Presented at the American Society of Hematology Annual Meeting; December 6–9, 2014; San Francisco, CA

Prognostic and Predictive Gene Expression Profiling Markers Confirmed in Carfilzomib, Lenalidomide, and Dexamethasone (KRd)–Treated, Newly Diagnosed Multiple Myeloma Patients

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Introduction

- The combination of carfilzomib (Kyprolis®; Onyx Pharmaceuticals, Inc., South San Francisco, CA), lenalidomide (Revlimid®; Celgene Corporation, Summit, NJ), and dexamethasone (KRd) has proven to be highly active with rapid and deep responses in patients with newly diagnosed multiple myeloma (NDMM)
  - In 2 KRd studies in which autologous stem cell transplantation (ASCT) was and was not incorporated into the KRd treatment plan (NCT01816971 and NCT01029054, respectively), the rate of near complete response (nCR) or better after 4 cycles of KRd was 17% and 38%, respectively; the stringent complete response (sCR) rate was 50% (after 8 cycles) and 61% (after at least 8 cycles), respectively[1,2]
    - In these studies, patients who achieved at least an nCR after 4 cycles of KRd were more likely to ultimately achieve an sCR than patients who did not achieve at least an nCR after 4 cycles
- The achievement of an sCR with KRd was associated with improved time-to-event outcomes in patients with NDMM[3]
  - The estimated 3-year progression-free survival (PFS) and overall survival (OS) rates trended higher in patients who achieved an sCR compared with those who did not (PFS: 87% vs 68%, respectively; OS: 100% vs 91%)[3]
- Because an early response to KRd predicts the subsequent depth of response and because the depth of response to KRd is associated with improved time-to-event outcomes, an early marker of response would be useful for predicting patient outcomes and identifying patients who would most likely benefit from treatment with KRd
- Previously, the MMprofiler (SkylineDx, Rotterdam, The Netherlands) gene expression profiling (GEP) assay identified a combination of 5 markers (SKY92, virtual gain[1q], virtual t[14;16]/t[14;20], MF, and CD2 clusters) that predicted treatment outcomes in patients who were treated with the proteasome inhibitor (PI) bortezomib[4]
  - Patients with NDMM received bortezomib in the HOVON-65/GMMG-HD4 randomized phase 3 trial, which evaluated bortezomib plus doxorubicin and dexamethasone (PAD) induction (before ASCT) with bortezomib maintenance (after ASCT) vs vincristine plus doxorubicin and dexamethasone (VAD) induction (before ASCT) with thalidomide maintenance (after ASCT)[5]
- Herein, we report the results of a retrospective analysis that evaluated the relationship of the MMprofiler GEP markers with best overall response after 4 cycles and PFS in patients with NDMM who were treated with KRd in the NCT01029054 and NCT01816971 studies

Materials and Methods

Study Designs

- The patient eligibility criteria, treatment schema, and response and safety assessments for the NCT01029054 and NCT01816971 studies have been described previously[1,2]
- The treatment schema for each study is shown in **Figure 1**
- In these 2 studies, patients received 4 cycles of induction, followed by extended KRd treatment with deferred ASCT (NCT01029054) or ASCT followed by extended treatment with KRd (NCT01816971)
- In both studies, patients received single-agent lenalidomide as maintenance after completing treatment with KRd

Figure 1. Schemas for (A) the KRd study with deferred ASCT (NCT01029054) and for (B) the KRd study with ASCT (NCT01816971), as well as (C) the dosing schedule for both studies

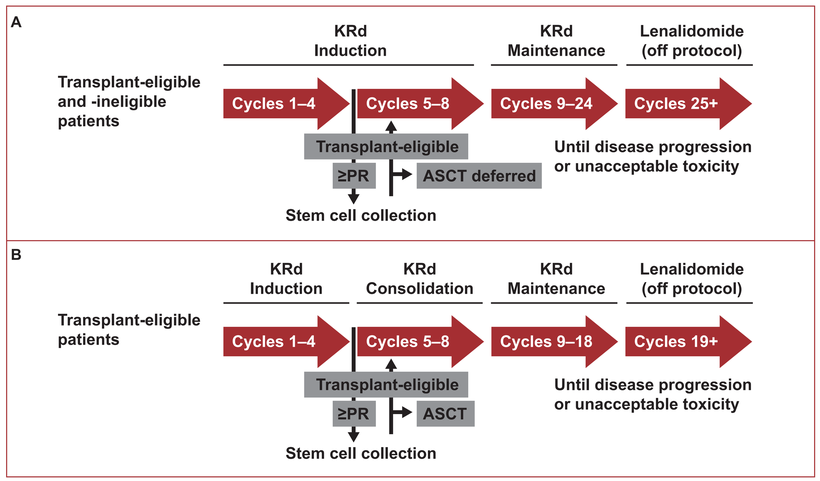


Figure 1, continued
C

KRd induction[a]	SCC + HD-ASCT[b]	KRd consolidation[a,c]	KRd maintenance[a]	LEN maintenance (off protocol)
• CFZ: days 1,[d] 2,[d] 8, 9, 15, and 16 (20, 27, or 36 mg/m²[e]) • LEN: days 1–21 (25 mg) • DEX: days 1, 8, 15, and 22 (40 mg)	• SCC with G-CSF ± plerixafor 2–4 weeks after day 21 of cycle 4 • HD melphalan 200 mg/m² with ASCT per the institution's standard practice	• CFZ: days 1, 2, 8, 9, 15, and 16 (last tolerated dose) • LEN: days 1–21 (15 mg for the first cycle with the option to increase to the last tolerated dose thereafter) • DEX: days 1, 8, 15, and 22 (20 mg or last tolerated dose)	• CFZ: days 1, 2, 15, and 16 (last tolerated dose) • LEN: days 1–21 (last tolerated dose) • DEX: days 1, 8, 15, and 22 (last tolerated dose)	• LEN: days 1–21 (last tolerated dose)

[a]Cycles were 28 days in duration.
[b]ASCT was part of the KRd treatment plan in the NCT01816971 study; patients had the option to defer ASCT in study NCT01029054.
[c]Consolidation was part of the KRd treatment plan in the NCT01816971 study; consolidation was not part of the KRd treatment plan in the NCT01029054 study.
[d]Starting dose of 20 mg/m² on days 1 and 2 of cycle 1 followed by escalation to the target dose thereafter.
[e]In the dose escalation study NCT01029054, the target dose level was 20, 27, or 36 mg/m²; in the NCT01816971 study, the target dose was 36 mg/m². The infusion time was 10 minutes for doses of 20 and 27 mg/m² and 30 minutes for the 36 mg/m² dose.

ASCT, autologous stem cell transplantation; CFZ, carfilzomib; DEX, dexamethasone; G-CSF, granulocyte colony-stimulating factor; HD, high-dose; HD-ASCT, high-dose autologous stem cell transplantation; KRd, carfilzomib with lenalidomide and dexamethasone; LEN, lenalidomide; SCC, stem cell collection.

GEP Assay

- The MMprofiler GEP assay was performed in a subset of patients with available high-quality RNA from CD138+ purified plasma cells by SkylineDx, using gene expression microarrays (GeneChip Human Genome U133 Plus 2.0; Affymetrix, Santa Clara, CA) and a prognostic 92-gene signature[6] (SKY92; SkylineDx, Rotterdam, The Netherlands)
- The set of MMprofiler markers investigated in this analysis included:
  - SKY92 signature[6]
  - A total of 7 virtual karyotyping markers (t[4;14], t[11;14], t[14;16]/t[14;20], gain[1q], del[13q], del[17p], hyperdiploid MM [H-MM; virtual gain(9q)])
  - Three clusters (MF, MS, and CD2)

GEP Analysis

- The GEP data were analyzed for associations between any of the MMprofiler markers and best overall response (≥nCR vs <nCR) after 4 cycles of KRd using the Fisher's exact test
- To analyze the associations between the MMprofiler markers and PFS, Kaplan–Meier curves were constructed, median survival estimates were calculated, and a Cox proportional hazards model was used to calculate hazard ratios (HRs) and the associated *P* values

Results

- The data cutoff date for this analysis was July 1, 2014
- Ninety-one patients were enrolled in the 2 KRd studies (NCT01029054: n=53; NCT01816971: n=38)
- CD138+ RNA was available from 26 patients (NCT01029054: n=15; NCT01816971: n=11)
  - Response data were available for all 26 patients
  - PFS data were available for 15 patients
    - All 15 were from study NCT01029054

Marker for Best Overall Response

- Patients with H-MM (virtual gain[9q]) were significantly less likely to achieve a best overall response of ≥nCR compared with those who did not have H-MM (virtual gain[9q]) (**Table 1**; *P*=.037)
  - None of the other MMprofiler markers had a statistically significant association with best overall response

Table 1. H-MM (virtual gain[9q]) Marker and Best Overall Response After 4 Cycles of KRd (n=26)

	Best overall response after 4 cycles of KRd	
H-MM (virtual gain[9q]) status	<nCR	≥nCR
H-MM (virtual gain[9q])–negative	8	9
H-MM (virtual gain[9q])–positive	8	1

H-MM; hyperdiploid multiple myeloma; KRd, carfilzomib plus lenalidomide and dexamethasone; nCR, near complete response.

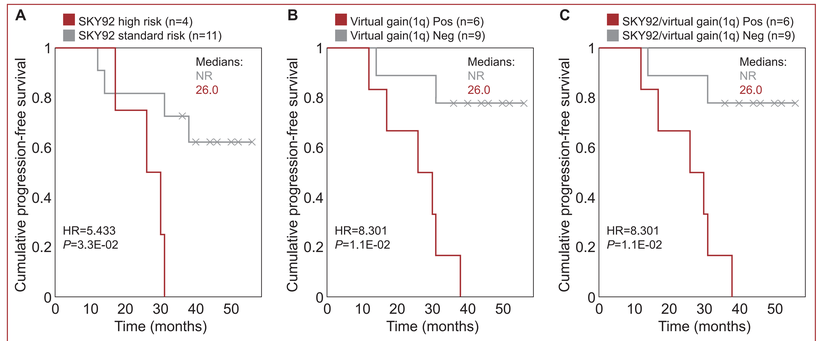
Individual Markers for PFS

- Two markers were found to have a statistically significant association with PFS: SKY92 (HR=5.4; *P*=.033; **Figure 2A**) and virtual gain(1q) (HR=8.3; *P*=.011; **Figure 2B**)
- The virtual t(4;14) marker was marginally associated with PFS (HR=3.4; *P*=.084)

The 5-Marker PI Predictor Signature for PFS

- Six of the 15 patients (40%) with PFS data were positive for SKY92 or virtual gain(1q), and none were positive for virtual t(14;16)/t(14;20), MF, or CD2 clusters
  - These patients constitute a high-risk subgroup with an inferior median PFS compared with standard-risk patients (26 months vs not reached, respectively; HR=8.3; *P*=.011; **Figure 2C**)
- Nine of the 15 patients (60%) were negative for all 5 markers
  - Their estimated 4-year PFS rate was 78%

Figure 2. Kaplan–Meier curves for progression-free survival by (A) SKY92 status,[6] (B) virtual gain(1q) status, and (C) combined SKY92/virtual gain(1q) status



HR, hazard ratio; Neg, negative; NR, not reached; Pos, positive.

Conclusions and Future Directions

- In patients with NDMM who were treated with KRd with or without ASCT, there were significant associations between MMprofiler GEP markers and response and PFS
  - The absence of H-MM (virtual gain[9q]) identified a subgroup of patients who were more likely to achieve at least an nCR at the end of 4 cycles of KRd
    - The achievement of at least an nCR after 4 cycles of KRd has been previously associated with the eventual achievement of an sCR
  - SKY92 and virtual gain(1q) were significantly associated with PFS
- The 5-marker PI predictor signature that previously predicted outcomes in patients treated with bortezomib (within the PAD regimen) was also associated with PFS in patients treated with carfilzomib within the KRd regimen
  - In patients treated with KRd, positivity for SKY92 or virtual gain(1q) was associated with a statistically significant reduction in the median PFS compared with patients who were negative for these markers
  - In the small subset of KRd-treated patients who were positive for SKY92 or virtual gain(1q), the median PFS appeared longer than the median PFS previously reported in patients who were treated with VAD followed by ASCT and thalidomide maintenance (26 vs 18 months, respectively)[5]
  - In KRd-treated patients who were negative for all 5 markers (n=9), the 4-year PFS rate appeared higher than the 4-year PFS rate previously reported for patients treated with PAD who were also negative for the 5-marker predictor signature (n=75) (78% vs 34%, respectively)[4]
- Enrollment is ongoing (planned enrollment of 70 patients) in the KRd with ASCT study (NCT01816971)

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Acknowledgments

This research was performed within the framework of CTMM, the Center for Translational Molecular Medicine, project BioCHIP grant 03O-102. Editorial assistance was provided by BlueMomentum, a division of KnowledgePoint360 Group, an Ashfield Company, and funded by Onyx Pharmaceuticals, Inc., an Amgen subsidiary.

Conflict of Interest Disclosure

van Vliet: *SkylineDx:* Employment. **Vij:** *Celgene:* Honoraria, Research Funding; *Onyx:* Honoraria, Research Funding; *Sanofi:* Honoraria; *Janssen:* Honoraria; *Novartis:* Honoraria; *Millennium:* Honoraria; *Array:* Honoraria. **Dumee:** *SkylineDx:* Employment. **Bosman:** *SkylineDx:* Employment. **deBest:** *SkylineDx:* Employment. **van Beers:** *SkylineDx:* Employment. **Jakubowiak:** *Bristol-Myers Squibb:* Consultancy, Honoraria, Membership on an entity's Board of Directors or advisory committees; *Celgene:* Consultancy, Honoraria, Membership on an entity's Board of Directors or advisory committees, Speakers Bureau; *Janssen:* Consultancy, Honoraria, Membership on an entity's Board of Directors or advisory committees, Speakers Bureau; *Millennium:* Consultancy, Honoraria, Membership on an entity's Board of Directors or advisory committees; *Novartis:* Honoraria, Membership on an entity's Board of Directors or advisory committees; *Onyx:* Consultancy, Honoraria, Membership on an entity's Board of Directors or advisory committees, Speakers Bureau; *SkylineDx:* Honoraria, Membership on an entity's Board of Directors or advisory committees.