Bronchoalveolar lavage fluid cytology by GMS stain for the diagnosis of invasive pulmonary aspergillosis in patients with hematologic malignancies: analysis of 67 episodes

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BACKGROUND

There is a paucity of studies on the yield of Gomori-methenamine-silver (GMS) stain in bronchoalveolar lavage (BAL) cytology and its comparison with fluorescent dyes for the diagnosis of invasive pulmonary aspergillosis (IPA) in patients with hematologic malignancies. To that end, we analysed the yield of direct fluorescent visualization in BAL cytology by GMS stain in a series of culture-positive IPA in patients with hematologic malignancies.

PATIENTS & METHODS

We retrospectively analysed all consecutive cases of culture–documented IPA (proven or probable according to the EORTC criteria) in adult patients with hematologic malignancy (September 1999-March 2015) at MD Anderson Cancer Center, a tertiary care cancer center in Houston, TX. All cases had concurrently available BAL cytology GMS. BAL was performed in the cytopathology laboratory by GMS was performed in the cytopathology laboratory by a pathologist (stained) and the Diff-Quik (Romanowsky stain).

GMS was performed in the cytopathology laboratory by centrifuging BAL samples using a high vacuum centrifuge. GMS stains were prepared using 5 - 6 ml of BAL per slide. BW samples were infrequently used for GMS staining. Cytopathology also routinely performed two modified Giemsa stains (Papanicolaou and the Diff-Quik (Romanowsky stain).

RESULTS

We identified 67 cases of IPA (proven in 2, probable in 65) in 66 patients.

- Most common underlying disease was AML. The majority of patients had active disease.
- 1/3 had a history of corticosteroid use, and recent severe neutropenia was present in 22%.
- Serum galactomannan levels were available in 30 cases and positive in 8.
- Most (78.7%) patients had CT chest imaging within a week of the BAL date.

Direct fungal visualization in BAL cytology based on GMS was positive in 41.8% cases, in contrast to only 3.6% positive direct smear Calcofluor White™ stain cases. Of note, one of the 2 cases with positive Calcofluor White™ stain had a negative GMS cytology.

BAL cytology was diagnostic for co-infections in 7 cases: 2 Pneumocystis jiroveci and 5 viral infections (cytopathic changes) (one had both).

BAL galactomannan was only available in 12 cases (17.6%), and was positive in 41.7. Of note, BAL cytology was positive in 1 case with negative BAL Aspergillus galactomannan, and Aspergillus galactomannan was positive in BAL in 5 cases with negative cytology.

Comparison between cases with positive and negative BAL cytology by GMS showed that cavitary lesions, history of SCT, and IPA caused by >1 Aspergillus species had more often a positive cytology. In addition, BAL GMS cytology was also more often positive when the positive culture sample was from a BAL aimed at the lesion compared to bilateral bronchial washing or when both cultures were positive.

In the multivariate analysis, only cavitary lesions were significantly associated with positive BAL cytology (p=0.045; OR 6.21, 95% CI 1.04-37.11).

There were no differences in the positivity rate of BAL GMS cytology according to Aspergillus species causing IPA or prior multidrug prophylaxis. No other significant associations were found between cytology and other variables.

CONCLUSIONS

GMS stain in (BAL) was positive in 42% of 67 cases and revealed co-infections in seven. In contrast, only 2/67 (3.6%) of BAL samples were positive by the direct smear fluorescent dye Calcofluor White™ stain.

Positive GMS was significantly more frequent in IPA with cavitary lesions and IPA caused by >1 Aspergillus species, but the proportion of positive cytology among Aspergillus species was not different.