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SHORT REPORT

Pulmonary *Aspergillus* colonization in humans and its impact on management of critically ill patients

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Summary. Samples of lung tissues were obtained and analysed for *Aspergillus* carriage in 56 patients undergoing thoracic surgical intervention and 18 people who had an unexpected death. Out of 74 samples, 46 (63%) had evidence of pulmonary fungal colonization. The surgery population had a rate of 62% of fungal growth, *Aspergillus* was present in 39%. The autopsy population had a rate of 61% of fungal colonization, *Aspergillus* was present in 41%. In these cases eradication of fungal spores residing in the lung prior to aggressive chemotherapy and prevention of further spore uptake during hospitalization is indispensable in preventing pulmonary aspergillosis.

Keywords: *Aspergillus* species, fungal colonization, aspergillosis prevention, neutropenia, antifungal therapy.

Fungal pathogens are recognized as a major and increasing source of life-threatening infections in the immunocompromised host (Denning *et al*, 1991). Infections due to *Aspergillus* species are among the most common causes of nosocomial pneumonia and associated with an extremely high mortality rate of 85% (Pfaller, 1992). The use of high-efficiency particulate air (HEPA) filtration sytems during hospitalization represents the current standard in invasive pulmonary aspergillosis (IPA) prevention (Rhame, 1991), but cases of *Aspergillus* pneumonia in this setting still occur. Rhame *et al* (1984) reported the results of bi-weekly nasal and pharyngeal cultures of 205 BMT recipients; 11 patients developed invasive aspergillosis without positive cultures. Since in an earlier study in which *Aspergillus* spore counts in hospitals and private homes were compared we found equal and sometimes even higher amounts of spores in private settings (unpublished information), we wanted to clarify whether patients were already loaded with *Aspergillus* spores in their lungs prior to hospitalization. In order to investigate in more detail the fungal colonization of the lower respiratory tract as a possible source of endogenous spread, we examined lung tissues received from surgical intervention and autopsy.

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PATIENTS AND METHODS

Patients. 56 patients who underwent surgical intervention such as lobectomy or pneumonectomy due to malignant tumours and 18 people who had a sudden unexpected death and were undergoing examination to clarify the cause of death, were investigated.

The patients had had no previous chemotherapy, no immediate foregoing hospitalization and no other underlying disease of the pulmonary system. Two samples of resected material with no damage to lung architecture were examined from each patient.

The primarily peripheral lungs, size 1 cm^3, were taken under aseptic conditions to avoid contamination; all the instruments and gloves were changed before obtaining the blocks of tissue.

Cultures. The samples were directly transferred into Sabouraud 4% fluid containing chloramphenicol and gentamycin (Merck, 6100 Darmstadt, Germany) and incubated at 35°C for 5 d. The test media were subjected a visual control for growth. The fungal isolates were plated onto Saboraud glucose agar (Merck, 6100 Darmstadt, Germany), incubated at 35°C for 4 d and identified according to morphology and culture characteristics.

For a quality control, a growth promotion test and growth control test were performed routinely.

Environment. Random monitoring of the air for microbial

content was performed in the autopsy area during excision; the clean areas in the operating theatres were routinely checked.

Statistics. 95% confidence intervals based on binomial distribution were calculated. Differences in fungal occurrence were evaluated by sex, study population and age using the two-sided Fisher's exact test and the *t*-test.

Table I. Patients and pulmonary fungal carriage.

Study group	Patients ($n=74$)	Fungal growth ($n=46$)	No fungal growth ($n=28$)
Autopsy patients	18	11 (61·1%)	7 (38·9%)
Surgical patients	56	35 (62·5%)	21 (37·5%)

RESULTS

Evidence of fungi was detecteded in 46 cases (95% confidence interval 50·1–73·2%), with *Aspergillus* species present in 30 individuals (95% confidence interval 49·75–78·65%) as shown in Table I. Fungal growth was seen in 62·5% in the surgical intervention group and in 61·1% ($P=1$) in the autopsy population. The age range of the population with fungal colonization in their lungs had a mean of $57·3 \pm 17·2$, and those without fungal growth a mean of $59·67 \pm 17·7$ ($P=0·57$). The spectrum of fungi cultured shown in Table II.

Table II. Presence of fungi detected.

	No. of patients with fungal colonization	
Species	Autopsy patients ($n=7$)	Surgery patients ($n=23$)
A. fumigatus	6	17
A. flavus	2	7
A. niger	1	3
A. terreus	1	1
A. glaucus	0	1
Mucor spp.	2	7
Penicillium spp.	2	5
Candida spp.	1	0

Twenty-two patients showed fungal growth in both lung tissues, 10 patients had more than one species detected.

The study group included 28 men and 46 women. Fungal contamination was higher in women than in men, but this difference was not statistically significant ($P=0·30$). No variables such as sex, age or study group were significantly associated with a culture positive for *Aspergillus* in univariate analysis.

No fungal contamination in the environment was detected during excision of lung tissue.

DISCUSSION

Our findings on pulmonary *Aspergillus* carriage are probably of clinical relevance. We speculate that fungal colonization of the lungs prior to hospitalization and aggressive cancer chemotherapy in patients at risk to IPA represents a possible source of endogenous spread starting with iatrogenic neutropenia.

A high concentration of fungal spores in the human natural environment is probably responsible for colonization of the lungs in 62·2% of all individuals. Since those people in whom we detected colonization had no clinical signs of disease, this fungal evidence appears to be harmless. Why, out of 74 individuals, only 46 were colonized is not clear. It is possible that those subjects showing no colonization could make use of a more efficient innate or acquired immunity.

Although *Aspergillus* spp. is an ubiquitous organism and contamination due to this airborn organism is commonly observed, we do not think that any samples were positive due to contamination: firstly, because the environmental screening detecteded no fungal contamination at the time of lung excision and, secondly, because comparable numbers of fungal carriers were seen in both the surgical and autopsy populations.

For practical purposes, however, several strategies for the prevention of invasive pulmonary aspergillosis must be investigated in order to prevent further spore uptake during hospitalization and to eliminate spores already taken up. HEPA-filtered facilities are considered the current standard in prevention of further spore uptake (Sherertz, 1987). Nevertheless, since several authors have reported IPA despite treatment in settings with HEPA filters, attention must be given to eradicating spores already taken up by patients at risk. If patients who are predisposed to aspergillosis are treated in laminar airflow units before starting severe immunosuppression, then this might allow the host defence to eliminate spores present by preventing the further acquisition of new spores. The time needed for such spore elimination is presently unknown. The empirical use of topical and systemic prophylaxis with antifungal agents with varying degrees of success has been described (Kibbler *et al*, 1997). On testing *in vitro* susceptibility we previously found that some isolates of *Aspergillus* spp. were probably amphotericin B resistant (Lass-Flörl *et al*, 1998). Treatment of patients with established *Aspergillus* infections seems to be ineffective; attention must be given to primary preventive measures.

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