



### THERAPEUTIC ANTIBODIES

# CD38 antibodies in multiple myeloma: back to the future

Niels W. C. J. van de Donk,<sup>1</sup> Paul G. Richardson,<sup>2</sup> and Fabio Malavasi<sup>3-5</sup>

<sup>1</sup>Department of Hematology, VU University Medical Center, Amsterdam, The Netherlands; <sup>2</sup>Jerome Lipper Myeloma Center, Department of Medical Oncology, Dana-Farber Cancer Institute, Harvard Medical School, Boston, MA; <sup>3</sup>Laboratory of Immunogenetics, Department of Medical Sciences and <sup>4</sup>Center for Experimental Research and Medical Studies (CeRMS), University of Turin, Turin, Italy; and <sup>5</sup>Department of Transplantation Immunology, University Hospital, Turin, Italy

**CD38 is highly and uniformly expressed on multiple myeloma (MM) cells, and at relatively low levels on normal lymphoid and myeloid cells, and in some tissues of nonhematopoietic origin. CD38 is a transmembrane glycoprotein with ectoenzymatic activity, and also functions as a receptor and adhesion molecule. Altogether, this has triggered the development of several CD38 antibodies including daratumumab (fully human), isatuximab (chimeric), and MOR202 (fully human). CD38 antibodies have pleiotropic mechanisms of action including Fc-dependent immune-effector mechanisms, direct apoptotic activity, and immunomodulatory effects by the elimination of CD38<sup>+</sup> immune-suppressor cells. CD38-targeting antibodies are generally well tolerated and induce partial response or better in ~30% of heavily pretreated MM patients as monotherapy. Based on their distinct mechanisms of action, favorable toxicity profile, and single-agent**

**activity, CD38 antibodies are attractive partners in combination regimens. Indeed, deep responses and prolonged progression-free survival can be achieved in relapsed/refractory MM patients when CD38 antibodies are combined with immunomodulatory agents or proteasome inhibitors. Infusion-related reactions, which typically occur during the first infusion, are the most frequent adverse events. Attention should also be paid to the interference of CD38 antibodies with certain laboratory assays, which may complicate response evaluation and blood compatibility testing. Several studies are currently examining the role of CD38-based therapies in newly diagnosed and high-risk smoldering MM. Furthermore, CD38 antibodies are currently also under investigation in other hematologic malignancies, including acute lymphoblastic leukemia, natural killer/T-cell lymphoma, and acute myeloid leukemia, as well as in solid tumors. (*Blood*. 2018;131(1):13-29)**

## Introduction

CD38 was identified in 1980 by E. L. Reinherz, S. Schlossman, and colleagues during their pioneering analysis of the human lymphocyte surface using monoclonal antibodies in search of the T-cell receptor.<sup>1,2</sup> One of the molecules identified, T10 (now CD38), initially served as marker for the study of thymocytes and activated T cells.<sup>3</sup> More than 35 years later, CD38-targeting antibodies are transforming multiple myeloma (MM) treatment because of their marked activity as single agents and in combinations, as well as their manageable toxicity profile. In this review, we will focus on CD38 function and tissue distribution, as well as on modes of action and mechanisms of resistance of CD38 antibodies. We will discuss the efficacy of CD38 antibodies both as single agents and in combination. The management of clinically relevant aspects of CD38 antibody therapy, including interference in laboratory assays, will also be discussed.

## CD38 tissue distribution

CD38 is highly expressed not only on plasma cells, but also on other lymphoid and myeloid cells, as well as on red blood cells and platelets.<sup>4</sup> A comparison of CD38 expression levels across these cellular populations shows that, after plasma cells, natural killer (NK) cells express the highest levels of CD38, followed by subpopulations of B and T cells.<sup>5</sup> The protein is also expressed in a

subset of hematological tumors, and shows especially broad and high expression levels in MM. CD38 is also expressed in tissues of nonhematopoietic origin including prostatic epithelial cells,<sup>6</sup> pancreatic islet cells, as well as in the perikarya and dendrites of some neurons. Other CD38<sup>+</sup> cells include airway-striated muscle cells,<sup>7</sup> renal tubules, retinal ganglionic cells, and corneal cells.<sup>8</sup>

## Control of CD38 expression

The promoter of CD38 is complex. The 5'-flanking promoter region of the gene contains cytosine-phosphate-guanine dinucleotide (CpG) islands and a binding site for transcription factor Sp1, but lacks canonical TATA and CAAT boxes. Upstream of these CpG islands are potential binding sites for various immunological transcription factors such as T-cell-specific transcription factor-1 $\alpha$  (TCF-1 $\alpha$ ), nuclear factor for interleukin-6 (NF-IL-6), and interferon-responsive factor-1 (IRF-1). A further level of control lies in the 5' end of intron 1, which contains a retinoic acid-responsive element (RARE) and a binding site for peroxisome proliferator-activated receptor  $\gamma$  (PPAR $\gamma$ ).<sup>9</sup>

## CD38 function

### CD38 as a receptor

CD38 was inferred to be a receptor after observations of signals induced by agonistic antibodies.<sup>10</sup> These signals were accessory

cell- and IL-2–dependent and additive with both CD3 and CD2. The CD38 molecule was later reported to be involved in triggering activation and proliferation signals that are lineage-unrestricted.<sup>11</sup> Since the identification of CD31 as a specific ligand,<sup>12</sup> CD38 has been generally recognized as a receptor, despite its intrinsic inability to act in this function, because of a very short cytoplasmic domain. Indeed, to act as a receptor, CD38 needs to be redirected to specialized microdomains of the cell membrane, in close proximity to professional receptors. This point has been confirmed by visually monitoring the formation of the immunological synapse in T lymphocytes.<sup>13</sup>

### CD38 as an ectoenzyme

CD38 is a multifunctional ectoenzyme involved in the catabolism of nicotinamide adenine dinucleotide (NAD<sup>+</sup>) and nicotinamide adenine dinucleotide phosphate (NADP), the 2 main substrates of the molecule identified so far.<sup>14,15</sup> This reaction leads to the generation of potent intracellular Ca<sup>2+</sup>-mobilizing compounds (cyclic adenosine diphosphate [ADP] ribose, ADP ribose, and nicotinic acid adenine dinucleotide phosphate). The balance between CD38 and its extracellular substrate determines cytoplasmic levels of NAD<sup>+</sup>. Consequently, inhibition of CD38 activity is followed by increased intracellular NAD<sup>+</sup> levels, independently of the actions of the other NAD<sup>+</sup>-consuming enzymes (CD157, poly ADP ribose polymerase, and sirtuins).<sup>16</sup> Recent studies suggest that CD38 is also involved in the production of adenosine, which has immunosuppressive effects.<sup>17</sup>

## Mechanism of action of CD38 antibodies

### Fc-dependent immune effector mechanisms

The relatively high expression of CD38 on MM cells, in combination with its role as receptor and ectoenzyme, suggested CD38 as a potential therapeutic antibody target for the treatment of MM. More than 25 years ago, several CD38-targeting antibodies were generated with activity against MM cells,<sup>18,19</sup> but these chimeric and humanized antibodies were not evaluated clinically (Figure 1). In 2008, the fully human immunoglobulin G1-κ (IgG1-κ) antibody daratumumab, was the first CD38 antibody that was administered in MM. Daratumumab was selected from a panel of 42 antibodies based on its unique ability to induce complement-dependent cytotoxicity (CDC) (Figure 2).<sup>20</sup> Daratumumab also kills MM cells via antibody-dependent cellular cytotoxicity (ADCC)<sup>20</sup> and antibody-dependent cellular phagocytosis (ADCP)<sup>21</sup> via antibody binding to activating Fcγ receptors (FcγRs) on immune effector cells. Furthermore, FcγR-mediated crosslinking of tumor-bound daratumumab induces programmed cell death.<sup>22</sup> Next to these Fc-dependent immune effector mechanisms, direct effects such as modulation of CD38 enzymatic function may also contribute to the antitumor activity of daratumumab.<sup>23</sup>

Similarly, isatuximab (IgG1-κ, chimeric) and MOR202 (IgG1-λ, fully human) have multiple mechanisms of action including CDC, ADCC, and ADCP.<sup>23,24</sup> Furthermore, isatuximab has strong proapoptotic activity independent of cross-linking and inhibits CD38 ectoenzyme function.<sup>23,25</sup> The relative contribution of these different mechanisms of action to MM cell killing differs among the 3 CD38 antibodies,<sup>23,25</sup> which can be explained by targeting different epitopes on the CD38 molecule (Figure 2).<sup>20,24</sup> It is currently

unknown whether these functional differences observed between different CD38 antibodies affect their therapeutic utility.

### Immunomodulatory effects

Interestingly, daratumumab also has immunomodulatory effects through the eradication of CD38-expressing regulatory T, regulatory B, and myeloid-derived suppressor cells (Figure 2).<sup>5</sup> This shift away from an immunosuppressive microenvironment results in CD4<sup>+</sup> and CD8<sup>+</sup> T-cell expansion, elevated antiviral and allergic functional T-cell responses, increased T-cell clonality, and, potentially, a better host-antitumor immune response.<sup>5</sup> Indeed, responders had higher expression levels of activation markers along with increased granzyme B production in CD8<sup>+</sup> T cells following daratumumab treatment.<sup>26</sup>

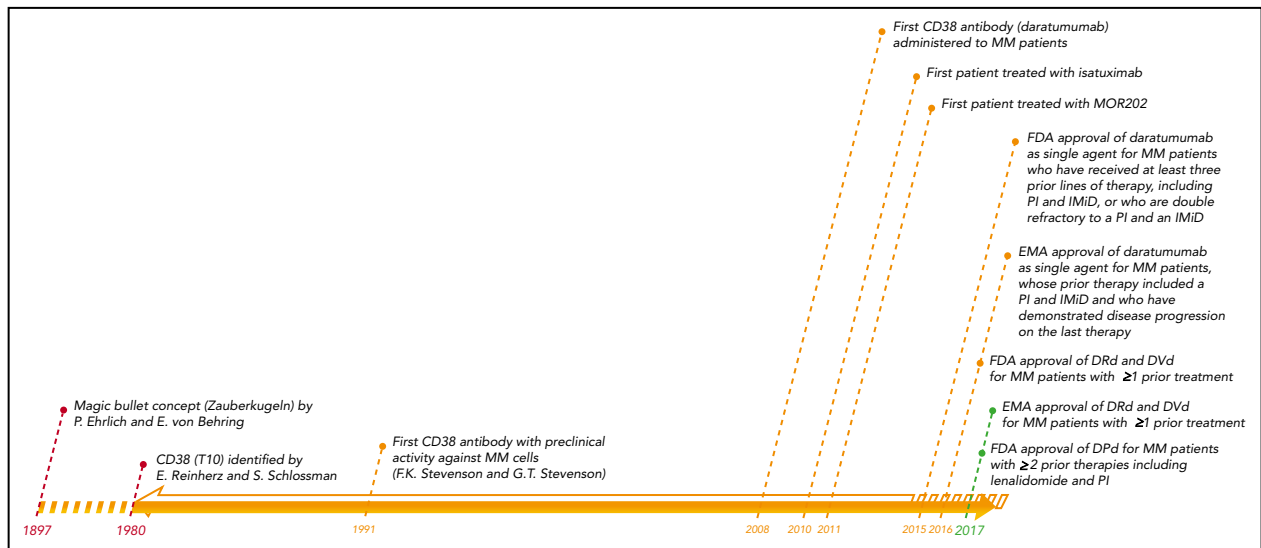
Preclinical experiments show that isatuximab also eliminates CD38<sup>+</sup> regulatory T cells, and thereby restores T-cell- and NK-cell-mediated antitumor immune responses.<sup>27</sup> It is currently unknown whether MOR202 has similar immunomodulatory activities.

## CD38 antibody monotherapy in MM

### Single-agent activity in MM patients

MM patients with disease refractory to immunomodulatory drugs (IMiDs) and proteasome inhibitors have a very poor outcome.<sup>28</sup> Based on the results from the GEN501<sup>29</sup> and Sirius<sup>30</sup> studies, daratumumab was approved as single agent in 2015 by the US Food and Drug Administration (FDA) and in 2016 by the European Medicines Agency (EMA) for relapsed/refractory MM. A pooled analysis of both monotherapy studies showed an overall response rate (ORR) of 31.1% (complete response [CR] in 4.7%), median duration of response of 7.6 months, median progression-free survival (PFS) of 4.0 months, and median overall survival (OS) of 20.1 months in patients treated with 16 mg/kg daratumumab (median 5 prior lines of therapy).<sup>31</sup> Responses were observed across all subgroups including patients with renal impairment, extramedullary plasmacytoma, and those with triple- or quadruple-refractory disease, advanced age, or high-risk cytogenetics.<sup>30-32</sup> Pharmacokinetic studies show that the current 16 mg/kg daratumumab administration schedule leads to rapid and sustained target saturation in the majority of patients.<sup>33,34</sup> Daratumumab has a manageable toxicity profile with 48% of patients experiencing infusion-related reactions (IRRs).<sup>31</sup> With the aim of shortening infusion time, a phase 1 study is currently investigating the subcutaneous delivery of daratumumab in combination with recombinant human hyaluronidase.<sup>35</sup> Preliminary data suggest that subcutaneous delivery is associated with a lower rate of IRRs as compared with IV daratumumab, with a comparable response rate and similar serum through concentrations in relapsed/refractory MM.<sup>35</sup>

These studies demonstrated the potent effects of targeting CD38 in MM, and supported the clinical development of additional agents in this class. MOR202 (4-16 mg/kg) combined with low-dose dexamethasone achieved partial response (PR) or better in 29% of patients (median 4 prior lines of therapy).<sup>36</sup> Also, isatuximab at a dose of ≥10 mg/kg has single-agent activity with an ORR of 24.3% (median PFS, 3.7 months; median OS, 18.6 months) in relapsed/refractory MM patients with a median of 5 prior therapies.<sup>37,38</sup>



**Figure 1. History of CD38 antibodies.** The timeline describes the discovery of CD38 protein, and the chronological introduction of CD38 antibodies in human trials. The timeline also shows the approved treatment options with CD38-targeted therapy in MM. DPd, daratumumab plus pomalidomide-dexamethasone; DRd, daratumumab plus lenalidomide-dexamethasone; DVd, daratumumab plus bortezomib-dexamethasone; EMA, European Medicines Agency; FDA, US Food and Drug Administration; IMiD, immunomodulatory drug; Pd, pomalidomide-dexamethasone; PI, proteasome inhibitor; Rd, lenalidomide-dexamethasone; Vd, bortezomib-dexamethasone.

## Determinants of response

There is a marked heterogeneity in the response of patients to CD38 antibodies as single agent. Mechanisms underlying the differential therapeutic efficacy probably include both host- and tumor-related factors.

**Tumor-related factors** Cell surface expression of CD38 on MM cells was associated with extent of daratumumab-mediated ADCC and CDC in MM cell lines and primary samples.<sup>39</sup> Similarly, daratumumab-treated patients who achieved at least PR, had higher baseline CD38 expression on their tumor cells, compared with patients with less than PR.<sup>40</sup> However, given the overlap in CD38 expression between responding and non-responding patients, CD38 levels alone do not explain the whole variability in response.<sup>40</sup> Baseline expression levels of complement inhibitors on MM cells or soluble CD38 molecules did not affect the activity of daratumumab.<sup>40</sup> Subgroup analysis suggests that CD38 antibodies are also active in patients with high-risk cytogenetics.<sup>30,38</sup> However, these subgroups involve relatively small numbers of patients, and further analysis is required to assess the impact of high-risk cytogenetics on outcome. The contribution of specific mutations and activation status of signaling pathways to the variability in response to CD38 antibodies is currently unknown.

**Bone marrow microenvironment** The bone marrow (BM) microenvironment confers protection against several anti-MM agents and also renders MM cells resistant to T cells.<sup>41</sup> We showed that BM stromal cells also protect MM cells against daratumumab-mediated ADCC.<sup>42</sup> This microenvironment-induced apoptosis resistance is possibly related to increased expression of antiapoptotic proteins such as survivin.<sup>42</sup>

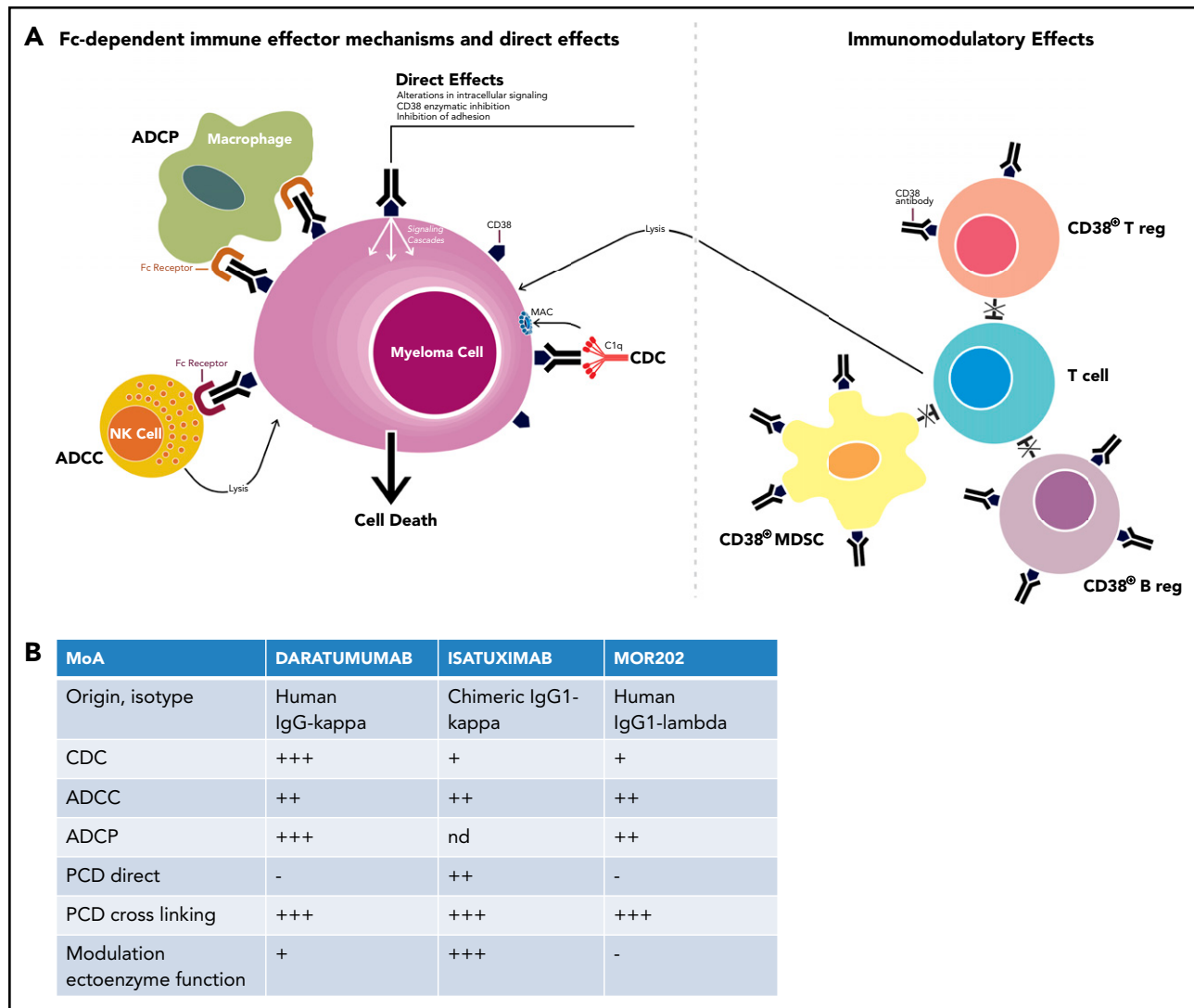
Differences in the frequency of NK cells or monocytes in the BM microenvironment also explained part of the variability in

daratumumab-mediated killing of primary MM cells.<sup>39</sup> Activity of these cells may be affected by previous or concomitant therapies. Furthermore, inhibitory signals transmitted to effector cells by MM cells may also result in effector cell dysfunction. The impact of genetic variation of activating and inhibitory receptors on response requires further investigation.<sup>43</sup>

## Acquired resistance

The majority of patients who initially respond to monotherapy eventually progress. Development of resistance toward daratumumab was associated with increased expression of the complement inhibitors CD55 and CD59 on MM cells, or the outgrowth of subpopulations with high expression of complement inhibitors.<sup>40</sup> Conversely, reduction of CD55 and CD59 sensitized MM cells to daratumumab-mediated CDC.<sup>40</sup>

CD38 reduction on nondepleted tumor cells may also confer protection against daratumumab.<sup>40</sup> However, significant CD38 reduction was already observed after the first daratumumab infusion, and in both responding and nonresponding patients including those with sustained clinical response,<sup>40</sup> which suggests that the continuous pressure to maintain MM cells in a CD38<sup>-/low</sup> state seems to offer a clinical benefit by altering MM cell adhesion and reducing immunosuppressive adenosine levels in the BM microenvironment.<sup>44,45</sup> Importantly, CD38 expression analysis was done by flow cytometry with a monoclonal antibody that binds to a different epitope than daratumumab, which excludes the possibility that binding of daratumumab masked the detection of CD38. The rapid daratumumab-mediated reduction of CD38 expression on surviving MM cells is probably the result of the selection of cells with low CD38 expression, release of tumor-derived CD38-carrying microvesicles,<sup>44,46</sup> and trogocytosis.<sup>47</sup> Interestingly, CD38 expression was preserved during treatment with MOR202.<sup>36</sup> No clinical data are yet available for isatuximab.



**Figure 2. Mechanism of action of CD38 antibodies.** (A) CD38-targeting antibodies have pleiotropic mechanisms of action, which can be subdivided into (i) Fc-dependent immune-effector mechanisms; (ii) direct effects; and (iii) immunomodulatory effects. The Fc-dependent immune-effector mechanisms include antibody-dependent cellular cytotoxicity (ADCC), antibody-dependent cellular phagocytosis (ADCP), and complement-dependent cytotoxicity (CDC). The process of ADCC is achieved through the activation of FcRs on NK cells and myeloid cells by tumor-cell attached CD38 antibodies. Subsequent release of perforin and granzymes from effector cells as well as interactions with death ligands FasL and tumor necrosis factor-related apoptosis-inducing ligand lead to MM cell death. In ADCP phagocytosis is mediated by monocytes, macrophages, neutrophils, and dendritic cells following interaction of the Fc tail of the therapeutic antibody with FcRs on these effector cells. CDC is initiated following the interaction of the antibody Fc domains with the classic complement-activating protein C1q, which leads to activation of downstream complement proteins, resulting in assembly of the membrane attack complex (MAC), which punches holes in MM tumor cells. The chemotactic complement molecules, C3a and C5a, are also produced during this process. These molecules can recruit and activate immune-effector cells. Direct effects include induction of apoptosis, as well as inhibition of CD38 ectoenzyme function, which may lead to reduced adenosine levels in the BM myeloma niche. Adenosine is an immunosuppressor that helps the tumor to evade the host immune response by promoting regulatory T cells (Tregs) and myeloid-derived suppressor cells (MDSCs), and depressing NK- and T-cell effectors. CD38 antibodies have immunomodulatory effects via the eradication of CD38<sup>+</sup> Tregs, regulatory B cells (Bregs), and MDSCs, which result in CD4<sup>+</sup> and CD8<sup>+</sup> T-cell expansion, and potentially a better host-antitumor immune response. Adapted from van de Donk et al<sup>78</sup> with permission. (B) The relative contribution of these different mechanisms of action to MM cell killing differs among daratumumab, isatuximab, and MOR202. The efficacy of these 3 CD38 antibodies was directly compared in preclinical studies in terms of direct induction of programmed cell death (PCD), induction of PCD after crosslinking, inhibition of CD38 ectoenzyme activity, and the induction of ADCC, ADCP, and CDC. The immunomodulatory effects of the CD38 antibodies were not compared in a head-to-head analysis, and therefore were not included in panel B. MoA, mechanism of action; nd, not determined.

Reduced biological activity mediated by anti-drug antibodies has not been observed with daratumumab or isatuximab up until now,<sup>29,30,48,49</sup> whereas it is a rare event with MOR202.<sup>50</sup>

Other factors that may contribute to development of resistance are changes in frequency and activity of effector cells. In this respect, NK cells have high CD38 expression and are rapidly reduced after infusion of daratumumab,<sup>51,52</sup> which may impair MM cell killing. However, NK-cell reduction was similar in responding and nonresponding patients.<sup>52</sup>

## Retreatment

Preliminary evidence from a study with 2 patients suggests that retreatment of patients with daratumumab is feasible and effective.<sup>53</sup> However, it is currently unknown whether a treatment-free interval is required in order to allow NK-cell recovery and CD38 expression levels to return to baseline on remaining MM cells.<sup>40,52</sup> Another strategy is to maintain daratumumab, and add other therapeutic agents to potentiate any anti-MM effects of daratumumab.

**Table 1. CD38 antibodies plus lenalidomide-dexamethasone in relapsed/refractory MM**

	NCT02076009 (POLLUX) <sup>59,62</sup>		NCT01749969 <sup>64</sup>	NCT01421186 <sup>36,65</sup>
Phase	Phase 3		Phase 1b	Phase 1/2a
Regimen	Rd	DRd	Isatuximab-Rd	MOR202-Rd
Treatments	Lenalidomide: 25 mg on days 1-21 per 28-d cycle (10 mg if CrCl 30-60 mL/min) Dexamethasone: 40 mg weekly (20 mg if age >75 y) Treatment until progression	Daratumumab (16 mg/kg): weekly for 8 wk, then Q2W for 16 wk, thereafter Q4W Lenalidomide: 25 mg on days 1-21 per 28-d cycle (10 mg if CrCl 30-60 mL/min) Dexamethasone: 40 mg weekly (20 mg if age >75 y) Treatment until progression	Isatuximab: 3, 5, or 10 mg/kg Q2W or 10, 20 mg/kg QW for 4 wk, and then Q2W Lenalidomide: 25 mg on days 1-21 per 28-d cycle (10 mg if CrCl 30-60 mL/min) Dexamethasone: 40 mg weekly Treatment until progression	MOR202 8-16 mg/kg, weekly Lenalidomide: 25 mg on days 1-21 per 28-d cycle Dexamethasone: 40 mg weekly (20 mg if age >75 y) Treatment until progression or a maximum of 2 y
No. of patients	283	286	57 (52 evaluable for response)	15 (13 evaluable for response)
Median age, y	65	65	61	64
Median no. of prior therapies	1	1	5	2
Len-refractory, %	Len-refractory patients were excluded	Len-refractory patients were excluded	82	13
Bort-refractory, %	16.3	19.9	65	27
IRR, %	Not applicable	47.7	56	6.7
≥PR, %	76.4	92.9	55.7 (Len-refractory: 52.4%)	84.6
≥VGPR, %	44.2	75.8	36.5 (Len-refractory: 35.7%)	23.1
≥CR, %	19.2	43.1	3.8 (Len-refractory: 0.0%)	0.0
MRD <sup>-</sup> disease (10 <sup>-5</sup> ), %	5.7	24.8	NA	NA
PFS	12-mo: 60.1%	12 mo: 83.2%	Median: 8.5 mo 12 mo: 38.9%	Median: not reached
OS	12 mo: 86.8%	12 mo: 91.2%	Not reported	Median: not reached

Bort, bortezomib; CR, complete response; CrCl, creatinine clearance; DRd, daratumumab plus lenalidomide-dexamethasone; IRR, infusion-related reaction; Len, lenalidomide; MRD, minimal residual disease; NA, not assessed; OS, overall survival; PFS, progression-free survival; PR, partial response; Q2W, once every 2 weeks; Q4W, once every 4 weeks; QW, once weekly; Rd, lenalidomide-dexamethasone; VGPR, very-good partial response.

## Combination therapy with CD38 antibodies

MM is a very heterogeneous disease with multiple subclones with different susceptibility to specific agents. Therefore, combining multiple agents with overlapping toxicities and synergistic mechanisms of action is a successful strategy in improving clinical outcome. Based on the efficacy of CD38 antibodies, these agents are very attractive as a component of combination regimens.

### IMiDs

The combination of a CD38 antibody with an IMiD such as lenalidomide and pomalidomide markedly enhanced NK-cell mediated ADCC.<sup>25,32,54-57</sup> This potentiation is mainly related to the

ability of IMiDs to increase NK-cell numbers and NK-cell activity.<sup>32,55</sup> In addition, IMiDs also promote tumoricidal activity of macrophages and enhance ADCC.<sup>58</sup> Furthermore, IMiDs increase the direct toxic effects of isatuximab<sup>25</sup> and increase CD38 expression on regulatory T cells, resulting in enhanced isatuximab-mediated elimination of these cells.<sup>27</sup> Interestingly, preclinical data show that IMiDs also synergize with daratumumab in IMiD-resistant tumors, indicating that the immune system of these patients can still respond to the immunomodulatory effects of these agents.<sup>32</sup> No improvement in CDC has been observed with IMiDs.<sup>55</sup>

Altogether, this formed the preclinical rationale for the randomized phase 3 Pollux study, in which 569 patients with at least 1 prior therapy were randomized to lenalidomide-dexamethasone (Rd) with or without daratumumab (Table 1). Patients treated with



**Table 2. CD38 antibodies plus pomalidomide-dexamethasone in relapsed/refractory MM**

	NCT01998971 <sup>66</sup>	NCT02283775 <sup>67,106</sup>	NCT01421186 <sup>36,65</sup>
Phase	1b	1b	1/2a
Regimen	Daratumumab-pom/dex	Isatuximab-pom/dex	MOR202-pom/dex
Treatments	Daratumumab (16 mg/kg): weekly for 8 wk, then Q2W for 16 wk, thereafter Q4W Pomalidomide: 4 mg on days 1-21 per 28-d cycle Dexamethasone: 40 mg weekly (20 mg if age >75 y) Treatment until progression	Isatuximab 5, 10, or 20 mg/kg, weekly during first cycle, then every 2 wk Pomalidomide: 4 mg on days 1-21/28 d cycle Dexamethasone: 40 mg weekly (20 mg if age ≥75 y) Treatment until progression	MOR202 8-16 mg/kg, weekly Pomalidomide: 4 mg on days 1-21 per 28-d cycle Dexamethasone: 40 mg weekly (20 mg if age >75 y) Treatment until progression or a maximum of 2 y
No. of patients	103	20 (14 evaluable patients)	11 (9 evaluable for response)
Median age, y	64	65.5	66
Median no. of prior therapies	4	5.0	3
Len-refractory, %	89	75	100
Bort-refractory, %	71	45	36
IRR, %	50 (mostly grade ≤ 2)	45 (all grade 1 or 2)	0
≥PR	60.2	64.3	55.6
≥VGPR	41.7	35.7	22.2
≥CR	16.5	7.1	22.2
MRD <sup>-</sup> disease (10 <sup>-5</sup> ) among patients with CR or better, %	29.4	NA	NA
Median PFS, mo	8.8	Not reported	Not reached
Median OS, mo	17.5	Not reported	Not reached

dex, dexamethasone; pom, pomalidomide. Other abbreviations are explained in Table 1.

daratumumab plus Rd (DRd) had higher rates of deeper responses, which translated into markedly better PFS (hazard ratio [HR], 0.37; PFS at 12 months, 83.2% vs 60.1%).<sup>59</sup> The PFS advantage was observed across all subgroups including patients ≥65 years and those with previous lenalidomide exposure.<sup>59,60</sup> Also, patients with high-risk cytogenetics [del(17p); t(4;14); t(14;16)] had better PFS with DRd compared with Rd (median PFS, 22.6 vs 10.2 months), but poor risk conferred by these cytogenetic abnormalities was not completely abrogated by adding daratumumab. Longer follow-up is needed to show potential OS benefit. Daratumumab also significantly increased minimal residual disease (MRD)-negativity rate (DRd vs Rd, 24.8% and 5.7%; threshold 10<sup>-5</sup>), which translated into significantly improved PFS.<sup>61,62</sup> In patients with high-risk cytogenetics, MRD<sup>-</sup> status was only achieved in those treated with daratumumab.<sup>62</sup> Other than IRRs, there was an increased incidence of grade 3/4 neutropenia, diarrhea, fatigue, and dyspnea when daratumumab was added to Rd, probably as a result of longer treatment exposure.<sup>59</sup> Similar to prior findings with daratumumab monotherapy, addition of daratumumab to Rd induced clonal expansion of T cells.<sup>63</sup> Furthermore, baseline T-cell richness was associated with improved PFS in the DRd arm.<sup>63</sup> Based on these

data, the FDA (2016) and EMA (2017) approved this triplet in MM patients with ≥1 prior therapy.

Isatuximab combined with Rd was evaluated in patients with more advanced MM (median of 5 prior lines of therapy; 82% lenalidomide-refractory). At least PR was achieved in 56% of the patients and median PFS was 8.5 months.<sup>64</sup> Fifty-two percent of lenalidomide-refractory patients obtained PR or better, indicating clinical synergy between these drugs.<sup>64</sup> Preliminary results of MOR202 combined with Rd also showed a good tolerability profile and evidence for long-lasting disease control.<sup>36,65</sup>

A phase 2 study showed that addition of daratumumab to pomalidomide-dexamethasone (Pd; DPd) is also effective with approximately a doubling of the response rate as compared with Pd alone (Table 2).<sup>66</sup> Based on these data, DPd was recently approved (June 2017) by the FDA for MM patients with ≥2 prior therapies including lenalidomide and a proteasome inhibitor (PI). Similar efficacy was observed when isatuximab or MOR202 was combined with Pd.<sup>36,67</sup> Phase 3 trials will further evaluate the value of adding daratumumab or isatuximab to Pd.

**Table 3. Bortezomib-dexamethasone with or without daratumumab in MM patients with at least 1 prior line of therapy (Castor study)**

	NCT02136134 (CASTOR) <sup>48,61</sup>	
Phase	Phase 3	
Regimen	Vd	DVd
Treatments	Bortezomib 1.3 mg/m <sup>2</sup> : days 1, 4, 8, 11 of 21-d cycle; 8 cycles Dexamethasone 20 mg: days 1, 2, 4, 5, 8, 9, 11,12; 8 cycles (20 mg once weekly if age >75 y)	Daratumumab (16 mg/kg): weekly ×10, Q3W until end of Vd, then Q4W until progression Bortezomib 1.3 mg/m <sup>2</sup> : days 1, 4, 8, 11 of 21-d cycle; 8 cycles Dexamethasone 20 mg: days 1, 2, 4, 5, 8, 9, 11,12; 8 cycles (20 mg once weekly if age >75 y)
No. of patients	247	251
Median age, y	64	64
Median no. of prior therapies	2	2
Previous IMiD therapy, %	80.2	71.3
Previous PI therapy, %	69.6	67.3
IRR, %	Not applicable	45.3
≥PR, %	63.2	82.9
≥VGPR, %	29.1	59.2
≥CR, %	9.0	19.2
MRD <sup>-</sup> disease (10 <sup>-5</sup> ), %	2.4	10.4
12-mo PFS, %	26.9	60.7
OS	Median OS, not evaluable; HR, 0.77	Median OS, not evaluable; HR, 0.77

DVd, daratumumab-bortezomib-dexamethasone; HR, hazard ratio; IMiD, immunomodulatory drug; PI, proteasome inhibitor; Q3W, once every 3 weeks; Vd, bortezomib-dexamethasone. Other abbreviations are explained in Table 1.

### Proteasome inhibitors


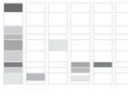

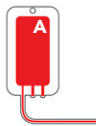

Preclinical studies also showed synergy when CD38-targeting agents were combined with PIs.<sup>54,68,69</sup> Similarly, in the Castor study, patients (≥1 prior line of therapy) treated with bortezomib-dexamethasone (Vd) with daratumumab (DVd) had a better response and PFS (HR, 0.39; 12-month PFS, 60.7% vs 26.9%), compared with Vd-treated patients (Table 3).<sup>48</sup> Subgroup analysis showed that adding daratumumab was beneficial in all subgroups including patients ≥65 years and those with prior bortezomib exposure.<sup>48,70</sup> Also, patients with high-risk cytogenetics achieved benefit from adding daratumumab to Vd (median PFS, 11.2 vs 7.2 months).<sup>71</sup> Achievement of MRD negativity resulted in excellent PFS in both treatment arms, but the MRD-negativity rate was significantly higher in the daratumumab group (DVd vs Vd, 10.4% and 2.4%; threshold 10<sup>-5</sup>).<sup>61</sup> MRD-negativity rate and PFS were superior with DRd compared with DVd.<sup>48,59</sup> Apart from type of standard of care to which daratumumab was added, differences in terms of study design (continuous treatment with Rd vs fixed duration treatment with Vd) and patient characteristics (more prior lines of treatment in Castor; greater proportion of patients with standard-risk cytogenetics in Pollux) may also explain

differences in depth and durability of response between both studies (Tables 1 and 3). Added toxicity of daratumumab consisted of IRRs.<sup>48</sup> Furthermore, the incidence of grade 3/4 thrombocytopenia, neutropenia, lymphopenia, diarrhea, dyspnea, and hypertension was higher in the daratumumab group, which may be partly explained by a larger proportion of patients in the DVd group receiving the maximum of 8 cycles of bortezomib treatment when compared with the control group.<sup>48</sup> The FDA (2016) and EMA (2017) approved DVd in patients with ≥1 prior treatment.

Furthermore, preliminary results show that CD38 antibodies can be effectively combined with the second-generation proteasome inhibitor carfilzomib.<sup>72,73</sup>

### Other combinations

The better understanding of host- and tumor-related factors that predict response has also resulted in the rational design of new daratumumab-based combinations. We showed that all-trans retinoic acid (ATRA) upregulates CD38 expression and reduces expression of complement inhibitors CD55 and CD59 on MM cells, leading to markedly improved daratumumab-mediated

	Management Aspect	Applicable to	Management and Developments
	Administration -Duration of infusion -Infusion-related reactions	All antibodies	Appropriate pre- and post-infusion medication; subcutaneous administration of daratumumab
	Interference of therapeutic antibody with SPEP/IFE assays	All antibodies but dependent on plasma concentrations	Shift assays; mass spectrometry
	Auto-immune adverse events	PD-1/PD-L1 neutralizing antibodies (not observed with CD38 antibodies)	Institution of appropriate treatment (including prompt treatment with prednisone)
	Interference with blood transfusion tests	CD38-targeting antibodies (and CD44-targeting antibodies)	DTT, anti-idiotypic, serotyping/genotyping before start therapy; provide patients with blood transfusion card
	Infections	Dependent on antibody type and other drugs in combination regimens	Herpes zoster prophylaxis is recommended for CD38 antibodies

**Figure 3. Practical aspects of management of CD38 antibody therapy.** The main adverse event associated with CD38 antibodies is the development of IRRs, which may prolong the duration of the CD38 antibody infusion. To prevent development of IRRs, it is important to administer appropriate pre- and postinfusion medication. We typically administer 1 to 3 hours prior to the daratumumab infusion preinfusion prophylaxis consisting of antihistamines (H1 receptor antagonist: eg, 2 mg of clemastine or 25-50 mg of diphenhydramine), acetaminophen (650-1000 mg), and corticosteroids (100 mg of methylprednisolone or equivalent for daratumumab monotherapy [following the second infusion, this can be lowered to 60 mg] and 20 mg of dexamethasone for combination therapy). Furthermore, we give all patients 10 mg of montelukast prior to the first daratumumab infusion. In case the patient experiences an IRR, we also consider montelukast in subsequent infusions. For patients on daratumumab monotherapy, we give as postinfusion medication corticosteroids (20 mg methylprednisolone or equivalent on each of the 2 days following the daratumumab infusion). For patients on daratumumab combination therapy, no additional methylprednisolone as postinfusion medication is needed if dexamethasone is administered the day after the daratumumab infusion as part of the background regimen (otherwise administration of 20 mg of methylprednisolone or equivalent can be considered on the day after the infusion). For patients with a history of chronic obstructive pulmonary disease, consider prescribing short- and long-acting bronchodilators and inhaled corticosteroids. For isatuximab and MOR202, which are currently not approved, we follow the study protocol for pre- and postinfusion prophylaxis. Herpes zoster prophylaxis should be considered in patients treated with CD38-targeted therapy. CD38 antibodies also interfere with laboratory tests such as serum protein electrophoresis (SPEP) and immunofixation electrophoresis (IFE), which may confound evaluation of response. CD38 antibodies also interfere with routine methods for compatibility testing for blood transfusion. Important management aspects of some other antibody classes are also shown.

ADCC and CDC.<sup>39</sup> Interestingly, ATRA also increases CD38 and reduces CD55 and CD59 levels on MM cells with acquired daratumumab resistance.<sup>40</sup> A phase 1/2 study is currently enrolling patients to evaluate the value of ATRA in patients with daratumumab-refractory disease. Similarly, the pan-histone deacetylase inhibitor panobinostat also induces upregulation of CD38 on MM cells, leading to increased ADCC with daratumumab.<sup>74</sup> CDC was not improved as a result of concomitant upregulation of CD55 and CD59.<sup>74</sup>

Various strategies to improve effector cell function, and thereby enhance ADCC/ADCP, are also evaluated in preclinical models. For example, NK-cell activity can be improved by blocking inhibitory killer immunoglobulin-like receptors (KIRs) with the anti-KIR antibody IPH2102 (lirilumab), which resulted in enhanced daratumumab-mediated lysis of MM cells.<sup>75</sup> Also, inhibition of the CD47-signal-regulatory protein  $\alpha$  (SIRP $\alpha$ ) antiphagocytic axis<sup>76</sup> or the programmed death-1 (PD-1)/programmed death ligand-1

(PD-L1) pathway, which delivers inhibitory signals to T cells, may enhance the activity of CD38 antibodies.<sup>77</sup>

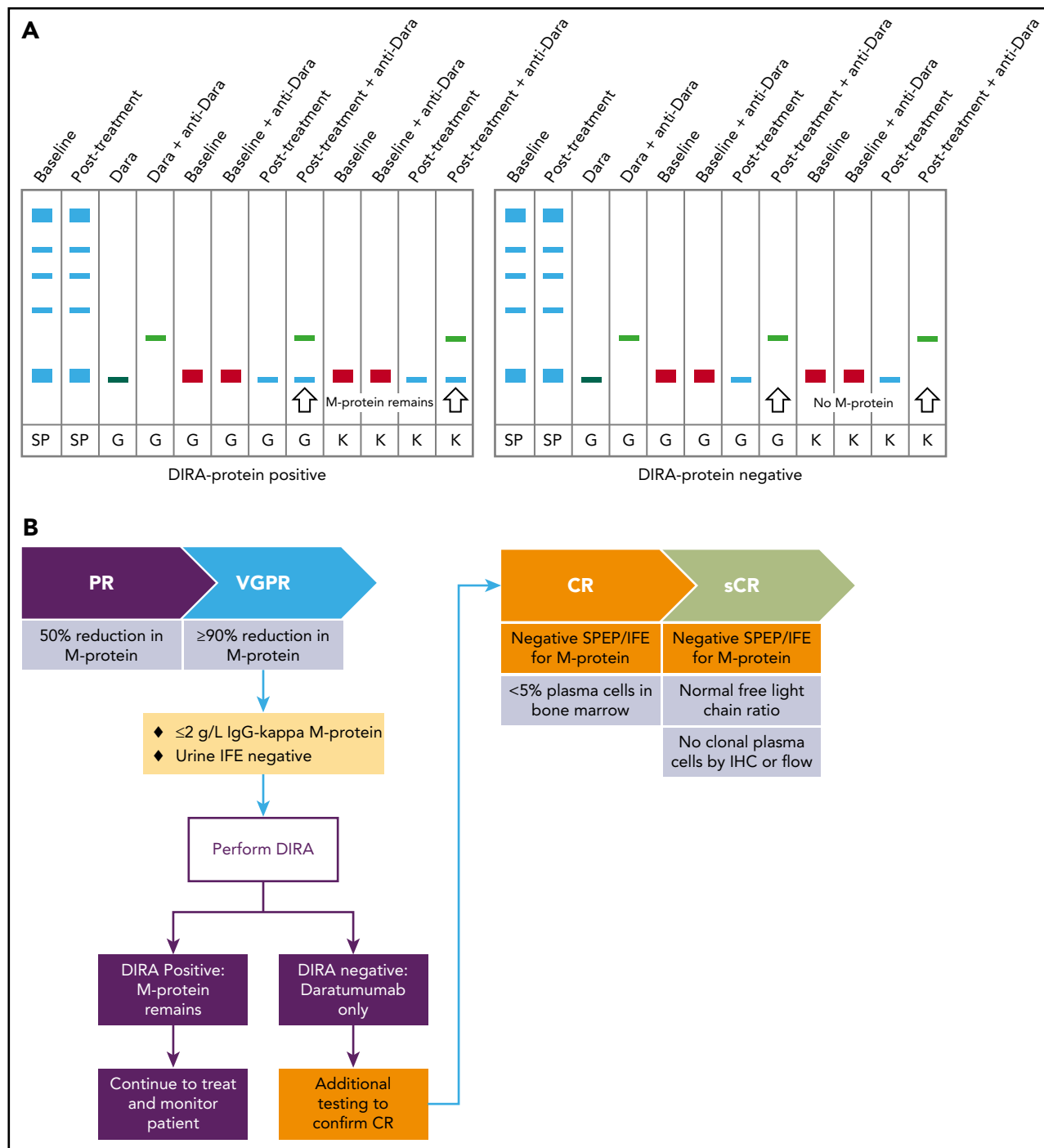
## Management aspects of CD38 antibodies

In the following section, we describe some practical management aspects that are characteristic for CD38 antibodies (Figure 3).<sup>78</sup>

### Infusion-related reactions

CD38 antibodies have a favorable toxicity profile with IRRs as the main side effect. Adequate and timely management of IRRs (including pre- and postinfusion prophylaxis) is important to prevent discomfort to patients, more serious toxicity, prolonged infusion time, and treatment discontinuation (Figure 3). Daratumumab-associated IRRs occur in ~50% of patients, are mostly grade 1/2, and consist mainly of respiratory conditions

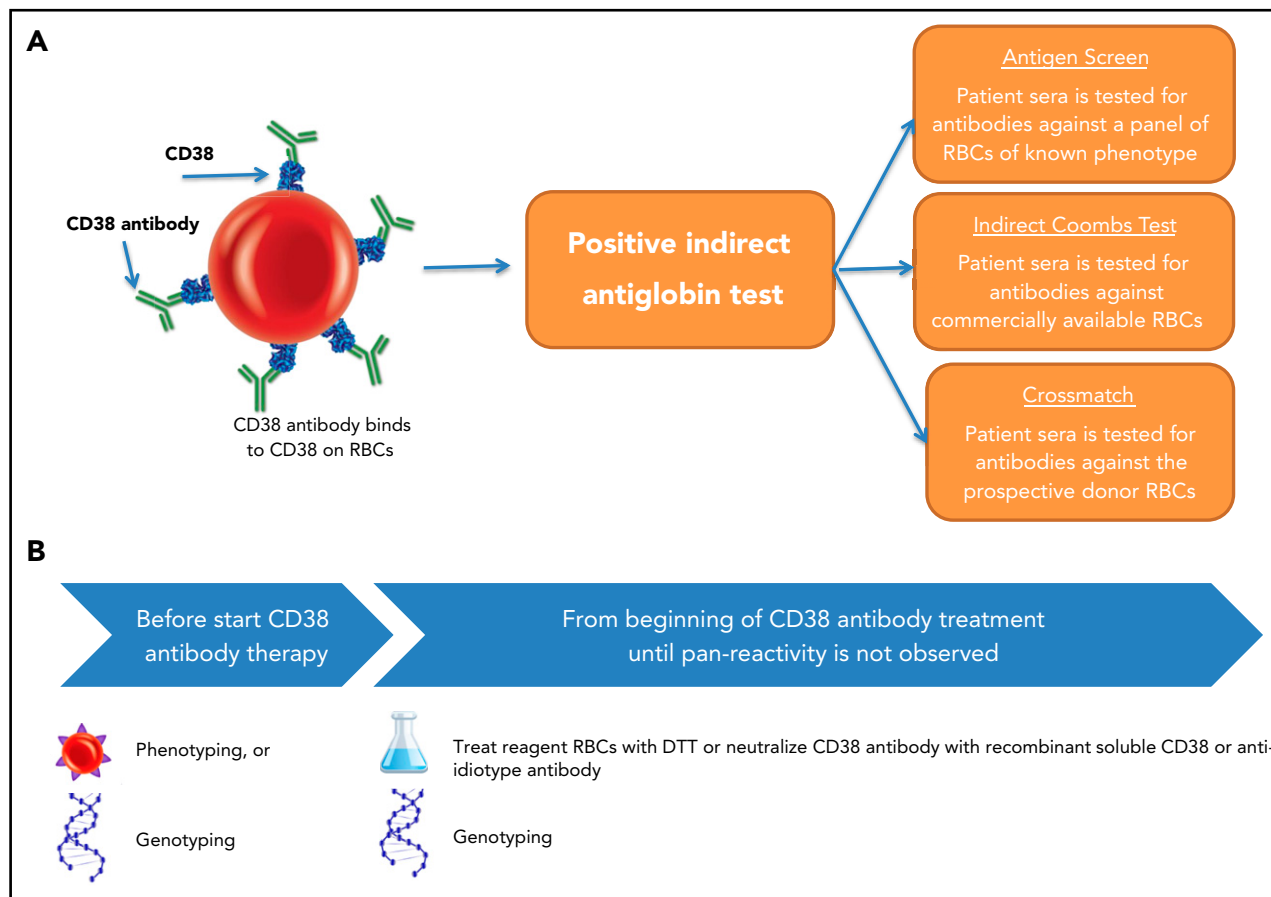




**Figure 4. Daratumumab interferes with the evaluation of response.** (A) The mean trough and peak serum daratumumab concentrations at the end of weekly 16 mg/kg dosing are 573  $\mu\text{g}/\text{mL}$  and 915  $\mu\text{g}/\text{mL}$ , respectively.<sup>33,34</sup> These concentrations decrease slightly when patients enter the less-intense dose periods.<sup>33,34</sup> These concentrations are greater than the sensitivity for most SPEP and serum IFE assays, which explains that daratumumab and also other therapeutic antibodies can be detected as a monoclonal band in SPEP and IFE when patients are assessed for response. Comigration of daratumumab (IgG- $\kappa$ ) with an IgG- $\kappa$  M-protein during SPEP and IFE can mask clearance of a patient's endogenous M protein in response to treatment. The daratumumab-specific IFE reflex assay (DIRA) uses a highly specific mouse anti-daratumumab antibody that binds daratumumab and shifts its migration away from endogenous M protein on IFE gels. Of note, because the anti-daratumumab antibody is of mouse origin, it is not precipitated with antibodies directed at human immunoglobulins.<sup>82</sup> The mouse anti-daratumumab antibody is therefore not detected as an additional band in the DIRA. (B) If an IgG- $\kappa$  band remains that is not shifted completely by the DIRA, the patient is considered to have residual M protein, and disease monitoring should continue. Patients with a single IgG- $\kappa$  band that is shifted completely by the DIRA are considered to have no remaining serum M protein and may have reached CR/sCR, and therefore are candidates for additional IMWG-required confirmatory testing (BM evaluation, free light-chain assay). Dara, daratumumab; G, IgG antisera; IHC, immunohistochemistry;  $\kappa$ ,  $\kappa$  antisera; sCR, stringent CR; SP, total serum protein fix; VGPR, very-good partial response.

such as nasal congestion, cough, allergic rhinitis, throat irritation, and dyspnea.<sup>31,48,59,79</sup> Nonrespiratory IRRs include chills and nausea.<sup>31</sup> Most IRRs (95.8%) occur during the first infusion,

and the incidence of IRRs decreases during second (7%) and subsequent infusions (7%).<sup>31</sup> There is no relationship between maximum daratumumab serum levels and development of IRRs.<sup>34</sup>



**Figure 5. CD38 antibodies interfere with blood group compatibility testing.** (A) CD38 antibodies bind to CD38 molecules present on reagent or donor red blood cells (RBCs), and thereby interfere with blood bank compatibility tests, such as antibody screening and crossmatching (both IATs). (B) Several strategies can be used to negate daratumumab interference with blood group compatibility testing to ensure that patients receive a timely transfusion. This includes a chemical method (dithiothreitol [DTT]) to denature CD38 so that daratumumab binding is reversed. Because the Kell blood group system is also sensitive to DTT treatment, K<sup>-</sup> units should be supplied after ruling out or identifying alloantibodies using DTT-treated RBCs. Approximately 9% of the population is reactive to the Kell blood group system, therefore, >90% of blood units will be Kell<sup>-</sup> and suitable for transfusion. Alternatively, binding of CD38 antibodies to RBCs can be prevented by using neutralizing agents such as anti-idiotype antibody or recombinant soluble CD38. Furthermore, extensive human erythrocyte antigen phenotyping or genotyping can be performed prior to starting CD38 antibody therapy. Genotyping is not impacted by CD38 antibodies and may therefore be performed at any time, also during CD38 antibody therapy. When patients require blood transfusions, blood products can be identified based on the results of phenotyping and genotyping. Importantly, once treatment with a CD38 antibody is discontinued, pan-agglutination may persist. The duration of this effect varies from patient to patient, but for daratumumab this effect may persist for up to 6 months. Mitigation methods should be used until pan-agglutination is no longer observed.

Isatuximab-induced IRRs have similar characteristics as those mediated by daratumumab, and occur in 55% to 56% of patients.<sup>38,64</sup> MOR202 treatment is associated with a lower incidence of IRRs (~10%), which may be explained by low CDC activity.<sup>36</sup> Therefore, MOR202 can be infused faster, compared with daratumumab or isatuximab.

IRRs are managed with temporary interruptions of the infusion, or extra antihistamines or corticosteroids.<sup>31</sup> Importantly, the leukotriene receptor antagonist, montelukast, probably mitigates CD38 antibody-associated IRRs.<sup>80</sup> Furthermore, inhalable corticosteroids and bronchodilators are recommended for patients with obstructive lung disorders.

### Response evaluation

Various therapeutic antibodies, including CD38-targeting antibodies, can be detected as a monoclonal band in serum protein electrophoresis (SPEP) and serum immunofixation electrophoresis (IFE),<sup>81,82</sup> which may confound interpretation of these assays and thus evaluation of response (Figure 4). As expected, CD38 antibodies as intact immunoglobulins do not interfere with the

serum free light chain (sFLC) assay.<sup>83</sup> Importantly, a recent update of the International Myeloma Working Group (IMWG) uniform response criteria clarified that CR requires disappearance of the original M protein associated with MM on IFE, and therefore CR is not affected by unrelated M proteins that are secondary to therapeutically administered monoclonal antibodies.<sup>84</sup>

However, M-protein monitoring is more complicated when the patient's M protein and therapeutic antibody have the same heavy- and light-chain isotype, and also co-migrate into the same region. In this situation, the 2 bands cannot be differentiated from each other, without using additional tests such as the daratumumab-specific IFE reflex assay (DIRA) to distinguish the therapeutic from the patient's M protein. This assay uses an anti-idiotype antibody that binds to daratumumab and alters its migration pattern to distinguish between endogenous M protein and therapeutic antibody.<sup>81,82</sup> The DIRA assay can be used to determine whether additional testing, including determination of the sFLC ratio and BM evaluation, is warranted in patients with IgG- $\kappa$  band and low measurable M protein ( $\leq 2$  g/L) to assess the presence of (stringent)CR. A commercially available automated

**Table 4. Selected ongoing or planned studies with CD38-targeting antibody-containing regimens in other conditions**

Study	Phase	Patients	Treatment
NCT02841033	1/2	Relapsed or refractory AL amyloidosis	Daratumumab as single agent
NCT02816476 (AMYDARA)	2	Patients with AL amyloidosis not in VGPR or better after previous treatment	Daratumumab as single agent
NCT03067571	2	AML or high-risk myelodysplastic syndrome (relapsed or refractory)	Daratumumab as single agent
NCT03011034	2	Transfusion-dependent patients with low or intermediate-1 risk myelodysplastic syndrome who are relapsed or refractory to erythropoiesis-stimulating agents	Daratumumab as single agent
NCT03095118	2	PGNMID and C3GN	Daratumumab as single agent
NCT03098550	1/2	Advanced or metastatic solid tumors	Daratumumab plus nivolumab
NCT02488759 (CheckMate358)	1/2	Virus-associated tumors such as squamous cell carcinoma of the head and neck, cervix, and anal canal	Nivolumab or nivolumab combinations, including nivolumab plus daratumumab
NCT02060188 (CheckMate142)	2	Recurrent and metastatic colon cancer	Nivolumab or nivolumab combinations, including nivolumab plus daratumumab
NCT03023423	1/2	Previously treated advanced or metastatic non-small cell lung cancer	Daratumumab plus atezolizumab vs atezolizumab
NCT02413489 (Carina)	2	Relapsed/refractory CD38 <sup>+</sup> mantle cell lymphoma, diffuse large B-cell lymphoma, and follicular lymphoma	Daratumumab as single agent
NCT02927925	2	Relapsed/refractory NKTCL, nasal type	Daratumumab as single agent
NCT01084252	1/2	Relapsed/refractory CD38 <sup>+</sup> hematological malignancies such as B-cell non-Hodgkin lymphoma, MM, AML, B-ALL, and CLL	Isatuximab as single agent
NCT02999633	2	Relapsed or refractory T-ALL and T-LBL	Isatuximab as single agent

AL, amyloid light chain; AML, acute myeloid leukemia; B-ALL, B-cell acute lymphoblastic leukemia; C3GN, C3 glomerulopathy associated with monoclonal gammopathy; CLL, chronic lymphocytic leukemia; NKTCL, NK-/T-cell lymphoma; PGNMID, proliferative glomerulonephritis with monoclonal immune deposit; T-ALL, T-cell acute lymphoblastic leukemia; T-LBL, T-lymphoblastic lymphoma. Other abbreviations are explained in Table 1.

and standardized DIRA test is now available.<sup>85</sup> Similar assays are under development for isatuximab and MOR202.<sup>86</sup>

### Blood transfusion

Daratumumab binds to CD38 on test red blood cells, explaining why daratumumab-treated patients have pan-reactive indirect antiglobulin tests (IATs) observed on antibody screens (Figure 5).<sup>87-90</sup> Positive IATs may persist for up to 6 months after cessation of daratumumab therapy.<sup>89,91</sup> Remarkably, daratumumab-treated patients generally have negative direct antiglobulin tests, which is explained by rapid removal of CD38 from the red

blood cell surface without altering levels of other proteins such as Kell or Duffy.<sup>89,90</sup> Furthermore, daratumumab does not affect ABO/Rh typing of patient red blood cells.<sup>88,92</sup>

The interference with routine methods for compatibility testing for blood transfusion puts patients who require transfusion at risk for delays in receiving compatible blood, as underlying allo-antibodies cannot be detected.<sup>93</sup> Standard serologic methods to eliminate pan-reactive antibodies fail to resolve the interference,<sup>93</sup> but blood banks can use a variety of mitigation methods to safely and timely provide blood products for patients treated with

**Table 5. Selected ongoing and planned studies with CD38-targeting antibody-containing regimens in newly diagnosed MM or smoldering MM**

Study	Phase	Patients	Treatment
NCT02316106 (Centaurus)	2	Intermediate or high-risk smoldering MM	3 dose schedules of daratumumab as single agent (short, intermediate, and long treatment duration)
NCT02960555	2	High-risk smoldering MM	Isatuximab as single agent
Cassiopeia (IFM 2015-01; HOVON 131; NCT02541383)	3	NDMM, transplant eligible	Randomization 1: VTD induction therapy, followed by high-dose melphalan plus autologous stem cell rescue, followed by VTD consolidation vs VTD with daratumumab induction therapy, followed by high-dose melphalan plus autologous stem cell rescue, followed by VTD with daratumumab consolidation Randomization 2: daratumumab as single agent in maintenance vs observation only
NCT02874742	2	NDMM, transplant eligible	RVD + daratumumab induction–high-dose melphalan plus autologous stem cell rescue, followed by RVD plus daratumumab consolidation, followed by daratumumab plus lenalidomide maintenance vs RVD induction–high-dose melphalan plus autologous stem cell rescue, followed by RVD consolidation, followed by lenalidomide maintenance
EMN study	3	NDMM, transplant eligible	Randomization 1: KRD induction therapy, followed by high-dose melphalan plus autologous stem cell rescue, followed by KRD consolidation vs KRD with daratumumab induction therapy, followed by high-dose melphalan plus autologous stem cell rescue, followed by KRD with daratumumab consolidation Randomization 2: daratumumab or daratumumab plus lenalidomide maintenance
NCT02955810	1	NDMM, transplant eligible	CyBorD plus daratumumab induction–high-dose melphalan plus autologous stem cell rescue, followed by CyBorD plus daratumumab consolidation, followed by maintenance with daratumumab (low-risk) or daratumumab plus bortezomib (high-risk)
NCT02951819	2	Untreated NDMM and relapsed MM	CyBorD plus daratumumab induction, followed by daratumumab plus dexamethasone maintenance
NCT02252172 (Maia)	3	NDMM, transplant ineligible	Rd vs DRd

CyBorD, cyclophosphamide-bortezomib-dexamethasone; DRd, daratumumab plus lenalidomide-dexamethasone; KR, carfilzomib and lenalidomide; KRD, carfilzomib-lenalidomide-dexamethasone; NDMM, newly diagnosed MM; Rd, lenalidomide-dexamethasone; Vd, bortezomib-dexamethasone; VMP, bortezomib-melphalan-prednisone; VRD or RVD, bortezomib-lenalidomide-dexamethasone; VTD, bortezomib-thalidomide-dexamethasone.

**Table 5. (continued)**

Study	Phase	Patients	Treatment
NCT02195479 (Alcyone)	3	NDMM, transplant ineligible	VMP vs VMP plus daratumumab
Hovon143	2	NDMM, transplant ineligible and frail or unfit	Ixazomib-dexamethasone plus daratumumab induction, followed by ixazomib plus daratumumab maintenance
NCT03012880	2	NDMM, 18 y and older	Ixazomib-Rd plus daratumumab
NCT01998971	1b	NDMM, irrespective of transplant eligibility for Vd, VTD, and KRd, and transplant ineligible for VMP	Daratumumab combined with Vd, VTD, KRd, or VMP
NCT02513186	1	NDMM, transplant ineligible	Isatuximab plus CyBorD or VRD
NCT03104842	2	NDMM, transplant eligible and high-risk cytogenetic abnormalities  NDMM, transplant ineligible and high-risk cytogenetic abnormalities	Isatuximab plus KRd induction, followed by high-dose melphalan plus autologous stem cell rescue, followed by isatuximab plus KRd consolidation, followed by isatuximab plus KR maintenance  Isatuximab plus KRd induction followed by isatuximab plus KR maintenance

CyBorD, cyclophosphamide-bortezomib-dexamethasone; DRd, daratumumab plus lenalidomide-dexamethasone; KR, carfilzomib and lenalidomide; KRd, carfilzomib-lenalidomide-dexamethasone; NDMM, newly diagnosed MM; Rd, lenalidomide-dexamethasone; Vd, bortezomib-dexamethasone; VMP, bortezomib-melphalan-prednisone; VRD or RVD, bortezomib-lenalidomide-dexamethasone; VTD, bortezomib-thalidomide-dexamethasone.

daratumumab.<sup>93</sup> First, denaturation of CD38 by dithiothreitol (DTT) negates daratumumab interference and allows identification of underlying clinically significant alloantibodies.<sup>88</sup> A disadvantage of the DTT method is the disruption of a limited number of other blood group antigens with the Kell antigen as the most relevant DTT-sensitive red blood cell antigen in routine clinical practice. Therefore, Kell<sup>-</sup> units should be provided when using the DTT method, unless the patient is known to be Kell<sup>+</sup>.<sup>87,88</sup> In a multicenter international study, this approach was shown to be robust and reproducible.<sup>87</sup> A neutralization method with the use of a daratumumab idiotype antibody or recombinant soluble CD38 is also effective, but currently not widely available.<sup>88,89</sup> A third approach is red blood cell phenotyping or genotyping in patients who will be treated with a CD38 antibody.<sup>92</sup> Phenotyping should be done before CD38 therapy is started, whereas genotyping can be performed after CD38 treatment has started. In case of transfusion, phenotypically or genotypically matched red blood cell units should be given. We also provide our patients with blood transfusion cards indicating that CD38 antibody therapy is being received. Although daratumumab interferes with blood typing and cross-matching, no adverse events associated with blood transfusions have been described.<sup>30,31,48,59,94,95</sup>

The other CD38 antibodies also interfere in the IAT,<sup>89</sup> and similar mitigation strategies can be used as clinically indicated.

## New applications for CD38 therapy

### Other plasma cell dyscrasias

CD38 antibodies are currently also being evaluated in smoldering MM and certain monoclonal gammopathies with renal significance (Table 4). Furthermore, preliminary results show that

daratumumab is effective and safe in patients with advanced immunoglobulin light-chain amyloidosis.<sup>96,97</sup>

### Other hematological malignancies

CD38 antibodies also have activity against lymphoma cell lines and in a lymphoma mouse model.<sup>20,21,24,69</sup> Furthermore, daratumumab exerts significant cytotoxicity against chronic lymphocytic leukemia cells via ADCC and ADCP, but without significant CDC, probably as a result of high complement inhibitor expression.<sup>98</sup> In addition, daratumumab interferes with CD38 signaling and reduces CLL adhesion, migration, and homing.<sup>98</sup>

CD38 is also strongly expressed on NK cells, which explains the sustained response with daratumumab in a patient with relapsed/refractory nasal-type extranodal NK-cell-T-cell lymphoma.<sup>99</sup> Preclinical studies have also shown activity of CD38 antibodies against T-cell acute lymphoblastic leukemia (ALL) and B-cell ALL,<sup>24</sup> Waldenström macroglobulinemia,<sup>100</sup> and acute myeloid leukemia (AML).<sup>101</sup> However, the BM microenvironment impairs the anti-AML activity of daratumumab.<sup>101</sup> Several studies with CD38 antibodies in other CD38<sup>+</sup> hematologic malignancies are currently ongoing (Table 4).

### Solid tumors

CD38 antibodies may also have therapeutic potential beyond the treatment of hematologic malignancies (Table 4). Because daratumumab has immunomodulatory effects via elimination of CD38<sup>+</sup> immune-suppressor cells,<sup>5</sup> CD38 antibodies may even be effective in CD38<sup>-</sup> tumors. Interestingly, preclinical lung cancer studies suggest that CD38 confers resistance to PD-L1 blockade by inhibiting CD8<sup>+</sup> T-cell function, which explains the marked efficacy



of combination therapy of PD-L1 blocker and CD38 antibody.<sup>77</sup> This forms the preclinical rationale of various ongoing studies with daratumumab plus PD-1/PD-L1-neutralizing antibodies.<sup>77</sup>

### Autoimmune and allergic diseases

CD38 antibodies may also be active in autoantibody-mediated diseases by eradicating autoantibody-producing plasma cells. Indeed, daratumumab was effective in the treatment of refractory autoimmune hemolytic anemia.<sup>102</sup> Similarly, daratumumab reduces total and allergen-specific IgE levels by depleting IgE-producing plasma cells, and this suggests potential value of CD38 antibodies in the management of severe allergy.<sup>103</sup>

## Conclusions and future prospects

Although CD38 is widely expressed, CD38-targeting antibodies have manageable toxicity. Furthermore, by virtue of distinct mechanisms of action, CD38 antibodies have impressive single-agent activity in heavily pretreated MM patients, including those refractory to newer agents such as pomalidomide and carfilzomib. CD38 antibodies can also safely be added to backbone regimens, markedly increasing their efficacy. There is currently a lack of direct comparisons among different treatment regimens for early relapsed MM. However, the PFS advantage observed with daratumumab added to lenalidomide and bortezomib is unprecedented. Furthermore, a meta-analysis has identified DRd as the most effective treatment option in relapsed/refractory MM, followed by other triplets including DVd and Rd plus elotuzumab, ixazomib, or carfilzomib.<sup>104</sup> Biomarkers need to be identified that predict response to these different combinations to further improve and personalize treatment. Choice of treatment of patients who become refractory to CD38 antibody-based therapy should be made on an individual basis taking into account tumor and patient characteristics, as well as treatment history. Because CD38 antibodies reduce NK-cell numbers,<sup>52</sup> it is currently unknown whether antibodies that depend on NK-cell-mediated ADCC, such as elotuzumab, are equally effective before and after CD38 antibody therapy. Furthermore, the concept of adding another backbone regimen to the CD38-targeting antibody should also be further explored.

We also expect that CD38 antibodies will increasingly be incorporated into first-line regimens over the next few years given their efficacy and manageable toxicity profile. Preliminary results demonstrate that incorporation of daratumumab in standard

regimens, such as Vd, bortezomib-thalidomide-dexamethasone, bortezomib-melphalan-prednisone, or carfilzomib-lenalidomide-dexamethasone, did not result in significant additional toxicity, other than IRRs.<sup>105</sup> Various studies are currently evaluating the role of CD38 antibodies in induction, consolidation, and maintenance in both transplant-eligible and elderly patients with newly diagnosed MM (Table 5). Furthermore, CD38 antibodies are also being evaluated in smoldering MM to prevent progression to active MM (Table 5). Also, the impact of combining CD38 antibodies with other immunotherapeutic antibodies is of particular interest going forward as the immuno-oncology field in MM expands. Finally, treatment of patients with CD38 antibodies also requires bidirectional communication between the clinician and laboratory/blood bank when assessing therapeutic responses to minimize errors or delays in providing compatible red blood cell units.

## Acknowledgments

The authors thank Victor Muñoz Sanz (Sanz Serif Research + Design Agency) for creating Figures 1, 2, and 3. The authors also thank Caline Sakabedoyan for help with the DIRA image and blood transfusion interference image.

## Authorship

Contribution: All authors performed literature searches and prepared the manuscript.

Conflict-of-interest disclosure: N.W.C.J.v.d.D. has received research support from Janssen Pharmaceuticals, Amgen, Celgene, and Bristol-Myers Squibb (BMS), and serves on advisory boards for Janssen Pharmaceuticals, Amgen, Celgene, BMS, Novartis, and Servier. P.G.R. has served on advisory boards for Millennium Pharmaceuticals, Celgene, Novartis, Janssen Pharmaceuticals, and BMS. F.M. has received research support from Janssen Pharmaceuticals, Celgene, Tusk Therapeutics, and Centrose, and serves on advisory boards for Centrose and Tusk Therapeutics.

Correspondence: Niels W. C. J. van de Donk, Department of Hematology, VU University Medical Center, De Boelelaan 1117, 1081HV Amsterdam, The Netherlands; e-mail: n.vandedonk@vumc.nl.

## Footnotes

Submitted 31 May 2017; accepted 17 July 2017. Prepublished online as *Blood* First Edition paper, 8 November 2017; DOI 10.1182/blood-2017-06-740944.

## REFERENCES

- Laubach JP, Richardson PG. CD38-targeted immunochemotherapy in refractory multiple myeloma: a new horizon. *Clin Cancer Res*. 2015;21(12):2660-2662.
- 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 USA*. 1980;77(3):1588-1592.
- Terhorst C, van Agthoven A, LeClair K, Snow P, Reinherz E, Schlossman S. Biochemical studies of the human thymocyte cell-surface antigens T6, T9 and T10. *Cell*. 1981;23(3):771-780.
- Deaglio S, Mehta K, Malavasi F. Human CD38: a (r)evolutionary story of enzymes and receptors. *Leuk Res*. 2001;25(1):1-12.
- Krejci J, Casneuf T, Nijhof IS, et al. Daratumumab depletes CD38+ immune regulatory cells, promotes T-cell expansion, and skews T-cell repertoire in multiple myeloma. *Blood*. 2016;128(3):384-394.
- Crowell PD, Goldstein AS. Functional evidence that progenitor cells near sites of inflammation are precursors for aggressive prostate cancer. *Mol Cell Oncol*. 2017;4(2):e1279723.
- Kotlikoff MI, Kannan MS, Solway J, et al. Methodologic advancements in the study of airway smooth muscle. *J Allergy Clin Immunol*. 2004;114(suppl 2):S18-S31.
- Horenstein AL, Sizzano F, Lusso R, et al. CD38 and CD157 ectoenzymes mark cell subsets in the human corneal limbus. *Mol Med*. 2009;15(3-4):76-84.
- Song EK, Lee YR, Kim YR, et al. NAADP mediates insulin-stimulated glucose uptake and insulin sensitization by PPAR $\gamma$  in adipocytes. *Cell Reports*. 2012;2(6):1607-1619.
- Alessio M, Roggero S, Funaro A, et al. CD38 molecule: structural and biochemical analysis on human T lymphocytes, thymocytes, and plasma cells. *J Immunol*. 1990;145(3):878-884.
- Malavasi F, Deaglio S, Funaro A, et al. Evolution and function of the ADP ribosyl cyclase/CD38 gene family in physiology and pathology. *Physiol Rev*. 2008;88(3):841-886.

12. Deaglio S, Morra M, Mallone R, et al. Human CD38 (ADP-ribosyl cyclase) is a counter-receptor of CD31, an Ig superfamily member. *J Immunol*. 1998;160(1):395-402.
13. Muñoz P, Mittelbrunn M, de la Fuente H, et al. Antigen-induced clustering of surface CD38 and recruitment of intracellular CD38 to the immunologic synapse. *Blood*. 2008;111(7):3653-3664.
14. Howard M, Grimaldi JC, Bazan JF, et al. Formation and hydrolysis of cyclic ADP-ribose catalyzed by lymphocyte antigen CD38. *Science*. 1993;262(5136):1056-1059.
15. Lee HC. Nicotinic acid adenine dinucleotide phosphate (NAADP)-mediated calcium signaling. *J Biol Chem*. 2005;280(40):33693-33696.
16. Cagnetta A, Cea M, Calimeri T, et al. Intracellular NAD<sup>+</sup> depletion enhances bortezomib-induced anti-myeloma activity. *Blood*. 2013;122(7):1243-1255.
17. Chillemi A, Quarona V, Antonioli L, Ferrari D, Horenstein AL, Malavasi F. Roles and modalities of ectonucleotidases in remodeling the multiple myeloma niche. *Front Immunol*. 2017;8:305.
18. Stevenson FK, Bell AJ, Cusack R, et al. Preliminary studies for an immunotherapeutic approach to the treatment of human myeloma using chimeric anti-CD38 antibody. *Blood*. 1991;77(5):1071-1079.
19. Goldmacher VS, Bourret LA, Levine BA, et al. Anti-CD38-blocked ricin: an immunotoxin for the treatment of multiple myeloma. *Blood*. 1994;84(9):3017-3025.
20. 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.
21. 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.
22. Overdijk MB, Jansen JH, Nederend M, et al. The therapeutic CD38 monoclonal antibody daratumumab induces programmed cell death via Fcγ receptor-mediated cross-linking. *J Immunol*. 2016;197(3):807-813.
23. Lammerts van Bueren J, Jakobs D, Kaldenhoven N, et al. Direct in vitro comparison of daratumumab with surrogate analogs of CD38 antibodies MOR03087, SAR650984 and Ab79 [abstract]. *Blood*. 2014;124(21). Abstract 3474.
24. Deckert J, Wetzell MC, Bartle LM, et al. SAR650984, a novel humanized CD38-targeting antibody, demonstrates potent antitumor activity in models of multiple myeloma and other CD38<sup>+</sup> hematologic malignancies. *Clin Cancer Res*. 2014;20(17):4574-4583.
25. Jiang H, Acharya C, An G, et al. SAR650984 directly induces multiple myeloma cell death via lysosomal-associated and apoptotic pathways, which is further enhanced by pomalidomide. *Leukemia*. 2016;30(2):399-408.
26. Adams H III, Stevenaert F, Krejcik J, et al. High-parameter mass cytometry (CyTOF) evaluation of relapsed/refractory multiple myeloma (MM) Pts (Pts) treated with daratumumab supports immune modulation as a novel mechanism of action [abstract]. *Blood*. 2016;128(22). Abstract 4521.
27. Feng X, Zhang L, Acharya C, et al. Targeting CD38 suppresses induction and function of T regulatory cells to mitigate immunosuppression in multiple myeloma. *Clin Cancer Res*. 2017;23(15):4290-4300.
28. Usmani SZ, Diels J, Ito T, Mehra M, Khan I, Lam A. Daratumumab monotherapy compared with historical control data in heavily pretreated and highly refractory patients with multiple myeloma: an adjusted treatment comparison. *Am J Hematol*. 2017;92(8):E146-E152.
29. Lokhorst HM, Plesner T, Laubach JP, et al. Targeting CD38 with daratumumab monotherapy in multiple myeloma. *N Engl J Med*. 2015;373(13):1207-1219.
30. Lonial S, Weiss BM, Usmani SZ, et al. Daratumumab monotherapy in patients with treatment-refractory multiple myeloma (SIRIUS): an open-label, randomised, phase 2 trial. *Lancet*. 2016;387(10027):1551-1560.
31. Usmani SZ, Weiss BM, Plesner T, et al. Clinical efficacy of daratumumab monotherapy in patients with heavily pretreated relapsed or refractory multiple myeloma. *Blood*. 2016;128(1):37-44.
32. Nijhof IS, Groen RW, Noort WA, et al. Preclinical evidence for the therapeutic potential of CD38-targeted immunotherapy in multiple myeloma patients refractory to lenalidomide and bortezomib. *Clin Cancer Res*. 2015;21(12):2802-2810.
33. Clemens PL, Yan X, Lokhorst HM, et al. Pharmacokinetics of daratumumab following intravenous infusion in relapsed or refractory multiple myeloma after prior proteasome inhibitor and immunomodulatory drug treatment. *Clin Pharmacokinet*. 2017;56(8):915-924.
34. Xu XS, Yan X, Puchalski T, et al. Clinical implications of complex pharmacokinetics for daratumumab dose regimen in patients with relapsed/refractory multiple myeloma. *Clin Pharmacol Ther*. 2017;101(6):721-724.
35. Usmani S, Nahi H, Mateos MV, et al. Open-label, multicenter, dose escalation phase 1b study to assess the subcutaneous delivery of daratumumab in patients (pts) with relapsed or refractory multiple myeloma (PAVO) [abstract]. *Blood*. 2016;128(22). Abstract 1149.
36. Raab M, Chatterjee M, Goldschmidt H, et al. A phase I/IIa study of the CD38 antibody MOR202 alone and in combination with pomalidomide or lenalidomide in patients with relapsed or refractory multiple myeloma [abstract]. *Blood*. 2016;128(22). Abstract 1152.
37. Martin TG III, Richter J, Vij R, Cole C, Atanackovic D, Zonder J. A dose finding phase II trial of isatuximab (SAR650984, anti-CD38 mAb) as a single agent in relapsed/refractory multiple myeloma [abstract]. *Blood*. 2015;126(23). Abstract 509.
38. Richter J, Martin TG III, Vij R, et al. Updated data from a phase II dose finding trial of single agent isatuximab (SAR650984, anti-CD38 mAb) in relapsed/refractory multiple myeloma (RRMM) [abstract]. *J Clin Oncol*. 2016;34(suppl 15):8005.
39. 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. *Leukemia*. 2015;29(10):2039-2049.
40. Nijhof IS, Casneuf T, van Velzen J, et al. CD38 expression and complement inhibitors affect response and resistance to daratumumab therapy in myeloma. *Blood*. 2016;128(7):959-970.
41. de Haart SJ, van de Donk NW, Minnema MC, et al. Accessory cells of the microenvironment protect multiple myeloma from T-cell cytotoxicity through cell adhesion-mediated immune resistance. *Clin Cancer Res*. 2013;19(20):5591-5601.
42. de Haart SJ, Holthof L, Noort WA, et al. Sepantronium bromide (YM155) improves daratumumab-mediated cellular lysis of multiple myeloma cells by abrogation of bone marrow stromal cell-induced resistance. *Haematologica*. 2016;101(8):e339-e342.
43. Marra J, Du J, Hwang J, Wolf J, Martin TG III, Venstrom J. KIR and HLA genotypes influence clinical outcome in multiple myeloma patients treated with SAR650984 (anti-CD38) in combination with lenalidomide and dexamethasone [abstract]. *Blood*. 2014;124(21). Abstract 2126.
44. Horenstein AL, Chillemi A, Quarona V, et al. NAD<sup>+</sup>-metabolizing ectoenzymes in remodeling tumor-host interactions: the human myeloma model. *Cells*. 2015;4(3):520-537.
45. An G, Jiang H, Acharya C, et al. SAR 650984, a therapeutic anti-CD38 monoclonal antibody, blocks CD38-CD31 interaction in multiple myeloma [abstract]. *Blood*. 2014;124(21). Abstract 4729.
46. Chillemi A, Quarona V, Zito A, et al. Generation and characterization of microvesicles after daratumumab interaction with myeloma cells [abstract]. *Blood*. 2015;126(23). Abstract 1849.
47. Taylor RP, Lindorfer MA. Fcγ-receptor-mediated trogocytosis impacts mAb-based therapies: historical precedence and recent developments. *Blood*. 2015;125(5):762-766.
48. Palumbo A, Chanan-Khan A, Weisel K, et al; CASTOR Investigators. Daratumumab, bortezomib, and dexamethasone for multiple myeloma. *N Engl J Med*. 2016;375(8):754-766.
49. Martin T, Strickland S, Glenn M, Zheng W, Daskalakis N, Mikhael J. SAR650984, a CD38 monoclonal antibody in patients with selected CD38<sup>+</sup> hematological malignancies: data from a dose-escalation phase I study [abstract]. *Blood*. 2013;122(21):284.
50. Raab M, Goldschmidt H, Agis H, Blau I, Einsele H, Engelhardt M. A phase I/IIa study

- of the human anti-CD38 antibody MOR202 (MOR03087) in relapsed or refractory multiple myeloma (rrMM) [abstract]. *J Clin Oncol*. 2015;33(suppl):8574.
51. Cherkasova E, Espinoza L, Kotecha R, Reger R, Berg M, Aue G. Treatment of ex vivo expanded NK cells with daratumumab F(ab')<sub>2</sub> fragments protects adoptively transferred NK cells from daratumumab-mediated killing and augments daratumumab-induced antibody dependent cellular toxicity (ADCC) of myeloma [abstract]. *Blood*. 2015;126(23). Abstract 4244.
  52. Casneuf T, Xu X, Adams H, et al. Pharmacodynamic relationship between natural killer cells and daratumumab exposure in relapsed/refractory multiple myeloma [abstract]. In: Proceedings from the EHA21 Congress; 10 June 2016; Copenhagen, Denmark. Abstract P286.
  53. Alici E, Chrobok M, Lund J, et al. Re-challenging with anti-CD38 monotherapy in triple-refractory multiple myeloma patients is a feasible and safe approach. *Br J Haematol*. 2016;174(3):473-477.
  54. 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.
  55. 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.
  56. Boxhammer R, Steidl S, Endell J. Effect of IMiD compounds on CD38 expression on multiple myeloma cells: MOR202, a human CD38 antibody in combination with pomalidomide [abstract]. *J Clin Oncol*. 2015;33(suppl 15):8588.
  57. Endell J, Boxhammer R, Steidl S. Synergistic in vitro activity of MOR202, a human CD38 antibody, in combination with pomalidomide [abstract]. *Blood*. 2014;124(21). Abstract 5712.
  58. Bruns H, Jitschin R, Mougiakakos D, et al. Lenalidomide enhances MOR202 dependent macrophage-mediated effector functions via the vitamin D pathway [abstract]. *Blood*. 2015;126(23). Abstract 2203.
  59. Dimopoulos MA, Oriol A, Nahi H, et al; POLLUX Investigators. Daratumumab, lenalidomide, and dexamethasone for multiple myeloma. *N Engl J Med*. 2016;375(14):1319-1331.
  60. Usmani S, Dimopoulos M, Belch A, et al. Efficacy of daratumumab, lenalidomide, and dexamethasone versus lenalidomide and dexamethasone in relapsed or refractory multiple myeloma patients with 1 to 3 prior lines of therapy: updated analysis of Pollux [abstract]. *Blood*. 2016;128(22). Abstract 1151.
  61. Avet-Loiseau H, Casneuf T, Chiu C, et al. Evaluation of minimal residual disease (MRD) in relapsed/refractory multiple myeloma (RRMM) patients treated with daratumumab in combination with lenalidomide plus dexamethasone or bortezomib plus dexamethasone [abstract]. *Blood*. 2016;128(22). Abstract 246.
  62. San-Miguel J, Dimopoulos MA, Usmani S, et al. Depth of response and MRD with daratumumab plus lenalidomide and dexamethasone (DRd) vs lenalidomide and dexamethasone (Rd) in RRMM: Pollux [abstract]. *Clin Lymphoma Myeloma Leuk*. 2017;17(suppl 1):e17-e18.
  63. Chiu C, Casneuf T, Axel A, et al. Daratumumab in combination with lenalidomide plus dexamethasone induces clonality increase and T-cell expansion: results from a phase 3 randomized study (POLLUX) [abstract]. *Blood*. 2016;128(22). Abstract 4531.
  64. Martin T, Baz R, Benson DM, et al. A phase 1b study of isatuximab plus lenalidomide and dexamethasone for relapsed/refractory multiple myeloma. *Blood*. 2017;129(25):3294-3303.
  65. Chatterjee M, Raab MS, Goldschmidt H, et al. A phase I/IIa study of the CD38 antibody MOR202 in combination with pomalidomide or lenalidomide in patients with relapsed or refractory multiple myeloma [abstract]. *Clin Lymphoma Myeloma Leuk*. 2017;17(suppl 1):e61.
  66. Chari A, Suvannasankha A, Fay J, et al. Phase 1b study of daratumumab plus pomalidomide and dexamethasone in relapsed and/or refractory multiple myeloma (RRMM) with  $\geq 2$  prior lines of therapy [abstract]. *Clin Lymphoma Myeloma Leuk*. 2017;17(suppl 1):e14-e15.
  67. Richardson P, Mikhael J, Usmani S, Raje N, Bensinger W, Campana F. Preliminary results from a phase 1b study of isatuximab in combination with pomalidomide and dexamethasone in relapsed and refractory multiple myeloma [abstract]. *Blood*. 2016;128(22):2123.
  68. Moreno L, Zabaleta A, Alignani D, et al. Critical analysis on the mechanism of action (MoA) of the anti-CD38 monoclonal antibody isatuximab in multiple myeloma (MM) [abstract]. *Blood*. 2016;128(22):2105.
  69. Boxhammer R, Weirather J, Steidl C, Endell J. MOR202, a human anti-CD38 monoclonal antibody, mediates potent tumoricidal activity in vivo and shows synergistic efficacy in combination with different antineoplastic compounds [abstract]. *Blood*. 2015;126(23):3015.
  70. Mateos MV, Estell J, Barreto W, et al. Efficacy of daratumumab, bortezomib, and dexamethasone versus bortezomib and dexamethasone in relapsed or refractory myeloma based on prior lines of therapy: updated analysis of castor [abstract]. *Blood*. 2016;128(22):1150.
  71. Weisel KC, San Miguel J, Cook G, et al. Efficacy of daratumumab in combination with lenalidomide plus dexamethasone (DRd) or bortezomib plus dexamethasone (DvD) in relapsed or refractory multiple myeloma (RRMM) based on cytogenetic risk status [abstract]. *J Clin Oncol*. 2017;35(suppl 15):8006.
  72. Martin TG III, Mannis G, Chari A, Munster P, Campana F, Hui A. Phase 1b study of isatuximab and carfilzomib in relapse and refractory multiple myeloma [abstract]. *Blood*. 2016;128(22):2111.
  73. Jakubowiak AJ, Chari A, Lonial S, et al. Daratumumab (DARA) in combination with carfilzomib, lenalidomide, and dexamethasone (KRd) in patients (pts) with newly diagnosed multiple myeloma (MMY1001): an open-label, phase 1b study [abstract]. *J Clin Oncol*. 2017;35(suppl 15):8000.
  74. Garcia-Guerrero E, Gogishvili T, Danhof S, et al. Panobinostat induces CD38 upregulation and augments the antimyeloma efficacy of daratumumab. *Blood*. 2017;129(25):3386-3388.
  75. 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.
  76. Jaiswal S, Jamieson CH, Pang WW, et al. CD47 is upregulated on circulating hematopoietic stem cells and leukemia cells to avoid phagocytosis. *Cell*. 2009;138(2):271-285.
  77. Chen L, Averett Byers L, Ullrich S, Wistuba I, Qin X, Gibbons D. CD38 as a novel immune checkpoint and a mechanism of resistance to the blockade of the PD-1/PD-L1 axis [abstract]. *J Clin Oncol*. 2017;35(suppl 7S). Abstract 79.
  78. van de Donk NW, Moreau P, Plesner T, et al. Clinical efficacy and management of monoclonal antibodies targeting CD38 and SLAMF7 in multiple myeloma. *Blood*. 2016;127(6):681-695.
  79. Voorhees PM, Weiss B, Usmani S, et al. Management of infusion-related reactions following daratumumab monotherapy in patients with at least 3 lines of prior therapy or double refractory multiple myeloma (MM): 54767414MMY2002 (Sirius) [abstract]. *Blood*. 2015;126(23):1829.
  80. Chari A, Mark T, Krishnan A, et al. Use of montelukast to reduce infusion reactions in an early access treatment protocol of daratumumab in United States patients with relapsed or refractory multiple myeloma [abstract]. *Blood*. 2016;128(22):2142.
  81. van de Donk NW, Otten HG, El Haddad O, et al. Interference of daratumumab in monitoring multiple myeloma patients using serum immunofixation electrophoresis can be abrogated using the daratumumab IFE reflex assay (DIRA). *Clin Chem Lab Med*. 2016;54(6):1105-1109.
  82. McCudden C, Axel AE, Slaets D, et al. Monitoring multiple myeloma patients treated with daratumumab: teasing out monoclonal antibody interference. *Clin Chem Lab Med*. 2016;54(6):1095-1104.
  83. Rosenberg AS, Bainbridge S, Pahwa R, Jialal I. Investigation into the interference of the monoclonal antibody daratumumab on the free light chain assay. *Clin Biochem*. 2016;49(15):1202-1204.
  84. Durie BG, Miguel JF, Blade J, Rajkumar SV. Clarification of the definition of complete response in multiple myeloma. *Leukemia*. 2015;29(12):2416-2417.

85. Caillon H, Irimia A, Simon J, et al. Overcoming the interference of daratumumab with immunofixation electrophoresis (IFE) using an industry-developed Dira test: hydrashift 2/4 daratumumab [abstract]. *Blood*. 2016;128(22):2063.
86. Murata K, McCash SI, Carroll B, et al. Treatment of multiple myeloma with monoclonal antibodies and the dilemma of false positive M-spikes in peripheral blood [published online ahead of print 21 September 2016]. *Clin Biochem*. doi:10.1016/j.clinbiochem.2016.09.015.
87. Chapuy CI, Aguad MD, Nicholson RT, et al; DARA-DTT Study Group\* for the BEST Collaborative. International validation of a dithiothreitol (DTT)-based method to resolve the daratumumab interference with blood compatibility testing. *Transfusion*. 2016; 56(12):2964-2972.
88. Chapuy CI, Nicholson RT, Aguad MD, et al. Resolving the daratumumab interference with blood compatibility testing. *Transfusion*. 2015;55(6 Pt 2):1545-1554.
89. Oostendorp M, Lammerts van Bueren JJ, Doshi P, et al. When blood transfusion medicine becomes complicated due to interference by monoclonal antibody therapy. *Transfusion*. 2015;55(6 Pt 2):1555-1562.
90. Sullivan HC, Gerner-Smidt C, Nooka AK, et al. Daratumumab (anti-CD38) induces loss of CD38 on red blood cells. *Blood*. 2017; 129(22):3033-3037.
91. Lintel NJ, Brown DK, Schafer DT, Tsimba-Chitsva FM, Koepsell SA, Shunkwiler SM. Use of standard laboratory methods to obviate routine dithiothreitol treatment of blood samples with daratumumab interference. *Immunohematology*. 2017;33(1):22-26.
92. Hannon JL, Clarke G. Transfusion management of patients receiving daratumumab therapy for advanced plasma cell myeloma. *Transfusion*. 2015;55(11):2770.
93. Murphy MF, Dumont LJ, Greinacher A; BEST Collaborative. Interference of new drugs with compatibility testing for blood transfusion. *N Engl J Med*. 2016;375(3):295-296.
94. Dimopoulos MA, Sonneveld P, Sun H. Daratumumab and blood-compatibility testing. *N Engl J Med*. 2016;375(25):2497-2498.
95. Chari A, Satta T, Tayal A, et al. Outcomes and management of red blood cell transfusions in multiple myeloma patients treated with daratumumab [abstract]. *Blood*. 2015; 126(23). Abstract 3571.
96. Sher T, Fenton B, Akhtar A, Gertz MA. First report of safety and efficacy of daratumumab in 2 cases of advanced immunoglobulin light chain amyloidosis. *Blood*. 2016;128(15): 1987-1989.
97. Kaufman G, Witteles R, Wheeler M, et al. Hematologic responses and cardiac organ improvement in patients with heavily pretreated cardiac immunoglobulin light chain (AL) amyloidosis receiving daratumumab [abstract]. *Blood*. 2016;128(22). Abstract 4525.
98. Matas-Céspedes A, Vidal-Crespo A, Rodríguez V, et al. The human CD38 monoclonal antibody daratumumab shows antitumor activity and hampers leukemia-microenvironment interactions in chronic lymphocytic leukemia. *Clin Cancer Res*. 2017;23(6):1493-1505.
99. Hari P, Raj RV, Olteanu H. Targeting CD38 in refractory extranodal natural killer cell-T-cell lymphoma. *N Engl J Med*. 2016;375(15): 1501-1502.
100. Paulus A, Akhtar S, Bashir Y, et al. Drug resistance alters CD38 expression and in vitro response to daratumumab in waldenstrom macroglobulinemia cells [abstract]. *Blood*. 2016;128(22). Abstract 3018.
101. Dos Santos C, Xiaochuan S, Chenghui Z, et al. Anti-leukemic activity of daratumumab in acute myeloid leukemia cells and patient-derived xenografts [abstract]. *Blood*. 2014; 124(21). Abstract 2312.
102. Tolbert V, Goldsby R, Huang J, et al. Daratumumab is effective in the treatment of refractory post-transplant autoimmune hemolytic anemia: a pediatric case report [abstract]. *Blood*. 2016;128(22). Abstract 4819.
103. Blankestijn MA, van de Donk N, Sasser K, Knulst AC, Otten HG. Could daratumumab be used to treat severe allergy? *J Allergy Clin Immunol*. 2017;139(5):1677-1678.
104. van Beurden-Tan CHY, Franken MG, Blommestein HM, Uyl-de Groot CA, Sonneveld P. Systematic literature review and network meta-analysis of treatment outcomes in relapsed and/or refractory multiple myeloma. *J Clin Oncol*. 2017; 35(12):1312-1319.
105. 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 [abstract]. *Blood*. 2014;124(21). Abstract 176.
106. Richardson P, Mikhael J, Usmani S, et al. Preliminary results from a phase 1b study of isatuximab in combination with pomalidomide (Pom) and dexamethasone (Dex) in relapsed and refractory multiple myeloma (RRMM). *Clin Lymphoma Myeloma Leuk*. 2017;17(suppl 1):e16-e17.