Phase 1 study of CART-ddBCMA for the treatment of subjects with relapsed and refractory multiple myeloma

Matthew J. Frigault,1,2 Michael R. Bishop,3 Jacalyn Rosenblatt,4 Elizabeth K. O'Donnell,1,2 Noopur Raje,1,2 Daniella Cook,1,2 Andrew J. Yee,1,2 Emma Logan,4 David E. Avigan,4 Andrzej Jakubowiak,3 Kit Shaw,5 Heather Daley,3 Sarah Nikiforow,3 Faith Griffin,6 Christine Cornwell,6 Angela Shen,6 Christopher Heery,6 and Marcela V. Maus1,2

1Massachusetts General Hospital Cancer Center, Boston, MA; 2Harvard Medical School, Boston, MA; 3David and Etta Jonas Center for Cellular Therapy, University of Chicago, Chicago, IL; 4Beth Israel Deaconess Medical Center, Boston, MA; 5Dana Farber Cancer Institute, Cell Manipulation Core Facility, Brookline, MA; and 6Arcellx, Inc., Gaithersburg, MD

Key Points

• CART-ddBCMA is safe for use in patients with relapsed or refractory multiple myeloma.
• CART-ddBCMA produces deep and durable responses in patients with poor prognostic factors.

Relapsed and refractory multiple myeloma (RRMM) is a plasma cell neoplasm defined by progressively refractory disease necessitating chronic and increasingly intensive therapy. Despite recent advances, limited treatment options exist for RRMM. This single-arm, open label phase 1 study aimed to evaluate the safety of novel B-cell maturation antigen (BCMA)-targeting chimeric antigen receptor (CAR) T construct that leverages a completely synthetic antigen-binding domain (CART-ddBCMA), which was specifically engineered to reduce immunogenicity and improve CAR cell surface stability. Thirteen patients ≥18 years with RRMM who received at least 3 prior regimens of systemic therapy were enrolled in the study. Patients received a single dose of 100 × 10^6 CART-ddBCMA (DL1) or 300 × 10^6 CART-ddBCMA (DL2) following standard lymphodepleting chemotherapy. The primary endpoints of the study were to evaluate the incidence of treatment emergent adverse events, including dose-limiting toxicities, and establish a recommended phase 2 dose. Results showed that CART-ddBCMA was well tolerated and demonstrated a favorable toxicity profile. Only 1 case of grade ≥3 cytokine release syndrome and 1 case of immune effector cell–associated neurotoxicity were reported; both were at DL2 and were manageable with standard treatment. No atypical neurological toxicities and Parkinson disease-like movement disorders were observed. The maximum tolerated dose was not reached. All infused patients responded to CART-ddBCMA, and 9/12 (75%) patients achieved complete response/stringent complete response. Responses deepened over time, and at the time of last data-cut (median follow-up 56 weeks), 8/9 (89%) evaluable patients achieved minimal residual disease negativity. In conclusion, the findings demonstrate the safety of CART-ddBCMA cells and document durable responses to CART-ddBCMA in patients with RRMM. This trial was registered at www.clinicaltrials.gov as #NCT04155749.
We developed anti-BCMA CAR T cells with a CAR composed of a ν-domain-based antigen binder fused to the CD8 hinge and transmembrane domain in tandem with the intracellular signaling domains of 4-1BB and CD3ζ and introduced into human T cells via lentiviral vector (CART-ddBCMA). Based on the encouraging efficacy seen in preclinical studies, we initiated a phase 1 clinical study of CART-ddBCMA patients with relapsed/refractory MM (#NCT04155749).

**Methods**

**CART-ddBCMA**

CART-ddBCMA drug product consists of autologous T cells genetically modified ex vivo to express a binding domain that specifically recognizes BCMA. The binding domain was identified in a library of randomized α3D sequences using standard phage-display technologies, and site-directed mutagenesis was used to enhance target affinity and minimize immunogenicity. The resulting sequence encoding a 73 amino acid ν-domain with nanomolar affinity for human BCMA was cloned into a lentiviral vector along with CD8 hinge and transmembrane region, 4-1-BB and CD3ζ intracellular signaling domains. Preclinical studies of CAR T cells using ν-domains showed the absence of tonic signaling, consistently high levels of cell surface expression, and low immunogenicity based on in silico modeling. CART-ddBCMA displayed reproducible BCMA-dependent NFAT signaling, cytokine (interleukin-2, interferon-γ) secretion, and induced BCMA-specific cytotoxicity in tumor cell lines. In the mouse-human xenograft models, CART-ddBCMA eradicated BCMA-expressing tumors within 2 weeks of single administration. Body weights of the mice were not impacted by CART-ddBCMA treatment, and no histopathological findings in the in vivo studies were attributable to ddBCMA exposure.

**Study design**

This open-label, multicenter phase 1 trial enrolled adults with RRMM to evaluate the safety of intravenous administration of CART-ddBCMA (supplemental Figure 1). The protocol was approved by the Institutional Review Board at each center, and the trial was performed in accordance with the principles of the Declaration of Helsinki. Eligible patients required treatment with at least 3 prior lines of systemic therapy, including a proteasome inhibitor, an IMiD, and an anti-CD38 antibody. Alternatively, patients were eligible if deemed to have “triple-refractory” disease following treatment with proteasome inhibitor, IMiD, and anti-CD38 antibody as part of the same or different regimens. Eligibility criteria also included adequate organ function (creatinine clearance ≥50 mL/min and left ventricular ejection fraction ≥45%) and an Eastern Cooperative Group Performance Status of 0-1. Patients with plasma cell leukemia or active central nervous system involvement were excluded but ongoing anticoagulation was allowed. Patients with prior BCMA-targeted therapy were eligible after medical monitor discussion.

After providing written informed consent, patients were enrolled and underwent leukapheresis. Bridging therapy was allowed, but a 2-week washout was required prior to lymphodepleting chemotherapy and cell infusion. Repeat baseline assessments were required prior to initiation of lymphodepleting chemotherapy. For lymphodepleting chemotherapy, patients received a regimen of...
fludarabine (30 mg/m²) and cyclophosphamide (300 mg/m²) daily on days −5, −4, and −3 prior to cell infusion. Cells were manufactured by the Connell and O’Reilly Families Cell Manipulation Core Facility of the Dana-Farber/Harvard Cancer Center. Patients received a dose of 100 × 10⁶ CART-ddBCMA (dose level 1, DL1) or 300 × 10⁶ CART-ddBCMA (dose level 2, DL2) on day 0. Blood was collected at days 0, 1 to 4, 7, 9, 11, 14, 21, and 28 after CART-ddBCMA infusion and then months 2 to 6, 9, 12, 15, 18, 21, and 24 to monitor CAR T-cell expansion and persistence. Additional blood was drawn to evaluate correlative pharmacodynamic effects. Patients were also monitored for disease progression up to 24 months. At the time of disease progression, or at 24 months if progression did not occur, patients were transferred to long-term follow-up phase of the study. Retreatment of patients was possible with FDA and institutional review board approval, and patients were dosed from material remaining from the initial manufacturing run.

**End points**

The primary end points of the study were to evaluate the incidence of treatment-emergent adverse events (AEs), including dose-limiting toxicities (DLTs), and establish the recommended phase 2 dose. DLTs were defined as any investigational study drug–related grade 3+ toxicity occurring within the first 28 days as well as any grade 4 life-threatening toxicity, grade ≥3 cytokine release syndrome (CRS) that did not improve to ≤grade 2 within 72 hours, any grade ≥3 neurotoxicity including any seizures, any grade ≥3 toxicity involving vital organs (eg, cardiac, pulmonary) that resulted in significant and irreversible organ damage, any grade ≥3 nonhematologic toxicity that did not improve to ≤grade 2 within 72 hours, and any death not attributed to underlying malignancy. Toxicity grading was performed according to the National Cancer Institute Common Terminology Criteria for Adverse Events version 5.0. CRS and immune effector cell-associated neurotoxicity (ICANS) were graded according to the American Society for Transplant and Cellular Therapy consensus criteria. Secondary endpoints included, but were not limited to, duration of response, PFS, OS, and correlative and exploratory studies. Response assessments were performed per the International Myeloma Working Group (IMWG) consensus criteria.

**Statistical analysis**

Sample size was based on a 3 + 3 dose escalation design. A total of 6 evaluable subjects were enrolled in each dose level to ensure adequate evaluation for potential DLT incidence in selecting a recommended phase 2 dose. Data are presented as the median and range for continuous variables and frequency for categorical variables. Time-to-event analyses and the associated 95% confidence intervals were estimated using the Kaplan-Meier method. Subjects were censored at the date of last follow-up. All patients who received CART-ddBCMA infusion were included in this analysis as was planned per protocol.

**Results**

**Patient disposition and characteristics**

Between 19 November 2019 and 14 April 2021, 13 subjects were enrolled, and 12 were infused with CART-ddBCMA, 6 at DL1 and 6 at DL2 (Figure 1). One subject discontinued prior to cell infusion due to disease-related complications unrelated to the investigational product. As of the data cut for this analysis, on 4 November 2021, the median follow-up was 56 weeks (range, 33-90). Drug products were successfully manufactured for all 13 patients with a median vein-to-vein time of 35 days (range, 33-42 days) for infused patients. CAR expression in the final product was consistent for all patients, and the variability in % CAR⁺ cells in the final product was low between patients. The median CAR⁺ cells in total CD3⁺ cells was 74.5% (range, 61% to 87%). The median patient age was 69 years (range, 44-76) for patients treated with 100 × 10⁶ CART-ddBCMA and 60 years (range, 52-65) for those treated with 300 × 10⁶ CART-ddBCMA (Table 1). The median time since diagnosis was 6.5 years (range, 1.8-11.8 years), and patients had received a median of 5 (range, 5-7), 4 (range, 3-16), and 5 (range, 3-16) prior lines of therapy in DL1, DL2, and overall, respectively. Nine of the 10 subjects with evaluable cytogenetics (90%) had high-risk features per IMWG, 7/12 subjects (58%) had extramedullary disease at time of treatment, and 10/12 subjects (83%) were considered penta-refractory at time of enrollment. One patient had progressed following treatment with a BCMA ADC. All enrolled patients received bridging therapy following leukapheresis for progressive and/or symptomatic disease.

**Safety**

All patients experienced grade 3 or 4 treatment-emergent AEs following CART-ddBCMA infusion, as shown in Table 2. The most common AEs were hematologic, including neutropenia (92%), anemia (83%), lymphocytopenia (87%), and decreased hemoglobin (75%). Most patients had cytopenias resolved to baseline or ≤grade 2 by 28 days. Of those patients who did not, all but 1 patient resolved to baseline or ≤grade 2 with standard interventions by month 5. Lymphocytopenia in 1 patient was resolved to baseline levels by month 12. Investigator attribution of these events was related to lymphodepletion chemotherapy plus underlying bone marrow function. In all cases of CR or sCR, improvement in cytopenias occurred compared with a screening of baseline value. The most common nonhematologic grade 3 or 4 AEs were hypertension (25%) and electrolyte imbalances (17%). There were no treatment-emergent grade 3 or 4 infections. CAR T-cell–associated toxicities occurred in all subjects, but most were low grade and manageable. CRS occurred in all patients, with a median onset of 2.5 days (range, 0-6 days) and duration of 7 days (range, 3-8 days) in DL1 and 1 day (range, 0-3 days) and 4.5 days (range, 3-6 days), respectively, in DL2. No patient in DL1 experienced grade 3+ CRS, but 1 patient experienced grade 3 CRS in DL2 that was partly attributed to a delay in defined tocilizumab administration (Table 3). Four subjects in DL1 and 5 subjects in DL2 (9/12 overall) required tocilizumab for the management of CRS (median dose; range, 1-2 doses). Two subjects in DL1 and 3 subjects in DL2 received 1 dose of dexamethasone for CRS management. ICANS occurred in 2 subjects: 1 in DL1 and 1 in DL2. The subject in DL1 experienced grade 2 ICANS that began on D+2 and resolved by D+5 with administration of steroids. The subject in DL2 experienced ICANS on D+6 as decreased mental status and decreasing immune effector cell-associated encephalopathy score of 7 consistent with grade 1 characteristics. Severe CRS was not seen in this subject, and the patient did not require tocilizumab. The severity of ICANS was grade 3 on D+9 based on clinical presentation and immune effector cell-associated encephalopathy score of 2, which was solely driven by global aphasia rather than decreased level of consciousness because the patient remained able to follow commands and intermittently...
respond. Following treatment with anakinra and steroids, the subject improved to grade 2 ICANS on D+19 and to grade 1 on D+20, and the AE was completely resolved on D+22. No long-term deficits or sequela have been identified in both subjects. Also, at the time of last data cut there were no cases of delayed-onset progressive movement disorders with features of Parkinson disease, as described in other investigational and commercially available BCMA-targeted CAR T-cell products.\textsuperscript{52,53} Given the low incidence of high-grade CAR T-cell–related AEs and only 1 observed DLT, a maximum tolerated dose of CART-ddBCMA was not reached.

**Efficacy**

At the time of data-cut, all subjects in the study had >200 days of follow-up. The median duration of follow-up was 12.6 months in all patients (15.6 months in DL1 and 8.3 months in DL2). The objective response rate was 100% across both dose levels of CART-ddBCMA, with 9 patients (75.0%) having CR/sCR, 1 (8.3%) having a very good partial response (PR), and 2 (16.7%) having a PR (Figure 2). The median time to response for all subjects was 28 days (range, 28-87) with deepening of responses over time. Median time to response was 28.5 days (range, 28-57) in DL1 and 28 days (range, 28-87) in DL2. Median duration of response, PFS, and OS was not reached at both DLs. Because the ORR was comparable between DL1 and DL2 and the toxicities were lower at DL1, additional subjects were enrolled in DL1.

Minimal residual disease (MRD) was evaluated by next-generation sequencing at day +28 in 9/12 patients. One month after CART-ddBCMA infusion, 5/9 patients were MRD negative ($10^{-5}, n=3$; $10^{-6}, n=2$) with further deepening of responses over time (Figure 2). At the time of last data cut, 5 subjects were MRD negative at $10^{-6}$ and 2 were MRD negative at $10^{-5}$. Of those who achieved MRD negativity at $10^{-6}$ ($n=5$), none have had progressive disease.

Disease progression was observed in 3 subjects. One subject treated on DL1 had progression at day +115 after a best response of PR at day +28. The subject was retreated with CART-ddBCMA at DL2 on day +136 and had further progression at day +205 from initial CAR T infusion. The second subject with disease progression was treated on DL2, reached PR at day 28, very good PR at month

---

**Table 1. Patient demographics**

| Characteristics | Dose level 1 100 million CAR-T (n = 6) | Dose level 2 300 million CAR-T (n = 6) |
|----------------|---------------------------------------|---------------------------------------|
| Age, y, median (min-max) | 73 (66-75) | 60 (53-65) |
| Sex | 3 male | 5 male |
| BMPC >50% | 3/6 | 4/6 |
| Extramedullary disease | 4/6 | 3/6 |
| High-risk cytogenetics per IMWG | 5/5\* | 4/5\* |
| Prior lines of therapy, median (min-max) | 5 (5-7) | 4 (3-16) |
| Prior HSCT | 3/6 | 4/6 |
| Penta-refractory† | 6/6 | 4/6 |
| IgG myeloma | 1 | 5 |
| IgA myeloma | 3 | 0 |
| Light chain only | 2 | 1 |

BMPC, bone marrow plasma cell; HSCT, hematopoietic stem cell transplant.
\*Some subjects were not evaluable or data were not available at time of data cutoff.
†Penta-refractory patients are refractory to bortezomib, carfilzomib, daratumumab, lenalidomide, and pomalidomide.
subject did not receive any retreatment at the time of reporting. For almost 1 year but had disease progression by day +336. The progression was treated on DL1 and had a PR, which was maintained following reinfusion. The third subject with disease progression was retreated at DL1. The subject had an ongoing PR at the time of ADC prior to enrollment in the study. After progression, the subject and rising M-protein (Figure 2). This subject had received a BCMA

| Table 3. CAR-T–associated adverse events |
|-----------------------------------------|
| **CART-T-associated AEs Per ASTCT criteria CRS** | **100 million (N = 6)** | **300 million (N = 6)** |
| | Grade 1/2 | Grade 3 | Grade 1/2 | Grade 3 |
| Median onset (min-max) | 2.5 d (0-4 d) | <24 h (0-1 d) | 2.5 d (0-4 d) | <24 h (0-1 d) |
| Median duration (min-max) | 5 d (2-7 d) | 3 d (1-9 d) | 5 d (2-7 d) | 3 d (1-9 d) |
| Neurotoxicity (ICANs) | 1 | 0 | 0 | 1 |

**Toxicity management**

- Tocilizumab
- Dexamethasone
- Anakinra

ASTCT, American Society for Transplantation and Cellular Therapy; ICANS, immune effector cell-associated neurotoxicity.

4, and sCR at month 9 (concurrently MRD negative at 10⁻⁶) but had progressive disease at day +320 with new lymphadenopathy and rising M-protein (Figure 2). This subject had received a BCMA ADC prior to enrollment in the study. After progression, the subject was retreated at DL1. The subject had an ongoing PR at the time of reporting following reinfusion. The third subject with disease progression was treated on DL1 and had a PR, which was maintained for almost 1 year but had disease progression by day +336. The subject did not receive any retreatment at the time of reporting.

**CAR T-cell expansion and persistence**

CAR-T-ddBCMA expansion was measured by transgene vector copy number in whole blood. The median time to peak expansion of CART-ddBCMA after infusion was 14 days (range, 9-21) in DL1, 10 days (range, 7-14) in DL2, and 11 days (range, 7-21) in all subjects. The median copies of vector transgene per microgram of genomic DNA at the peak level was 71 992 (range, 10 068-5 026 634) in DL1, 91 829 (range, 43 785-3 026 634) in DL2, and 111 829 (range, 42 785-3 026 634) in all subjects. Median persistence of CART-ddBCMA was 59 days (range, 21-180) in DL1, 42 days (range, 28-180) in DL2, and 42 days (range, 21-180) in all subjects. CART-ddBCMA kinetics, including peak level, time to peak expansion, and persistence, were similar for DL1 and DL2 (Figure 3).

Soluble BCMA (s-BCMA) levels in the serum, a marker for plasma cells and myeloma cells, decreased in all subjects following CART-ddBCMA treatment. The fall in s-BCMA levels in the peripheral blood continued even after CART-ddBCMA was undetectable in the peripheral blood (supplemental Figures 2-3). The recovery of s-BCMA levels was relatively slow in patients with ongoing response, and most patients had lower s-BCMA levels for >6 months, suggesting a slower recovery of plasma cells.

**Discussion**

This study demonstrated that the maximally tolerated dose of CART-ddBCMA was not exceeded at a flat dose of 300 × 10⁶ CAR⁺ cells. Evaluation of secondary endpoints indicates an ORR of 100%, with 75% of those responses being CR or better collectively and ≥67% being CR or better in each dose level. The AEs observed in this trial were consistent with previously observed AEs in BCMA CAR T-cell trials, including the pivotal study that led to ide-cel approval. In this study, only 1 patient (8.3%) had grade 3 neurotoxicity occurring at DL2 within the first week of treatment, which was the only DLT observed in the study. Importantly, no grade ≥3 CRS or ICANS occurred in DL1, and no cases of delayed-onset Parkinson-like progressive movement disorders were observed at either dose level. At DL1, the lack of grade ≥3 CRS and ICANS occurred in the context of 100% ORR (6/6) and 66.7% (4/6) sCR. No patients experienced atypical neurotoxicity despite a median follow-up of 12.6 months. Ten of the 12 subjects dosed with CART-ddBCMA (83.3%) remained in ongoing response at time of data cut (median follow-up 395 days). Additionally, of the patients who were evaluable, 88.9% were MRD-negative within 1 month of treatment, and many (5/6 patients who were tested multiple times) maintained or developed a deeper response to treatment over time based on their MRD status.

These responses occurred in patients with relatively poor prognostic indicators, including 7/12 (58.3%) with high tumor burden (bone marrow plasma cell >50%), 7/12 (58.3%) with extramedullary disease, and 9/10 evaluable (90%) with high-risk cytogenetics. They were also heavily pretreated, with 7/12 (58.3%) patients having prior hematopoietic stem cell transplant and 10/12 (83.3%) having penta-refractory disease. Given the comparable ORRs between DL1 and DL2 and the comparatively lower CAR T-related toxicities in patients treated with 100 × 10⁶ CART-ddBCMA, we have continued enrollment of the expansion cohort at DL1. If the response rate observed in this study continues in a larger cohort, this dose level will be used in a pivotal trial that is currently in development.

CART-ddBCMA kinetics, including median time to reach peak expansion (10 days) and median time to onset of response (28 days), were similar and consistent with kinetics of CAR T-cell therapies, including ide-cel and ciltacel. The ORR and CR observed with CART-ddBCMA was comparable to ORR observed with ide-cel and ciltacel. These results are promising...
when compared with ide-cel and cilta-cel given the higher rates of high-risk cytogenetics (90% vs 35% and 24%, respectively), extra-medullary involvement (58% vs 39% and 13%, respectively) and penta-refractory disease (83% vs 26% and 42%, respectively). After a median follow-up of 12.3 months, median duration of response, PFS, and OS have not been reached at either dose level. More importantly, CART-ddBCMA responses deepened over time, and 6 subjects (of 8 evaluable) remained relapse-free beyond 12-month evaluation, including 3 subjects remaining relapse-free beyond 20 months (Figure 2A), indicating the durability of the efficacy.

The persistence of CART-ddBCMA cells in the peripheral blood was also similar to most BCMA targeting CAR T-cell therapies, which noted a drop in peripheral CAR⁺ cells within 60 days and lack of detectable CAR⁺ cells in peripheral blood within 120 days in most subjects. We believe the drop in CART-ddBCMA levels is mainly due to lack of antigen stimulation following tumor elimination. Intriguingly, durability of efficacy was not found to correlate with presence of detectable CAR T cells in multiple myeloma in previous studies. In our study, although CART-ddBCMA cells were not detectable after 120 days, responses were durable for >12 months in 6/8 evaluable subjects. Furthermore, s-BCMA levels remained low in all subjects with ongoing response, and the recovery rate was slow, suggesting a slower recovery of BCMA-expressing plasma cells in the peripheral blood. Further studies and additional data are needed to verify if slower recovery of BCMA-expressing plasma cells is due to ongoing

Figure 2. Objective responses in patients treated with CART-ddBCMA. Responses were assessed according to the IMWG consensus criteria. MRD status is also indicated along with extent of MRD, presented as the number of multiple myeloma cells detected in the bone marrow per 1 × 10⁴, 1 × 10⁵, or 1 × 10⁶ total nucleated cells. An MRD of ≤1 × 10⁻⁴ is considered MRD-negative. (A) The best responses for each patient are shown, grouped by dose cohorts. (B) OR and sCR/CR rate over time.
immunosurveillance against BCMA expressing plasma cells by CART-ddBCMA cells.

This trial is the first to demonstrate the use of D-domain antigen-binding domain-based CAR T cells. D-domains have distinct advantages, such as triple α-helical bundle stabilized by a hydrophobic core with no disulfide bonds or N-linked glycosylation sites, and are easily manipulatable, allowing for removal of immunogenic epitopes and modulation of the target binding specificities. Therefore, the production of D-domain based CAR T cells is expected to provide consistent manufacturing with lower interpatient variability and decreased tonic signaling, which may improve the durability of BCMA CART responses. The current study provides the first evidence on clinical application of D-domains. The durable responses, consistent CAR⁺ expression rate per cell (median vector copy number of 2.33; range, 1.33-3.55), and low interpatient variability (median CAR⁺CD3⁺ cells in the product of 74%; range, 61% to 87%) noted in the current study are encouraging and support the development of binding domains for other targets.

This study is limited by a small sample size and is mainly designed to evaluate the initial safety of CART-ddBCMA administration. This limitation of the study should be considered during the interpretation of the findings on safety and efficacy. Further studies in a larger cohort are needed to confirm the safety and efficacy of CART-ddBCMA cells for treatment of RRMM.

In conclusion, we characterized the safety of CART-ddBCMA at doses of 100 and 300 × 10⁶ cells per patient. We further showed that CART-ddBCMA administration can induce deep and durable responses in patients with high-risk RRMM.

**Acknowledgments**

The authors thank Trisha R. Berger for drafting the manuscript, Anand Rotte for editorial assistance, Sujatha Kuruvakkat and Sigal Shachar for data analyses, Jerome Ritz for CART-ddBCMA support with cell manufacturing, and Scott Currence and Travis Quigley for clinical operations support.

**Authorship**

Contribution: M.J.F., M.R.B., J.R., E.K.O., N.R., D.C., A.J.Y., E.L., D.E.A., A.J., K.S., H.D., and S.N. contributed to the study design, collection of data, and analyses and interpretation of data; F.G. and C.C. contributed to study design and data analyses; and A.S., C.H., and M.V.M. contributed to the study design and interpretation of data.

Conflict-of-interest disclosure: M.J.F. reported consultancy fees from Celgene, consultancy and research funding from Novartis, consultancy fees from Arcellx, and consultancy fees and research funding from Gilead/Kite. M.R.B. reported honoraria, membership on an entity’s board of directors or advisory committees, research funding, and speakers bureau fees from Kite; honoraria and speakers bureau fees from Incyte; membership on an entity’s board of directors or advisory committees from Autolus; membership on an entity’s board of directors or advisory committees and research funding from Novartis; membership on an entity’s board of directors or advisory committees and research funding from CRSPPR Therapeutics; honoraria and speakers bureau fees from Bristol Myers Squibb. E.K.O. reported consultancy fees from Celgene. J.R. reported consultancy fees from...
Attivare Therapeutics; research funding from Bristol Myers Squibb; consultancy fees from Parexel; consultancy fees and patents and royalties from Wolters Kluwer Health; consultancy fees from Imaging Endpoints; membership on an entity’s board of directors or advisory committees from Karyopharm. N.R. reported consultancy fees from Celgene. A.J.Y. reported consultancy fees from Adaptive, Bristol Myers Squibb, GSK, Oncopetptides, Karyopharm, Amgen, Takeda, Sanofi, and Janssen. A.S. reported former employment at and current equity holder in private company Arcellx. D.E.A. reported membership on an entity’s board of directors or advisory committees and research funding from Celgene; research funding from Pharmacyclics; consultancy fees and research funding from Kite Pharma; membership on an entity’s board of directors or advisory committees from Juno, Partner Tx, Karyopharm, Bristol Myers Squibb, Avi MedTech Ltd., Takeda, Legend Biotech, and Chugai, consultancy fees from Janssen, Parexcel, Takeda, and Sanofi. A.J. reported membership on an entity’s board of directors or advisory committees from Bristol Myers Squibb, Celgene, AbbVie, Gracell, GSK, Janssen, Karyopharm, Amgen, and Sanofi. K.S. reported current equity holder in publicly traded company Orchard Therapeutics, Ltd. S.N. reported ad hoc advisory boards for Kite/Gilead, Novartis, lovance, and GlaxoSmithKline. F.G. reported current employment at and current equity holder in private company Arcellx. C.C. reported current employment at and current equity holder in private company Arcellx. M.V.M. reported consultancy fees and research funding from Arcellx, Kite, and Novartis. The remaining authors declare no competing financial interests.

ORCID profiles: M.J.F., 0000-0002-6774-5694; K.S., 0000-0002-1293-8050; M.V.M., 0000-0002-7578-0393.

Correspondence: Matthew J. Frigault, Massachusetts General Hospital, 55 Fruit St, Boston, MA 02114; email: mfrigault@partners.org.

References

1. Usmani S, Ahmadi T, Ng Y, et al. Analysis of real-world data on overall survival in multiple myeloma patients with ≥3 prior lines of therapy including a proteasome inhibitor (PI) and an immunomodulatory drug (IMiD), or double refractory to a PI and an IMiD. Oncologist. 2016;21(11):1355-1361.

2. Jagannath S, Lin Y, Goldschmidt H, et al. KarMMa-RW: comparison of idecabtagene vicleucel with real-world outcomes in relapsed and refractory multiple myeloma. Blood Cancer J. 2021;11(6):116.

3. Pick M, Vainstein V, Goldschmidt N, et al. Daratumumab resistance is frequent in advanced-stage multiple myeloma patients irrespective of CD38 expression and is related to dismal prognosis. Eur J Haematol. 2018;100(5):494-501.

4. Gandhi UH, Cornell RF, Lakshman A, et al. Outcomes of patients with multiple myeloma refractory to CD38-targeted monoclonal antibody therapy. Leukemia. 2019;33(9):2266-2275.

5. Yu B, Jiang T, Liu D. BCMA-targeted immunotherapy for multiple myeloma. J Hematol Oncol. 2020;13(1):125.

6. Tai YT, Anderson KC. B cell maturation antigen (BCMA)-based immunotherapy for multiple myeloma. Expert Opin Biol Ther. 2019;19(11):1143-1156.

7. Moreaux J, Legouffe E, Jourdan E, et al. BAFF and APRIL protect myeloma cells from apoptosis induced by interleukin 6 deprivation and dexamethasone. Blood. 2004;103(8):3148-3157.

8. Novak AJ, Darce JR, Arendt BK, et al. Expression of BCMA, TACI, and BAFF-R in multiple myeloma: a mechanism for growth and survival. Blood. 2004;103(2):689-694.

9. Tai YT, Acharya C, An G, et al. APRIL and BCMA promote human multiple myeloma growth and immunosuppression in the bone marrow microenvironment. Blood. 2016;127(25):3225-3236.

10. Godara A, Zhou P, Kugelmass A, et al. Presence of soluble and cell-surface B-cell maturation antigen in systemic light-chain amyloidosis and its modulation by gamma-secretase inhibition. Am J Hematol. 2020;95(5):E110-E113.

11. Trudel S, Lendvai N, Popat R, et al. Antibody-drug conjugate, GSK2857916, in relapsed/refractory multiple myeloma: an update on safety and efficacy from dose expansion phase I study. Blood Cancer J. 2019;9(4):37.

12. Topp MS, Duell J, Zugmaier G, et al. Evaluation of AMG 420, an anti-BCMA bispecific T-cell engager (BiTE) immunotherapy, in R/R multiple myeloma (MM) patients: updated results of a first-in-human (FIH) phase I dose escalation study. J Clin Oncol. 2019;37(suppl 15):8007.

13. Madduri D, Rosko A, Brayer J, et al. 291 REGN5458, a BCMA x CD3 bispecific monoclonal antibody, induces deep and durable responses in patients with relapsed/refractory multiple myeloma (RRMM). Paper presented at the 62nd American Society of Hematology Annual Virtual Meeting; 5-8 December 2020.

14. Bahlis NJ, Raje NS, Costello C, et al. Efficacy and safety of elranatamab (PF-06863135), a B-cell maturation antigen (BCMA)-CD3 bispecific antibody, in patients with relapsed or refractory multiple myeloma (MM). Paper presented at American Society of Clinical Oncology Annual Virtual Meeting; 5-8 December 2021.

15. Moreau P, Usmani SZ, Garfall AL, et al. 896 Updated results from MajesTEC-1: phase 1/2 study of teclistamab, a B-cell maturation antigen x CD3 bispecific antibody, in relapsed/refractory multiple myeloma. Paper presented at the 62nd American Society of Hematology Annual Virtual Meeting; 5-8 December 2020.

16. Schirmacher V. Cancer vaccines and oncolytic viruses exert profoundly lower side effects in cancer patients than other systemic therapies: a comparative analysis. Biomedicines. 2020;8(3):E61.
17. Golubovskaya V, Zhou H, Li F, et al. Novel CS1 CAR-T cells and bispecific CS1-BCMA CAR-T cells effectively target multiple myeloma. *Biomedicines.* 2021;9(10):142-122.

18. Firor AE, Jares A, Ma Y. From humble beginnings to success in the clinic: chimeric antigen receptor-modified T-cells and implications for immunotherapy. *Exp Biol Med (Maywood).* 2015;240(8):1087-1098.

19. Larson RC, Maus MV. Recent advances and discoveries in the mechanisms and functions of CAR T cells. *Nat Rev Cancer.* 2021;21(3):145-161.

20. Zmievskaya E, Valullina A, Ganeeva I, Petukhov A, Rizvanov A, Bulatov E. Application of CAR-T cell therapy beyond oncology: autoimmune diseases and viral infections. *Biomedicines.* 2021;9(1):59.

21. Styczynski J. A brief history of CAR-T cells: from laboratory to the bedside. *Acta Haematol Pol.* 2020;51(1):2-5.

22. Munshi NC, Anderson LD Jr, Shah N, et al. Idecabtagene vilocileucel in relapsed and refractory multiple myeloma. *N Engl J Med.* 2021;384(8):705-716.

23. Raje N, Berdeja J, Lin Y, et al. Anti-BCMA CAR T-cell therapy bb2121 in relapsed or refractory multiple myeloma. *N Engl J Med.* 2019;380(18):1726-1737.

24. Berdeja JG, Madduri D, Usmani SZ, et al. Ciltacabtagene autoleucel, a B-cell maturation antigen-directed chimeric antigen receptor T-cell therapy in patients with relapsed or refractory multiple myeloma (CAR TITUDE-1): a phase 1b/2 open-label study. *Lancet.* 2021;398(10297):314-324.

25. Zhao WH, Liu J, Wang BY, et al. A phase 1, open-label study of LCAR-B38M, a chimeric antigen receptor T cell therapy directed against B cell maturation antigen, in patients with relapsed or refractory multiple myeloma. *J Hematol Oncol.* 2018;11(1):141.

26. Xu J, Chen LJ, Yang SS, et al. Exploratory trial of a biepitopic CAR T-targeting B cell maturation antigen in relapsed/refractory multiple myeloma. *Proc Natl Acad Sci USA.* 2019;116(19):9543-9551.

27. Roex G, Timmers M, Wouters K, et al. Safety and clinical efficacy of BCMA CAR-T-cell therapy in multiple myeloma. *J Hematol Oncol.* 2020;13(1):164.

28. Qu X, An G, Sui W, et al. 1830 Updated phase 1 results of C-CAR088, an anti-BCMA CAR T-cell therapy in relapsed or refractory multiple myeloma. Presented at the 63rd American Society of Hematology Annual Meeting; 11-14 December 2021; Atlanta, GA.

29. de Larrea CF, Gonzalez-Calle V, Cabañas V, et al. 2837 Results from a pilot study of AR1002oh, an academic BCMA-directed CAR-T cell therapy with fractionated initial infusion and booster dose in patients with relapsed and/or refractory multiple myeloma. Presented at the 63rd American Society of Hematology Annual Meeting; 11-14 December 2021; Atlanta, GA.

30. Mailankody S, Liedtke M, Sidana S, et al. 651 Universal updated phase 1 data validates the feasibility of allogeneic Anti-BCMA ALLO-715 therapy for relapsed/refractory multiple myeloma. Presented at the 63rd American Society of Hematology Annual Meeting; 11-14 December 2021; Atlanta, GA.

31. Li C, Wang D, Song Y, et al. 547 A phase 1/2 study of a novel fully human B-cell maturation antigen-specific CAR T cells (CT103A) in patients with relapsed and/or refractory multiple myeloma. Presented at the 63rd American Society of Hematology Annual Meeting; 11-14 December 2021; Atlanta, GA.

32. Teoh PJ, Chng WJ. CAR T-cell therapy in multiple myeloma: more room for improvement. *Blood Cancer J.* 2021;11(4):84.

33. Da Viá MC, Dietrich O, Truger M, et al. Homozygous BCMA gene deletion in response to anti-BCMA CAR T cells in a patient with multiple myeloma. *Nat Med.* 2021;27(4):616-619.

34. Brudno JN, Maric I, Hartman SD, et al. T cells genetically modified to express an anti-B-cell maturation antigen chimeric antigen receptor cause remissions of poor-prognosis relapsed multiple myeloma. *J Clin Oncol.* 2018;36(22):2267-2280.

35. Ajina A, Maher J. Strategies to address chimeric antigen receptor tonic signaling. *Mol Cancer Ther.* 2018;17(9):1795-1815.

36. Frigault MJ, Lee J, Basil MC, et al. Identification of chimeric antigen receptors that mediate constitutive or inducible proliferation of T cells. *Cancer Immunol Res.* 2015;3(4):356-367.

37. Long AH, Haso WM, Shern JF, et al. 4-1BB costimulation ameliorates T cell exhaustion induced by tonic signaling of chimeric antigen receptors. *Nat Med.* 2015;21(6):581-590.

38. Wagner DL, Fritsche E, Pulsipher MA, et al. Immunogenicity of CAR T cells in cancer therapy. *Nat Rev Clin Oncol.* 2021;18(6):379-393.

39. Siegler E, Li S, Kim YJ, Wang P. Designed ankyrin repeat proteins as Her2 targeting domains in chimeric antigen receptor-engineered T cells. *Hum Gene Ther.* 2017;28(9):726-736.

40. Han X, Cinay GE, Zhao Y, Guo Y, Zhang X, Wang P. Adnectin-based design of chimeric antigen receptor for T cell engineering. *Mol Ther.* 2017;25(11):2466-2476.

41. Zajic CU, Dobersberger M, Schaffner I, et al. A conformation-specific ON-switch for controlling CAR T cells with an orally available drug. *Proc Natl Acad Sci USA.* 2020;117(26):14926-14935.

42. Salzer B, Schueller CM, Zajc CU, et al. Engineering AvidCARs for combinatorial antigen recognition and reversible control of CAR function. *Nat Commun.* 2020;11(1):4166.

43. Walsh ST, Cheng H, Bryson JW, Roder H, DeGrado WF. Solution structure and dynamics of a de novo designed three-helix bundle protein. *Proc Natl Acad Sci USA.* 1999;96(10):5486-5491.

44. Zhu Y, Alonso DO, Maki K, et al. Ultrafast folding of alpha3D: a de novo designed three-helix bundle protein. *Proc Natl Acad Sci USA.* 2003;100(26):15486-15491.

45. Qin H, Edwards JP, Zařítkayš L, et al. Chimeric antigen receptors incorporating D domains targeting CD123 direct potent mono- and bi-specific antitumor activity of T cells. *Mol Ther.* 2019;27(7):1262-1274.

46. Rotte A, Heery C, Gliner B, Tice D, Hilbert D. BCMA targeting CAR T cells using a novel D-domain binder for multiple myeloma: clinical development update. *Immuono-Oncology Insights.* 2022;3:13-24.
47. Buonato JM, Edwards J, Zaritskaya L, et al. Design and demonstration of potent in vitro and in vivo activity for CART ddBCMA, a BCMA targeted CAR T cell therapy incorporating a non scFv binding domain. Presented at the American Society of Gene & Cell Therapy Virtual Meeting; 11-14 May 2021.
48. Buonato JM, Edwards JP, Zaritskaya L, et al. Preclinical efficacy of BCMA directed CAR-T cells incorporating a novel D domain antigen recognition domain. *Mol Cancer Ther.* 2022;21(7):1171-1183.
49. Lee DW, Santomasso BD, Locke FL, et al. ASTCT consensus grading for cytokine release syndrome and neurologic toxicity associated with immune effector cells. *Biol Blood Marrow Transplant.* 2019;25(4):625-638.
50. Kumar S, Paiva B, Anderson KC, et al. International Myeloma Working Group consensus criteria for response and minimal residual disease assessment in multiple myeloma. *Lancet Oncol.* 2016;17(8):e328-e346.
51. Le Tourneau C, Lee JJ, Siu LL. Dose escalation methods in phase I cancer clinical trials. *J Natl Cancer Inst.* 2009;101(10):708-720.
52. Van Oekelen O, Aleman A, Upadhyaya B, et al. Neurocognitive and hypokinetic movement disorder with features of parkinsonism after BCMA-targeting CAR-T cell therapy. *Nat Med.* 2021;27(12):2099-2103.
53. Abecma. Package insert. Celgene; 2021.
54. Madduri D, Usmani SZ, Jagannath S, et al. Results from CARTITUDE-1: a phase 1b/2 study of JNJ-4528, a CAR-T cell therapy directed against B-cell maturation antigen (BCMA), in patients with relapsed and/or refractory multiple myeloma (R/R MM). *Blood.* 2019;134(suppl 1):577.
55. Ali SA, Shi V, Maric I, et al. T cells expressing an anti-B-cell maturation antigen chimeric antigen receptor cause remissions of multiple myeloma. *Blood.* 2016;128(13):1688-1700.
56. Cohen AD, Garfall AL, Stadtmauer EA, et al. B cell maturation antigen-specific CAR T cells are clinically active in multiple myeloma. *J Clin Invest.* 2019;129(6):2210-2221.
57. Frigault MJ, O'Donnell E, Raje NS, et al. Phase 1 study of CART-ddBCMA, a CAR-T therapy utilizing a novel synthetic binding domain, for the treatment of subjects with relapsed and refractory multiple myeloma. *J Clin Oncol.* 2021;39(suppl 15):8015.