrating in vitro stability, and ADC selectivity for CD30-positive cells. More importantly, BV successfully induced cell cycle arrest and apoptosis. In xenograft mouse models, BV treatment resulted in partial tumor regression in an HL model and complete tumor regression in an ALCL model, with a significant difference in tumor responses when compared to the 'naked' antibody. These treatment responses held true in mouse models of both subcutaneous and disseminated ALCL.78

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Brentuximab vedotin—clinical studies. Based on the promising results of preclinical studies, a phase I trial of BV was conducted in 45 patients with relapsed/refractory CD30-positive hematologic malignancies, including 42 patients with HL, two with systemic ALCL, and one with angioimmunoblastic T-cell lymphoma. The median number of prior therapies in this group was three (range one to seven), including 33 patients (73%) who had undergone an autologous stem cell transplant (auSCT); 17 patients had objective responses, including 11 CR, with a median duration of response of 9.7 months. Tumor regression was seen in 36 of 42 patients (86%) evaluable for radiologic response. The main side effects were fatigue, nausea, diarrhea, neutropenia, and peripheral neuropathy, with the latter seen in 16 patients. The recommended phase 2 dose was 1.8 mg/kg.82

Phase II and III studies using BV followed thereafter, as summarized in Table 1.

Pro et al. reported results for a cohort with relapsed/refractory systemic ALCL. 58 patients were dosed with 1.8 mg/kg every three weeks; 50 patients (86%) achieved objective responses, including 33 patients attaining CR and 17 patients attaining PR. The median duration of overall response was 12.6 months, and the median CR duration was 13.2 months. The most common grade 3 or 4 side effects included neuropathy, thrombocytopenia, and sensory neuropathy in 12% of patients.83 Younes et al. reported results for 102 patients with HL who had relapsed after auSCT. The objective response rate (ORR) was 75%, with a CR rate of 34%. Median duration of response was 6.7 months in all responders, and 20.5 months in those patients achieving CR. These findings are important given that the median number of prior treatments excluding auSCT was three, with a median time to relapse after transplant of 6.7 months, suggesting relatively chemo-refractory disease. Side effects were similar to those reported by Pro et al.; twenty patients continued therapy due to adverse events, predominantly sensory or motor neuropathies.84 A smaller study by Horwitz et al. in 35 patients with PTCL showed an ORR of 41%, with eight patients achieving CR. While the median progression-free survival (PFS) was 6.7 months in patients with angioimmunoblastic T-cell lymphoma, median PFS was a disappointing 1.6 months in the PTCL-NOS subgroup.85

In CTCL, phase II trials have been conducted in patients with Sezary syndrome and mycosis fungoides (MF), as well as in primary cutaneous ALCL and lymphomatoid papulosis. Kim et al. reported objective responses in 21 of 30 patients with either Sezary syndrome or MF, with seven patients having >90% clearance of cutaneous disease. It is notable that clinical response was not limited to those patients with >10% CD30 expression by IHC.27 Duvic et al. reported an ORR of 73% in their cohort of 48 patients, including CR in 17 patients. Mirroring the findings of Kim et al., responses were seen even in MF/Sezary syndrome patients with low CD30 expression, defined as <10% by IHC. All 11 patients with either primary cutaneous ALCL or lymphomatoid papulosis responded to BV; however, the median duration of response in this group was only 26 weeks.86

Prince et al. have recently reported the results of a phase III trial in CTCL of BV versus physician’s choice of either methotrexate or bexarotene. This study of 128 patients with CTCL included 97 patients with MF and 31 with primary cutaneous ALCL. The cut-off for CD30 positivity in this study was defined as 10% in at least one tumor sample; multiple biopsies could be submitted for central review, and not all biopsies had to meet this cut-off requirement. Primary endpoints were overall response rates of four months or longer (ORR4) and PFS. Both ORR4 and PFS strongly favored BV, with ORR4 of 56 versus 13% (P < 0.0001) and median PFS of 16.7 versus 3.5 months (HR 0.27, P < 0.0001), with a median follow-up of 17.5 months.87

Expanding the treatment spectrum, Jacobsen et al. reported an ORR of 44% in a phase II trial of BV in relapsed refractory B-cell lymphoma, including a majority cohort with diffuse large B-cell lymphoma (49 of 68 patients). Eight patients attained CR, with median response duration of 16.6 months. Median PFS was four months (0.6+ – 24+ months).72 Bartlett et al. reported on the use of BV in a cohort of 52 diffuse large B-cell lymphoma patients designated CD30 negative with conventional IHC, with an ORR of 31% and CR rate of 12%. With computer-assisted digital image analysis, 11 of 16 responders were designated as low level positive for CD30 (CD30 ⩾ 1%).88

Brentuximab vedotin—additional treatment options. Bartlett et al. demonstrated that retreatment is feasible, treating 21 patients with HL and eight with ALCL who had previously achieved at least PR to BV. Overall response rate was 60% in the HL cohort and 88% in the ALCL cohort, with CR rates of 30 and 63%, respectively. As expected, sensory and motor neuropathy rates were higher than in upfront treatment, but the regimen was otherwise well tolerated.89

As with other antibody therapies, the next logical step was to use BV in combination. The use of BV with ABVD or AVD for upfront treatment of HL demonstrated significant additive pulmonary toxicity in the bleomycin-exposed cohort. Subsequent withdrawal of the bleomycin from the combination regimen did not appear to compromise efficacy.90 The phase III trial comparing ABVD and AVD with BV has completed recruitment; however, final results are pending. Promising results have been reported in a phase I trial in CD30-positive PTCL, using either BV administered sequentially with CHOP, or BV in combination with CHP. ORR was 85% in the sequential treatment arm, and 100% in the combination arm, with CR rates of 62 and 88%, respectively.91

Additionally, early reports of a phase I/II trial of BV and bendamustine in patients with relapsed HL or ALCL suggest the combination is well tolerated, with an ORR of 67%.92

Other treatment options include the use of BV as consolidation rather than salvage. Moskovitz et al. reported on their double-blind, randomized controlled trial assessing BV consolidation after auSCT for patients with relapsed HL. There was a significant improvement in risk of progression in the BV-treated cohort, with...
| Trial          | Phase | Disease subgroup | Patient number | Median age in years (range) | Response rates | Response duration | Survival        |
|---------------|-------|-----------------|----------------|-----------------------------|----------------|------------------|-----------------|
| Pro et al.    | II    | sALCL           | 58             | 52 (14–76)                  | ORR 86% (95% CI 74.6–93.9) | Median DOR 12.6 months (95% CI 5.7–NE) | Median PFS 13.3 months (95% CI 6.9–NE) |
| Younes et al. | II    | HL              | 102            | 31 (15–77)                  | ORR 75% (95% CI 64.9–82.6%) | Median DOR 6.7 months (95% CI 3.6–14.8) | Median PFS 5.6 months (95% CI 5.0–9.0) |
| Horwitz et al.| II    | PTCL-NOS        | 35             | 64 (33–83)                  | ORR 41% (95% CI 24.6–59.3)  | Median DOR 7.6 months (95% CI 1.3–14+)  | Median PFS 2.6 months |
| Duvic et al.  | II    | CTCL (MF, pcALCL, LyP) | 48 (28 with MF, 9 with LyP, 2 with pcALCL) | 59.5 (31–77) | ORR 73% | Not reported |
| Kim et al.    | II    | CTCL (MF and SS) | 32 (30 evaluable for efficacy) | 62 (20–87) | ORR in 21/30 (70%, 90% CI 53–83) CR in 1/30 PR in 20/30 (SD in 4/30) | Not reported |
| Jacobsen et al.| II    | B-cell lymphoma | 68 (48 with DLBCL, 19 other B-cell lymphomas) | 62 (17–85) in DLBCL cohort 36 (16–68) in other lymphoma cohort | ORR 44% in DLBCL cohort (95% CI 29.5–58.8) CR 17% PR 27% ORR 26% in other lymphoma cohort (95% CI 9.1–51.2) CR 16% PR 11% | Median DOR in DLBCL cohort 5.6 months (0–22.7+ months) |
| Prince et al. | III   | CTCL            | 128            | 62 (22–83) in BV group 58 (22–83) in PC group | ORR4 56.3% (BV group) versus 12.5% (PC group), with P < 0.0001 ORR 67% in BV group CR 16% ORR in 20% in PC group CR 2% | Not reported |
| Bartlett et al.| II    | DLBCL           | 52             | Med PFS 14.7 months (0.4–15.6) |

Abbreviations: AITL, angioimmunoblastic T-cell lymphoma; CI, confidence interval; CR, complete response; CTCL, cutaneous T-cell lymphoma; DLBCL, diffuse large B-cell lymphoma; DOR, duration of objective response; EFS, event-free survival; HL, Hodgkin lymphoma; LyP, lymphomatoid papulosis; MF, mycosis fungoides; NE, not evaluable; ORR, objective response rate; ORR4, objective response rate at 4 months; OS, overall survival; PC, physician’s choice; pcALCL, primary cutaneous anaplastic large cell lymphoma; PFS, progression-free survival; PR, partial response; PTCL, peripheral T-cell lymphoma; PTCL-NOS, peripheral T-cell lymphoma, not otherwise specified; sALCL, systemic anaplastic large cell lymphoma; SD, stable disease; SS, Sezary syndrome.
a median PFS of 42.9 months compared to 24.9 months in the placebo-treated cohort. By independent review, the estimated 2-year PFS was 63% in the BV cohort and 51% in the placebo group. There was, however, no significant difference in overall survival between BV and placebo arms. 93

The results of these studies raise some interesting questions, one of which is whether the efficacy of BV lies solely in its capacity to affect directed cell death. This does not seem to be the case, with evidence of enhanced T-cell activation as well as dendritic cell priming and maturation in mouse models treated with microtubule-depolymerizing agents akin to that in BV. 87 In this context, the combination of BV with immune checkpoint inhibitors is a rational one, and indeed, has been the focus of a recent phase I/II trial. The other question is whether the predicted inhibitory effects is a rational one, and indeed, has been the focus of a recent phase I/II trial. The question of dose-limiting toxicity of the compound, as discussed earlier.

Brentuximab vedotin—side effects and resistance. While BV is undoubtedly more effective than naked mAbs and other conjugate therapies, it does not entirely overcome the problems seen with earlier immunotoxin compounds. Despite the documented cellular specificity of BV, the recognition of dose-limiting and cumulative peripheral neuropathy with treatment suggests the compound has off-target effects. This is likely mediated in part through the anti-tubulin effects of MMAE. 79 Hansen et al. have recently shown that HL cells can release CD30-containing extracellular vesicles. These vesicles then bind to CD30L-expressing cells in the tumor microenvironment, subsequently causing off-target binding of SGN-35 in vitro. 64 In addition to the side effects reported in the major studies, subsequent case series have highlighted some concerning BV toxicities. Cases of pancreatitis—fatal at least one instance—have been reported. 82 Progressive multifocal leukoencephalopathy, an incurable and often fatal CNS infection, has also been reported associated with BV. 86

Resistance can be an issue with repeated BV exposure. Chen et al. have demonstrated diverse resistance mechanisms in HL and ALCI cell lines, including increased expression of drug transporter proteins, and MMAE resistance. While CD30 downregulation is a putative mechanism, and was seen in vitro, this finding was not seen in the tissue samples examined in this study, mirroring prior case reports of persistent CD30 expression even in the setting of reduced BV efficacy. 82 However, cases of relapsed/ refractory ALCI with downregulation of CD30 following BV treatment have also been reported. 99–101

Other therapeutic approaches
Phase I and II trials have been performed using bi-specific molecules incorporating anti-CD30 mAbs; however, the utility of these molecules has been hampered by the invariable development of antibodies against the agents. One such molecule combined an anti-CD30 mAb with the bi-specific mAb HRS-3/A9, directed against CD16, aiming to enhance recruitment of natural killer cells and phagocytic cells. A phase I/II clinical trial was conducted in 15 patients with relapsed/ refractory HL, with one CR and one PR lasting six and three months respectively. Nine of the 15 patients developed antibodies to the bi-specific molecule, leading to treatment discontinuation. 102

Similarly, an anti-CD64/anti-CD30 molecule H22 x Ki-4 was developed in the late 1990s. This combined the antigen-binding region of Ki-4 with a humanized CD64-specific mAb, with the theoretical rationale being that CD64, as part of the high affinity immunoglobulin receptor, should enhance antibody-dependent cell-mediated cytotoxicity. While it was well-tolerated in a phase I trial, with objective responses in four of 10 patients with multiply relapsed/ refractory HL, interest in these agents has waned. 103

The most recent efforts in targeting CD30 have focused on the use of chimeric antigen receptor T (CAR T) cells. Hombach et al. reported on the safety of CD30-specific CAR T therapy, demonstrating in mouse models that CD30/CD34-positive normal hematopoietic cells and activated lymphocytes were not targeted by CD30 CAR T cells. 104 More importantly, Wang et al. have recently published preliminary results for a phase I trial, showing safety and efficacy, with PR in seven of the 18 patients treated, and stable disease in six. 105

**FUTURE DIRECTIONS**
We are yet to fully define the significance of tumoral CD30 expression, that is, whether the molecule merely signifies cell of origin, for example, ‘stimulated lymphocyte’, whether its expression plays some role in perpetuating a malignant phenotype, or whether it reflects the recruitment of an inflammatory milieu that enhances tumor growth and survival. Indeed, CD30 expression may reflect a combination of all of these possibilities, and the relative balance of each may inform our thinking about tumor pathogenesis as well as how best to target CD30 therapeutically. Certainly it seems that merely demonstrating CD30 expression does not guarantee responses to anti-CD30 therapies, as demonstrated by the disappointing results of BV in PTCL-NOS, and the recently reported failure to achieve the ORR endpoint in a phase II study of BV in primary mediastinal B-cell lymphoma, despite strong CD30 expression in this tumor type. 106 Conversely, the efficacy of BV in some tumors even with low-level CD30 expression is intriguing, suggestive of off-target immune modulatory effects of the compound, as discussed earlier.

The pleiotropic responses to CD30 stimulation are additionally of interest. Some of the studies outlined earlier would suggest the variable responses reflect differing mechanisms by which CD30 is upregulated in different tumor types, with consequent diverse effects on the signaling pathways mediated through CD30. The unanswered question is whether varying the type or mechanism of CD30 ligand binding may allow us to specifically target pro-apoptotic cellular signaling pathways, or to counteract cellular survival mechanisms. The finding that apoptosis may be augmented with antibody cross-linkage suggests this might be the case; however, this has not been studied in depth.

From a therapeutic standpoint, the failure to achieve substantive benefits with monoclonal antibodies and other antibody–drug combinations is disappointing. It is clear that BV overcomes many of these issues; however, the neurotoxicity of the MMAE may limit its long-term use. This raises the issue of why ‘naked’ antibody therapies are not as successful as expected. Some possibilities have been addressed, for example, it does not appear to be due to an excess of antibody binding to soluble CD30. 107 The paucity of available structural data limits our understanding of the way in which CD30-antibody interactions occur, such as the possibility that the interaction between CD30 and anti-CD30 antibodies may orient the antibody in a way that is unable to properly induce antibody-dependent cell-mediated cytotoxicity. X-ray or cryo-electron microscopy assessment of the structure of CD30 alone, CD30 in complex with CD30L, and CD30 bound to SGN-30 and SGN-35, would be invaluable in investigating these possibilities.

It has additionally been suggested that the differential response to anti-CD30 antibodies in HL and ALCI may in fact reflect a pro-survival benefit conferred by, and specific to, the HL microenvironment. 108,109 Supporting this postulate is the observation of a ‘bystander’ effect on surrounding, non-CD30-positive cells exposed to BV, which may alter the microenvironment immune signaling in HL. 81,94 Clinical trials of the combination of nivolumab and BV are currently under way, with
preliminary results suggesting that the combination is safe and well-tolerated.\textsuperscript{110} Trials combining the naked antibody with immune-checkpoint inhibitors would be an interesting question for future study.

Some of the therapeutic approaches targeting CD30 currently under investigation in clinical trials are summarized in Table 2. These trials are focused predominantly around the use of BV in combination with other agents, or exploring other therapeutic niches for BV, such as salvage as a bridge to transplant. Of note, however, is the use of anti-CD30 chimeric antigen receptor T cells, the results of which are awaited with interest.

The limited expression of CD30 to certain tumor types remains a tantalizing target in an era of purpose-designed therapies. Historical attempts at targeting CD30 may have failed due to a lack of clear understanding of the mechanisms and significance of CD30 expression in both health and in disease. Answering the question of how to maximize an anti-CD30 strategy into the future may require us to return to the laboratory bench, using contemporary structural techniques, molecular profiling and immunological modeling.

**CONFLICT OF INTEREST**

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**REFERENCES**

1. Schwab U, Stein H, Gerdes J, Lemke H, Kirchner H, Schaadt M et al. Production of a monoclonal antibody specific for Hodgkin's and Sterneberg-Reed cells of Hodgkin's disease and a subset of normal lymphoid cells. *Nature* 1982; 299: 65–67.
2. Stein H, Gerdes J, Schwab U, Lemke H, Mason DY, Ziegler A et al. Identification of Hodgkin and Sterneberg-reed cells as a unique cell type derived from a newly-detected small-cell population. *Int J Cancer* 1982; 30: 445–459.
3. Durkop H, Latza U, Hummel M, Eitelbach F, Seed B, Stein H. Molecular cloning and expression of a new member of the nerve growth factor receptor family that is characteristic for Hodgkin's disease. *Cell* 1992; 68: 421–427.
4. Smith CA, Gruss HJ, Davis T, Anderson D, Farrah T, Baker E et al. CD30 antigen, a marker for Hodgkin's lymphoma, is a receptor whose ligand defines an emerging family of cytokines with homology to TNF. *Cell* 1993; 73: 1349–1360.
5. Smith CA, Farrah T, Goodwin RG. The TNF receptor superfamily of cellular and viral proteins: activation, costimulation, and death. *Cell* 1994; 76: 959–962.
6. Dong L, Hulsmeyer M, Durkop H, Hansen HP, Schneider-Mergener J, Ziegler A et al. CD30 biology and targeted therapies. *Blood Cancer Journal* 2013; 3: e280.
7. Younes A, Consoli U, Zhao S, Snell V, Thomas E, Gruss HJ et al. CD30 ligand expression on resting normal and malignant human B lymphocytes. *Br J Haematol* 1996; 93: 562–571.
8. Gruss HJ, DaSilva N, Hu ZB, Uphoff CC, Goodwin RG, Drexler HG. Expression and regulation of CD30 ligand and CD30 in human leukemia-lymphoma cell lines. *Leukemia* 1994; 8: 2083–2094.
9. Younes A, Consoli U, Zhao S, Snell V, Thomas E, Gruss HJ et al. CD30 ligand is expressed on resting normal and malignant human B lymphocytes. *Br J Haematol* 1996; 93: 562–571.
10. Gruss HJ, Pinto A, Gloghini A, Wehnes E, Wright B, Boiani N et al. CD30 ligand expression in nonmalignant and Hodgkin's disease-involved lymphoid tissues. *Am J Pathol* 1996; 149: 469–481.
11. Hsu PL, Hsu SM. Autocrine growth regulation of CD30 ligand in CD30-expressing Reed–Sterneberg cells: distinction between Hodgkin's disease and anaplastic large cell lymphoma. *Lab Invest* 2000; 80: 1111–1119.
12. Gattin V, Degani M, Gloghini A, De Jullio A, Imparato S, Rossi FM et al. CD30 ligand is frequently expressed in human hematopoietic malignancies of myeloid and lymphoid origin. *Blood* 1997; 89: 2048–2059.
13. Josimovic-Alasevic O, Durkop H, Schwarting R, Beca E, Stein H, Diamantstein T. Ki-1 (CD30) antigen is released by Ki-1-positive tumor cells in vitro and in vivo. I. Partial characterization of soluble Ki-1 antigen and detection of the antigen in cell culture supernatants and in serum by an enzyme-linked immunosorbent assay. *Eur J Immunol* 1989; 19: 157–162.
14. Stein H, Mason DY, Gerdes J, O'Connor N, Wainscoat J, Pallesen G et al. The expression of the Hodgkin's disease associated antigen Ki-1 in reactive and
neoplastic lymphoid tissue: evidence that Reed-Sternberg cells and histiocytic malignancies are derived from activated lymphoid cells. Blood 1985; 66: 848–858.

15 Durkop H, Foss HD, Eitelbach F, Anagnostopoulou I, Latza U, Pileri S et al. Expression of the CD30 antigen in non-lymphoid tissues and cells. J Pathol 2000; 190: 613–618.

16 Beverley PC. Activation antigens: new and previously defined clusters. In: McMichael AJ (ed). Leucocyte Typing III. Oxford University Press; Oxford: 1987. pp 516–528.

17 Schwarting R, Gerdes J, Durkop H, Falini B, Pileri S, Stein H. BER-H2: a new anti-Ki-1 (CD30) monoclonal antibody directed at a formol-resistant epitope. Blood 1989; 74: 1678–1689.

18 Ellis TM, Simms PE, Slivnick DJ, Jack HM, Fisher RL. CD30 is a signal-transducing molecule that defines a subset of human activated CD45RO+ T cells. J Immunol 1993; 151: 2380–2389.

19 Savage KJ, Harris NL, Vose JM, Ullrich F, Jaffe ES, Conners JM et al. ALK-anaplastic large-cell lymphoma is clinically and immunophenotypically different from both ALK+ ALCCL and peripheral T-cell lymphoma, not otherwise specified: report from the International Peripheral T-Cell Lymphoma Project. Blood 2008; 111: 5496–5504.

20 Kim WY, Nam SJ, Kim S, Kim TM, Heo DS, Kim CW et al. Prognostic implications of CD30 expression in extranodal natural killer/T-cell lymphoma according to treatment modalities. Leuk Lymphoma 2015; 56: 1778–1786.

21 Jacobsen ED, Sharman JP, Oki Y, Advani RH, Winter RN, Bello CM et al. Brentuximab vedotin demonstrates objective responses in a phase 2 study in relapsed/refractory DLBCL with variable CD30 expression. Blood 2015; 125: 1394–1402.

22 Montes-Moreno S, Odqvist L, Diaz-Perez JA, Lopez AB, de Villамиososa SG, Mazorra F et al. EBV-positive diffuse large B-cell lymphoma of the elderly is an aggressive post-germinal center B-cell neoplasm characterized by prominent nuclear factor-kB activation. Mod Pathol 2012; 25: 968–982.

23 Blatt K, Cerny-Reiterer S, Schwabj J, Sotlar K, Eisenwort G, Stefanzi G et al. Identification of the Ki-1 antigen (CD30) as a novel therapeutic target in systemic mastocytosis. Blood 2015; 126: 2832–2841.

24 Sotlar K, Cerny-Reiterer S, Petat-Dutter K, Hessel H, Berezowska S, Mullauer L et al. Aberrant expression of CD30 in neoplastic mast cells in high-grade mastocytosis. Mod Pathol 2011; 24: 585–595.

25 Bossard C, Dobay MP, Parrens M, Lamant L, Missiaglia E, Haison C et al. Immunohistochemistry as a valuable tool to assess CD30 expression in peripheral T-cell lymphomas: high correlation with mRNA levels. Blood 2014; 124: 2983–2986.

26 Onaindia A, Martinez N, Montes-Moreno S, Almaraz C, Rodriguez-Moreno S, Cerecida L et al. CD30 expression by B and T cells: a frequent finding in angiomedioblastic t-cell lymphoma and peripheral T-cell lymphoma—not otherwise specified. Am J Surg Pathol 2016; 40: 378–385.

27 Kim YH, Tavallae M, Sundram U, Salva KA, Wood GS, Li S et al. Phase II interim analysis of the activated protein kinase signaling activates the CD30 promoter in anaplastic large cell lymphoma. Clin Cancer Res 2011; 17: 463–469.

28 Kim WY, Nam SJ, Kim S, Kim TM, Heo DS, Kim CW et al. Impaired negative selection of T cells in Hodgkin's disease antigen depleted mice. Cell 1996; 84: 551–562.

29 Gaspal FM, Kim MY, McConnell FM, Raykundalia C, Bekiaris V, Lane PJ. Mice deficient in Ox40 and CD30 signals lack memory antibody responses because of deficient CD4 T cell memory. J Immunol 2005; 174: 3891–3896.

30 Kennedy MK, Willis CT, Armitage R. Deciphering CD30 ligand biology and its role in humoral immunity. Immunology 2006; 118: 143–152.

31 Buckel CS, Thompson CB. CD30-dependent degradation of TRAF2: implications of the Ki-1 antigen (CD30) as a novel therapeutic target in systemic mastocytosis. Blood 2002; 100: 1355–1366.

32 Buckel CS, Gedrich RW, Gillilalan ME, Thompson CB. Induction of nuclear factor kappaB by the CD30 receptor is mediated by TRAF1 and TRAF2. Mol Cell Biol 1997; 17: 1535–1542.

33 Zheng B, Fiumara P, Li YV, Georgakis G, Snell V, Younes M et al. MEK/ERK pathway is aberrantly active in Hodgkin disease: a signaling pathway shared by CD30, CD40, and RANK that regulates cellular proliferation and survival. Blood 2003; 102: 1019–1027.

34 Krysov SV, Rowley TF, Al-Shamkahi A. Inhibition of p38 mitogen-activated protein kinase unmaska CD30-triggered apoptotic pathway in anaplastic large cell lymphoma cells. Mol Cancer Ther 2007; 6: 703–711.

35 Watanabe M, Nakano K, Togano T, Nakashima M, Hishigahara M, Kadin ME et al. Targeted repression of overexpressed CD30 downregulates NF-kappaB and ERK1/2 pathway in Hodgkin lymphoma cell lines. OncoRes 2011; 19: 463–469.

36 Mir SS, Richter BW, Duckett CS. Differential effects of CD30 activation in anaplastic large cell lymphoma. Clin Cancer Res 2006; 12: 1391.

37 Nishikori M, Ohno H, Haga H, Uchiyama T. Stimulation of CD30 in anaplastic large cell lymphoma leads to production of nuclear factor-kappaB p52, which is associated with hyperphosphorylated Bcl-2. Cancer Sci 2005; 96: 487–497.

38 Horie R, Watanabe T, Morishita Y, Ito K, Ishida T, Kanegae Y et al. Ligand-independent signaling by overexpressed CD30 drives NF-kappaB activation in Hodgkin–Reed–Sternberg cells. Oncogene 2001; 21: 2493–2503.

39 Hirsch B, Hummel M, Bentink S, Hapel A, von Bubnau S, Al-Shamkahi A. Mechanistic defects in the intracellular domain of human CD30 differentially activate canonical and alternative transcription factor NF-kappaB signaling. PLoS One 2012; 7: e52424.

40 Naka T, Ohtake K, Hishigahara M, Umezawa K, Kadin ME et al. JunB induced by constitutive CD30-extracellular signal-regulated kinase 1/2 mitogen-activated protein kinase signaling activates the CD30 promoter in anaplastic large cell lymphoma and reed-sternberg cells of Hodgkin lymphoma. Cancer Res 2005; 65: 7628–7634.

41 Bodickler RL, Kip NS, Xing X, Zeng Y, Zhang ZZ, Lee JH et al. The Oncogenic transcription factor IRF4 is regulated by a novel CD30/NF-kappaB positive feedback loop in peripheral T-cell lymphoma. Blood 2015; 125: 3118–3127.

42 Patanakul P, Vedervad A, Prochazka-Carlova M, Laharrarre E, Vergier B, Jouary T, Bleyl-Marr Y et al. IRF4 gene rearrangements define a subgroup of CD30-positive cutaneous T-cell lymphomas: a study of 54 cases. J Invest Dermatol 2010; 130: 816–825.

43 Watanabe M, Itoh K, Togano T, Kadin ME, Watanabe T, Hishigahara M et al. Ets-1 activates overexpression of JunB and CD30 in Hodgkin's lymphoma and anaplastic large-cell lymphoma. Am J Pathol 2012; 180: 831–838.

44 Watanabe M, Ogawa Y, Ito K, Hishigahara M, Kadin ME, Abraham LJ et al. ets-1 mediated relief of repressive activity of the CD30 promoter microsatellite in Hodgkin and Reed-Sternberg cells. Am J Pathol 2003; 163: 633–641.

45 Guss HJ, Boiani N, Williams DE, Armitage R, Smith CA, Goodwin RG. Pleiotropic effects of the CD30 ligand on CD30-expressing cells and lymphoma cell lines. Blood 1994; 83: 2045–2056.

46 Wright CW, Rumble JM, Buckel CS. Differential regulation of alternative NF-kappaB pathways in anaplastic large cell lymphoma cells. J Biol Chem 2007; 282: 10252–10262.

47 Mir SS, Richter BW, Buckel CS. Differential effects of CD30 activation in anaplastic large cell lymphoma and Hodgkin lymphoma cell lines. Blood 2000; 96: 4307–4312.

48 Staber PB, Noehammer C, Durkop H, Schauer S, Kenner L, Linkes W et al. Overexpression patterns indicate CD30 mediated activation of different apoptosis pathways in anaplastic large cell lymphoma but not in Hodgkin's lymphoma. Leuk Res 2006; 30: 343–348.

49 Pfeifer W, Levi E, Petrogiannis-Haliotis T, Lehmann L, Wang Z, Kadin ME. A murine xenograft model for human CD30+ anaplastic large cell lymphoma. Successful growth inhibition with an anti-CD30 antibody (Hefi-1). Am J Pathol 1999; 155: 1353–1359.
95 Tian ZG, Longo DL, Funakoshi S, Asai O, Ferris DK, Widmer M et al. In vivo antitumor effects of unconjugated CD30 monoclonal antibodies on human anaplastic large-cell lymphoma xenografts. Cancer Res 1995; 55: 5335–5341.

96 Maeda N, Huta, Oflazoglu E, Yoshikai Y. Susceptibility of human T-cell leukemia virus type I-infected cells to humanized anti-CD30 monoclonal anti-

97 bodies in vitro and in vivo. Cancer Sci 2010; 101: 224–230.

98 Wahl M, Kluussman K, Thompson JD, Chen JH, Francisco LV, Schnell R, Staak O et al. The anti-CD30 monoclonal antibody SGN-30 promotes growth arrest and DNA fragmentation in vitro and affects antitumor activity in models of Hodgkin’s disease. Cancer Res 2002; 62: 3736–3742.

99 Cerveny CG, Law CL, McCormick RS, Lenox JS, Hamblett KJ, Westendorf LE et al. Antitumor activity of anti-CD30 immunotoxin in human leukemia xenografts. Blood 2003; 102: 3757–3742.

100 Barth S, Huhn M, Matthey B, Schnell R, Lange H, Huhn M et al. Antitumor efficacy of unconjugated CD30 monoclonal antibodies in Hodgkin’s lymphoma xenografts. Blood Cancer J 2016; 10 (9): 2897–2902.

101 Asai O, Ferris DK, Widmer M et al. In vivo antitumor effects of unconjugated CD30 monoclonal antibodies on human anaplastic large-cell lymphoma xenografts. Cancer Res 1995; 55: 5335–5341.

102 Asai O, Ferris DK, Widmer M et al. In vivo antitumor effects of unconjugated CD30 monoclonal antibodies on human anaplastic large-cell lymphoma xenografts. Cancer Res 1995; 55: 5335–5341.
100 Nielson C, Fischer R, Fraga G, Aires D. Loss of CD30 expression in anaplastic large cell lymphoma following brentuximab therapy. *J Drugs Dermatol* 2016; 15: 894–895.

101 Al-Rohil RN, Torres-Cabala CA, Patel A, Tetzlaff MT, Ivan D, Nagarajan P et al. Loss of CD30 expression after treatment with brentuximab vedotin in a patient with anaplastic large cell lymphoma: a novel finding. *J Cutan Pathol* 2016; 43: 1161–1166.

102 Hartmann F, Renner C, Jung W, Deisting C, Juwana M, Eichentopf B et al. Treatment of refractory Hodgkin’s disease with an anti-CD16/CD30 bispecific antibody. *Blood* 1997; 89: 2042–2047.

103 Borghmann P, Schnell R, Fuss I, Manzke O, Davis T, Lewis LD et al. Phase 1 trial of the novel bispecific molecule H22xKi-4 in patients with refractory Hodgkin lymphoma. *Blood* 2002; 100: 3101–3107.

104 Hombach AA, Gorgens A, Chmielewski M, Murke F, Kimpel J, Giebel B et al. Superior therapeutic index in lymphoma therapy: CD30+ CD34+ hematopoietic stem cells resist a chimeric antigen receptor T-cell attack. *Mol Ther* 2016; 24: 1423–1434.

105 Wang CM, Wu ZQ, Wang Y, Guo YL, Dai HR, Wang XH et al. Autologous T cells expressing CD30 chimeric antigen receptors for relapsed or refractory Hodgkin lymphoma: an open-label phase I trial. *Clin Cancer Res* 2017; 23: 1156–1166.

106 Zinzani PL, Pellegrini C, Chiappella A, Di Rocco A, Salvi F, Cabras MG et al. Brentuximab vedotin in relapsed primary mediastinal large B-cell lymphoma: results from a phase 2 clinical trial. *Blood* 2017; 129: 2328–2330.

107 Nagata S, Ise T, Onda M, Nakamura K, Ho M, Raubitschek A et al. Cell membrane-specific epitopes on CD30: Potentially superior targets for immunotherapy. *Proc Natl Acad Sci USA* 2005; 102: 7946–7951.

108 Aldinucci D, Gloghini A, Pinto A, De Filippi R, Carbone A. The classical Hodgkin’s lymphoma microenvironment and its role in promoting tumour growth and immune escape. *J Pathol* 2010; 221: 248–263.

109 Poppema S, van den Berg A. Interaction between host T cells and Reed–Sternberg cells in Hodgkin lymphomas. *Semin Cancer Biol* 2000; 10: 345–350.

110 Herrera AF, Bartlett NL, Ramchandren R, Vose JM, Moskowitz AJ, Feldman TA et al. Preliminary results from a phase 1/2 study of brentuximab vedotin in combination with nivolumab in patients with relapsed or refractory Hodgkin lymphoma. *Blood* 2016; 128: 1105.