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BACKGROUND

- Air quality is of utmost importance in a hospital setting to prevent infection by inhalation of fungal conidia, especially in units with immunocompromised or critically ill patients.¹
- Many outbreaks of nosocomial aspergillosis were linked to contaminated ventilation systems, including some in pediatric centers.²
- Air cultures, as opposed to particulate counts, are not routinely recommended to monitor air quality in hospitals.³
- There is no well established relationship between air particulate counts and the presence of fungal spores.^{4,5}

OBJECTIVES

- To evaluate the relationship between particulate counts and the amount of fungal spores in air samples.
- To determine if a particulate count threshold could help predict fungal aerocontamination.

MATERIALS AND METHODS

- Single-center retrospective study comparing particulate counts per ft³ with air fungal cultures per m³.
- Samples were obtained in HEPA-filtered patient rooms on selected wards : Oncology/Solid Organ Transplant (SOT), Intensive Care (PICU), Neonatology (NICU) and Hematology/Stem-Cell Transplant (HSCT), Infectious Disease (ID).
- Samples were obtained during daytime, in summer, fall and winter
- Particulate counts were obtained with MetOne HHP6 Handheld Airborne Particulate Counter (Hach Ultra Analytics, Grants Pass, USA (figure 1a)
- Air cultures were made using IDEAL microbiological air sampler (bioMérieux, Marcy l'Etoile, France), (figure 1b) on inhibitory mold agar incubated for five days.
- Statistical analysis included :
 - Pearson correlation coefficient between particulate counts and number of fungal CFU/m³
 - Logistic regression to assess odds of positive cultures at three pre-determined particulate count thresholds based on ISO 14644: 3 000, 10 000 and 20 000
 - ROC curve analysis to assess difference between culture-positive and culture-negative samples.



Figure 1a : MetOne HHP6 Handheld Airborne Particulate Counter



Figure 1b : IDEAL microbiological air sampler, Biomérieux

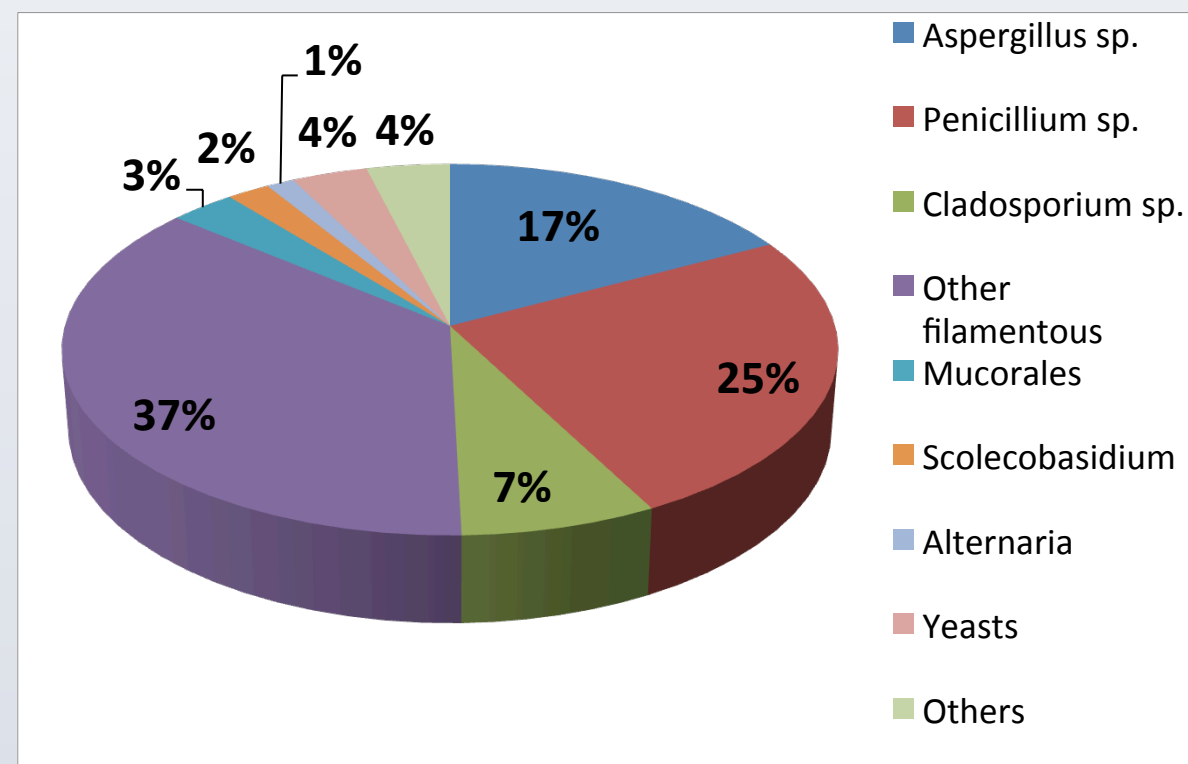
RESULTS

- Between March 2009 and August 2010, 254 air samples for particulate count and fungal culture were obtained.

Cultures

- 159 samples were positive for fungi cultures (62.6%)
- Among all isolated colonies, the most frequently identified organisms were *Penicillium sp.* (25%) and *Aspergillus sp.* (17%) (figure 2)
- *Aspergillus spp.* was isolated in 16.5% of all samples and, notably, from two rooms harboring less than 100 particulates per m³

Figure 2 : Proportions of isolated fungi on overall colonies (n=839)



Particulate counts

- Median was 3787, interquartile range was 1416 - 8672. Mean was 11 000 and min/max were 40 and 1 019 547, respectively.
- Particulate counts were higher in winter (median=7780) but fungal aerocontamination was higher in summer (average=4.1 CFU/m³) (table 1 & figure 3)

Figure 3 : Average fungal CFU and median particulate counts through time

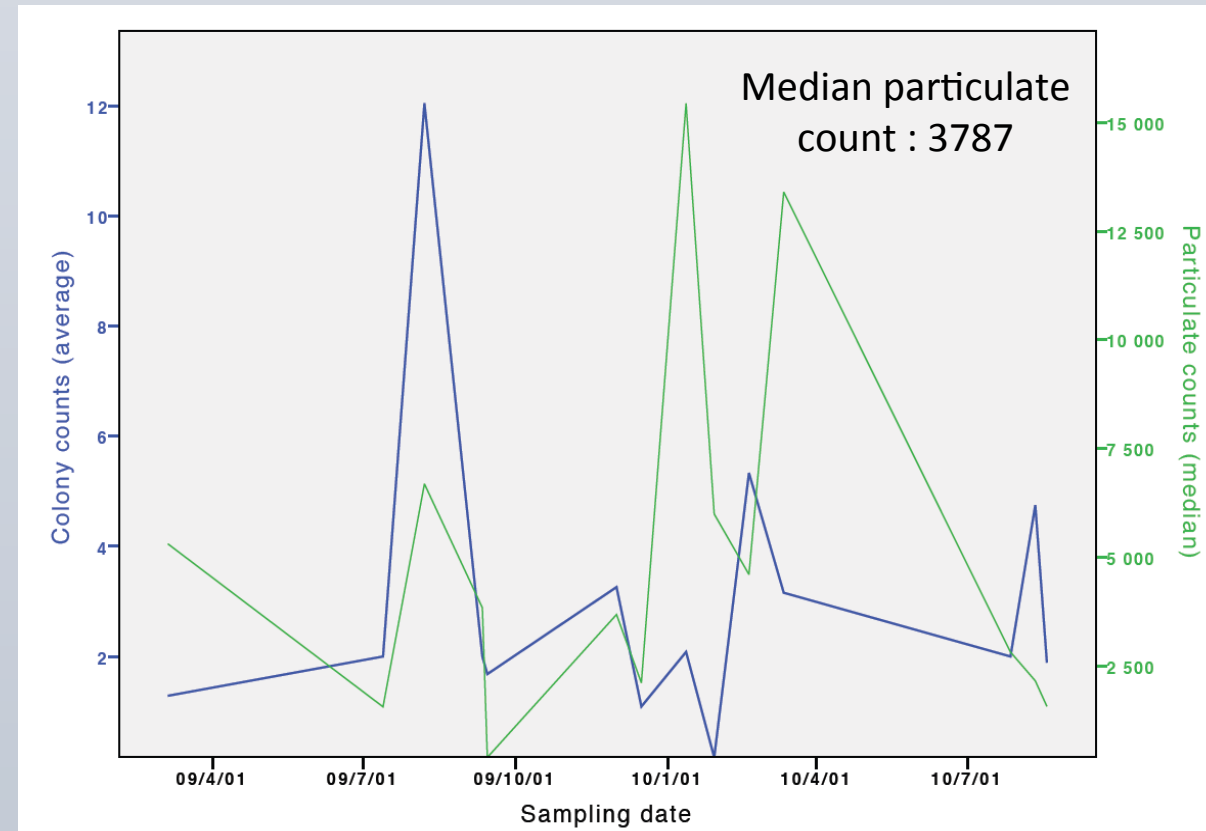


Table 1 : Seasonal distribution of positive cultures

Season	Particulate counts (median)	Average CFU/m ³	Positive cultures n (%)	<i>Aspergillus sp.</i> cultures n (%)	Total n
Winter	7 780	2.5	49 (52)	13 (14)	94
Summer	2 034	4.1	86 (75)	22 (19)	115
Fall	3 000	2.2	24 (53)	6 (13)	45

Table 2 : Ward distribution of positive cultures

Ward	Particulate counts (median)	Average CFU/m ³	Positive cultures n (%)	<i>Aspergillus sp.</i> cultures n (%)	Total n
HSCT	3 166	2.0	36 (54)	4 (6)	67
PICU	3 480	3.8	13 (76)	2 (12)	17
NICU	1 938	2.8	36 (57)	5 (8)	63
ID	13 807	5.5	37 (80)	17 (37)	46
SOT	2 951	3.0	37 (61)	13 (21)	61

- Both particulate counts and fungal CFU were higher in the ID ward (table 2), which uses negative-pressure ventilation.
- When restricting the analysis to positive-pressured rooms (n=208), median particulate counts dropped to 2698, but air fungal cultures remained positive 58.7% of the time.

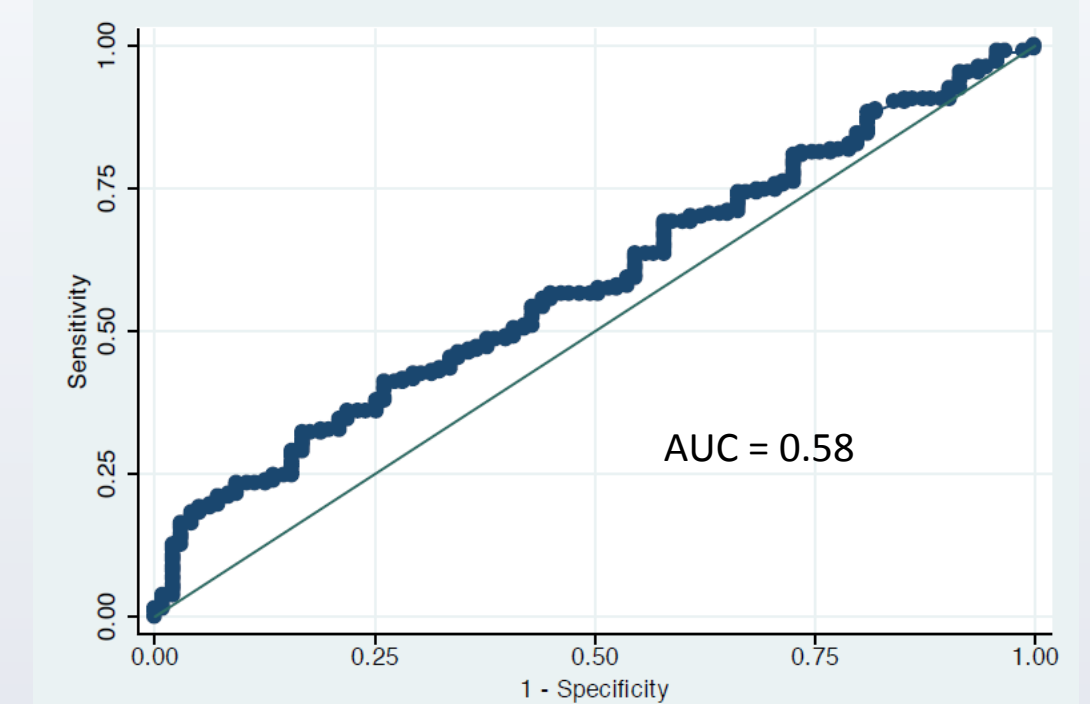
Correlation between particulate counts and positive cultures

- Pearson correlation coefficient between log₁₀ of particulate count and log₁₀ of fungal CFU/m³ was 0.235 (p<0.01) showing a poor correlation.
- Culture-negative samples had a mean log₁₀ particulate count of 3.4 vs 3.58 for positive cultures (p=0.03)
- By logistic regression, a one log increase in particulate count was associated with a significant increase in the odds for positive cultures (OR=1.55; 95% CI 1.03-2.35; p=0.035)
- A particulate count higher than 20 000 was significantly associated with a greater odd of positive culture (OR=6.7; 95% CI 1.5-29.3; p=0.012)
- However, 60% of samples with a particulate count lower than 20 000 and 60% of samples with a particulate count lower than 3 000 harbored positive cultures. (table 3)

Table 3 : Culture positivity using different particulate count thresholds

Particulate counts thresholds	Positive cultures/total (%)	<i>Aspergillus sp.</i> cultures n (%)	OR (95% CI) for positive culture	p value
< 3000	68/114 (60)	12 (11)	1.0	-
≥ 3000	91/140 (65)	29 (21)	1.3 (0.8-2.1)	0.381
≥ 10000	42/57 (74)	12 (21)	1.9 (1.0-3.7)	0.052
≥ 20000	20/22 (91)	7 (32)	6.7 (1.5-29.3)	0.012
Total	159/254 (63)	41 (16)		

Figure 4 : ROC analysis for particulate count thresholds



- No sensitive/specific threshold was identified by ROC curve analysis to predict positive cultures. (figure 4)

DISCUSSION

- In this study, we found a significant association between a particulate count over 20 000 and positive fungal air cultures.
- Many non-pathogenic fungi were identified on cultures, but they could themselves represent an indirect marker of aerocontamination.¹
- Higher particulate counts during winter might be explained by heating devices used in the hospital.
- Particulate count is highly variable even upon multiple testing under stable conditions. Thus, particulate count is not an accurate predictor of air fungal contamination.
- No clinical correlation was established during the surveillance period, and this would require further studies.

CONCLUSION

In this study, a particulate count above 20 000 was significantly associated with a positive air fungal culture. However, the majority of samples with lower particulate counts were also positive. Thus, particulate count does not accurately predict the presence of fungal conidia in air and cannot be used to rule out fungal aerocontamination.

REFERENCES

1. Alberti C, et al. J Hosp Infect 2001;48:198-206.
2. Anderson K, et al. Thorax 1996;51:256-61.
3. Sehulster L, Chinn RY, Cdc, Hicpac. MMWR Recommendations and reports : Morbidity and mortality weekly report Recommendations and reports / Centers for Disease Control 2003;52:1-42.
4. Overberger PA, et al. Am Ind Hyg Assoc J 1995;56:706-12.
5. Raisi LL, M.; Katsivela, E. Global NEST Journal 2010;12:84-91.

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