Monoclonal Antibodies in Myeloma

Pia Sondergeld, PhD, Niels W. C. J. van de Donk, MD, PhD, Paul G. Richardson, MD, and Torben Plesner, MD

Dr Sondergeld is a medical student at the University of Giessen in Giessen, Germany. Dr van de Donk is a hematologist in the department of hematology at the VU University Medical Center in Amsterdam, The Netherlands. Dr Richardson is the R.J. Corman Professor of Medicine at Harvard Medical School, and clinical program leader and director of clinical research at the Jerome Lipper Myeloma Center, division of hematologic malignancy, department of medical oncology, Dana-Farber Cancer Institute in Boston, MA. Dr Plesner is a professor of hematology at the University of Southern Denmark and a consultant in the department of hematology at Vejle Hospital in Vejle, Denmark.

Corresponding author:
Torben Plesner, MD
Professor of Hematology
University of Southern Denmark
Institute of Regional Health Science
Department of Hematology
Section of Internal Medicine
Vejle Hospital
Kabbeltoft 25
DK-7100 Vejle
Denmark
Tel: +(45) 79406313 / +(45) 20887692
Fax: +(45) 79406851
E-mail: torben.plesner@rsyd.dk

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Abstract: The development of monoclonal antibodies (mAbs) for the treatment of disease goes back to the vision of Paul Ehrlich in the late 19th century; however, the first successful treatment with a mAb was not until 1982, in a lymphoma patient. In multiple myeloma, mAbs are a very recent and exciting addition to the therapeutic armamentarium. The incorporation of mAbs into current treatment strategies is hoped to enable more effective and targeted treatment, resulting in improved outcomes for patients. A number of targets have been identified, including molecules on the surface of the myeloma cell and components of the bone marrow microenvironment. Our review focuses on a small number of promising mAbs directed against molecules on the surface of myeloma cells, including CS1 (elotuzumab), CD38 (daratumumab, SAR650984, MOR03087), CD56 (lorvotuzumab mertansine), and CD138/syndecan-1 (BT062/indatuximab ravtansine).

Introduction

Under normal circumstances, the immune system has finely tuned mechanisms of defense and is able to keep malignant transformations in check, thus ensuring the healthy balance of renewal that is characteristic of life. The circumstances under which these processes go awry and result in cancerous growth are unknown. Immunotherapy, the application of agents aimed at engaging or augmenting the immune system to target cancer cells, is recognized as a crucial strategy and a growing area in tumor therapy.[1] Immunotherapeutic strategies can be classified as active modalities, which involve vaccination or adjuvant therapy to actively promote antitumor effector mechanisms, or passive methods, among which monoclonal antibodies (mAbs) and the adoptive transfer of genetically modified specific T cells are the most widely investigated.[1]

The concept of using antibodies as magic bullets to treat disease goes back to Paul Ehrlich, who in 1899 developed the side chain theory.[2] He postulated the existence of receptors (ie, side chains) on

immune cells that are specific for particular substances (ie, antigens) that bind and activate the cell to produce more receptors to be released into the blood stream to neutralize the antigen. His landmark immunologic insights were rewarded with the Nobel Prize in Physiology or Medicine in 1908 and provided the inspiration for multiple groundbreaking studies into the treatment of human disease.[3,4]

Another milestone in the development of mAbs for therapy was the invention of hybridoma technology by Georges Köhler and César Milstein in 1975. They fused immortal myeloma cells to a specific antibody–expressing B cell that was derived from murine spleen cells of immunized mice, thus creating immortal cells able to produce a specific antibody. In their landmark article published in *Nature*, they hypothesized that "such cultures could be valuable for medical and industrial use."[5] The significance of the work was recognized with the Nobel Prize in Physiology or Medicine, which the 2 researchers received in 1984.

As predicted by Köhler and Milstein, soon large numbers of new antibodies, including antibodies against leukocyte markers, were being produced in many laboratories around the world. Thus, it became increasingly difficult to know which antibodies were directed against the same molecules. This prompted the organization of the first Human Leucocyte Differentiation Antigens workshop in Paris in 1982, during which many of these antibodies were evaluated and compared. The aim of this and subsequent workshops was the independent validation of antibody specificity and usability for research, diagnosis, and therapy. The workshop resulted in the creation of the CD nomenclature, in which antibodies with a similar reaction pattern nominated as "Clusters of Differentiation" (CD) are grouped together; the characterized molecules starting with CD1 originated from these workshops.[6,7] The system therefore represents a classification of the many mAbs generated by different laboratories around the world against epitopes on the surface molecules of leukocytes.

The CD nomenclature is, in its origin, a nomenclature of antibodies, and the prefix of *anti* is not required. This is a common mistake that was already predicted by Milstein during an early workshop. Since its conception, the use of the classification system has been expanded to many other cell types. The CD system is commonly used as a method to identify cells by allowing them to be defined and discriminated based on the composition of molecules on their surface.

Following the development of the hybridoma technology, it was not until 1982 that the first lymphoma patient was successfully treated with a mAb, which was a patient-specific anti-idiotype antibody therapy.[8] In 1993, the CD20 mAb rituximab (Rituxan, Genentech/Biogen Idec), a chimeric mAb, was used for the first time to treat lymphoma. In 1997, rituximab became the first licensed mAb to treat cancer when it was approved by the US Food and Drug Administration (FDA) for lymphoma; a year later it was approved by the European Medicines Agency.[9] CD20 was identified as an attractive target in lymphoma because of its expression on more than 90% of B-cell lymphomas.[10-12] CD20 is expressed on normal B cells from the pre–B-cell stage to the activated B-cell stage, but is not expressed on stem cells, plasma cells, or cells of other lineages.[12] Rituximab was initially investigated as monotherapy in lymphoma,[13] but was soon combined with various chemotherapy regimens and included as maintenance therapy.[14-18] Rituximab is now considered a standard therapy in lymphoma and has had an overwhelming impact on survival.[19]

Rituximab also has been investigated in multiple myeloma (MM); however, results were disappointing. This may be explained by the small percentage of patients (approximately 20%) with CD20 expression on their myeloma cells. Even in patients with CD20 expression, the response to rituximab is poor.[20-23] Of note, patients with translocations t(11;14) frequently have high levels of CD20 expression on myeloma cells.[24] In addition, Treon and colleagues have shown in vitro that interferon gamma induces CD20 expression on MM bone marrow plasma cells and B cells, and facilitates rituximab binding to MM bone marrow plasma cells.[25] In an era in which personalized therapy is increasingly being investigated as the optimal approach to therapy, these observations may provide a rationale for the further investigation of rituximab in selected MM patients.

Targets for mAbs in MM

A number of targets have been identified for mAb therapy in myeloma. These include components of the bone marrow microenvironment and molecules on the surface of the myeloma cell, where mAbs target the tumor cells directly or the interaction between the myeloma and the bone marrow stromal cells. The table provides an overview of targets and mAbs currently undergoing investigation. The impressive list shows the enormous research activity in the field. Many of these mAbs are still in preclinical or early clinical development, and a number of excellent review articles exist to provide an overview of these.[23,26-29] For our review, we will focus on a limited number of promising mAbs directed against molecules on the surface of myeloma cells.

Mechanism of Action of mAbs

The majority of mAbs belong to the immunoglobulin G (IgG) isotype, for which 4 subtypes exist. IgG constitutes the main immunoglobulin in serum and has a half-life of 23 days. IgG is a key player in the humoral immune response, with central roles in opsonization, complement activation, and antibody-dependent cytotoxicity (ADCC).

Table. mAbs and Their Targets in Myeloma

Target	mAb
Myeloma cell surface molecules	
CD20	Rituximab
CD38	Daratumumab, SAR650984, MOR03087
CD40	Dacetuzumab
CD54 (ICAM-1)	BI-505
CD56	Lorvotuzumab
CD70	SGN-70
CD74	Milatuzumab
CD138	BT062
CD200	Samalizumab
CD317 (HM1.24)	HM1.24 mAb
CS1	Elotuzumab
IGF-1R	AVE1642, figitumumab, IMC-A12
Components of the bone marrow milieu	
IL-6	Siltuximab
VEGF	Bevacizumab
BAFF	LY2127399
DKK1	BHQ880
RANKL	Denosumab
NK cell surface molecules	
KIR	IPH2101
PD-L1	CT-011

BAFF, B-cell activating factor; DKK1, Dickkopf-1; IGF-1R, insulin-like growth factor 1 receptor; IL-6, interleukin 6; KIR, killer cell immunoglobulin-like receptor; mAb, monoclonal antibody; NK, natural killer; PD-L1, programmed cell death ligand 1; RANKL, receptor activator of the NF-κB ligand; VEGF, vascular endothelial growth factor.

mAbs can exert their cytotoxic effects through a number of mechanisms, involving the use of immune system cells or complement proteins, or by acting independently of the host's immune system (see the figure).[30,31] mAbs are able to transmit death signals by binding to and cross-linking surface receptors on the target cancer cell or by blocking an activation signal that is necessary for continued cancer growth or viability, thereby inducing apoptosis. Mechanisms that are dependent on the immune system include ADCC, the antibody-induced lysis of a target cell by an activated natural killer (NK) cell. This relies on the Fc domain of the target cell–bound mAb binding to Fc receptors on the surface of NK cells, which activates the NK cells and results in the killing of the tumor cell. The Fc domain of the target cell–bound mAb also can bind to Fc receptors on macrophages, resulting in antibody-dependent cellular phagocytosis (ADCP). In addition, the activation of the complement system is a key mechanism of action of mAbs. Binding of the Fc domain of the mAb to the classic complement-activating protein C1q activates the complement cascade, which leads to the lysis of the target cells (complement-dependent cytotoxicity [CDC]). Moreover, complement fragments that are released upon activation of the cascade can attract and activate immune cells, also linking innate and adaptive immunity.

Overview of Promising Targets and mAbs in MM

CS1

CS1 (also known as CRACC, CD319, and SLAMF7) is a member of the signaling lymphocytic activation molecule (SLAM) family, which is involved in normal immune regulation, but has also been linked to immunodeficiency and autoimmune diseases.[32,33] CS1 is a cell surface glycoprotein and its expression is limited to hematopoietic cells; it is found on NK cells, NK-like T cells, CD8+ T cells, activated monocytes, dendritic cells, and plasma cells, with the expression on plasma cells being the highest among the immune cells.[34] As a key regulator of normal immune cell function, CS1 activates NK cells and is thought to have a growth-promoting role in normal B-cell development and an inhibitory role in T-cell development.[35] CS1 is highly expressed in more than 95% of cases of MM[34,36] and may have a tumor-stimulating effect by promoting cell adhesion.[36] The high expression levels observed in myeloma make CS1 an attractive target for mAb therapy.

Elotuzumab

Elotuzumab (HuLuc63) is a humanized IgG1 mAb that was first developed as a mouse antibody and then humanized.[34] Elotuzumab binds CS1 but does not interact with other members of the SLAM family.[35] In preclinical studies, elotuzumab induced lysis of human MM cell lines and primary human MM cells; efficacy also was seen in vivo in preclinical models of myeloma.[34,36,37] Elotuzumab may exert anti-MM efficacy via NK cell–mediated ADCC, because NK cells are required for elotuzumab activity.[34] CS1 also is found on NK cells, and elotuzumab can directly enhance NK cell function by ligation of CS1 on NK cells, thereby augmenting the ADCC.[38,39] Preclinical studies have shown that elotuzumab-mediated ADCC can be enhanced by combining the agent with bortezomib (Velcade, Millennium Pharmaceuticals) or lenalidomide (Revlimid, Celgene), which provides the rationale for clinical combination studies.[37,39]

The preclinical activity of elotuzumab monotherapy could not be observed clinically.[40] In the first-in-human phase 1 trial involving 35 heavily pretreated patients with relapsed/refractory myeloma, a dose-escalation scheme was implemented with intravenous elotuzumab administered at doses ranging from 0.5 to 20 mg/kg every 2 weeks. No objective responses were seen, but stable disease (SD) was noted in 26.5% of patients. The most common adverse

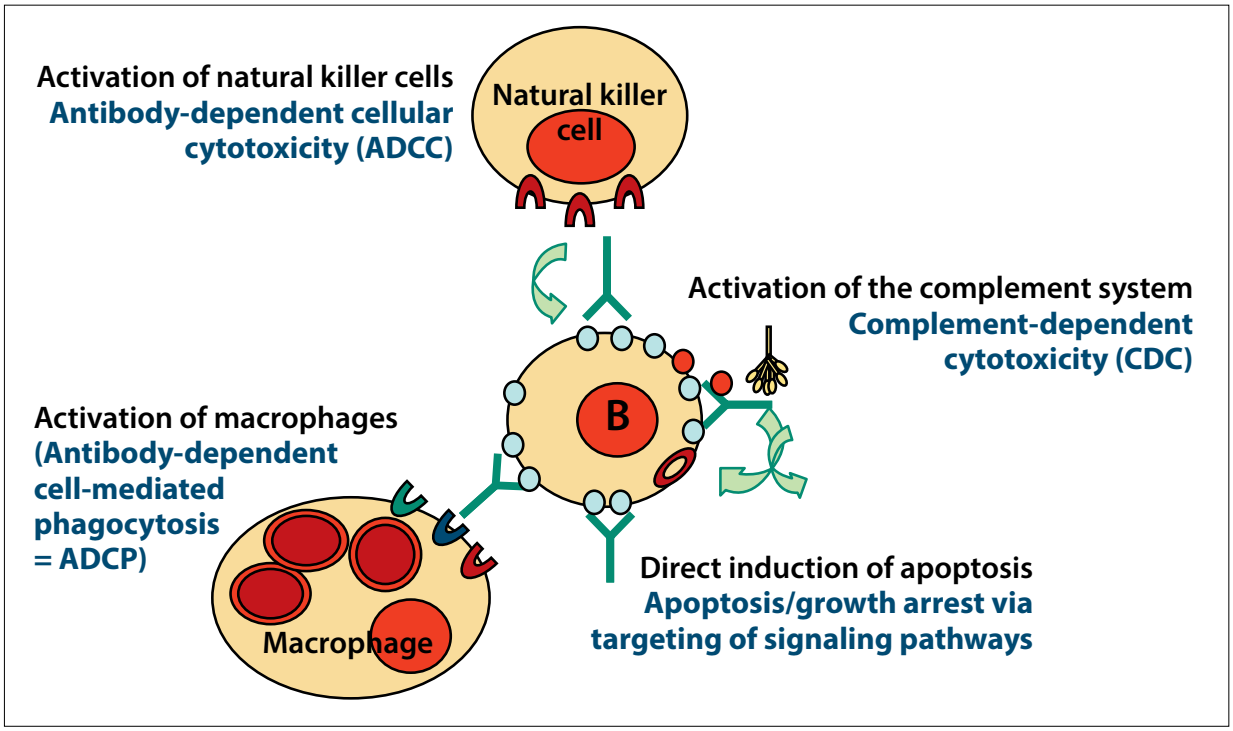

Figure. Mechanism of action of monoclonal antibodies.

Adapted with permission from Golay J et al. *Arch Biochem Biophys.* 2012;526(2):146-153.[31]

events (AEs) included cough, headache, back pain, fever, and chills. In general, AEs were mild to moderate in severity. The infusion-related AEs diminished once a premedication regimen was implemented.

In 2 following phase 1 dose-escalation trials, elotuzumab was combined with lenalidomide and dexamethasone or bortezomib, and encouraging results were obtained.[41,42] In the study by Lonial and colleagues, 28 patients with advanced MM who had received a median of 3 prior MM therapies were treated with elotuzumab, lenalidomide, and dexamethasone.[41] The combination showed encouraging activity, with an objective response rate of 82%. At a median follow-up of 16.4 months, the median time to progression (TTP) was not reached for patients in the 20-mg/kg cohort, who were treated until disease progression. Two patients experienced serious infusion reactions during cycle 1, and the most frequent grade 3 or 4 toxicities were neutropenia (36%) and thrombocytopenia (21%). In the study by Jakubowiak and colleagues, 28 patients received elotuzumab in combination with bortezomib.[42] The objective response rate was 48%, and the median TTP was 9.46 months. Notably, two of 3 bortezomib-refractory patients responded. Lymphopenia and fatigue were the most frequent grade 3 or 4 AEs, at 25% and 14%, respectively.

The positive results observed in these trials were the basis for a phase 2 study investigating the combination of elotuzumab, lenalidomide, and low-dose dexamethasone in patients with relapsed/refractory MM following 1 to 3 prior regimens. The final results of the study were presented at the 2014 annual meeting of the American Society of Hematology (ASH).[43] Seventy-three patients were randomly assigned to receive elotuzumab at 10 or 20 mg/kg plus lenalidomide at 25 mg on days 1 through 21 and dexamethasone at 40 mg once weekly in 28-day dosing cycles. Treatment was continued until disease progression. The overall response rate (ORR) was 84% in the overall group, and 92% and 76% in the 10-mg/kg and 20-mg/kg groups, respectively. The median progression-free survival (PFS) also was longer in the 10-mg/kg group (32.5 months) compared with the 20-mg/kg group (25 months); the PFS for the overall group was 28.8 months. Based on these results, the 10-mg/kg dose will be taken forward in trials. The most common grade 3 or 4 AEs were diarrhea, anemia, hyperglycemia, lymphopenia, thrombocytopenia, and neutropenia. Infusion reactions were noted in 11% of patients; these were of grade 1 or 2 severity and consisted mainly of pyrexia, nausea, and rash. In an attempt to reduce the infusion time of elotuzumab, the flow rate was increased to 5 mL/min, resulting in an infusion time of less than 1 hour. This was possible for 33% of the infusions.

Recently, Lonial and colleagues reported the results of a large phase 3 trial in which the combination of elotuzumab, lenalidomide, and dexamethasone was shown

to be significantly more effective in PFS and ORR than lenalidomide and dexamethasone alone in patients with relapsed or refractory MM following 1 to 3 prior lines of therapy (ELOQUENT-2).[44] The median PFS was 19.4 months in the elotuzumab group (n=321), compared with 14.9 months in the control group (n=325) (P<.001), and the ORR was 79% vs 66%, respectively (P<.001). In both groups, common grade 3 or 4 AEs were lymphopenia, neutropenia, fatigue, and pneumonia. Infusion reactions were observed in 10% of patients in the elotuzumab group; these were mainly grade 1 or 2 and only 2 patients discontinued treatment owing to infusion reactions. The combination of elotuzumab, lenalidomide, and dexamethasone also is being investigated in a randomized phase 3 trial in the front-line setting (NCT01335399), and a number of other combination studies are ongoing, both in the relapsed/refractory and front-line setting.

CD38

Human CD38 is a 45-kDa single-chain transmembrane glycoprotein with a short amino-terminal cytoplasmic tail, a single membrane-spanning region, and a long extracellular carboxy-terminal domain.[45] CD38 was first described in 1980 and was initially used as a phenotypic marker of differentiation in normal and leukemic blood cells because of its distinct pattern of expression, being predominantly expressed by progenitors and early hematopoietic cells, then lost during maturation, only to be re-expressed during cell activation.[46] However, CD38 is now recognized to combine a multitude of activities, including adhesion, receptor function, and enzymatic functions.[44,47,48] CD38 is an ectoenzyme, a membrane protein exerting its catalytic function on the external surface of the cell membrane.[49] CD38 catalyzes the conversion of nicotinamide adenine dinucleotide (NAD)+ to adenosine diphosphate ribose (ADPR) and nicotinic acid adenine dinucleotide phosphate (NAADP), and thus is involved in the mobilization of calcium. As a receptor, it is involved in immune processes, mediating the production of cytokines by effector cells, the proliferation of T lymphocytes, and the protection of mature B lymphocytes and dendritic cells from apoptosis.[50]

CD38 is expressed at high levels by committed progenitor bone marrow cells, B lymphocytes in germinal centers, terminally differentiated plasma cells, and activated tonsils. Early bone marrow cells are CD38-negative, and mature virgin and memory B lymphocytes express low levels of the molecule.[51] Myeloma cells express CD38 in a large majority of patients, although at varying surface densities.[51-53] CD38 also is involved in a pathway leading to the production of adenosine, which is an important regulator of multiple biological functions in the tumor microenvironment, including local immunologic tolerance. CD38 is thus implicated to be part of the local survival strategy of the neoplastic plasma cell in the bone marrow milieu.[49,51] Because of its almost universal expression and its importance for the survival of the plasma cells, CD38 has been identified as an attractive target for mAb therapy in myeloma, not only because of the induction of antibody-mediated effects, but also because of the potential effects of a functional block of its enzymatic activity.[51] Interestingly, CD38 also is overexpressed by the majority of acute lymphoblastic leukemias and by a proportion of acute myeloid leukemias, non-Hodgkin lymphomas, and chronic lymphocytic leukemias (CLLs). In CLL, CD38 expression also can carry prognostic information.[51]

Daratumumab

Daratumumab (HuMax-CD38) is a human CD38 IgG1 mAb that was generated by immunizing transgenic mice possessing human immunoglobulin genes. Daratumumab can effectively kill tumor cells isolated from patients with MM and myeloma-derived cells lines by ADCC, ADCP, and CDC.[54,55] Another mechanism of action is induction of apoptosis upon secondary crosslinking.[56] Daratumumab also was active at low concentrations in a severe combined immunodeficiency (SCID) mouse xenograft tumor model.[54]

Preclinical studies demonstrated a clear synergy between lenalidomide or bortezomib and daratumumab in inducing ADCC.[57,58] Furthermore, the addition of daratumumab to RVD (lenalidomide, bortezomib, and dexamethasone) or MPV (melphalan, prednisone, and bortezomib) significantly increased the activity of these combinations in lysis assays. Of note, a recent study provided the first preclinical evidence for the benefit of combining daratumumab and lenalidomide in patients with myeloma refractory to lenalidomide and bortezomib.[59] Daratumumab induced lysis of lenalidomide- and bortezomib-resistant cell lines and of primary myeloma cells derived from patients with disease refractory to lenalidomide and/or bortezomib. Moreover, lenalidomide synergistically enhanced the daratumumab-mediated lysis of MM cells through the activation of NK cells. Recently, Nijhof and colleagues showed that the efficacy of daratumumab-induced NK cell–mediated ADCC may be further enhanced by the modulation of NK-cell regulatory signals transmitted via the inhibitory and activating NK receptors (killer cell immunoglobulin-like receptors [KIRs]).[60] They used the human monoclonal anti-KIR antibody IPH2102 to block NK cell inhibitory receptors while activating NK cells with lenalidomide, and demonstrated synergistically improved myeloma cell lysis.

Another interesting observation from preclinical studies is that CD38 expression levels can be induced by all-trans retinoic acid (ATRA).[61-63] In a recent study, Nijhof and colleagues showed that ATRA enhanced daratumumab-mediated ADCC and CDC through an increase in CD38 expression levels, providing a rationale for the further evaluation of combining daratumumab and ATRA.[64]

Daratumumab is currently undergoing clinical investigation, and results from ongoing trials confirm the positive results obtained in vitro. In a phase 1/2 trial, patients with relapsed/refractory MM who had received 2 or more prior lines of therapy were treated with single-agent daratumumab administered in a dose-escalation scheme.[65] Patients had received a median of 5.5 prior therapies, and 75% of patients were refractory to both lenalidomide and bortezomib. In part 1 of the study, a dose escalation of daratumumab from 0.005 mg/kg to 24 mg/kg was implemented, administered intravenously (IV) once weekly. Daratumumab was well tolerated, and in 12 patients who received the agent at 4 to 24 mg/kg, a partial response (PR) as best response was seen in 5 patients. In the second part of the study, 2 dosing cohorts were selected: 8 mg/kg and 16 mg/kg; 30 and 20 patients were treated, respectively. The ORR was higher in the 16-mg/kg cohort compared with the 8-mg/kg cohort (35% vs 10%, respectively). Of note, no severe infusion-related reactions were seen (all grade 1 and 2) and the infusion times could be reduced to approximately 3.4 hours with the third infusion. The most frequent grade 3 or 4 AEs were thrombocytopenia (13%), neutropenia (10%), pneumonia (10%), and hyperglycemia (7%), and the most frequent serious AEs were pneumonia (10%) and lymphopenia (10%).

At the 2015 annual meeting of the American Society of Clinical Oncology (ASCO), Lonial and colleagues presented the results of an international phase 2 study investigating daratumumab monotherapy in 106 patients who had received a median of 5 prior lines of therapy and with disease refractory to a proteasome inhibitor and an immunomodulatory drug (IMiD).[66] Daratumumab dosed at 16 mg/kg resulted in an ORR of 29% (3 stringent complete responses [sCRs], 10 very good partial responses [VGPRs], 18 PRs) with a median PFS of 3.7 months and a 1-year survival rate of 65%. Daratumumab was well tolerated, with no discontinuations due to daratumumab. Infusion-related reactions occurred predominantly during the first infusion; they were usually grade 1 or 2 and were manageable.

In another phase 1/2 study involving patients with relapsed and refractory MM following 2 to 4 prior lines of therapy, daratumumab was combined with lenalidomide and dexamethasone.[67] Daratumumab was dose-escalated from 2 to 16 mg/kg in part 1 of the study (n=13) and then given at 16 mg/kg in the expansion cohort (n=30). With a mean follow-up duration of 12.9 months for part 1 and 5.6 months for part 2, the overall best response was 100% in part 1 (31% complete response [CR] rate, 46% VGPR rate, 23% PR rate) and 86.7% in part 2 (6.7% CR rate, 43% VGPR rate, 37% PR rate). Responses in part 2 improved over time, with 75% of patients treated for at least 6 months reaching VGPR or better. The combination showed a favorable safety profile, and an accelerated infusion was tolerable but associated with a higher incidence of grade 1 or 2 AEs.

Daratumumab also has been investigated in a small phase 1b study in combination with currently used regimens.[68] Patients with newly diagnosed MM received daratumumab plus bortezomib and dexamethasone; bortezomib, melphalan, and prednisone; or bortezomib, thalidomide (Thalomid, Celgene), and dexamethasone. Patients with relapsed/refractory MM were treated with the combination of pomalidomide (Pomalyst, Celgene), dexamethasone, and daratumumab. In 18 patients treated at the time of reporting, the ORR was 100% in the newly diagnosed group and 50% in the relapsed group. In addition, the combinations were well tolerated, and daratumumab did not result in significant additional toxicity. Furthermore, daratumumab did not have a negative impact on stem cell mobilization.

Based on the promising results from early clinical trials, daratumumab was granted breakthrough drug status by the FDA in 2013.[69] Phase 3 trials in the relapsed/refractory and upfront setting are currently ongoing, and the results of these studies are eagerly awaited (NCT02076009, NCT02076009, NCT02195479).

SAR650984 (SAR, Isatuximab)

SAR is a chimeric CD38 IgG1 mAb. In vitro, SAR possesses potent antimyeloma activity against primary MM cells and MM tumor cell lines, acting through ADCC, CDC, ADCP, and direct apoptosis.[70] In addition, SAR inhibits the ectoenzymatic activity of CD38 and tumor growth in xenograft tumor models.[70,71]

In a first-in-human phase 1 dose-escalation trial, 40 patients with heavily pretreated relapsed/refractory MM received SAR at doses ranging from 0.0001 to 20 mg/kg IV every week or every 2 weeks.[72] Patients had received a median of 6.5 prior therapies, including bortezomib and IMiDs. In the preliminary report, 33% of patients obtained a clinical benefit and the ORR was 27% in the overall patient group (all doses). At SAR doses of 10 mg/kg or greater, 37% of patients obtained a clinical benefit (≥ minimal response [MR]) and the ORR was 32%. Overall, SAR was well tolerated. The most common treatment-emergent AEs were fatigue and nausea, and the most common grade 3 or 4 AE was pneumonia. Infusion reactions were predominantly grade 1 or 2 and, following the implementation of prophylactic treatment for infusion reactions, these were limited to the first cycle.

SAR also has been investigated in combination with lenalidomide and dexamethasone in a study that was updated at the 2014 ASH annual meeting.[73] The combination of lenalidomide with SAR offers potential synergistic activity through concerted direct antimyeloma activity and an increase in ADCC mediated through lenalidomide.[74] The phase 1b study included 31 patients with relapsed/refractory MM who had received a median of 7 prior therapies and of

whom 84% had relapsed/refractory disease following IMiD therapy.[73] In the dose-escalation phase of the trial, patients received SAR at 3, 5, or 10 mg/kg plus lenalidomide and dexamethasone. With mandatory prophylactic treatment, most infusion reactions were grade 1 or 2 and occurred in cycle 1. Two patients had to discontinue owing to grade 3 infusion reactions, which resolved in both patients. Treatment-emergent AEs were predominantly grade 1 or 2, with the exception of hematologic AEs and pneumonia. Overall, the safety findings were consistent with those of the individual agents. In the overall group, the ORR was 58% (6% sCR, 23% VGPR, 29% PR), and at SAR dosing levels of 10 mg/kg (n=24), the ORR was 63% (8% sCR, 29% VGPR, 25% PR). Of note, in patients with disease relapsed and refractory to IMiDs (n=26), the ORR was 50%. With a median follow-up of 9 months, the overall median PFS was 6.2 months. The encouraging activity seen in this heavily pretreated population is the basis for ongoing trials, including combinations with carfilzomib (Kyprolis, Onyx) and pomalidomide (NCT02332850, NCT02283775).

MOR03087 (MOR202; MOR)

MOR (HuCAL) is a fully human CD38 IgG1 mAb that has demonstrated potent antimyeloma activity in preclinical studies.[75,76] MOR was found to kill CD38-expressing cell lines and primary MM cells from patients by ADCC in a concentration-dependent manner.[75] In addition, ADCP is a potent effector mechanism of MOR. The addition of lenalidomide to MOR enhanced ADCP-mediated killing in vitro in a synergistic manner, providing the rationale for combination of the 2 drugs.[76] Furthermore, MOR was shown to inhibit tumor growth in SCID-mouse xenograft models.[75] MOR is currently being evaluated in a phase 1/2 trial in relapsed/refractory MM. At the 2015 ASCO meeting, preliminary results of a dose-escalating phase 1/2 trial of MOR in patients with relapsed/refractory MM who had a median of 4 prior therapies were presented.[77] In 42 patients treated so far, encouraging activity with acceptable toxicity has been reported, and the results of further dosing cohorts are awaited.

CD56

CD56, also termed neuronal cell adhesion molecule (N-CAM), is a membrane glycoprotein that is found on muscle cells and neurons and is thought to be involved in cell adhesion, migration, invasion, and survival.[78-81] Expression of CD56 has been noted on a variety of cancer cells, including small cell lung cancer, neuroblastoma, other neuroendocrine tumors, and ovarian cancer.[82,83] Within the hematopoietic compartment, CD56 is normally restricted to NK cells and a subset of T lymphocytes, and is not found on normal plasma cells.[84,85] However, CD56 is expressed on 70% to 90% of myeloma cells,[20,86-88] where it is involved in disease progression and correlates with the extent of bone disease.[89,90]

Lorvotuzumab Mertansine (IMGN901)

A humanized CD56 mAb-maytansinoid conjugate has been constructed, comprised of a tumor-targeting antibody coupled by a linker to a potent antimicrotubular cytotoxic agent (DM1).[79,91] Maytansine, originally derived from the Ethiopian shrub *Maytenus serrata*, inhibits tubulin polymerization and is approximately 200- to 1000-fold more cytotoxic than the vinca alkaloids.[92,93] The narrow therapeutic window of maytansine precludes the application of the agent on its own; however, some have hypothesized that conjugation to an antibody to facilitate intracellular delivery would expand the therapeutic window and enable the exploitation of the cytotoxic potency, leading to effective tumor cell killing with reduced toxicity. After binding to the target tumor cell, the antibody-maytansinoid conjugate is internalized and metabolized, and active maytansinoid metabolites are released, which results in the killing of the tumor cell.[94]

In preclinical studies, lorvotuzumab demonstrated antimyeloma activity in MM cell line assays, patient MM samples, and human MM xenograft models.[93] Of note, the adhesion of MM cell lines and patient MM cells to bone marrow stromal cells did not protect against lorvotuzumab cytotoxicity. In preclinical studies, additive or synergistic activity was observed when lorvotuzumab was combined with lenalidomide, bortezomib, or melphalan, providing the rationale for clinical combination studies.[95,96]

In a phase 1 single-agent study, lorvotuzumab was investigated in patients with heavily pretreated (median, 6 lines) relapsed/refractory CD56-positive MM.[97] The agent was given IV at doses ranging from 40 to 140 mg/m^2/week for 2 consecutive weeks, with cycles repeating every 3 weeks. At the last reporting, 37 patients had been treated. Four grade 3 drug-related toxicities were reported: fatigue, renal failure, weakness, and absence of deep tendon reflexes. No grade 4 drug-related toxicities were noted and no patients demonstrated a humoral response against either the antibody or the DM1 component. Clinical benefit (≥SD for at least 3 months) was seen in 46% of patients (2 PRs, 3 MRs).

Based on the observation of synergistic activity in preclinical studies, lorvotuzumab also has been combined with lenalidomide and dexamethasone in a phase 1 study in 44 patients with CD56+ relapsed/refractory MM.[98] Lorvotuzumab was given in escalating doses (75 mg/m^2, 90 mg/m^2, and 112 mg/m^2). In 39 patients evaluable for response, the ORR (≥PR) was 56.4% (≥MR, 64.1%; 1 sCR, 11 VGPRs, 10 PRs, 3 MRs). The TTP was 7.7 months in the dosing group selected for further development (75 mg/m^2). The most common treatment-related AEs were peripheral neuropathy (PN), fatigue, neutropenia, thrombocytopenia,

nausea, and diarrhea. Most reports of PN were grade 2 or less, with the majority of patients having a grade 1 PN at baseline. Nonetheless, PN was the single most common cause of dose reduction. It has been suggested that the neuropathy may be due to low systemic levels of cytotoxin, which are released following the cellular catabolism of the antibody-drug conjugates and may thus be a class effect of such conjugates.[91]

Taken together, the positive results seen in these early clinical studies support the continued evaluation of lorvotuzumab in relapsed/refractory MM.[91]

CD138

CD138 (syndecan-1) is a member of the transmembrane heparan sulfate proteoglycan family. As an extracellular matrix receptor, it is involved in cell-cell and cell-matrix adhesion. CD138 is typically found on mature epithelial cells.[99,100] Within the hematopoietic system, CD138 is restricted to plasma cells with no expression on hematopoietic stem cells.[101,102]

BT062 (Indatuximab Ravtansine)

BT062 is a murine/human chimeric CD138 IgG4 mAb that is conjugated with a highly cytotoxic maytansinoid derivative (DM4), a potent microtubule-targeted compound that inhibits the proliferation of cells at the stage of mitosis.[79,103] Once bound to CD138 on target cells, the mAb-maytansinoid conjugate is internalized and the cytotoxic agent released, leading to the death of the target cell. In preclinical experiments, BT062 inhibited the growth of MM cell lines and primary tumor cells from MM patients, without cytotoxicity against peripheral blood mononuclear cells from healthy volunteers.[104] The drug also inhibited tumor growth in xenograft mouse models of human MM.[104]

In the first-in-human phase 1 study, 32 patients with heavily pretreated relapsed/refractory MM received BT062 once every 3 weeks at different dose levels.[105] SD or better was noted in 52% of patients (1 PR, 2 MRs, 11 SDs). Most AEs were mild to moderate and were typical of the disease and the patient population. As expected, owing to the expression of CD138 by tissues of epithelial origin, toxicities involving the skin and/or mucosa and the eye were observed. The vast majority of these were grade 1 or 2, although at higher dose levels, toxicity involving the skin or mucosa was also of grade 2 or 3. In a subsequent phase 1/2 study, a more frequent dosing schedule with BT062 was investigated.[106] In this study, BT062 was administered IV on days 1, 8, and 15 of a 4-week cycle using different dosing levels in heavily pretreated patients (n=31) who had received a median of 5 prior therapies. The dosing schedule with a higher frequency was well tolerated. The most frequently reported AEs were anemia, diarrhea, and fatigue. Overall, toxicity was mild to moderate. Also in this study, substantial antitumor activity was observed, with SD or better achieved by 56% (1 PR, 3 MRs, 11 SDs) and a median PFS of 121 days.

BT062 also has been combined with lenalidomide and dexamethasone in a phase 1/2 dose-escalation study in patients with relapsed/refractory MM who had failed at least 1 prior therapy.[107] In this study, 45 patients received BT062 at 80 mg/m^2, 100 mg/m^2, or 120 mg/m^2 plus lenalidomide and dexamethasone. At the time of reporting at the 2014 ASH annual meeting, the ORR in 36 evaluable patients was 78% (1 sCR, 2 CRs, 10 VGPRs, 15 PRs). Of note, responses were seen in lenalidomide-refractory patients. The most common AEs were diarrhea, fatigue, nausea, and hypokalemia. These positive results require confirmation in larger clinical trials, but suggest that BT062 could offer a novel effective therapeutic option in MM.

Discussion

The introduction of mAbs presents the most significant advancement of myeloma therapy in recent years. It is hoped that their incorporation into current treatment strategies for MM will emulate the progress of mAb addition to lymphoma therapy. Currently, few results from clinical trials are available, with elotuzumab being the mAb that has progressed farthest in clinical development. All of the mAbs discussed in this review have demonstrated encouraging activity when combined with current standard therapies, and some of the challenges are how best to incorporate the mAbs into existing strategies (as part of combinations or as monotherapy) and for which stages (whether they should be used in the maintenance setting or even for the treatment of smoldering myeloma). Based on the impressive activity seen in the relapsed/refractory setting, it is expected that more impressive efficacy results will be obtained in the frontline setting, where the likelihood of resistance or escape is much lower than in later lines of treatment.[23] The results of the phase 1 study by Moreau and colleagues of daratumumab administered upfront as part of MPV or RVD regimens in a very limited number of patients suggest that this may be the case.[67] One of the attractive features of the mAbs is the good tolerability that has been described in many studies, making the mAbs feasible therapeutic options for those patient groups for whom tolerability considerations often limit the application of active treatment. In some of the studies, a reduction of the infusion time, which presents a substantial practical challenge, was found to be feasible. This is an area that will require further optimization.

One of the issues in mAb therapy is certainly the limited understanding of some of the detailed mechanistic processes. For example, despite an almost universal expression of a surface molecule, single-agent activity of a specific mAb is limited, as is the case with elotuzumab.[40]

This may be related to differences among the mAbs in the relative importance of CDC, ADCC, ADCP, and direct effects of antibody-mediated killing.

It is well known that soluble forms of syndecan-1 (CD138), CS1, CD38, and CD56 exist.[36,45,109-111] For example, syndecan-1 is cleaved from the myeloma cell surface by heparanases, and the level of soluble syndecan-1 is a powerful prognostic factor in myeloma.[109-111] Soluble CD38 levels are elevated in MM patients.[108] Soluble CD56 carries prognostic information and can be used to differentiate between myeloma and paraproteinemias from other causes.[112,113] The presence of soluble forms of antigens may impact the outcome of treatment with mAbs. Binding of the mAb to the soluble form of the antigen may cause reduced activity of the mAb, and the formation of soluble antigen-antibody complexes may cause side effects due to the deposition of these immune-complexes at sites other than in the MM cells. These factors may explain unexpected results regarding efficacy or toxicity.

An important difference among the reviewed agents is the role of the mAb. For example, elotuzumab and the CD38 mAbs engage and augment the immune response; lorvotuzumab and BT062 deliver a cytotoxic agent into the tumor cell. This is reflected in the different effects that result from binding of the mAb to the target molecule. With elotuzumab, daratumumab, SAR, and MOR, the target molecules (CS1 for elotuzumab and CD38 for the others) are retained on the surface following ligand binding, whereas in the case of lorvotuzumab (CD56) and BT062 (CD138/syndecan-1), they are internalized together with the bound antibody conjugate. Therefore, it can be expected that elotuzumab and the CD38 mAbs continue to trigger immunologic antitumor effects following binding, whereas lorvotuzumab and BT062 are limited in their ability to engage the immune system itself. However, an important feature of antibody-drug conjugates is their ability to produce metabolites that are capable of diffusing into neighboring cells and thereby causing bystander killing.[79] Intense research efforts are ongoing to identify novel targets with optimal internalization kinetics and intracellular trafficking properties. Of note, some immunoconjugates are already finding application in the clinic. For example, brentuximab vedotin (Adcetris, Seattle Genetics), a CD30 mAb conjugated to the cytotoxic agent monomethyl auristatin E, is approved in Europe and the United States for the treatment of Hodgkin lymphoma and systemic anaplastic large cell lymphoma.

In myeloma therapy, the development of resistance to treatment presents an obstacle that severely limits treatment options. Cell adhesion–mediated drug resistance through the interaction of adhesion receptors with their ligands on bone marrow stromal cells and extracellular matrix proteins within the bone marrow presents an important de novo and acquired resistance mechanism.[114,115] mAbs may present an important strategy to evade this mechanism of resistance. For example, elotuzumab interferes with cell adhesion–mediated drug resistance.[29]

Taken together, mAbs present an exciting addition to current myeloma treatment strategies, and it is hoped that their incorporation will enable a more effective and targeted treatment of myeloma, resulting in improved outcomes for myeloma patients.

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References

1. Kazemi T, Younesi V, Jadidi-Niaragh F, Yousefi M. Immunotherapeutic approaches for cancer therapy: an updated review [published online March 24, 2015]. *Artif Cells Nanomed Biotechnol*. doi:10.3109/21691401.2015.1019669.
2. Ehrlich P. Croonian Lecture: On Immunity with Special Reference to Cell Life. *Proc. R. Soc. Lond.* 1899;66:424-433
3. Strebhardt K, Ullrich A. Paul Ehrlich's magic bullet concept: 100 years of progress. *Nat Rev Cancer*. 2008;8(6):473-480.
4. Zielinska E. Side-chain theory, circa 1900. *Scientist*. July 1, 2013.
5. Köhler G, Milstein C. Continuous cultures of fused cells secreting antibody of predefined specificity. *Nature*. 1975;256(5517):495-497.
6. Bernard A, Boumsell L. The clusters of differentiation (CD) defined by the First International Workshop on Human Leucocyte Differentiation Antigens. *Hum Immunol*. 1984;11(1):1-10.
7. Zola H, Swart B, Banham A, et al. CD molecules 2006—human cell differentiation molecules. *J Immunol Methods*. 2007;319(1-2):1-5.
8. Miller RA, Maloney DG, Warnke R, Levy R. Treatment of B-cell lymphoma with monoclonal anti-idiotype antibody. *N Engl J Med*. 1982;306(9):517-522.
9. Grillo-López AJ, White CA, Dallaire BK, et al. Rituximab: the first monoclonal antibody approved for the treatment of lymphoma. *Curr Pharm Biotechnol*. 2000;1(1):1-9.
10. Stashenko P, Nadler LM, Hardy R, Schlossman SF. Characterization of a human B lymphocyte-specific antigen. *J Immunol*. 1980;125(4):1678-1685.
11. Anderson KC, Bates MP, Slaughenhoupt BL, Pinkus GS, Schlossman SF, Nadler LM. Expression of human B cell-associated antigens on leukemias and lymphomas: a model of human B cell differentiation. *Blood*. 1984;63(6):1424-1433.
12. Zhou L-J, Tedder TF. CD20 Workshop Panel Report. In: Schlossman SF, Boumsell L, Gilks W, et al, eds. *Leucocyte Typing V. White Cell Differentiation Antigens.* Oxford, United Kingdom: Oxford University; 1995:511-514.
13. McLaughlin P, Grillo-López AJ, Link BK, et al. Rituximab chimeric anti-CD20 monoclonal antibody therapy for relapsed indolent lymphoma: half of patients respond to a four-dose treatment program. *J Clin Oncol*. 1998;16(8):2825-2833.
14. Coiffier B, Lepage E, Briere J, et al. CHOP chemotherapy plus rituximab compared with CHOP alone in elderly patients with diffuse large-B-cell lymphoma. *N Engl J Med*. 2002;346(4):235-242.
15. Pfreundschuh M, Trümper L, Osterborg A, et al; MabThera International Trial Group. CHOP-like chemotherapy plus rituximab versus CHOP-like chemotherapy alone in young patients with good-prognosis diffuse large-B-cell lymphoma: a randomised controlled trial by the MabThera International Trial (MInT) Group. *Lancet Oncol*. 2006;7(5):379-391.
16. van Oers MH, Klasa R, Marcus RE, et al. Rituximab maintenance improves clinical outcome of relapsed/resistant follicular non-Hodgkin lymphoma in patients both with and without rituximab during induction: results of a prospective randomized phase 3 intergroup trial. *Blood*. 2006;108(10):3295-3301.
17. Marcus R, Imrie K, Solal-Celigny P, et al. Phase III study of R-CVP compared with cyclophosphamide, vincristine, and prednisone alone in patients with previously untreated advanced follicular lymphoma. *J Clin Oncol*. 2008;26(28):4579-4586.

18. Salles G, Seymour JF, Offner F, et al. Rituximab maintenance for 2 years in patients with high tumour burden follicular lymphoma responding to rituximab plus chemotherapy (PRIMA): a phase 3, randomised controlled trial. *Lancet*. 2011;377(9759):42-51.
19. Sehn LH, Donaldson J, Chhanabhai M, et al. Introduction of combined CHOP plus rituximab therapy dramatically improved outcome of diffuse large B-cell lymphoma in British Columbia. *J Clin Oncol*. 2005;23(22):5027-5033.
20. Harada H, Kawano MM, Huang N, et al. Phenotypic difference of normal plasma cells from mature myeloma cells. *Blood*. 1993;81(10):2658-2663.
21. Zojer N, Kirchbacher K, Vesely M, Hübl W, Ludwig H. Rituximab treatment provides no clinical benefit in patients with pretreated advanced multiple myeloma. *Leuk Lymphoma*. 2006;47(6):1103-1109.
22. Kapoor P, Greipp PT, Morice WG, Rajkumar SV, Witzig TE, Greipp PR. Anti-CD20 monoclonal antibody therapy in multiple myeloma. *Br J Haematol*. 2008;141(2):135-148.
23. Richardson PG, Lonial S, Jakubowiak AJ, Harousseau JL, Anderson KC. Monoclonal antibodies in the treatment of multiple myeloma. *Br J Haematol*. 2011;154(6):745-754.
24. Robillard N, Avet-Loiseau H, Garand R, et al. CD20 is associated with a small mature plasma cell morphology and t(11;14) in multiple myeloma. *Blood*. 2003;102(3):1070-1071.
25. Treon SP, Pilarski LM, Belch AR, et al. CD20-directed serotherapy in patients with multiple myeloma: biologic considerations and therapeutic applications. *J Immunother*. 2002;25(1):72-81.
26. Yang J, Yi Q. Therapeutic monoclonal antibodies for multiple myeloma: an update and future perspectives. *Am J Blood Res*. 2011;1(1):22-33.
27. van de Donk NW, Kamps S, Mutis T, Lokhorst HM. Monoclonal antibody-based therapy as a new treatment strategy in multiple myeloma. *Leukemia*. 2012;26(2):199-213.
28. Allegra A, Penna G, Alonci A, et al. Monoclonal antibodies: potential new therapeutic treatment against multiple myeloma. *Eur J Haematol*. 2013;90(6):441-468.
29. Kuroda J, Nagoshi H, Shimura Y, Taniwaki M. Elotuzumab and daratumumab: emerging new monoclonal antibodies for multiple myeloma. *Expert Rev Anticancer Ther*. 2013;13(9):1081-1088.
30. Weiner GJ. Monoclonal antibody mechanisms of action in cancer. *Immunol Res*. 2007;39(1-3):271-278.
31. Golay J, Introna M. Mechanism of action of therapeutic monoclonal antibodies: promises and pitfalls of in vitro and in vivo assays. *Arch Biochem Biophys*. 2012;526(2):146-153.
32. Cannons JL, Tangye SG, Schwartzberg PL. SLAM family receptors and SAP adaptors in immunity. *Annu Rev Immunol*. 2011;29:665-705.
33. Veillette A. SLAM-family receptors: immune regulators with or without SAP-family adaptors. *Cold Spring Harb Perspect Biol*. 2010;2(3):a002469.
34. Hsi ED, Steinle R, Balasa B, et al. CS1, a potential new therapeutic antibody target for the treatment of multiple myeloma. *Clin Cancer Res*. 2008;14(9):2775-2784.
35. Veillette A, Guo H. CS1, a SLAM family receptor involved in immune regulation, is a therapeutic target in multiple myeloma. *Crit Rev Oncol Hematol*. 2013;88(1):168-177.
36. Tai YT, Dillon M, Song W, et al. Anti-CS1 humanized monoclonal antibody HuLuc63 inhibits myeloma cell adhesion and induces antibody-dependent cellular cytotoxicity in the bone marrow milieu. *Blood*. 2008;112(4):1329-1337.
37. van Rhee F, Szmania SM, Dillon M, et al. Combinatorial efficacy of anti-CS1 monoclonal antibody elotuzumab (HuLuc63) and bortezomib against multiple myeloma. *Mol Cancer Ther*. 2009;8(9):2616-2624.
38. Collins SM, Bakan CE, Swartzel GD, et al. Elotuzumab directly enhances NK cell cytotoxicity against myeloma via CS1 ligation: evidence for augmented NK cell function complementing ADCC. *Cancer Immunol Immunother*. 2013;62(12):1841-1849.
39. Balasa B, Yun R, Belmar NA, et al. Elotuzumab enhances natural killer cell activation and myeloma cell killing through interleukin-2 and TNF-α pathways. *Cancer Immunol Immunother*. 2015;64(1):61-73.
40. Zonder JA, Mohrbacher AF, Singhal S, et al. A phase 1, multicenter, open-label, dose escalation study of elotuzumab in patients with advanced multiple myeloma. *Blood*. 2012;120(3):552-559.
41. Lonial S, Vij R, Harousseau JL, et al. Elotuzumab in combination with lenalidomide and low-dose dexamethasone in relapsed or refractory multiple myeloma. *J Clin Oncol*. 2012;30(16):1953-1959.
42. Jakubowiak AJ, Benson DM, Bensinger W, et al. Phase I trial of anti-CS1 monoclonal antibody elotuzumab in combination with bortezomib in the treatment of relapsed/refractory multiple myeloma. *J Clin Oncol*. 2012;30(16):1960-1965.
43. Richardson PG, Jagannath S, Moreau P, et al. Final results for the 1703 phase 1b/2 study of elotuzumab in combination with lenalidomide and dexamethasone in patients with relapsed/refractory multiple myeloma [ASH abstract 302]. *Blood*. 2014;124(21)(suppl).
44. Lonial S, Dimopoulos M, Palumbo A, et al; ELOQUENT-2 Investigators. Elotuzumab Therapy for Relapsed or Refractory Multiple Myeloma [published online June 2, 2015]. *N Engl J Med*. doi:10.1056/NEJMoa1505654.
45. Mehta K, Shahid U, Malavasi F. Human CD38, a cell-surface protein with multiple functions. *FASEB J*. 1996;10(12):1408-1417.
46. Reinherz EL, Kung PC, Goldstein G, Levey RH, Schlossman SF. Discrete stages of human intrathymic differentiation: analysis of normal thymocytes and leukemic lymphoblasts of T-cell lineage. *Proc Natl Acad Sci U S A*. 1980;77(3):1588-1592.
47. Funaro A, Malavasi F. Human CD38, a surface receptor, an enzyme, an adhesion molecule and not a simple marker. *J Biol Regul Homeost Agents*. 1999;13(1):54-61.
48. Deaglio S, Mehta K, Malavasi F. Human CD38: a (r)evolutionary story of enzymes and receptors. *Leuk Res*. 2001;25(1):1-12.
49. Quarona V, Ferri V, Chillemi A, et al. Unraveling the contribution of ectoenzymes to myeloma life and survival in the bone marrow niche. *Ann N Y Acad Sci*. 2015;1335:10-22.
50. Fedele G, Di Girolamo M, Recine U, et al. CD38 ligation in peripheral blood mononuclear cells of myeloma patients induces release of protumorigenic IL-6 and impaired secretion of IFNγ cytokines and proliferation. *Mediators Inflamm*. 2013;2013:564687.
51. Chillemi A, Zaccarello G, Quarona V, et al. Anti-CD38 antibody therapy: windows of opportunity yielded by the functional characteristics of the target molecule. *Mol Med*. 2013;19:99-108.
52. Chan CS, Wormsley SB, Pierce LE, Peter JB, Schechter GP. B-cell surface phenotypes of proliferating myeloma cells: target antigens for immunotherapy. *Am J Hematol*. 1990;33(2):101-109.
53. Lin P, Owens R, Tricot G, Wilson CS. Flow cytometric immunophenotypic analysis of 306 cases of multiple myeloma. *Am J Clin Pathol*. 2004;121(4):482-488.
54. de Weers M, Tai YT, van der Veer MS, et al. Daratumumab, a novel therapeutic human CD38 monoclonal antibody, induces killing of multiple myeloma and other hematological tumors. *J Immunol*. 2011;186(3):1840-1848.
55. Overdijk MB, Verploegen S, Bögels M, et al. Antibody-mediated phagocytosis contributes to the anti-tumor activity of the therapeutic antibody daratumumab in lymphoma and multiple myeloma. *MAbs*. 2015;7(2):311-321.
56. Jansen JH, Boross P, Overdijk MB, van Bueren JJ, Parren PW, Leusen JH. Daratumumab, a human CD38 antibody induces apoptosis of myeloma tumor cells via Fc receptor-mediated crosslinking [ASH abstract 2974]. *Blood*. 2012;120(21)(suppl).
57. van der Veer MS, de Weers M, van Kessel B, et al. Towards effective immunotherapy of myeloma: enhanced elimination of myeloma cells by combination of lenalidomide with the human CD38 monoclonal antibody daratumumab. *Haematologica*. 2011;96(2):284-290.
58. van der Veer MS, de Weers M, van Kessel B, et al. The therapeutic human CD38 antibody daratumumab improves the anti-myeloma effect of newly emerging multi-drug therapies. *Blood Cancer J*. 2011;1(10):e41.
59. Nijhof IS, Groen RW, Noort WA, et al. Preclinical evidence for the therapeutic potential of CD38-targeted immuno-chemotherapy in multiple myeloma patients refractory to lenalidomide and bortezomib. *Clin Cancer Res*. 2015;21(12):2802-2810.
60. Nijhof IS, Lammerts van Bueren JJ, van Kessel B, et al. Daratumumab-mediated lysis of primary multiple myeloma cells is enhanced in combination with the human anti-KIR antibody IPH2102 and lenalidomide. *Haematologica*. 2015;100(2):263-268.
61. Mehta K, Ocanas L, Malavasi F, Marks JW, Rosenblum MG. Retinoic acid-induced CD38 antigen as a target for immunotoxin-mediated killing of leukemia cells. *Mol Cancer Ther*. 2004;3(3):345-352.
62. Uruno A, Noguchi N, Matsuda K, et al. All-trans retinoic acid and a novel synthetic retinoid tamibarotene (Am80) differentially regulate CD38 expression in human leukemia HL-60 cells: possible involvement of protein kinase C-delta. *J Leukoc Biol*. 2011;90(2):235-247.
63. Malavasi F. Editorial: CD38 and retinoids: a step toward a cure. *J Leukoc Biol*. 2011;90(2):217-219.
64. Nijhof IS, Groen RW, Lokhorst HM, et al. Upregulation of CD38 expression on multiple myeloma cells by all-trans retinoic acid improves the efficacy of daratumumab [published online May 15, 2015]. *Leukemia*. doi:10.1038/leu.2015.123.
65. Lokhorst HM, Laubach J, Nahi H, et al. Dose-dependent efficacy of daratumumab (DARA) as monotherapy in patients with relapsed or refractory multiple myeloma (RR MM) [ASCO abstract 8513]. *J Clin Oncol*. 2014:32(15)(suppl).
66. Lonial S, Weiss BM, Usmani SZ, et al. Phase II study of daratumumab (DARA) monotherapy in patients with ≥ 3 lines of prior therapy or double refractory multiple myeloma (MM): 54767414MMY2002 (Sirius) [ASCO abstract LBA8512]. *J Clin Oncol*. 2015;33(suppl).
67. Plesner T, Arkenau HT, Lokhorst HM, et al. Safety and efficacy of daratumumab with lenalidomide and dexamethasone in relapsed or relapsed, refractory multiple myeloma [ASH abstract 84]. *Blood*. 2014;124(21)(suppl).
68. Moreau P, Mateos MV, Bladé J, et al. An open-label, multicenter, phase 1b study of daratumumab in combination with backbone regimens in patients with multiple myeloma [ASH abstract 176]. *Blood*. 2014;124(21)(suppl).
69. Laubach JP, Tai YT, Richardson PG, Anderson KC. Daratumumab granted breakthrough drug status. *Expert Opin Investig Drugs*. 2014;23(4):445-452.

70. Deckert J, Wetzel MC, Bartle LM, et al. SAR650984, a novel humanized CD38-targeting antibody, demonstrates potent antitumor activity in models of multiple myeloma and other CD38+ hematologic malignancies. *Clin Cancer Res*. 2014;20(17):4574-4583.
71. Hann B, Cai T, Wang D, Hom YK, Aftab BT, Martin T, Wolf J. SAR650984, an anti-CD38 antibody, shows anti-tumor activity in a preclinical model of multiple myeloma. Poster presented at: 104th annual meeting of the American Association for Cancer Research; April 6-10 2013; Washington, DC. Abstract 5469.
72. Martin T, Strickland S, Glenn M, Hsu K, Charpentier E, Mikhael J. A phase I trial of SAR650984, a CD38 monoclonal antibody, in relapsed or refractory multiple myeloma [ASCO abstract 8532]. *J Clin Oncol.* 2014;32(suppl).
73. Martin TG, Baz R, Benson DM, Lendvai N, Campana F, Charpentier E, Vij R. A phase Ib dose escalation trial of SAR650984 (anti-CD-38 mAb) in combination with lenalidomide and dexamethasone in relapsed/refractory multiple myeloma [ASH abstract 83]. *Blood.* 2014;124(21)(suppl).
74. Wu L, Adams M, Carter T, et al. lenalidomide enhances natural killer cell and monocyte-mediated antibody-dependent cellular cytotoxicity of rituximab-treated CD20+ tumor cells. *Clin Cancer Res*. 2008;14(14):4650-4657.
75. Tesar M. Fully human antibody MOR202 against CD38 for the treatment of multiple myeloma and other blood-borne malignancies [ASCO abstract 8106]. *J Clin Oncol.* 2007;25(18)(suppl).
76. Endell J, Boxhammer R, Wurzenberger C, Ness D, Steidl S. The activity of MOR202, a fully human anti-CD38 antibody, is complemented by ADCP and is synergistically enhanced by lenalidomide in vitro and in vivo [ASH abstract 4018]. *Blood.* 2012;120(21)(suppl).
77. Raab MS, Goldschmidt H, Agis H, et al. A phase I/IIa study of the human anti-CD38 antibody MOR202 (MOR03087) in relapsed or refractory multiple myeloma (rrMM) [ASCO abstract 8574]. *J Clin Oncol*. 2015;33(suppl).
78. Mechtersheimer G, Staudter M, Möller P. Expression of the natural killer (NK) cell-associated antigen CD56(Leu-19), which is identical to the 140-kDa isoform of N-CAM, in neural and skeletal muscle cells and tumors derived therefrom. *Ann N Y Acad Sci*. 1992;650:311-316.
79. Lutz RJ, Whiteman KR. Antibody-maytansinoid conjugates for the treatment of myeloma. *MAbs*. 2009;1(6):548-551.
80. Amoureux MC, Cunningham BA, Edelman GM, Crossin KL. N-CAM binding inhibits the proliferation of hippocampal progenitor cells and promotes their differentiation to a neuronal phenotype. *J Neurosci*. 2000;20(10):3631-3640.
81. Prag S, Lepekhin EA, Kolkova K, et al. NCAM regulates cell motility. *J Cell Sci*. 2002;115(Pt 2):283-292.
82. Patel K, Moore SE, Dickson G, et al. Neural cell adhesion molecule (NCAM) is the antigen recognized by monoclonal antibodies of similar specificity in small-cell lung carcinoma and neuroblastoma. *Int J Cancer*. 1989;44(4):573-578.
83. Ohishi Y, Kaku T, Oya M, Kobayashi H, Wake N, Tsuneyoshi M. CD56 expression in ovarian granulosa cell tumors, and its diagnostic utility and pitfalls. *Gynecol Oncol*. 2007;107(1):30-38.
84. Griffin JD, Hercend T, Beveridge R, Schlossman SF. Characterization of an antigen expressed by human natural killer cells. *J Immunol*. 1983;130(6):2947-2951.
85. Lanier LL, Le AM, Civin CI, Loken MR, Phillips JH. The relationship of CD16 (Leu-11) and Leu-19 (NKH-1) antigen expression on human peripheral blood NK cells and cytotoxic T lymphocytes. *J Immunol*. 1986;136(12):4480-4486.
86. Van Camp B, Durie BG, Spier C, et al. Plasma cells in multiple myeloma express a natural killer cell-associated antigen: CD56 (NKH-1; Leu-19). *Blood*. 1990;76(2):377-382.
87. Rawstron AC, Owen RG, Davies FE, et al. Circulating plasma cells in multiple myeloma: characterization and correlation with disease stage. *Br J Haematol*. 1997;97(1):46-55.
88. Sahara N, Takeshita A, Shigeno K, et al. Clinicopathological and prognostic characteristics of CD56-negative multiple myeloma. *Br J Haematol*. 2002;117(4):882-885.
89. Ely SA, Knowles DM. Expression of CD56/neural cell adhesion molecule correlates with the presence of lytic bone lesions in multiple myeloma and distinguishes myeloma from monoclonal gammopathy of undetermined significance and lymphomas with plasmacytoid differentiation. *Am J Pathol*. 2002;160(4):1293-1299.
90. Damgaard T, Knudsen LM, Dahl IM, Gimsing P, Lodahl M, Rasmussen T. Regulation of the CD56 promoter and its association with proliferation, anti-apoptosis and clinical factors in multiple myeloma. *Leuk Lymphoma*. 2009;50(2):236-246.
91. Berdeja JG. Lorvotuzumab mertansine: antibody-drug-conjugate for CD56+ multiple myeloma. *Front Biosci (Landmark Ed)*. 2014;19:163-170.
92. Remillard S, Rebhun LI, Howie GA, Kupchan SM. Antimitotic activity of the potent tumor inhibitor maytansine. *Science*. 1975;189(4207):1002-1005.
93. Tassone P, Gozzini A, Goldmacher V, et al. In vitro and in vivo activity of the maytansinoid immunoconjugate huN901-N2′-deacetyl-N2′-(3-mercapto-1-oxopropyl)-maytansine against CD56+ multiple myeloma cells. *Cancer Res*. 2004;64(13):4629-4636.
94. Erickson HK, Park PU, Widdison WC, et al. Antibody-maytansinoid conjugates are activated in targeted cancer cells by lysosomal degradation and linker-dependent intracellular processing. *Cancer Res*. 2006;66(8):4426-4433.
95. Lutz R, Ab O, Foley K, et al. Efficacy of the huN901-DM1 conjugate in combination with antineoplastic agents against multiple myeloma cells in preclinical studies. Paper presented at: 98th Annual Meeting of the American Association for Cancer Research; April 14-18, 2007; Los Angeles, CA. Abstract 5577.
96. Whiteman K, Ab O, Bartle L, Foley K, Goldmacher V, Lutz R. Efficacy of IMGN901 (huN901-DM1) in combination with bortezomib and lenalidomide against multiple myeloma cells in preclinical studies. Paper presented at: 99th Annual Meeting of the American Association for Cancer Research; April 12-16, 2008; San Diego, CA. Abstract 2146.
97. Chanan-Khan A, Wolf J, Garcia J, et al. Efficacy analysis from phase I study of lorvotuzumab mertansine (IMGN901), used as monotherapy, in patients with heavily pre-treated CD56-positive multiple myeloma—a preliminary efficacy analysis [ASH abstract 1962]. *Blood.* 2011;118(21)(suppl).
98. Berdeja JG, Hernandez-Ilizaliturri F, Chanan-Khan A, et al. Phase I study of lorvotuzumab mertansine (LM, IMGN901) in combination with lenalidomide (Len) and dexamethasone (Dex) in patients with CD56-positive relapsed or relapsed/refractory multiple myeloma (MM) [ASH abstract 728]. *Blood.* 2012;120(21)(suppl).
99. Elenius K, Salmivirta M, Inki P, Mali M, Jalkanen M. Binding of human syndecan to extracellular matrix proteins. *J Biol Chem*. 1990;265(29):17837-17843.
100. Mali M, Jaakkola P, Arvilommi AM, Jalkanen M. Sequence of human syndecan indicates a novel gene family of integral membrane proteoglycans. *J Biol Chem*. 1990;265(12):6884-6889.
101. Sanderson RD, Lalor P, Bernfield M. B lymphocytes express and lose syndecan at specific stages of differentiation. *Cell Regul*. 1989;1(1):27-35.
102. Wijdenes J, Vooijs WC, Clément C, et al. A plasmocyte selective monoclonal antibody (B-B4) recognizes syndecan-1. *Br J Haematol*. 1996;94(2):318-323.
103. Oroudjev E, Lopus M, Wilson L, et al. Maytansinoid-antibody conjugates induce mitotic arrest by suppressing microtubule dynamic instability. *Mol Cancer Ther*. 2010;9(10):2700-2713.
104. Ikeda H, Hideshima T, Fulciniti M, et al. The monoclonal antibody nBT062 conjugated to cytotoxic Maytansinoids has selective cytotoxicity against CD138-positive multiple myeloma cells in vitro and in vivo. *Clin Cancer Res*. 2009;15(12):4028-4037.
105. Jagannath S, Chanan-Khan A, Heffner LT, et al. BT062, an antibody-drug conjugate directed against CD138, shows clinical activity in patients with relapsed or relapsed/refractory multiple myeloma [ASH abstract 305]. *Blood.* 2011;118(21)(suppl).
106. Heffner LT, Jagannath S, Zimmerman TM, et al. BT062, an antibody-drug conjugate directed against CD138, given weekly for 3 weeks in each 4 week cycle: safety and further evidence of clinical activity [ASH abstract 4042]. *Blood*. 2012;120(21)(suppl).
107. Kelly KR, Chanan-Khan A, Heffner LT, et al. Indatuximab ravtansine (BT062) in combination with lenalidomide and low-dose dexamethasone in patients with relapsed and/or refractory multiple myeloma: clinical activity in patients already exposed to lenalidomide and bortezomib [ASH abstract 4736]. 2014;124(21)(suppl).
108. Funaro A, Horenstein AL, Calosso L, et al. Identification and characterization of an active soluble form of human CD38 in normal and pathological fluids. *Int Immunol*. 1996;8(11):1643-1650.
109. Lovell R, Dunn JA, Begum G, et al; Working Party on Leukaemia in Adults of the National Cancer Research Institute Haematological Oncology Clinical Studies Group. Soluble syndecan-1 level at diagnosis is an independent prognostic factor in multiple myeloma and the extent of fall from diagnosis to plateau predicts for overall survival. *Br J Haematol*. 2005;130(4):542-548.
110. Yang Y, Yaccoby S, Liu W, et al. Soluble syndecan-1 promotes growth of myeloma tumors in vivo. *Blood*. 2002;100(2):610-617.
111. Yang Y, Macleod V, Bendre M, et al. Heparanase promotes the spontaneous metastasis of myeloma cells to bone. *Blood*. 2005;105(3):1303-1309.
112. Kaiser U, Oldenburg M, Jaques G, Auerbach B, Havemann K. Soluble CD56 (NCAM): a new differential-diagnostic and prognostic marker in multiple myeloma. *Ann Hematol*. 1996;73(3):121-126.
113. Ong F, Kaiser U, Seelen PJ, et al. Serum neural cell adhesion molecule differentiates multiple myeloma from paraproteinemias due to other causes. *Blood*. 1996;87(2):712-716.
114. Hideshima T, Mitsiades C, Tonon G, Richardson PG, Anderson KC. Understanding multiple myeloma pathogenesis in the bone marrow to identify new therapeutic targets. *Nat Rev Cancer*. 2007;7(8):585-598.
115. Chesi M, Bergsagel PL. Many multiple myelomas: making more of the molecular mayhem. *Hematology Am Soc Hematol Educ Program*. 2011:344-353.