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a multi-center study in the era of novel myeloma therapies

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Invasive fungal infections in patients with multiple myeloma: a multi-center study in the era of novel myeloma therapies

Running title: Invasive fungal infection in patients with myeloma

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Key words: fungal infection; myeloma; novel agents
Multiple myeloma (MM) is a hematological malignancy with increasing prevalence in older populations. Infections, particularly pyogenic infections and reactivation of latent viral infections, are a leading cause of morbidity and mortality in patients with MM. Previously, invasive aspergillosis (IA) has been found to be a significant opportunistic infection in patients with myeloma managed with intensive conventional combination chemotherapeutic regimens with nearly 50% attributable mortality. IA tended to occur early in the treatment course, in patients with higher disease stage. However, the treatment of myeloma has undergone a paradigm shift with the use of immunomodulatory drugs (IMiDs), proteasome inhibitors (PI) and autologous hematopoietic stem cell transplant (ASCT) as the new standard of care.

The epidemiology and outcomes of invasive fungal infections (IFI) are well defined in other hematology populations, and are supported by guidelines for antifungal prophylaxis. However, the disease burden and characteristics of IFI in patients with MM in the era of novel agents have not been previously reported. In addition, the contribution and impact of these novel agents and early use of ASCT on risk of IFI is uncertain. Therefore our study aimed to define the role of antifungal prophylaxis, the epidemiology, risk factors and outcomes of IFI in patients with MM receiving novel agents.

The study was conducted at the Peter MacCallum Cancer Centre (PMCC) and Austin Hospital (AH) which are tertiary referral centers for MM in Victoria, Australia. All patients with MM and an IFI managed at both centers from January 2009 to December 2011 were identified retrospectively from electronic medical management and discharge records, antimicrobial restricted approval and pharmacy dispensing systems and included in this study. Clinical, microbiology and radiology records of the identified patients were further reviewed using a standardized tool to capture the following: patient demographics, myeloma type, stage and treatment received (number of treatment cycles, lines of therapy), known predispositions for IFI (neutropenia, receipt of corticosteroids or chemotherapy), use of antifungal prophylaxis, clinical features, type and site of IFI, antifungal treatment received and outcomes (need for intensive care management and all-cause mortality at 30 days).

During the study period, patients with MM received treatment consistent with prevailing international practices including the use of IMiDs, (lenalidomide or thalidomide) and/or PI (bortezomib) in combination with corticosteroids. Fluconazole prophylaxis was routinely used for the period of ASCT (up to 3 months post transplant) for all patients at PMCC and in patients with past and/or expected history of mucositis at AH during the period of neutropenia. Patients suspected of having an IFI or with persistent febrile neutropenia with unclear source were investigated with high-resolution computer tomography (CT) of the chest and sinuses or position emission tomography-CT scan followed by directed tissue sampling for microscopy and fungal culture. Molecular testing with Aspergillus polymerase chain reaction and galactomannan testing on serum and BAL were routinely used at PMCC.

A line of therapy as defined by the International Myeloma Workshop consensus panel recommendations was used to provide an estimate of disease burden, treatment and number of relapses. IFI was defined and classified according to the European Organisation for Research and Treatment of Cancer (EORTC)/Mycoses Study Group
We received approval from human research ethics committees at both institutions.

Overall 372 patients received treatment for MM at both centers and were followed for a median of 24 months. Nine patients (2.4%) were diagnosed with 9 episodes of IFI; 3 were proven, 2 probable and 4 possible according to EORTC/MSG criteria. Rates of invasive mould infection and IA were 0.8% (3/372) and 0.3% (1/372), respectively. The IFI rate was 2.2% (3/135) following ASCT and 2.5% (6/237) for patients who had not received an ASCT. The rate of IFI in patients who received ≥ 3 lines of therapy was 15.0%.

Patients with an IFI had a median age of 62 (IQR, 59 to 64) years and were predominantly male (6/9). A majority of patients (6/9) were International staging system stage 1 at myeloma diagnosis. Most IFI episodes (8/9) occurred with myeloma progression (n=5) or after a second or third autograft (n=3) at a median of 35 (26 to 60) months following initial disease diagnosis after having received a median of 5 lines of therapy. Predisposing factors for IFI were present in the majority of instances: neutropenia < 0.5 X 10^9/L for ≥ 10 days temporally related to onset of an IFI (8/9 cases), corticosteroid therapy (≥0.5mg/kg/day of prednisolone equivalent over 4 weeks in 3/9), and T cell suppressive chemotherapy prior to diagnosis of IFI (6/9).

The sites of involvement were pulmonary (6/9) and disseminated (3/9). Isolated fungal pathogens included Candida albicans, Candida parapsilosis, Scedosporium prolificans (proven), Aspergillus fumigatus, and Scopulariopsis sp. (probable). All patients with an IFI received antifungal treatment. The majority of episodes 5/9 (55.6%) (2 proven, 1 probable and 2 possible) required management in the intensive care unit and overall 30-day all-cause mortality was (4/9) 44.4%. Characteristics of IFI episodes and outcomes are summarized in Table 2.

Univariate and multivariate regression analysis of risk factors was performed, using IFI as the evaluable outcome. Univariate analysis demonstrated that receipt of bortezomib (p=0.01), and ≥3 lines of therapy for MM (p < 0.01) were associated with IFI (Table 3). Multivariate logistic regression analysis demonstrated that receipt of ≥ 3 lines of therapy within 3 years was independently associated with an increased risk of IFI (p = 0.02) (Table 3).

In our study, we observed an overall low IFI rate of 2.4% with an invasive mould infection rate of 0.8%. Despite the lack of use of mould active prophylaxis (2%), the rates of invasive mould infection and IA (0.8% and 0.3%, respectively) in this study are comparable to rates (0.5-0.7%) reported in other studies of MM patients during the last decade.(7, 8) The use of fluconazole prophylaxis in the study was consistent with Australian antifungal guidelines.(4) In this context, our IFI rate of 2.2% following ASCT remains within the anticipated range expected following ASCT for patients with other hematological malignancies.(9, 10) The uptake of a diagnostic driven approach to persistent neutropenic fever following ASCT does not appear to have led to a higher IA rate through increased detection in our study in comparison to other centers using a similar approach.(11)
We observed that IFI occurred mostly during the period of disease progression (85.7%) with a median of 35 months between initial myeloma diagnosis and episode of IFI and a median of 5 lines of treatment. This is in contrast to older studies, where a much earlier onset of IFI has been reported.(3, 8) This shift could be because IMiDs and PI are inherently less myelosuppressive compared with conventional chemotherapy, which now tend to be used in combination for patients with refractory or progressive disease.(12) Cumulative deficits in various arms of the immune system, in particular of cell-mediated immunity due to progressive disease could also account for our findings.(2)

The impact of novel agents and use of ASCT on risk of IFI has not been previously evaluated. In our study the type of treatment, including receipt of ASCT and bortezomib did not appear to be independently associated with risk of developing an IFI. However, the number of lines of therapy was significantly associated with an increased risk of developing an IFI. With ≥3 lines of therapy, the IFI rate was 15.0%, which would warrant consideration of antifungal prophylaxis or surveillance. This finding suggests cumulative exposure to immunosuppressive treatment and disease burden is a greater determinant of IFI risk than type of individual therapy.

Mortality rates up to 60% were reported in the earlier era of conventional therapy.(3) IFI in our patients was still associated with significant morbidity and mortality with nearly 60% of episodes requiring ICU management and a 30-day all-cause mortality rate of 44%. These outcomes underscore the need for improvement in early diagnosis and treatment.

Our study has several limitations. Intensity of diagnostic evaluation for IFI was determined by the treating physician, which may have resulted in selection bias favouring diagnosis of IFI in symptomatic and unwell patients. Although the number of detected cases of IFI was limited, these were drawn from a large heterogeneously treated myeloma patient population and our data have allowed us to define the relationship between antifungal prophylaxis, IFI and outcomes.

In our dedicated study of IFI in patients with MM, the first in the era of novel anti-myeloma agents, rates of IFI and IA, including following ASCT remain low. This is in the context of fluconazole prophylaxis during ASCT, overall low use of mould active prophylaxis and diagnostic driven strategies for neutropenic fever. It appears cumulative treatment exposure and disease burden (≥3 lines of therapy in 3 years) is a greater determinant of IFI risk than type of individual therapy. Therefore, a high-risk group has been identified which could benefit from mould active prophylaxis or enhanced surveillance for IFI.
**Authorship**
BT was the primary investigator, developed the study protocol and was involved in data collection and review. JT and KU were involved in data collection and review. All authors were involved in manuscript writing and critical review.

**Acknowledgement**
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**Conflict of interest**
SJH has received honoraria and research grant funding from Celgene corporation and Janssen Cilag. AG is on the advisory board for Merck Sharpe & Dohme (MSD). MAS has received grant funding from and sat on advisory boards for Gilead, MSD and Pfizer. All other authors have no conflicts of interest to declare.
References


<table>
<thead>
<tr>
<th>Patient characteristics</th>
<th>Overall N = 372 (%)</th>
<th>PMCC N = 251 (%)</th>
<th>AH N = 121 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (median; IQR): years</td>
<td>65 (58-72)</td>
<td>64 (56-71)</td>
<td>67 (59-75)</td>
</tr>
<tr>
<td>Sex:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>226 (61)</td>
<td>148 (59)</td>
<td>78 (64)</td>
</tr>
<tr>
<td>Female</td>
<td>146 (39)</td>
<td>103 (41)</td>
<td>43 (36)</td>
</tr>
<tr>
<td>Treatment received:</td>
<td>316 (85)</td>
<td>225 (90)</td>
<td>91 (75)</td>
</tr>
<tr>
<td>---------------------</td>
<td>----------</td>
<td>----------</td>
<td>--------</td>
</tr>
<tr>
<td>Immunomodulatory drug</td>
<td>93 (25)</td>
<td>63 (25)</td>
<td>30 (25)</td>
</tr>
<tr>
<td>Bortezomib</td>
<td></td>
<td>101 (40)</td>
<td>34 (28)</td>
</tr>
<tr>
<td>ASCT</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anti-fungal prophylaxis:</td>
<td>118 (32)</td>
<td>103 (41)</td>
<td>15 (12)</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>6* (2)</td>
<td>6 (2)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Mould active agent*#</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Invasive fungal infection:</td>
<td>N = 9 (%)</td>
<td>N = 7 (%)</td>
<td>N = 2 (%)</td>
</tr>
<tr>
<td>Proven</td>
<td>3 (33)</td>
<td>3 (43)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Probable</td>
<td>2 (22)</td>
<td>2 (29)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Possible</td>
<td>4 (44)</td>
<td>2 (29)</td>
<td>2 (100)</td>
</tr>
</tbody>
</table>

Table 1: Overall characteristics of patients with myeloma (2009 to 2011)

PMCC: Peter MacCallum Cancer Centre; AH: Austin Hospital

# mould active agents include voriconazole, posaconazole and liposomal amphotericin

*Of the 6 patients who received mould active antifungal prophylaxis, 2 received posaconazole following allogeneic stem cell transplant and 4 received mould active agent due to previous isolation of *Aspergillus* sp. in sputum and multiple lines of previous therapy (median = 3).
### Table 2: Clinical features and outcomes of patients with myeloma and an invasive fungal infection

<table>
<thead>
<tr>
<th>Patient</th>
<th>Disease status</th>
<th>Number of lines of therapy</th>
<th>Treatment regimen prior to IFI</th>
<th>Antifungal prophylaxis prior to IFI</th>
<th>EORTC/MSG</th>
<th>Prolonged neutropenia</th>
<th>High dose steroids</th>
<th>Clinical presentation</th>
<th>Site of infection</th>
<th>Microbiology Results</th>
<th>Treatment</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Progression</td>
<td>6</td>
<td>DT PACE</td>
<td>Fluconazole</td>
<td>Proven</td>
<td>Yes</td>
<td>Yes</td>
<td>Febrile neutropenia</td>
<td>Sinus</td>
<td>Blood culture: Scedosporium prolificans</td>
<td>Liposomal amphotericin</td>
<td>Death from disseminated infection</td>
</tr>
<tr>
<td>2</td>
<td>Progression</td>
<td>5</td>
<td>Melphalan</td>
<td>Fluconazole</td>
<td>Proven</td>
<td>Yes</td>
<td>No</td>
<td>Febrile neutropenia following second ASCT for progressive disease</td>
<td>Not applicable</td>
<td>Blood culture: G. parapsilosis</td>
<td>Caspofungin followed by voriconazole</td>
<td>Survived past 30 days</td>
</tr>
<tr>
<td>3</td>
<td>Progression</td>
<td>5</td>
<td>Bortezomib and romidepsin</td>
<td>None</td>
<td>Proven</td>
<td>No</td>
<td>No</td>
<td>Persistent fevers, Respiratory failure from concurrent influenza A infection</td>
<td>Not applicable</td>
<td>Blood culture: G. ahlbicans</td>
<td>Caspofungin</td>
<td>Death from respiratory failure</td>
</tr>
<tr>
<td>4</td>
<td>Progression</td>
<td>3</td>
<td>DVPACE</td>
<td>Fluconazole</td>
<td>Possible</td>
<td>Yes</td>
<td>Yes</td>
<td>Persistent fever Respiratory symptoms</td>
<td>Chest</td>
<td>BAL: Scapularia sp.</td>
<td>Caspofungin and voriconazole</td>
<td>Death from respiratory failure</td>
</tr>
<tr>
<td>5</td>
<td>Progression</td>
<td>6</td>
<td>Melphalan</td>
<td>Fluconazole</td>
<td>Possible</td>
<td>Yes</td>
<td>No</td>
<td>Persistent fever Respiratory symptoms following third ASCT for progressive disease</td>
<td>Chest</td>
<td>BAL: Aspergillus fumigatus</td>
<td>Voriconazole followed by posaconazole</td>
<td>Survived past 30 days</td>
</tr>
<tr>
<td>6</td>
<td>Progression</td>
<td>5</td>
<td>Cyclophosphamide and bortezomib</td>
<td>None</td>
<td>Possible</td>
<td>Yes</td>
<td>No</td>
<td>Persistent febrile neutropenia</td>
<td>HRCT: nodules in upper lobe</td>
<td>BAL Culture negative</td>
<td>Voriconazole</td>
<td>Survived past 30 days</td>
</tr>
<tr>
<td>7</td>
<td>Induction</td>
<td>1</td>
<td>Lenalidomide and dexamethasone</td>
<td>None</td>
<td>Possible</td>
<td>Yes</td>
<td>Yes</td>
<td>Febrile neutropenia Respiratory symptoms</td>
<td>HRCT: multiple cavitatory nodules</td>
<td>BAL Culture negative; Aspergillus PCR positive</td>
<td>Voriconazole</td>
<td>Survived past 30 days</td>
</tr>
<tr>
<td>8</td>
<td>Progression</td>
<td>4</td>
<td>Melphalan</td>
<td>None</td>
<td>Possible</td>
<td>Yes</td>
<td>No</td>
<td>Febrile neutropenia following second ASCT for progressive disease</td>
<td>HRCT: Right middle lobe nodules</td>
<td>BAL Culture negative</td>
<td>Posaconazole</td>
<td>Survived past 30 days</td>
</tr>
<tr>
<td>9</td>
<td>Progression</td>
<td>2</td>
<td>Lenalidomide</td>
<td>None</td>
<td>Possible</td>
<td>Yes</td>
<td>No</td>
<td>Febrile with evolving skin lesions for investigation</td>
<td>HRCT: multiple scattered nodules</td>
<td>BAL Culture negative</td>
<td>Posaconazole followed by liposomal amphotericin</td>
<td>Death from disseminated bacterial infection</td>
</tr>
</tbody>
</table>

IFI: invasive fungal infection; EORTC: European organization for research and treatment of cancer; MSG: Mycology study group; ASCT: autologous hematopoietic stem cell transplantation; HRCT: High resolution computer tomography; BAL: broncho-alveolar lavage DT PACE: dexamethasone-thalidomide-cisplatin-doxorubicin-cyclophosphamide and etoposide; DVPACE: dexamethasone-bortezomib-cisplatin-doxorubicin-cyclophosphamide and etoposide
Table 3: Impact of multiple myeloma treatment factors on risk of developing invasive fungal infection

IFI: invasive fungal infection; OR: odds ratio; CI: confidence interval; AF: anti-fungal; ASCT: autologous hematopoietic stem cell transplant

*Covariates were included in the model if univariate analysis demonstrated p<0.20. All analyses were performed using Stata (version 9.0, StataCorp Inc., Texas, USA) and p ≤ 0.05 was deemed statistically significant.