Title

Proceedings from the Blood and Marrow Transplant Clinical Trials Network Myeloma Intergroup Workshop on Immune and Cellular Therapy in Multiple Myeloma.

Journal

Transplantation and Cellular Therapy, 28(8)

Authors

Holstein, Sarah
Asimakopoulos, Fotis
Azab, Abdel
et al.

Publication Date

2022-08-01

DOI

10.1016/j.jtct.2022.05.019

Peer reviewed

HHS Public Access

Author manuscript

Transplant Cell Ther. Author manuscript; available in PMC 2023 August 01.

Published in final edited form as:

Transplant Cell Ther. 2022 August ; 28(8): 446–454. doi:10.1016/j.jtct.2022.05.019.

Proceedings from the BMT CTN Myeloma Intergroup Workshop on Immune and Cellular Therapy in Multiple Myeloma

Sarah A. Holstein[1], **Fotis Asimakopoulos**[2], **Abdel Kareem Azab**[3], **Giada Bianchi**[4], **Manisha Bhutani**[5], **Leslie A. Crews**[2], **Tom Cupedo**[6], **Hannah Giles**[7], **Sarah Gooding**[8], **Jens Hillengass**[9], **Lukas John**[10], **Shari Kaiser**[11], **Lydia Lee**[12], **Kylee Maclachlan**[13], **Marcelo C. Pasquini**[14], **Flavia Pichiorri**[15,16], **Nina Shah**[17], **Monica Shokeen**[3], **Brian R. Shy**[17], **Eric L. Smith**[18], **Raluca Verona**[19], **Saad Z. Usmani**[13], **Philip L. McCarthy**[9]

[1]University of Nebraska Medical Center, Omaha, NE, USA

[2]Moores Cancer Center, University of California San Diego, La Jolla, CA, USA

[3]Washington University School of Medicine, St. Louis, MO, USA

[4]Brigham and Women's Hospital, Harvard Medical School, Boston, MA, USA

[5]Levine Cancer Institute, Charlotte, NC, USA

[6]ErasmusMC Cancer Institute Rotterdam, Rotterdam, The Netherlands

[7]University Hospitals Birmingham NHS Foundation Trust, Birmingham, UK

[8]MRC Molecular Hematology Unity, Weatherall Institute of Molecular Medicine, University of Oxford, Oxford, UK

[9]Roswell Park Comprehensive Cancer Center, Buffalo, NY, USA

[10]University Hospital Heidelberg, Heidelberg, Germany

[11]Bristol Myers Squibb, Seattle, WA, USA

[12]University College London, London, UK

[13]Memorial Sloan Kettering Cancer Center, New York, NY, USA

[14]Medical College of Wisconsin, Milwaukee, WI, USA

[15]Judy and Bernard Briskin Center for Multiple Myeloma Research, Department of Hematology and Hematopoietic Cell Transplantation, City of Hope, Duarte, CA, USA

[16]Department of Hematologic Malignancies Translational Science, Beckman Research Institute, City of Hope, Duarte, CA, USA

[17]University of California San Francisco, San Francisco, CA, USA

[*]Corresponding author: Sarah A. Holstein, MD, PhD, Division of Oncology and Hematology, Department of Internal Medicine, University of Nebraska Medical Center, Omaha, NE 68198, Academic Office Phone: 402-559-8500, Academic Office Fax: 402-559-6520, sarah.holstein@unmc.edu.

[18]Dana-Farber Cancer Institute, Boston, MA, USA

[19]Janssen Research & Development, Spring House, PA, USA

Abstract

The Blood and Marrow Transplant Clinical Trials Network (BMT CTN) Myeloma Intergroup conducted a workshop on Immune and Cellular Therapy in Multiple Myeloma on January 7, 2022. This workshop included presentations by basic, translational and clinical researchers with expertise in plasma cell dyscrasias. Four main topics were discussed: 1) platforms for myeloma disease evaluation; 2) insights into pathophysiology; 3) therapeutic target and resistance mechanisms; and 4) cellular therapy for multiple myeloma. In the present report, we provide a comprehensive summary of these workshop presentations.

Keywords

multiple myeloma; imaging; CAR T-cell; tumor microenvironment; resistance

Introduction:

The Blood and Marrow Transplant Clinical Trials Network (BMT CTN) Myeloma Intergroup has conducted an annual workshop focused on minimal residual disease (MRD) and immune profiling (IP) assessment in multiple myeloma (MM) since 2016.[1–5] To accommodate the evolution of scientific research in this field as well as the implementation of novel immune-based therapeutic strategies, the most recent workshop shifted focus towards immune and cellular therapy research. Due to the ongoing COVID-19 pandemic, the sixth annual workshop was held virtually on January 7, 2022.

The virtual workshop included 16 presentations as well as four live question and answer sessions (the full agenda is shown in the Supplemental Material). In this report we provide a comprehensive summary of the workshop, which focused on four main topics: platforms for MM disease evaluation, insights into pathogenesis, therapeutic target and resistance mechanisms, and cellular therapy for MM. This workshop included presentations by basic science, translational and clinical investigators with expertise in the field of plasma cell dyscrasias (PCDs) and encouraged discussion amongst other investigators and trainees interested in cutting edge research in MM. Key takeaways from each presentation are shown in Table 1.

Session 1: Platforms for Myeloma Disease Evaluation:

Previous workshops focused on minimal residual disease (MRD) assessment via more traditional techniques such as multiparametric flow cytometry and next generation sequencing evaluation of the bone marrow (BM).[1–5] However, there is increasing interest in alternative methodologies that are capable of assessing for the presence of clonal plasma cells and/or targeting those cells, including mass spectrometry (MS) and theranostics. In this session, research focused on developing novel platforms for MM disease evaluation was discussed.

Abdel Kareem Azab (Washington University) discussed his group's research focused on tumor microenvironment (TME)-targeted nanoparticles. There is substantial data in the literature demonstrating the role that the BM niche plays in MM pathophysiology. His group's prior work has demonstrated that the interaction between the tumor cells and other cells such as stromal cells and endothelial cells not only induce proliferation of the tumor cells but also induce drug resistance.[6–10] Disruption of these interactions can lead to enhanced sensitivity of the tumor cells to therapies. Both tumor-directed therapies and TME-directed therapies can be limited due to non-specific distribution and off-target toxicities. To overcome this problem and deliver the drugs selectively to the tumor, nanoparticles have been developed that combine these drugs and take advantage of the endothelial cell layer associated with MM cells. For example, they determined that the bone marrow endothelial cells isolated from patients with MM had overexpression of P-selectin relative to those obtained from healthy donors.[11] Similarly, using a mouse MM model, they demonstrated that higher levels of P-selectin were detected in bone marrow endothelial cells relative to healthy mice.[11] They engineered nanoparticles with PSGL-1 (the ligand of P-selectin) on the surface and demonstrated enhanced uptake of these nanoparticles in the tumor of these MM-bearing mice compared to non-targeted nanoparticles.[11] These nanoparticles were then loaded with the proteasome inhibitor bortezomib and the ROCK inhibitor Y-27362, with the latter being used to disrupt interactions between the MM cells and stromal/endothelial cells.[11] These targeted, combination drug-loaded nanoparticles significantly delayed tumor growth and survival relative to single agent-loaded nanoparticles or to free drugs (used alone or in combination).[11] Thus these studies form the basis for future studies evaluating the strategy of more selectively targeting anti-MM therapies to the BM microenvironment in order to minimize systemic toxicity, overcome potential drug resistance mechanisms and improve anti-tumor efficacy.

Hannah Giles (University Hospitals Birmingham NHS Foundation Trust) reported on MS testing of samples obtained from patients enrolled in the Myeloma XI trial. MS methodologies are emerging as a sensitive tool for the monitoring of monoclonal proteins in patients with PCDs. The International Myeloma Working Group (IMWG) has recently approved the use of intact light chain assays using the matrix-assisted laser desorption/ionization time-of-flight MS (MALDI-TOF MS) platform, such as the MASS-FIX assay, in lieu of immunofixation.[12] In the presented work, samples were tested using an intact light chain MALD-TOF MS assay using reagent supplied by the Binding Site who is in the process of developing a commercially available MS assay. This method involves the enrichment for immunoglobulins in serum samples using anti-sera specific for IgG, IgA, IgM, total kappa, total lambda, free kappa and free lambda. Samples were washed to remove non-immunoglobulin proteins, eluted and reduced for subsequent analysis by MALDI-TOF MS. This assay has been reported to have a lower limit of measuring interval of 15 mg/L for intact immunoglobulin monoclonal proteins as well as linearity ranging from 0.015–100 g/L.[13] MALDI-TOF MS was used to evaluate the serum of 22 patients with non-secretory MM and 25 patients with oligo-secretory MM enrolled in the Myeloma XI trial.[14] They found that 20/22 of the non-secretory patients had detectable monoclonal proteins using MALDI-TOF MS. MALDI-TOF successfully identified monoclonal protein in all 25 of the patients originally classified as having oligo-secretory disease, with concordant findings

between the MS approach and immunofixation electrophoresis. They also evaluated all available follow-up samples of these non-secretory/oligo-secretory patients (post-cycle 1, post-induction, day +42 post-ASCT and day +100 post-ASCT) and found that MALDI-TOF MS detected residual monoclonal proteins in a greater proportion of patients than standard assays (immunofixation electrophoresis and serum free light chain assessments). Examples were presented demonstrating that liquid-chromatography MS (LC-MS) could detect very small monoclonal proteins that were undetectable by MALDI-TOF MS. However, it was noted that LCMS had much lower throughput. Overall, it was suggested that MALDI-TOF MS may be particularly useful for monitoring patients traditionally classified as having non-secretory or oligo-secretory disease. Dr. Giles concluded with the recommendation that bone marrow biopsies to confirm complete response and MRD status be performed once MALDI-TOF MS negativity is achieved in these patients.

Flavia Pichiorri (City of Hope) discussed the development of daratumumab radio-labeled antibodies. Previously published work using ^{64}Cu-DOTA-daratumumab demonstrated superior detection of MM cells *in vivo* compared to traditional ^{18}F-FDG-PET/CT.[15] These results were translated into the clinic in the form of a phase I trial in which 12 daratumumab-naïve patients were imaged following injection of varying doses of ^{64}Cu-DOTA-daratumumab.[16] While ^{18}F-FDG was mainly taken up in the brain, the ^{64}Cu-DOTA-daratumumab had much stronger uptake in the spine and other sites of bony disease. As CD38 is expressed on non-MM cells, additional work was done in order to decrease the background by increasing the concentrations of unlabeled daratumumab co-administered with the labeled antibody. When the ^{64}Cu-DOTA-daratumumab was administered in the absence of unlabeled daratumumab, there was strong uptake in the liver and spleen. With increasing doses of unlabeled daratumumab, the uptake in the liver and spleen diminished while retaining uptake in the spine and pelvis. With higher doses of unlabeled daratumumab, the uptake of the ^{64}Cu-DOTA-daratumumab was lost.[16] These studies established a dose of 45 mg of unlabeled daratumumab as the optimal dose allowing for increased MM targeting by the labeled antibody.[16] This research group is also exploring therapeutic radiolabeled antibodies using beta- or alpha-emitter radiolabeled DOTA-daratumumab. Mouse MM studies with ^{225}Ac-DOTA-daratumumab showed dose-dependent efficacy and less whole-body toxicity compared with targeted β-therapy (^{177}Lu-DOTA-daratumumab).[17]

Monica Shokeen (Washington University School of Medicine in Saint Louis) discussed imaging in MM. She noted that advanced whole-body imaging provides timely and accurate diagnosis and staging as well as assessment of treatment response and spatial assessment of disease. Imaging modalities may be classified as anatomical (e.g., radiographic skeletal surveys or CT), functional (MRI) or molecular (PET).[18, 19] Each modality is associated with pros and cons. For anatomical modalities, the advantages include being widely available while the disadvantages include limited functional information and limited sensitivity. For the functional modality, an advantage is that MRI may be considered to be a gold standard for BM involvement (high spatial resolution), however, a disadvantage is that assessment of response in the BM can be challenging. For the molecular modality, advantages include sensitivity as well as accurate molecular/cellular level detection while disadvantages include rapid handling of short-lived isotopes. Currently 2-[^{18}F]-fluoro-2-deoxy-D-glucose (FDG) is the most commonly used PET tracer for MM. However, FDG has certain limitations in this

disease including that background marrow uptake may obscure focal lesions and that there may be low hexokinase-2 enzyme expression and inconsistent Glut-1 transporter expression. Thus, there is a need for the development of new PET tracers that are specific for diffuse disease involvement in the BM and are taken up by non-FDG avid lesions. Immuno-PET imaging combines the specificity of targeted antibodies with the sensitivity of PET and may be a promising strategy for stratifying patients for immune- and cell-based therapies. One such example is [^{89}Zr]-daratumumab which has been studied in both pre-clinical and clinical settings.[20, 21] These studies demonstrated the ability of the immuno-PET to detect both medullary and extramedullary sites of disease, including labeling areas that were not identified by traditional FDG-PET imaging. ^{89}Zr-labeled elotuzumab is also being studied in preclinical MM models, where the ^{89}Zr-elotuzumab outperformed FDG in detecting sites of MM disease.[22] In addition to antibody-based tracers, small molecule-based tracers are also being studied. One example is the PET radiotracer ^{64}Cu-LLP2A which detects very late antigen-4 (VLA4) (overexpressed in MM cells) and was evaluated in a preclinical MM model.[23, 24] First-in-human studies with this agent are ongoing (NCT03804424), but thus far no adverse effects have been reported. Overall, imaging is an important tool as it has the potential to lead to precision therapies (theranostics), provide information regarding MRD and improve understanding of TME and clonal heterogeneity.

Session 2: Insights into Pathogenesis:

This session focused on recent research exploring the underlying genetic and TME changes that are associated with PCD pathogenesis. Fotis Asimakopoulos (University California San Diego) discussed MM matrix remodeling and immune microenvironment. The composition of tumor matrix that physically hosts and envelops tumor cells in their cellular immune microenvironment is an important regulator of their interactions and holds prognostic and predictive value.[25] His lab has been focused on a tumor matrix proteoglycan called versican (VCAN). VCAN is a complex macromolecule composed of a protein core and branching sugar chains that collectively take the shape of a bottle brush.[26] His group has proposed that VCAN is an instigator of sterile inflammation in the MM microenvironment. VCAN, acting as endogenous damage associated molecular patterns, activates toll-like receptors on myeloid cells leading to the release of inflammatory mediators that promote inflammatory stromal cells.[27] He discussed the evidence supporting an immunosuppressive role of VCAN within the TME. Acting through toll like receptor 2, versican renders antigen presenting cells dysfunctional or tolerogenic.[28] Myeloid derived versican correlates with effector dysfunction in the MM microenvironment.[29] In solid tumors, VCAN is associated with immunoregulatory Th2 and Treg responses as well as loss of cytotoxic effector functions.[30] In addition, high VCAN levels correlate with resistance to checkpoint inhibitors.[31] The Asimakopoulos group has reported that proteolysis of VCAN generates a bioactive N-terminal fragment called versikine that has opposing pro-immunogenic roles.[32] Versikine promotes the activation and survival of tumor antigen presenting dendritic cells, particularly the cDC1 (Batf3-dependent) dendritic cell lineage.[33] These cells internalize dead tumor cells and cross-present tumor antigens to prime anti-tumor effector T cells. VCAN proteolysis to versikine is observed both focally and diffusely in the MM BM, while in solid tumors it is predominantly localized within the peri-tumoral stroma.[33, 34] This location is significant as VCAN proteolysis appears to reverse immune exclusion, promoting

entry of CD8-positive T-cells within the tumor mass. The stoichiometry between VCAN and versikine is context specific. In contrast to solid tumors, in MM post-autologous stem cell transplant (ASCT), VCAN is always in excess and high VCAN proteolysis is associated with inferior progression free survival (PFS) and overall survival (OS).[34] In non-ASCT settings, the stoichiometry may differ and this is currently being investigated. Overall, there is evidence that VCAN and its matrikines fine-tune the immune microenvironments in both MM and solid tumors.

Kylee Maclachlan (Memorial Sloan Kettering Cancer Center (MSKCC)) discussed chromothripsis in MM and its precursor disease. Chromothripsis is a complex structural variant in which a single catastrophic event results in chromosomal shattering and random rejoining. This can be observed in clustering of chromosomal breakpoints and oscillation between two or more copy number states. The prevalence of chromothripsis is highly variable across malignancies. In general, the prevalence is lower in hematological malignancies when compared with solid organ cancers, but in MM the prevalence is estimated at 24%, which is the highest of all hematological malignancies.[35–37] Chromothripsis is associated with inferior PFS and OS in newly diagnosed MM.[36] The MSKCC research group has demonstrated that chromothripsis can be reliably predicted using copy number signatures in whole genome sequencing (WGS) data (AUC 0.9).[38] The copy number signatures can also be used to predict chromothripsis using whole exome sequencing data (AUC 0.82).[38] Multivariate analyses for PFS/OS revealed that the best factors for prediction were copy number signature, international staging system (ISS) stage, age and APOBEC mutational activity.[38] This group has also reported that chromothripsis is associated with progression in MM precursors. WGS of 32 patients with precursors disease showed significant differences in copy number aberration, APOBEC mutation activity and chromothripsis in patients whose disease was stable over time compared to those whose disease ultimately progressed to MM.[39] The significance of this finding is that current prognostic scores, which primarily assess bulk of disease, may be improved by the inclusion of genomic information defining underlying disease biology.

Tom Cupedo (ErasmusMC Cancer Institute Rotterdam) discussed the immune and stromal microenvironment in MM. The development of MM from precursor states involves not only the accumulation of mutations within the tumor cells but also remodeling of the TME, particularly BM stromal cells (BMSC). While there has been significant interest in targeting these stromal cells, the identity of the MM-supportive stromal cells has been uncertain. To address this gap in knowledge, a workflow was developed to isolate and purify non-hematopoietic cells from cryopreserved BM aspirates and then interrogate them via single cell RNA (scRNA) sequencing using 10X Genomics.[40] This analysis was performed on patient samples from 13 newly diagnosed MM patients as well as 7 control samples from individuals without hematological disease.[40] Combining the data from all samples, uniform manifold approximation and projection (UMAP) presentation demonstrated 5 clusters of mesenchymal stromal cells, a cluster of endothelial cells, a small cluster of osteo-lineage cells as well as megakaryocytes. When the samples were then segregated by MM vs control, it was revealed that there are two clusters which are unique to the MM samples. These clusters are characterized by transcription of MM survival genes, including *IL6* and *LIF*, as well as by an inflammatory transcriptome with enrichment for *CXCL2, CXCL3, CXCL5,*

CXCL8, CLL2 and *PTGS2*. Thus, these inflammatory mesenchymal stromal cells (iMSCs) suggest the presence of BM inflammation, which is supported by data showing elevated levels of IL-6, CXCL8 and CCL2 protein levels in BM plasma from MM patients compared with control.[40] These data suggested that these iMSC could be MM niche cells, which is supported by co-localization studies showing that CD44+ iMSC co-localize with MM cells *in situ*. Similar studies were performed on 9 paired diagnosis and post-induction therapy BM samples from patients on the CASSIOPEIA trial. This analysis revealed that induction therapy (with bortezomib-thalidomide-dexamethasone +/− daratumumab) failed to normalize the BM inflammation signals.[40] In summary, these data reveal the presence of activated stromal cells that form a MM-supportive BM niche and that the presence of iMSC is associated with BM inflammation. However, induction therapy did not normalize BM inflammation or iMSC presence, raising the hypothesis that iMSC may contribute to disease relapse/progression.

Lukas John (University of Heidelberg) discussed spatial heterogeneity in the MM microenvironment. It was noted that modern imaging techniques have demonstrated that MM is distributed heterogeneously throughout the skeleton. It has been hypothesized that MM cells within focal lesions might not only differ from more diffuse involvement but also may contain the origins of progressive disease. Prior research has demonstrated that these focal lesions harbor unique genomic clones and that the presence of 3 or more large focal lesions is associated with inferior PFS.[41–44] This work led to the hypothesis that the critical steps toward disease aggressiveness and treatment failure are initiated in restricted areas in the skeletal system. To address this hypothesis, studies were performed to delineate the spatial subclonal architecture and examine the cellular composition and transcriptional state within focal lesions. Paired aspirates from CT-guided biopsies of focal lesions as well as from a diagnostic random iliac crest site were compared using WGS (n=16), scRNAseq (n=7) and T-cell receptor sequencing (TCRseq) (n=7). Both site-specific and clone-specific genetic and transcriptional heterogeneity was demonstrated in the majority of the patients. scRNAseq was performed on CD138-negative cells from 6 patients with genetic heterogeneity to interrogate the immune microenvironment. A decrease in macrophages/macrophage progenitor cells was observed in the focal lesion samples relative to the iliac crest BM aspirate. A trend towards increased Tregs and CD8 effector cells in the focal lesions was also observed. It is hypothesized that genetic heterogeneity of the MM cells could lead to different neoantigens in different sites, which would lead to different T-cell clones and functional states. To characterize the T-cell compartment, TCR sequencing was performed at the single cell level. These studies failed to demonstrate site-specific expanded T-cell clones but did reveal differences in proportions of the T-cell clones across sites. In aggregate, these studies have shown that focal lesions are a hotspot for genetic and transcriptional heterogeneity and revealed differences in the TME, including different prevalence of expanded T-cell clones.

Session 3: Therapeutic Target and Resistance Mechanisms:

This session focused on novel therapeutic targets as well as recent research exploring resistance mechanisms underlying current and investigational MM therapies. Sarah Gooding (University of Oxford) discussed mechanisms of resistance to immunomodulatory drugs

(IMiDs). It is recognized that there are multiple causes of failure of IMiD-based therapies, including side effects and tolerability issues, tumor-mediated mechanisms of resistance allowing selection of resistant clones and immune-mediated mechanisms such as loss of T and NK cell stimulatory effects. Her group has investigated the mechanisms by which subclonal genetic events may confer drug resistance by identifying genetic changes which drive clonal selection in patients receiving IMiD-based therapy. All IMiDs and cereblon E3 ligase modulators (CELMoDs) bind cereblon (CRBN), an E3 ligase substrate recognition adaptor protein. IMiDs/CELMoDs change the substrate protein preference of the E3 ligase complex such that essential transcription factors (e.g., IKZF1) are degraded leading to cell cycle arrest and MM cell death. The CRBN complex requires neddylation for E3 ligase activity. In order to maintain a homeostatic level of ubiquitination, deneddylation (mediated by the COP9 signalosome) constantly occurs. The deneddylated E3 ligase complex dissociates and then requires complex reassembly and neddylation to return to an active state. Large WGS datasets including samples from diagnosis, lenalidomide-resistant and pomalidomide-resistant time points, were evaluated to identify genetic aberrations in this pathway that were selected during IMiD resistance acquisition.[45] Mutations, copy loss, structural variant and exon 10 splicing abnormalities in CRBN were determined. The incidence of all of these mechanisms of functional CRBN loss increased significantly as the MM became refractory to IMiDs, from 7.5% at new diagnosis to 20.7% at lenalidomide-resistance and 29.6% at pomalidomide-resistance.[45] They also demonstrated that the presence of any *CRBN* aberration was associated with 50% reduction in PFS in those treated with pomalidomide after lenalidomide refractoriness. Of these aberrations, heterozygous copy loss of the *CRBN* locus on chromosome 3p was most common. *In vitro* CRISPR screens have identified other genes whose loss is associated with IMiD resistance, including those involved in the COP9 signalosome and there is early evidence that losses or mutations of these genes may also have relevance in clinical IMiD resistance.[45, 46] In aggregate these studies demonstrate that regional copy loss of *CRBN* at 3p and COPS7b/8 at 2q37 may drive IMiD therapy-specific clonal advantage in MM and that gene copy loss may play a wider role than gene mutation in the selection of IMiD-resistant clones. One key question that remains unanswered is how CELMoDs perform in IMiD-refractory disease characterized by CRBN loss-associated mechanisms.

Leslie Crews (UCSD) discussed her group’s work targeting interferon regulatory factor-4 (IRF4) in MM. IRF4 is a key plasma cell differentiation factor and driver of MM cell survival.[46] High levels of IRF4 correlate with advanced disease stage and inferior OS outcomes.[47–49] In addition, IRF4 collaborates with the cancer oncogene and stem cell reprogramming gene MYC, driving its expression in a positive feedback loop, enhancing MM cell survival and disease progression.[50] Therefore, they hypothesized that IRF4 might function as a central reprogramming gene in malignant plasma cells and that inhibition might help prevent disease relapse and drug resistance. In whole transcriptome sequencing studies, they found that IRF4 mRNA levels were higher in high-risk MM and plasma cell leukemia samples compared to newly diagnosed cases.[51] They characterized IRF4 protein expression levels in primary samples and patient-derived xenograft (PDX) models. They developed an intranuclear flow cytometry assay to enable quantification of human IRF4 in the tumors isolated from engrafted mice using a panel of MM cell surface antibodies

and an IRF4 antibody.[52] This assay enabled them to confirm the high levels of IRF4 in the primary MM samples as well as demonstrate sustained IRF4 expression after *in vivo* transplantation. IRF4 has not been considered a druggable target, but the use of antisense oligonucleotide (ASO) strategies has allowed for selective targeting. Ongoing advances in ASO chemistry have led to improved stability and bioavailability properties. In collaboration with Ionis Pharmaceuticals, they have evaluated ASO-mediated inhibition of IRF4. As IRF4 expression varies amongst human MM cell lines, they tested 9 genetically diverse cell lines and evaluated effects on cell viability and IRF4 expression.[51] In addition to observing a significant decrease in IRF4 expression, they were also able to demonstrate a significant decrease in MYC expression following treatment with the IRF4 ASO.[51] Using an aggressive MM PDX model[53], they evaluated the impact of daily ASO treatment on tumor burden and demonstrated significant reduction in tumor burden and IRF4 expression.[51] Using NanoString analysis, they found that IRF4 target genes such as CXCR4 as well as microenvironment associated genes were downregulated in the BM of the PDX mice treated with the ASO.[51] They have also found that knockdown of IRF4 via ASO treatment induces cell cycle arrest and potentiates the cytotoxic effects of lenalidomide and bortezomib.[51] Their results led to the identification of a lead therapeutic candidate (ION251) that has now advanced to a phase I study testing the selective inhibition of human IRF4 in patients with relapsed/refractory MM (NCT04398485).

Raluca Verona (Janssen) discussed potential mechanisms associated with bispecific antibody (BiAb) therapies. Two of these agents currently under development are teclistamab and talquetamab that dually bind to myeloma targets (BCMA and GPRC5D, respectively) and CD3 receptors on T-cells. This dual binding to both T-cells and MM cells leads to T-cell recruitment, activation and killing of the tumor cells. Thus far the ongoing trials have demonstrated durable responses that deepen over time in patients with refractory MM.[54, 55] Studies evaluating determinants of response have involved *ex vivo* assessment of BM aspirate samples incubated with the drugs. High effector:target (E:T) ratios and high T-cell frequencies were associated with enhanced *ex vivo* killing of MM cells by teclistamab.[56] Similarly, with talquetamab, improved killing of MM cells *ex vivo* was associated with higher E:T ratios, higher GPRC5D expression, lower Tregs and lower frequencies of T-cells expressing PD-1 and HLA-DR.[57] One potential mechanism of resistance for T-cell BiAbs may be loss of antigen expression. For example, a recent publication reported BCMA loss due to a biallelic BCMA deletion following exposure to AMG-420, another BCMA-directed bispecific agent.[58] These authors reported the presence of heterozygous deletions of common immunotherapy target genes such as CD38, GPRC5D in both newly diagnosed and relapsed/refractory MM, but that frequencies were higher in the relapsed/refractory setting.[58] While the heterozygous deletion would not impact the overall level of expression, it would predispose to complete loss of antigen in the setting of therapeutic selection leading to the loss of the other allele. There may also be T-cell intrinsic mechanisms of resistance, including low CD4/CD8 ratio, reduced frequencies of stem cell memory and central memory, increased T-cell exhaustion/senescence profile, Treg expansion and changes in TCR clonality/repertoire. These data are consistent with a model where the BiAbs are engaging with existing T-cells as opposed to recruiting new T-cells and therefore the profile of the existing T-cells may be a critical determinant of response and

resistance.[59, 60] In summary, there are likely a myriad of intrinsic and acquired resistance mechanisms to CD3 BiAbs, including both tumor-dependent (e.g., tumor burden, genetic mutation/loss, antigen expression, survival/apoptosis pathways, IFN-γ signaling) and T-cell/TME-dependent mechanisms (e.g., immune fitness, T-cell exhaustion/senescence, T-cell memory subsets, TME-mediated suppression).[58, 60–66] Understanding these mechanisms will be critical for the successful development of next-generation approaches.

Giada Bianchi (Brigham and Women's Hospital) discussed her group's work focusing on targeting histone deacetylase (HDAC)-3 to modulate the bone marrow microenvironment (BME). Previous studies have demonstrated that silencing of HDAC3 using shRNA or using an HDAC3 inhibitor impairs MM proliferation and prevents tumor growth in a xenograft model.[67] Her group was interested in determining whether targeting HDAC3 in the BM niche could impact MM growth. HDAC3 is highly expressed in BMSC in MM but loss of HDAC3 (either via knockout or pharmacological inhibition) did not alter their survival.[68] However knockout or knockdown of HDAC3 in the BMSC did limit MM cell proliferation, both *in vitro* and *in vivo*.[68] The mechanism underlying these effects was determined to be contact independent, as conditioned supernatant from the HDAC3 knockout BMSC also impaired MM cell growth. They identified increased levels sgp130 levels in the supernatant. Sgp130 functions as a decoy receptor, trapping the IL-6/IL6R complex, and they showed that this resulted in inhibition of the IL-6 trans-signaling pathway.[68] They also identified both quantitative and qualitative changes in exosomes. MicroRNA (miRNA) sequencing and MS analysis showed significant changes in miRNA and protein content of exosomes derived from the HDAC3 knockout BMSC co-cultured with MM cells. Thus, HDAC3 appears to be a promising target to block the BM niche-MM interaction and limit MM cell survival.

Session 4: Cellular Therapy for Multiple Myeloma:

As of early 2022, there are two commercially available anti-BCMA CAR T-cell products for patients with relapsed/refractory MM. While these therapies are associated with deep responses in heavily refractory patient populations[69, 70], subsequent relapse appears inevitable and there is substantial ongoing research dedicated towards understanding mechanisms of resistance and identifying alternative cellular therapy strategies.

Shari Kaiser (Bristol Myers Squibb) discussed correlative data from the KarMMa study which evaluated idecabtagene vicleucel (Ide-cel). It was noted that Ide-cel expansion was a strong correlate for PFS. Peak CAR T-cell expansion was higher in responders than non-responders ($p<0.001$), demonstrating the need for adequate exposure to achieve a durable response.[71] While both CD4+ and CD8+ T-cells were expanded, there was a bias towards a higher proportion of CD8+ T-cells at T_{max} in responders vs non-responders. In addition, effector memory CAR T populations for both CD4+ and CD8+ CAR T subsets were predominant at T_{max} ($p<0.001$).[71] Their studies demonstrated that early soluble BCMA (sBCMA) clearance at month 2 post-infusion is associated with durable responses.[69] In addition, early MRD negativity (at 10^{-5}) at month 3 was found to be critical for durability; if MRD-positive at month 3, then achievement of subsequent complete response (CR) was unlikely. Low sBCMA levels (<20 ng/mL) may be an indirect measure of ongoing CAR T functional persistence, and duration of CAR T function correlates with duration of

response.[69] A retrospective analysis of BCMA expression on CD138+ cells was performed and it was determined that all enrolled subjects had BCMA expression at baseline and that the percentage of BCMA-positive cells (as determined by immunohistochemistry) did not different between responders and non-responders.[72] However, patients experiencing early progression (within 6 months of Ide-cel infusion) tended to have lower BCMA expression at time of progression, thus raising the hypothesis that preferential killing of high-expressing BCMA MM cells occurs first, leaving cells with lower expression that evade the CAR T-cells. Their studies have found that complete antigen loss was rare in relapsing subjects (<5% of evaluable patients).[72]

Brian Shy (University California San Francisco) discussed non-viral knockin strategies for BCMA CAR T-cell manufacturing. The approach that is used to generate knockin in primary human T-cells involves T-cell isolation and activation with beads and cytokines. Two days later, the cells are electroporated with Cas9 ribonucleoproteins. Cas9 is an endonuclease and this is formulated with a guide RNA (gRNA) that targets it to a specific site in the genome to make a double-stranded break. Also included is the homology directed repair (HDR) template which includes the transgene to be introduced (e.g., BCMA CAR).[73–75] The T-cells are then expanded. This non-viral approach is advantageous, as in contrast to lentivirus, the system allows the targeting of a specific site in the genome which could potentially have safety benefits as well as functional benefits. Previous work demonstrated that for a CD19 CAR, the CAR T-cells that had knockin to the TRAC locus (the endogenous TCR gene) had superior function compared to randomly integrated CAR constructs.[76] In those studies, adeno-associated virus was used as the virus to deliver the HDR template, and while it is very efficient, it has a complicated manufacturing process which can be problematic from a clinical grade material perspective.[76, 77] Thus, a completely non-viral approach was pursued that used only naked DNA. One of the challenges with DNA is that it is toxic to cells, particularly long double-stranded DNAs at high concentrations. However, single stranded DNA (ssDNA) is less toxic to cells but is harder to get into cells at high enough concentrations to achieve efficient knockin. They developed a modification of long ssDNA templates with Cas9 target sites added to the ends that allows for the co-electroporated Cas9 RNPs to bind directly to the template and help deliver it to the cell nucleus. Using these ssDNA templates, they observed higher knockin efficiencies, less toxicity and higher yields relative to dsDNA templates. They have adapted this approach to manufacture cells using a GMP-compatible process at clinical scale. Thus far, they have obtained CAR T-cell counts that are well above the anticipated dose levels required for future clinical trials.

Eric Smith (DFCI) presented his group’s research focused on GPRC5D CAR T-cells and dual targeting CARs. He noted that BCMA escape occurs at the DNA and non-genetic level after BCMA CAR T therapy.[78–80] In particular, BCMA-low cells may serve as a reservoir for BCMA+ relapse. They identified GPRC5D as a potential target for immunotherapy for MM and demonstrated that GPRC5D is expressed independently of BCMA.[81] Preclinical studies showed that GPRC5D-targeted CAR T-cells rescued mice from BCMA-negative tumor escape.[81] This work led to two clinical trials involving GPRC5D targeted T-cell products (MCARH109 (NCT04555551) and CC-95266 (NCT04674813)). The phase I trial of the MCARH109 product has thus far reported on the outcomes of 16 patients treated with 25–450 × 10^6 CAR T-cells.[82] On-target toxicities such as nail changes, rash and dysgeusia

were observed, but all grade 1.[82] The overall response rate (ORR) was 69% with 50% of patients achieving MRD-negativity.[82] Of note, responses were observed in patients with prior BCMA therapy (ORR of 80%) as well as patients who previously had received BCMA CAR T therapy (ORR of 75%). Another potential strategy involves the use of dual-targeted CAR T-cells.[83] This may be achieved by co-administration of two mono-specific CAR T-cell populations, by mixing viruses to result in co-expression of two vectors, by creating a vector that expresses two CARs (bicistronic vector), or by creating a single stalk CAR (tandem CAR).[83] Several of these strategies have been investigated by the Smith group with respect to simultaneously targeting BCMA and GPRC5D and appear promising.[84]

Lydia Lee (University College London) discussed work exploring APRIL CAR T therapy. It was noted that APRIL binds transmembrane activator and cyclophilin ligand interactor (TACI), BCMA and proteoglycan, but a truncated form of APRIL binds only TACI and BCMA. The hypothesis was that an APRIL CAR would increase targetable tumor antigen and reduce the risk of antigen-negative tumor escape.[85] A phase I trial with APRIL CAR was conducted (the AUTO2 trial). The results of 11 enrolled patients were previously reported and included an ORR of 43% and a median PFS of 5 months.[85] CAR T persistence was brief—by 40 days post-infusion, cells were no longer detectable in the majority of patients. Phenotype analysis of the CD8+ CAR T-cells showed a primarily exhausted phenotype. Subsequent studies evaluating the *in vitro* characteristics of the APRIL CAR with bb2121 or LCAR-B38M CARs showed comparable activity, including IFNγ production and expansion *in vitro*. However, it was also noted that there was activity of the APRIL CARs in absence of BCMA, suggesting basal activation. As APRIL naturally trimerizes, they then conducted studies to look for evidence of oligomerization of the CAR construct on the CAR T-cell surface. 293T cells were transduced with the APRIL CAR and then reduced and nonreduced cell lysates were analyzed by western blot analysis for CD3ζ. In nonreduced lysate, while APRIL CAR was detected in the monomeric form, the majority was present in the oligomeric form. They postulated that the natural interaction of APRIL may contribute to the phenotype of basal T cell activation. They observed failure to control tumor by the APRIL CAR T-cells, but not the bb2121 or LCAR-B38M CAR T-cells, in *in vitro* studies utilizing restimulation with MM.1S cells. They then assessed APRIL CARs in various different constructs and found similar failure to control tumor with prolonged stimulation, suggesting that the binding of tumor with APRIL CAR is suboptimal. Thus, the physiological requirements of natural ligand interaction may differ from those optimal for CAR activation.

Future directions:

The ongoing work highlighted in this workshop exploring the MM TME will shed light not only on the pathogenesis of MM during the evolution from a normal plasma cell/BM environment to the malignant state, but also elucidate therapeutic resistance mechanisms. In turn, this work will enable novel therapeutic strategies meant to bypass or overcome these resistance mechanisms, leading to more effective eradication of the disease. Understanding the TME and MM pathophysiology also has implications from an imaging and theranostics perspective, which in turn will enable alternative strategies to detect MRD and target the disease. Consensus remains lacking as to the optimal approach to interrogate the immune

microenvironment in this disease, but clearly these studies are critical for the identification of biomarkers for response to novel immune- and cell-based therapies such as BiAbs and CAR T-cells. As we have previously advocated, consistent incorporation of immune profiling studies in MM therapeutic intervention trials could prove cost-effective in the long-run, should phenotypes be identified that predict for sensitivity (or resistance) to anti-MM therapies.[5] Finally, the rapid expansion of immune and cell-based therapies coupled with the ever-increasing sophistication of the genomic analyses of MM cells, holds the promise of eventual personalized medicine approaches for this disease as well as complete eradication of the malignant clone.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgements:

Support for the Blood and Marrow Transplant Clinical Trials Network was provided by grant #U10HL069294 and #U24HL138660 from the National Heart, Lung, and Blood Institute (NHLBI) and the National Cancer Institute (NCI). Support for the FA laboratory was provided by R01 CA252937 from the NCI. Support for the MS laboratory was provided by R01 CA248493 from the NCI. The content is solely the responsibility of the authors and do not reflect the views or the official policy or position of the National Institutes of Health.

Declaration of Competing Interest

SAH: has served as a consultant for Bristol Myers Squibb/Celgene, Genentech, GlaxoSmithKline, Janssen, Oncopeptides, Sanofi, SecuraBio, Takeda; has received research funding from Oncopeptides.

FA: is listed as an inventor on US patent US20170258898A1: “Versikine for inducing or potentiating an immune response”

AKA: has a pending US patent for the TME-targeting nanoparticles

GB: has served as a consultant for Karyopharm

MB: has received institutional research funding from Celgene/Bristol Myers Squibb, Cerecor, Cellularity, Janssen, MedImmune, Millennium Pharmaceuticals, C4 Therapeutics

LC: has received laboratory service contract funding from Ionis Pharmaceuticals

TC: has nothing to disclose

HG: has received sponsorship for her PhD tuition fees from the Binding Site Ltd.

SG: has received research funding from Bristol Myers Squibb

JH: has received honoraria from Janssen; has served on advisory boards for Adaptive Biotechnologies, Amgen, AXXESS Network, Bristol Myers Squibb, Celgene, GlaxoSmithKline, Janssen, Oncopeptides, Oncotracker, Sanofi and Skyline; is serving on an independent data safety monitoring board for Janssen.

LJ: has nothing to disclose

SK: is an employee of Bristol Myers Squibb and holds stock in the company

LL: has nothing to disclose

KM: has nothing to disclose

MCP: has served as a consultant for Bristol Myers Squibb and Amgen; has received research support from Kite, Novartis, Janssen, GSK, Crispr and Bristol Myers Squibb

FP: has nothing to disclose

NS: has received research funding from Celgene/Bristol Myers Squibb, Janssen, Bluebird Bio, Sutro Biopharma, Teneobio, Poseida, Nektar, Precision Biosciences; has served as an advisor for GlaxoSmithKline, Amgen, Indapta Therapeutics, Sanofi, CareDx, Kite, Karyopharm, Oncopeptides and CSL Behring.

MS: is co-founder and owner of Sarya, LLC.

BRS: is a holder of patents pertaining to this work

ELS: has served on scientific advisory boards for Bristol Myers Squibb, Novartis and Chimeric Therapeutics; served as a consultant for Secura Bio; has licensed patents/royalties from BMS and Sanofi; and has received research funding from BMS.

RV: is an employee of Janssen

SZU: has received research funding from Amgen, Array Biopharma, Bristol Myers Squibb, Celgene, GlaxoSmithKline, Janssen, Merck, Pharmacyclics, Sanofi, Seattle Genetics, SkylineDX, Takeda; has received consulting fees from Abbvie, Amgen, Bristol Myers Squibb, Celgene, EdoPharma, Genentech, Gilead, GlaxoSmithKline, Janssen, Oncopeptides, Sanofi, Seattle Genetics, SecuraBio, SkylineDx, Takeda, TeneoBio and has received speaking fees from Amgen, Celgene, Janssen and Takeda.

PLM: has served as a consultant for Bluebird Bio, Bristol Myers Squibb, Celgene, Juno, Fate Therapeutics, Hikmo, Janssen, Juno, Karyopharm, Magenta Therapeutics, Novartis, Oncopeptides, Sanofi, Starton Therapeutics, and Takeda.

References

1. Holstein SA, Avet-Loiseau H, Hahn T, Ho CM, Lohr JG, Munshi NC et al. BMT CTN Myeloma Intergroup Workshop on Minimal Residual Disease and Immune Profiling: Summary and Recommendations from the Organizing Committee. Biol Blood Marrow Transplant 2018; 24(4): 641–648. e-pub ahead of print 2017/12/16; doi: 10.1016/j.bbmt.2017.12.774 [PubMed: 29242112]
2. Holstein SA, Ye JC, Howard A, Bhutani M, Gormley N, Hahn T et al. Summary of the Second Annual BMT CTN Myeloma Intergroup Workshop on Minimal Residual Disease and Immune Profiling. Biol Blood Marrow Transplant 2019; 25(3): e89–e97. e-pub ahead of print 2018/11/09; doi: 10.1016/j.bbmt.2018.11.001 [PubMed: 30408566]
3. Holstein SA, Al-Kadhimi Z, Costa LJ, Hahn T, Hari P, Hillengass J et al. Summary of the Third Annual Blood and Marrow Transplant Clinical Trials Network Myeloma Intergroup Workshop on Minimal Residual Disease and Immune Profiling. Biol Blood Marrow Transplant 2020; 26(1): e7–e15. e-pub ahead of print 2019/09/19; doi: 10.1016/j.bbmt.2019.09.015 [PubMed: 31526843]
4. Holstein SA, Howard A, Avigan D, Bhutani M, Cohen AD, Costa LJ et al. Summary of the 2019 Blood and Marrow Transplant Clinical Trials Network Myeloma Intergroup Workshop on Minimal Residual Disease and Immune Profiling. Biol Blood Marrow Transplant 2020; 26(10): e247–e255. e-pub ahead of print 2020/06/27; doi: 10.1016/j.bbmt.2020.06.011 [PubMed: 32589921]
5. Holstein SA, Bahlis N, Bergsagel PL, Bhutani M, Bolli N, Brownstein C et al. The 2020 BMT CTN Myeloma Intergroup Workshop on Immune Profiling and Minimal Residual Disease Testing in Multiple Myeloma. Transplant Cell Ther 2021; 27(10): 807–816. e-pub ahead of print 2021/06/10; doi: 10.1016/j.jtct.2021.05.027 [PubMed: 34107340]
6. Azab AK, Quang P, Azab F, Pitsillides C, Thompson B, Chonghaile T et al. P-selectin glycoprotein ligand regulates the interaction of multiple myeloma cells with the bone marrow microenvironment. Blood 2012; 119(6): 1468–1478. e-pub ahead of print 2011/11/19; doi: 10.1182/blood-2011-07-368050 [PubMed: 22096244]
7. de la Puente P, Muz B, Azab F, Azab AK. Cell trafficking of endothelial progenitor cells in tumor progression. Clin Cancer Res 2013; 19(13): 3360–3368. e-pub ahead of print 2013/05/15; doi: 10.1158/1078-0432.CCR-13-0462 [PubMed: 23665736]
8. Azab AK, Sahin I, Moschetta M, Mishima Y, Burwick N, Zimmermann J et al. CXCR7-dependent angiogenic mononuclear cell trafficking regulates tumor progression in multiple myeloma. Blood 2014; 124(12): 1905–1914. e-pub ahead of print 2014/08/01; doi: 10.1182/blood-2014-02-558742 [PubMed: 25079359]

9. Muz B, Azab F, de la Puente P, Rollins S, Alvarez R, Kawar Z et al. Inhibition of P-Selectin and PSGL-1 Using Humanized Monoclonal Antibodies Increases the Sensitivity of Multiple Myeloma Cells to Bortezomib. Biomed Res Int 2015; 2015: 417586. e-pub ahead of print 2015/11/06; doi: 10.1155/2015/417586 [PubMed: 26539491]
10. Muz B, Azab F, Fiala M, King J, Kohnen D, Fogler WE et al. Inhibition of E-Selectin (GMI-1271) or E-selectin together with CXCR4 (GMI-1359) re-sensitizes multiple myeloma to therapy. Blood Cancer J 2019; 9(9): 68. e-pub ahead of print 2019/08/23; doi: 10.1038/s41408-019-0227-3 [PubMed: 31431613]
11. Federico C, Alhallak K, Sun J, Duncan K, Azab F, Sudlow GP et al. Tumor microenvironment-targeted nanoparticles loaded with bortezomib and ROCK inhibitor improve efficacy in multiple myeloma. Nat Commun 2020; 11(1): 6037. e-pub ahead of print 2020/11/29; doi: 10.1038/s41467-020-19932-1 [PubMed: 33247158]
12. Murray DL, Puig N, Kristinsson S, Usmani SZ, Dispenzieri A, Bianchi G et al. Mass spectrometry for the evaluation of monoclonal proteins in multiple myeloma and related disorders: an International Myeloma Working Group Mass Spectrometry Committee Report. Blood Cancer J 2021; 11(2): 24. e-pub ahead of print 2021/02/11; doi: 10.1038/s41408-021-00408-4 [PubMed: 33563895]
13. Multi-site verification of the automated EXTENT MALDI-TOF-MS system and immunoglobulin isotypes assay for the identification and quantification of monoclonal immunoglobulins. AACC Annual Scientific Meeting; Atlanta, GA, 2021.
14. Giles HV, Cook MA, Drayson MT, Cook G, Wright NJ, North SJ et al. Redefining nonmeasurable multiple myeloma using mass spectrometry. Blood 2022; 139(6): 946–950. e-pub ahead of print 2021/12/07; doi: 10.1182/blood.2021013794 [PubMed: 34871382]
15. Caserta E, Chea J, Minnix M, Poku EK, Viola D, Vonderfecht S et al. Copper 64-labeled daratumumab as a PET/CT imaging tracer for multiple myeloma. Blood 2018; 131(7): 741–745. e-pub ahead of print 2018/01/06; doi: 10.1182/blood-2017-09-807263 [PubMed: 29301755]
16. Krishnan A, Adhikarla V, Poku EK, Palmer J, Chaudhry A, Biglang-Awa VE et al. Identifying CD38+ cells in patients with multiple myeloma: first-in-human imaging using copper-64-labeled daratumumab. Blood Adv 2020; 4(20): 5194–5202. e-pub ahead of print 2020/10/24; doi: 10.1182/bloodadvances.2020002603 [PubMed: 33095874]
17. Minnix M, Adhikarla V, Caserta E, Poku E, Rockne R, Shively JE et al. Comparison of CD38-Targeted alpha- Versus beta-Radionuclide Therapy of Disseminated Multiple Myeloma in an Animal Model. J Nucl Med 2021; 62(6): 795–801. e-pub ahead of print 2020/11/01; doi: 10.2967/jnumed.120.251983 [PubMed: 33127621]
18. Hillengass J, Usmani S, Rajkumar SV, Durie BGM, Mateos MV, Lonial S et al. International myeloma working group consensus recommendations on imaging in monoclonal plasma cell disorders. Lancet Oncol 2019; 20(6): e302–e312. e-pub ahead of print 2019/06/05; doi: 10.1016/s1470-2045(19)30309-2 [PubMed: 31162104]
19. Vij R, Fowler KJ, Shokeen M. New Approaches to Molecular Imaging of Multiple Myeloma. J Nucl Med 2016; 57(1): 1–4. e-pub ahead of print 2015/11/07; doi: 10.2967/jnumed.115.163808 [PubMed: 26541780]
20. Ghai A, Maji D, Cho N, Chanswangphuwana C, Rettig M, Shen D et al. Preclinical Development of CD38-Targeted [(89)Zr]Zr-DFO-Daratumumab for Imaging Multiple Myeloma. J Nucl Med 2018; 59(2): 216–222. e-pub ahead of print 2017/10/14; doi: 10.2967/jnumed.117.196063 [PubMed: 29025987]
21. Ulaner GA, Sobol NB, O'Donoghue JA, Kirov AS, Riedl CC, Min R et al. CD38-targeted Immuno-PET of Multiple Myeloma: From Xenograft Models to First-in-Human Imaging. Radiology 2020; 295(3): 606–615. e-pub ahead of print 2020/04/08; doi: 10.1148/radiol.2020192621 [PubMed: 32255416]
22. Ghai A, Zheleznyak A, Mixdorf M, O'Neal J, Ritchey J, Rettig M et al. Development of [(89)Zr]DFO-elotuzumab for immunoPET imaging of CS1 in multiple myeloma. Eur J Nucl Med Mol Imaging 2021; 48(5): 1302–1311. e-pub ahead of print 2020/11/13; doi: 10.1007/s00259-020-05097-y [PubMed: 33179150]
23. Ghai A, Fettig N, Fontana F, DiPersio J, Rettig M, Neal JO et al. In vivo quantitative assessment of therapeutic response to bortezomib therapy in disseminated animal models of multiple myeloma

with [(18)F]FDG and [(64)Cu]Cu-LLP2A PET. EJNMMI Res 2021; 11(1): 97. e-pub ahead of print 2021/09/30; doi: 10.1186/s13550-021-00840-4 [PubMed: 34586539]

24. Hathi D, Chanswangphuwana C, Cho N, Fontana F, Maji D, Ritchey J et al. Ablation of VLA4 in multiple myeloma cells redirects tumor spread and prolongs survival. Sci Rep 2022; 12(1): 30. e-pub ahead of print 2022/01/09; doi: 10.1038/s41598-021-03748-0 [PubMed: 34996933]
25. Nakamura K, Smyth MJ, Martinet L. Cancer immunoediting and immune dysregulation in multiple myeloma. Blood 2020; 136(24): 2731–2740. e-pub ahead of print 2020/07/10; doi: 10.1182/blood.2020006540 [PubMed: 32645135]
26. Papadas A, Arauz G, Cicala A, Wiesner J, Asimakopoulos F. Versican and Versican-matrikines in Cancer Progression, Inflammation, and Immunity. J Histochem Cytochem 2020; 68(12): 871–885. e-pub ahead of print 2020/07/07; doi: 10.1369/0022155420937098 [PubMed: 32623942]
27. Hope C, Ollar SJ, Heninger E, Hebron E, Jensen JL, Kim J et al. TPL2 kinase regulates the inflammatory milieu of the myeloma niche. Blood 2014; 123(21): 3305–3315. e-pub ahead of print 2014/04/12; doi: 10.1182/blood-2014-02-554071 [PubMed: 24723682]
28. Tang M, Diao J, Gu H, Khatri I, Zhao J, Cattral MS. Toll-like Receptor 2 Activation Promotes Tumor Dendritic Cell Dysfunction by Regulating IL-6 and IL-10 Receptor Signaling. Cell Rep 2015; 13(12): 2851–2864. e-pub ahead of print 2015/12/30; doi: 10.1016/j.celrep.2015.11.053 [PubMed: 26711349]
29. Bailur JK, McCachren SS, Doxie DB, Shrestha M, Pendleton K, Nooka AK et al. Early alterations in stem-like/resident T cells, innate and myeloid cells in the bone marrow in preneoplastic gammopathy. JCI Insight 2019; 5. e-pub ahead of print 2019/04/24; doi: 10.1172/jci.insight.127807
30. Pearce OMT, Delaine-Smith RM, Maniati E, Nichols S, Wang J, Bohm S et al. Deconstruction of a Metastatic Tumor Microenvironment Reveals a Common Matrix Response in Human Cancers. Cancer Discov 2018; 8(3): 304–319. e-pub ahead of print 2017/12/03; doi: 10.1158/2159-8290.CD-17-0284 [PubMed: 29196464]
31. Cheng S, Li Z, Gao R, Xing B, Gao Y, Yang Y et al. A pan-cancer single-cell transcriptional atlas of tumor infiltrating myeloid cells. Cell 2021; 184(3): 792–809 e723. e-pub ahead of print 2021/02/06; doi: 10.1016/j.cell.2021.01.010 [PubMed: 33545035]
32. Mushtaq MU, Papadas A, Pagenkopf A, Flietner E, Morrow Z, Chaudhary SG et al. Tumor matrix remodeling and novel immunotherapies: the promise of matrix-derived immune biomarkers. J Immunother Cancer 2018; 6(1): 65. e-pub ahead of print 2018/07/05; doi: 10.1186/s40425-018-0376-0 [PubMed: 29970158]
33. Hope C, Emmerich PB, Papadas A, Pagenkopf A, Matkowskyj KA, Van De Hey DR et al. Versican-Derived Matrikines Regulate Batf3-Dendritic Cell Differentiation and Promote T Cell Infiltration in Colorectal Cancer. J Immunol 2017; 199(5): 1933–1941. e-pub ahead of print 2017/07/30; doi: 10.4049/jimmunol.1700529 [PubMed: 28754680]
34. Dhakal B, Pagenkopf A, Mushtaq MU, Cunningham AM, Flietner E, Morrow Z et al. Versican proteolysis predicts immune effector infiltration and post-transplant survival in myeloma. Leuk Lymphoma 2019; 60(10): 2558–2562. e-pub ahead of print 2019/03/09; doi: 10.1080/10428194.2019.1585836 [PubMed: 30845856]
35. Cortes-Ciriano I, Lee JJ, Xi R, Jain D, Jung YL, Yang L et al. Comprehensive analysis of chromothripsis in 2,658 human cancers using whole-genome sequencing. Nat Genet 2020; 52(3): 331–341. e-pub ahead of print 2020/02/07; doi: 10.1038/s41588-019-0576-7 [PubMed: 32025003]
36. Rustad EH, Yellapantula VD, Glodzik D, Maclachlan KH, Diamond B, Boyle EM et al. Revealing the impact of structural variants in multiple myeloma. Blood Cancer Discov 2020; 1(3): 258–273. e-pub ahead of print 2021/01/05; doi: 10.1158/2643-3230.BCD-20-0132 [PubMed: 33392515]
37. Maura F, Boyle EM, Rustad EH, Ashby C, Kaminetzky D, Bruno B et al. Chromothripsis as a pathogenic driver of multiple myeloma. Semin Cell Dev Biol 2022; 123: 115–123. e-pub ahead of print 2021/05/08; doi: 10.1016/j.semcdb.2021.04.014 [PubMed: 33958284]
38. Maclachlan KH, Rustad EH, Derkach A, Zheng-Lin B, Yellapantula V, Diamond B et al. Copy number signatures predict chromothripsis and clinical outcomes in newly diagnosed multiple myeloma. Nat Commun 2021; 12(1): 5172. e-pub ahead of print 2021/08/29; doi: 10.1038/s41467-021-25469-8 [PubMed: 34453055]

39. Oben B, Froyen G, Maclachlan KH, Leongamornlert D, Abascal F, Zheng-Lin B et al. Whole-genome sequencing reveals progressive versus stable myeloma precursor conditions as two distinct entities. Nat Commun 2021; 12(1): 1861. e-pub ahead of print 2021/03/27; doi: 10.1038/s41467-021-22140-0 [PubMed: 33767199]
40. de Jong MME, Kellermayer Z, Papazian N, Tahri S, Hofste Op Bruinink D, Hoogenboezem R et al. The multiple myeloma microenvironment is defined by an inflammatory stromal cell landscape. Nat Immunol 2021; 22(6): 769–780. e-pub ahead of print 2021/05/22; doi: 10.1038/s41590-021-00931-3 [PubMed: 34017122]
41. Rasche L, Chavan SS, Stephens OW, Patel PH, Tytarenko R, Ashby C et al. Spatial genomic heterogeneity in multiple myeloma revealed by multi-region sequencing. Nat Commun 2017; 8(1): 268. e-pub ahead of print 2017/08/18; doi: 10.1038/s41467-017-00296-y [PubMed: 28814763]
42. Rasche L, Angtuaco EJ, Alpe TL, Gershner GH, McDonald JE, Samant RS et al. The presence of large focal lesions is a strong independent prognostic factor in multiple myeloma. Blood 2018; 132(1): 59–66. e-pub ahead of print 2018/05/23; doi: 10.1182/blood-2018-04-842880 [PubMed: 29784643]
43. Zamagni E, Patriarca F, Nanni C, Zannetti B, Englaro E, Pezzi A et al. Prognostic relevance of 18-F FDG PET/CT in newly diagnosed multiple myeloma patients treated with up-front autologous transplantation. Blood 2011; 118(23): 5989–5995. e-pub ahead of print 2011/09/09; doi: 10.1182/blood-2011-06-361386 [PubMed: 21900189]
44. Hillengass J, Fechtner K, Weber MA, Bauerle T, Ayyaz S, Heiss C et al. Prognostic significance of focal lesions in whole-body magnetic resonance imaging in patients with asymptomatic multiple myeloma. J Clin Oncol 2010; 28(9): 1606–1610. e-pub ahead of print 2010/02/24; doi: 10.1200/JCO.2009.25.5356 [PubMed: 20177023]
45. Gooding S, Ansari-Pour N, Towfic F, Ortiz Estevez M, Chamberlain PP, Tsai KT et al. Multiple cereblon genetic changes are associated with acquired resistance to lenalidomide or pomalidomide in multiple myeloma. Blood 2021; 137(2): 232–237. e-pub ahead of print 2021/01/15; doi: 10.1182/blood.2020007081 [PubMed: 33443552]
46. Shaffer AL, Emre NC, Lamy L, Ngo VN, Wright G, Xiao W et al. IRF4 addiction in multiple myeloma. Nature 2008; 454(7201): 226–231. e-pub ahead of print 2008/06/24; doi: 10.1038/nature07064 [PubMed: 18568025]
47. Wang L, Yao ZQ, Moorman JP, Xu Y, Ning S. Gene expression profiling identifies IRF4-associated molecular signatures in hematological malignancies. PLoS One 2014; 9(9): e106788. e-pub ahead of print 2014/09/11; doi: 10.1371/journal.pone.0106788 [PubMed: 25207815]
48. Bai H, Wu S, Wang R, Xu J, Chen L. Bone marrow IRF4 level in multiple myeloma: an indicator of peripheral blood Th17 and disease. Oncotarget 2017; 8(49): 85392–85400. e-pub ahead of print 2017/11/22; doi: 10.18632/oncotarget.19907 [PubMed: 29156727]
49. Lopez-Girona A, Heintel D, Zhang LH, Mendy D, Gaidarova S, Brady H et al. Lenalidomide downregulates the cell survival factor, interferon regulatory factor-4, providing a potential mechanistic link for predicting response. Br J Haematol 2011; 154(3): 325–336. e-pub ahead of print 2011/06/29; doi: 10.1111/j.1365-2141.2011.08689.x [PubMed: 21707574]
50. Agnarelli A, Chevassut T, Mancini EJ. IRF4 in multiple myeloma-Biology, disease and therapeutic target. Leuk Res 2018; 72: 52–58. e-pub ahead of print 2018/08/12; doi: 10.1016/j.leukres.2018.07.025 [PubMed: 30098518]
51. Mondala PK, Vora AA, Zhou T, Lazzari E, Ladel L, Luo X et al. Selective antisense oligonucleotide inhibition of human IRF4 prevents malignant myeloma regeneration via cell cycle disruption. Cell Stem Cell 2021; 28(4): 623–636 e629. e-pub ahead of print 2021/01/22; doi: 10.1016/j.stem.2020.12.017 [PubMed: 33476575]
52. Vora AA, Mondala PK, Costello C, MacLeod AR, Crews LA. Sensitive intranuclear flow cytometric quantification of IRF4 protein in multiple myeloma and normal human hematopoietic cells. STAR Protoc 2021; 2(2): 100565. e-pub ahead of print 2021/06/18; doi: 10.1016/j.xpro.2021.100565 [PubMed: 34136833]
53. Lazzari E, Mondala PK, Santos ND, Miller AC, Pineda G, Jiang Q et al. Alu-dependent RNA editing of GLI1 promotes malignant regeneration in multiple myeloma. Nat Commun 2017; 8(1): 1922. e-pub ahead of print 2017/12/06; doi: 10.1038/s41467-017-01890-w [PubMed: 29203771]

54. Krishnan AY, Minnema MC, Berdeja JG, Oriol A, van de Donk NWCJ, Rodriguez-Otero P et al. Updated Phase 1 Results from MonumenTAL-1: First-in-Human Study of Talquetamab, a G Protein-Coupled Receptor Family C Group 5 Member D x CD3 Bispecific Antibody, in Patients with Relapsed/Refractory Multiple Myeloma. Blood 2021; 138(Supplement 1): 158–158. doi: 10.1182/blood-2021-146868
55. Moreau P, Usmani SZ, Garfall AL, van de Donk NWCJ, Nahi H, San-Miguel J et al. 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. Blood 2021; 138(Supplement 1): 896–896. doi: 10.1182/blood-2021-147915
56. Frerichs KA, Broekmans MEC, Marin Soto JA, van Kessel B, Heymans MW, Holthof LC et al. Preclinical Activity of JNJ-7957, a Novel BCMAxCD3 Bispecific Antibody for the Treatment of Multiple Myeloma, Is Potentiated by Daratumumab. Clin Cancer Res 2020; 26(9): 2203–2215. e-pub ahead of print 2020/01/24; doi: 10.1158/1078-0432.CCR-19-2299 [PubMed: 31969333]
57. Verkleij CPM, Broekmans MEC, van Duin M, Frerichs KA, Kuiper R, de Jonge AV et al. Preclinical activity and determinants of response of the GPRC5DxCD3 bispecific antibody talquetamab in multiple myeloma. Blood Adv 2021; 5(8): 2196–2215. e-pub ahead of print 2021/04/24; doi: 10.1182/bloodadvances.2020003805 [PubMed: 33890981]
58. Truger MS, Duell J, Zhou X, Heimeshoff L, Ruckdeschel A, John M et al. Single- and double-hit events in genes encoding immune targets before and after T cell-engaging antibody therapy in MM. Blood Adv 2021; 5(19): 3794–3798. e-pub ahead of print 2021/09/03; doi: 10.1182/bloodadvances.2021004418 [PubMed: 34471932]
59. Friedrich M, Neri P, Leblay N, Kehl N, Michel J, Lee H et al. Mapping the Multiple Myeloma T Cell Landscape By Immunotherapeutic Perturbation Reveals Mechanism and Determinants of Response to Bispecific T Cell Engagers. Blood 2021; 138(Supplement 1): 731–731. doi: 10.1182/blood-2021-152149
60. Leblay N, Maity R, Barakat E, McCulloch S, Duggan P, Jimenez-Zepeda V et al. Cite-Seq Profiling of T Cells in Multiple Myeloma Patients Undergoing BCMA Targeting CAR-T or Bites Immunotherapy. Blood 2020; 136(Supplement 1): 11–12. doi: 10.1182/blood-2020-137650 [PubMed: 32276273]
61. Meermeier EW, Welsh SJ, Sharik ME, Du MT, Garbitt VM, Riggs DL et al. Tumor burden limits bispecific antibody efficacy through T cell exhaustion averted by concurrent cytotoxic therapy. *Blood* Cancer Discov 2021; 2(4): 354–369. e-pub ahead of print 2021/07/15; doi: 10.1158/2643-3230.BCD-21-0038
62. Liu SQ, Grantham A, Landry C, Granda B, Chopra R, Chakravarthy S et al. A CRISPR Screen Reveals Resistance Mechanisms to CD3-Bispecific Antibody Therapy. Cancer Immunol Res 2021; 9(1): 34–49. e-pub ahead of print 2020/11/13; doi: 10.1158/2326-6066.CIR-20-0080 [PubMed: 33177106]
63. Leblay N, Maity R, Hasan F, Neri P. Deregulation of Adaptive T Cell Immunity in Multiple Myeloma: Insights Into Mechanisms and Therapeutic Opportunities. Front Oncol 2020; 10: 636. e-pub ahead of print 2020/05/21; doi: 10.3389/fonc.2020.00636 [PubMed: 32432039]
64. Ahn S, Leblay N, Neri P. Understanding the Mechanisms of Resistance to T Cell-based Immunotherapies to Develop More Favorable Strategies in Multiple Myeloma. Hemasphere 2021; 5(6): e575. e-pub ahead of print 2021/06/08; doi: 10.1097/HS9.0000000000000575 [PubMed: 34095759]
65. Wei AH, Ribera JM, Larson RA, Ritchie D, Ghobadi A, Chen Y et al. Biomarkers associated with blinatumomab outcomes in acute lymphoblastic leukemia. Leukemia 2021; 35(8): 2220–2231. e-pub ahead of print 2021/02/06; doi: 10.1038/s41375-020-01089-x [PubMed: 33542479]
66. Arenas EJ, Martinez-Sabadell A, Rius Ruiz I, Roman Alonso M, Escorihuela M, Luque A et al. Acquired cancer cell resistance to T cell bispecific antibodies and CAR T targeting HER2 through JAK2 down-modulation. Nat Commun 2021; 12(1): 1237. e-pub ahead of print 2021/02/25; doi: 10.1038/s41467-021-21445-4 [PubMed: 33623012]
67. Minami J, Suzuki R, Mazitschek R, Gorgun G, Ghosh B, Cirstea D et al. Histone deacetylase 3 as a novel therapeutic target in multiple myeloma. Leukemia 2014; 28(3): 680–689. e-pub ahead of print 2013/08/06; doi: 10.1038/leu.2013.231 [PubMed: 23913134]

68. Ho M, Chen T, Liu J, Dowling P, Hideshima T, Zhang L et al. Targeting histone deacetylase 3 (HDAC3) in the bone marrow microenvironment inhibits multiple myeloma proliferation by modulating exosomes and IL-6 trans-signaling. Leukemia 2020; 34(1): 196–209. e-pub ahead of print 2019/05/31; doi: 10.1038/s41375-019-0493-x [PubMed: 31142847]
69. Munshi NC, Anderson LD Jr., Shah N, Madduri D, Berdeja J, Lonial S et al. Idecabtagene Vicleucel in Relapsed and Refractory Multiple Myeloma. N Engl J Med 2021; 384(8): 705–716. e-pub ahead of print 2021/02/25; doi: 10.1056/NEJMoa2024850 [PubMed: 33626253]
70. Martin T, Usmani SZ, Berdeja JG, Jakubowiak A, Agha M, Cohen AD et al. Updated Results from CARTITUDE-1: Phase 1b/2Study of Ciltacabtagene Autoleucel, a B-Cell Maturation Antigen-Directed Chimeric Antigen Receptor T Cell Therapy, in Patients With Relapsed/Refractory Multiple Myeloma. Blood 2021; 138(Supplement 1): 549–549. doi: 10.1182/blood-2021-146060
71. Martin N, Thompson EG, Brown W, Finney O, Rytlewski J, Jiang Y et al. Idecabtagene Vicleucel (ide-cel, bb2121) Responses Are Characterized By Early and Temporally Consistent Activation and Expansion of CAR T Cells with a T Effector Phenotype. Blood 2020; 136(Supplement 1): 17–18. doi: 10.1182/blood-2020-134378
72. Correlation of tumor BCMA expression with response and acquired resistance to idecabtagene vicleucel in the KARMMA study in relapsed and refractory multiple myeloma. EHA 2020, 2020.
73. Nguyen DN, Roth TL, Li PJ, Chen PA, Apathy R, Mamedov MR et al. Polymer-stabilized Cas9 nanoparticles and modified repair templates increase genome editing efficiency. Nat Biotechnol 2020; 38(1): 44–49. e-pub ahead of print 2019/12/11; doi: 10.1038/s41587-019-0325-6 [PubMed: 31819258]
74. Roth TL, Puig-Saus C, Yu R, Shifrut E, Carnevale J, Li PJ et al. Reprogramming human T cell function and specificity with non-viral genome targeting. Nature 2018; 559(7714): 405–409. e-pub ahead of print 2018/07/12; doi: 10.1038/s41586-018-0326-5 [PubMed: 29995861]
75. Schumann K, Lin S, Boyer E, Simeonov DR, Subramaniam M, Gate RE et al. Generation of knock-in primary human T cells using Cas9 ribonucleoproteins. Proc Natl Acad Sci U S A 2015; 112(33): 10437–10442. e-pub ahead of print 2015/07/29; doi: 10.1073/pnas.1512503112 [PubMed: 26216948]
76. Eyquem J, Mansilla-Soto J, Giavridis T, van der Stegen SJ, Hamieh M, Cunanan KM et al. Targeting a CAR to the TRAC locus with CRISPR/Cas9 enhances tumour rejection. Nature 2017; 543(7643): 113–117. e-pub ahead of print 2017/02/23; doi: 10.1038/nature21405 [PubMed: 28225754]
77. Dobrowsky T, Gianni D, Pieracci J, Suh J. AAV manufacturing for clinical use: Insights on current challenges from the upstream process perspective. Current Opinion in Biomedical Engineering 2021; 20: 100353. doi: 10.1016/j.cobme.2021.100353
78. Samur MK, Fulciniti M, Aktas Samur A, Bazarbachi AH, Tai YT, Prabhala R et al. Biallelic loss of BCMA as a resistance mechanism to CAR T cell therapy in a patient with multiple myeloma. Nat Commun 2021; 12(1): 868. e-pub ahead of print 2021/02/10; doi: 10.1038/s41467-021-21177-5 [PubMed: 33558511]
79. Brudno JN, Maric I, Hartman SD, Rose JJ, Wang M, Lam N 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. e-pub ahead of print 2018/05/31; doi: 10.1200/JCO.2018.77.8084 [PubMed: 29812997]
80. Cohen AD, Garfall AL, Stadtmauer EA, Melenhorst JJ, Lacey SF, Lancaster E et al. B cell maturation antigen-specific CAR T cells are clinically active in multiple myeloma. J Clin Invest 2019; 130: 2210–2221. e-pub ahead of print 2019/03/22; doi: 10.1172/jci126397
81. Smith EL, Harrington K, Staehr M, Masakayan R, Jones J, Long TJ et al. GPRC5D is a target for the immunotherapy of multiple myeloma with rationally designed CAR T cells. Sci Transl Med 2019; 11(485). e-pub ahead of print 2019/03/29; doi: 10.1126/scitranslmed.aau7746
82. Mailankody S, Diamonte C, Fitzgerald L, Kane P, Wang X, Sikder DS et al. Phase I First-in-Class Trial of MCARH109, a G Protein Coupled Receptor Class C Group 5 Member D (GPRC5D) Targeted CAR T Cell Therapy in Patients with Relapsed or Refractory Multiple Myeloma. Blood 2021; 138(Supplement 1): 827–827. doi: 10.1182/blood-2021-153204 [PubMed: 34075408]

83. Simon S, Riddell SR. Dual Targeting with CAR T Cells to Limit Antigen Escape in Multiple Myeloma. Blood Cancer Discov 2020; 1(2): 130–133. e-pub ahead of print 2020/08/03; doi: 10.1158/2643-3230.BCD-20-0122 [PubMed: 34661143]

84. Fernandez de Larrea C, Staehr M, Lopez AV, Ng KY, Chen Y, Godfrey WD et al. Defining an Optimal Dual-Targeted CAR T-cell Therapy Approach Simultaneously Targeting BCMA and GPRC5D to Prevent BCMA Escape-Driven Relapse in Multiple Myeloma. Blood Cancer Discov 2020; 1(2): 146–154. e-pub ahead of print 2020/10/23; doi: 10.1158/2643-3230.BCD-20-0020 [PubMed: 33089218]

85. Lee L, Draper B, Chaplin N, Philip B, Chin M, Galas-Filipowicz D et al. An APRIL-based chimeric antigen receptor for dual targeting of BCMA and TACI in multiple myeloma. Blood 2018; 131(7): 746–758. e-pub ahead of print 2017/12/30; doi: 10.1182/blood-2017-05-781351 [PubMed: 29284597]

Highlights

- Summary of the BMT CTN Myeloma Intergroup workshop on immune and cellular therapy
- Mass spectrometry and theranostics are emerging platforms for disease evaluation
- Immune and stromal microenvironment play key roles in myeloma development
- Deciphering resistance mechanisms leads to novel therapeutic strategies
- Alternative strategies to BCMA-directed CAR T-cell therapy are under development

Table 1.

Key takeaway points from the workshop presentations.

A summarizing sentence is provided for each of the 16 presentations along with the surname of the presenter.

Session Title	Presentation Summaries
Platforms for Myeloma Disease Evaluation	• P-selectin-targeted therapies can aid in delivery of agents to the bone marrow microenvironment (Azab) • MALDI-TOF can detect small monoclonal proteins in patients considered to have oligo-secretory or non-secretory disease (Giles) • Daratumumab radio-labeled antibodies have diagnostic and therapeutic potential (Pichiorri) • Immuno-PET imaging may be a promising strategy for stratifying patients for immune- and cell-based therapy (Shokeen)
Insights into Pathogenesis	• The tumor matrix proteoglycan versican modulates the immune microenvironment (Asimakopoulos) • Chromothripsisis is associated with progression in myeloma precursors (Maclachlan) • Inflammatory mesenchymal stromal cells could be myeloma niche cells and their presence is associated with bone marrow inflammation (Cupedo) • Focal lesions are hotspots for genetic and transcriptional heterogeneity (John)
Therapeutic Targets & Resistance Mechanisms	• Regional copy loss of *CRBN* at 3p and COPS7b/8 at 2q37 may mediate IMiD resistance (Gooding) • Inhibition of IRF4 represents a novel therapeutic strategy (Crews) • Resistance to CD3-directed bispecific antibodies is likely a combination of intrinsic and acquired mechanisms, including tumor-dependent and T-cell/tumor microenvironment-dependent mechanisms (Verona) • HDAC3 may be a promising target to block the bone marrow niche-myeloma interaction and limit myeloma cell survival (Bianchi)
Cellular Therapy	• Early clearance of soluble BCMA correlates with durable responses in patients who received idecabtagene vicleucel in the KarMMa trial (Kaiser) • Non-viral knockin strategies using single stranded DNA can be applied to BCMA CAR T-cell manufacturing (Shy) • CAR T-cells targeting GPRC5D +/− BCMA appear promising (Smith) • Studies evaluating CAR T-cells targeting APRIL have been disappointing and may be a reflection of differences between the natural ligand interaction and that required for CAR activation (Lee)