ORIGINAL ARTICLE

Microbiological surveillance of hospital ventilation systems in departments at high risk of nosocomial infections

P. CRIMI, F. ARGELLATI, G. MACRINA, C. TINTERI, L. COPELLO, D. REBORA, L. ROMANIA, R. RIZZETTO Department of Health Sciences, University of Genoa, Italy

Key words

Surveillance • Ventilation systems • Airborne infections

Summary

The air in hospital wards with patients at high risk (Surgeries, Intensive Care Units and Bone Marrow Transplant Centers) has been surveyed less than the one in Operating Rooms. Therefore in this study we considered useful to verify the microbic contamination of the air of those wards evaluating the consistency of ventilation systems in relation also to the presence and location of HEPA absolute filters. Seven departments of Genoese San Martino Hospital at high risk of infection were taken into account. In there, environmental investigations have been performed by air samplings and by analyzing bacterial and fungal growth on plates after an incubation period. Almost 60% of all samples taken in wards yielded a positive result and the average values of bacterial and aspergillar charges measured at air flow emission openings decisively exceed the ones considered standard in operating rooms. Still, the average values of airborne bacterial charges were significantly higher in those wards equipped with central filters (p < 0.001), while as far as the aspergillar charge is concerned, no statistically relevant dif-

Introduction

The health-care environment contains a diverse population of bacteria, but only a few are significant pathogens for susceptible humans; bacteria are present in great numbers in moist and organic environments, but can also persist in air, water and on fomites.

Nosocomial infections are above all due to health-care workers practices [1-3], but also the contamination of the environment could lead to a rise in health-care facilities [4-5].

In the past 20 years the incidence of nosocomial infections has increased due, among other causes, to a substantial raise in the number of immuno-compromised patients not only for the illness itself, but also for the particularly aggressive diagnostic-therapeutic treatments [6]. These patients are often gathered in hospital areas declared at "high risk" of infection such as Intensive Care Unit, High Risk Surgery, Hematology and Bone Marrow Transplant ward.

As far as healthcare workers procedures are concerned, numerous guidelines have been issued [7-9], and the infection incidence seems often related to the absence or unsound application of the rules. Far less evident is ferences were noticed. In wards with ventilation system, the bacterial charge value raises from the emission grids to the middle of the room and to the aspiration grids, while the ward not equipped with a ventilation system presents in the middle of the room an average bacterial charge 2 to 10 times higher than the one in other wards. The average values regarding bacterial and aspergillar charges resulted quite high in all the departments surveyed. Nevertheless, if we take into account ventilation systems equipped with absolute filters HEPA located centrally or peripherally, it can be outlined that the air quality from the point of view of both microbic and aspergillar contamination turns out to be decisively better in systems with peripheral filters. Moreover, a compared analysis of the three Hematology wards allows us to infer that the presence of artificial ventilation systems can lower the bacterial and fungal compared with a ward with natural ventilation.

the correlation between bacterial charge and Nosocomial Infections (N.I.) in hospital patients: this is particularly true for airborne bacterial charges. As a matter of fact, only patients who underwent endoprothetic surgeries seemed exposed to a certain risk of infection in operating rooms with laminar air flow [10]. However, in wards at high risk, a tight relation between the presence of aspergillar charge in the air and the incidence of aspergillar illness has been observed in Hematological patients, in the Marrow Bone transplanted, or in those who were somehow immuno-compromised [11]. Despite a lack of evidence-based correlation between air quality and N.I., in these and other high risk wards are nevertheless requested systems capable of keeping environmental and specifically air contamination at minimum. For this reason, high risk wards are equipped with mechanical ventilation systems with adequate air change, HEPA filters and, possibly, positive air pressure.

Therefore, in the study described in this paper, we decided to survey the microbiological contamination of the air of a number of departments of a big hospital in Genoa, where patients subjected to a higher risk of hospital infections are hospitalized; in particular we want-

.....

105

ed to evaluate the consistency of the ventilation systems in relation to the presence and location of HEPA absolute filters.

Methods

In this study, 7 departments of San Martino Hospital, all at high risk of infection were taken into account, namely: two departments of Hematology, the Bone Marrow Transplant Centre, the Emergency Intensive Care Unit, the First Anesthesia and Resuscitation Service (I SAR) which is divided in the east and west side, and the Neurosurgical Intensive Therapy ward. In all these areas environmental investigations have been performed, from May 2001 until the end of 2003 and more intensively during the year 2002.

Six departments of the seven analyzed were equipped with mechanical ventilation systems and all were examined in proximity of the grids of the ventilation openings.

As regards the Hematology area, in the two departments equipped with mechanical ventilation systems, air sampling was carried out not only at the air flow emission, but also in the middle of the room and near the aspiration opening; moreover, in the Hematology G, lacking of mechanical ventilation systems, the air sampling was taken only in the middle of the hospital rooms at an approximate height of 1.70 m from the floor.

The departments were also divided in two groups, namely Group 1 with peripheral HEPA filters (including the Hematology Dept. A and the Bone Marrow Transplant Centre B) and Group 2 with central or inside main air ducts HEPA filters (including the Intensive Care Unit of Emergency C, the 1st Anesthesia and Resuscitation Service on the west D and east side E and the Neurosurgical Intensive Therapy ward F), moreover, the other Hematology ward (G), in which ventilation is natural, was also taken into account.

SAMPLING

The air sampling was carried out by means of a Surface Air System (SAS) device, extending the aspiration for 5 minutes, in order to evaluate a total volume of 1000 liters of air [12].

The device was prepared by inserting a plate with a diameter of 55 mm containing growing PCA agar suitable for bacteria growth and a plate containing the same amount of Sabouraud agar, suitable for fungal growth; then the instrument would be placed in the very proximity of the grids of ventilation and aspiration openings, but not at direct contact with it to avoid a deposit of extraneous materials on the plate that could alter the results. Once the device was activated, the air flow would be automatically directed towards the plate surface, where a deposit of airborne particles with a diameter suitable to pass through the 219 holes of the head of the instrument would be created.

After the air flow exposing time (5') expired, the plate would be removed to be incubated as soon as possible

.....

to avoid altering the microbiological characteristics of the aspirated microorganisms.

ENVIRONMENTAL SAMPLING ANALYSIS

Both types of plates have been incubated into a thermostat at a constant temperature of 37 °C \pm 1 °C: the incubation time has been set to 3 days for the plates with growing PCA agar (aimed to bacteria growth), while the plates on Sabouraud agar (aimed to fungal growth) have been kept into a thermostat at a temperature of 30 °C \pm 1 °C for seven days.

Mycelium, conidia and spores morphologies have been observed in dry blucotton lactophenol (LPCB) using both low (100x) and high (430x) enlargements.

In order to obtain the Colony Forming Units per cubic meter of air (CFU/m³), the number of colonies grown on selective agar was related to the cubic meter of air taken.

Values of bacterial and aspergillar charge higher than 0 CFU/m³ were considered positive, not only in case of samples taken near emission and aspiration openings, but also in case of samples taken in the middle of the room [13].

Finally, the global results of 2002 (year in which the greatest number of samples was taken) were correlated to seasonal changes: for this reason the samples were divided in four quarters.

STATISTICAL ANALYSIS

The percentage of positive samples, data average, the highest values, observed in departments with peripheral and central filters, were compared by means of the t-Student test. In addition, the average frequency of positive samples in departments provided with, respectively, peripheral and central filters, was evaluated by means of the χ^2 test.

Results

In Table I, results concerning bacterial charge in environmental samples taken at the emission openings, in the middle room and at the aspiration openings are reported; table shows for the departments surveyed, the number of samples taken, the number and percentage of the positive ones, and the average, standard deviation and maximum values of CFU/m³ detected.

It can be noted that the percentage of positive readings at the emission openings is quite high, as 60% (147/224) of all samples taken in the six departments yielded a positive result. The actual value of airborne bacterial charge near air flow emissions is quite erratic not only within the single department (high values of standard deviation) but also between different departments. Even when we analyze the maximum value of bacterial charge it can be observed occasional increases. It can be also noted in Table I that in the two departments of Hematology area equipped with ventilation system, the bacterial charge raises from the air emission spot to the middle of the room. The raise is much higher in department A which has lower value at the emission. At the aspiration the values of bacterial charge were substantially unchanged with respect the ones recorded in the middle of the rooms.

In Table II, results regarding aspergillar charge measured near air flow emission openings, middle room and aspiration openings are reported, but in this case, average, standard deviation and maximum values are not shown whenever the positive samples detected were too few or missing at all. At the air flow emission, but also in the middle of the room and near the aspiration opening, the percentage of positive readings is decisively lower compared to the one related to bacterial charge as it is for average and maximum values. Moreover, we can observe that aspergillar charge pattern seems quite irregular even if the average values in the department with natural ventilation are decisively higher than the ones recorded in departments with a mechanical ventilation system.

In Table III the percentage of positive samples is

arranged separately between departments with central HEPA filters and departments with peripheral HEPA filters. It can be noted that if we analyze the whole set of samples of bacterial charges taken in departments equipped with central filters (136 in number) and the set of those taken in departments with peripheral filters (88 in numbers), the percentage of positive readings is quite higher in the former (83%) than in the latter (38.6%) ($\chi^2 = 9.778$, p = 0.002) the same can be said for the sets of samples of aspergillar charges taken in departments with central filters (4.4%) and with peripheral filters (1.2%), even though a statistical significance has not been achieved (p = n.s.). Moreover, it was found that the average values of bacterial charge introduced in departments equipped with central filters were significantly higher (58.2 CFU/m³) with respect the departments with peripheral filters (21.4 CFU/m³) (t-Test = 5.033, p < 0.001), while considering the aspergillar charge, no statistical significant variation can be inferred from samples data. On the other hand, we

Tab. I. Results concerning bacterial charge in environmental samples taken at the emission openings, in the middle room and at the aspiration openings.

Air sampling points	Dpt.	samples n.	positive s.		Bacteric charge		
				%	average CFU/m ³	s.d. CFU/m ³	max CFU/m ³
Emission							
Hematology	А	73	25	34,2	23,2	19,8	80
Bone Marrow Transplant Centre	В	57	40	70	55,1	83,9	510
Emergency Intensive Care Unit	С	15	9	60	17,7	11,2	45
I SAR West	D	25	24	96	75	54,7	190
I SAR East	Е	25	24	96	56	63,2	265
Neurosurgical Intensive Therapy	F	29	25	62	57,4	60,8	310
Middle-room							
Hematology	А	37	37	100	125	81	305
Bone Marrow Transplant Centre	В	11	11	100	26	13,5	50
Hematology	G	21	21	100	233,7	140	575
Aspiration							
Hematology	А	25	24	96	117,7	83,9	310
Bone Marrow Transplant Centre	В	35	32	91,4	97	141	675

 Tab. II. Results regarding aspergillar charge measured near air flow emission openings, middle room and aspiration openings.

Air sampling points	Dpt.	samples n.	positive s.	%	As average CFU/m ³	spergillar char s.d. CFU/m³	•
Emission							
Hematology	А	71	0	0	0	/	0
Bone Marrow Transplant Centre	В	57	3	5,3	10	8,66	20
Emergency Intensive Care Unit	С	15	1	6,66	5	/	5
I SAR West	D	25	1	4	5	/	5
I SAR East dpt.E		25	1	4	3	/	3
Neurosurgical Intensive Therapy Middle-room	F	29	1	3,44	10	/	10
Hematology	А	41	1	2,4	5	/	5
Bone Marrow Transplant Centre	В	11	0	0	0	/	0
Hematology	G	21	2	9,5	10	0	10
Aspiration							
Hematology	А	29	0	0	0	/	/
Bone Marrow Transplant Centre	В	35	6	17	15	9,4	20

Tab. III. Percentage of positive samples, average and maximum values between departments with central HEPA filters and departments with peripheral HEPA filters.

	Dpt. with peripheral filters	Dpt. with central filters	Statistical analysis		р
	Bacteric	charge			
% positive samples	38,6	83,1	χ²-test		n.s.
average CFU/m ³	21,4	58,2	t-test	5,033	< 0,001
max CFU/m ³	80	510	t-test		n.s.
	Aspergilla	ır charge			
% positive samples	1,2	4,4	χ²-test		n.s.
average CFU/m ³	1,3	8,4	Pt-test		n.s.
max CFU/m ³	5	20	t-test		n.s.

must emphasize that in one of the two wards equipped with peripheral filters (dept. A), no positive sample to aspergillar charge was ever recorded.

Finally from the analysis of the samplings taken by the air flow emission in 2002 it can be outlined that both for bacterial (B) and aspergillar (A) charges the values raise from the first quarter (winter: B = 47% A = 0%), to the second and third (spring and summer: B = 93% A = 13%) and then decline in the fourth quarter (autumn: B = 65% A = 7%).

Discussion

The air composition inside nosocomial closed environments has been subjected to a number of studies, partly dedicated to micro-biological aspects [14-18]. Airborne microbes and fungi, in subjects already weakened by constitutional or acquired immune-depression, can be responsible of severe infections against the respiratory district [19 20], or of systemic infections [21]. Among patients who are the most exposed to such risk, we must consider prioritarly those who undergo invasive and weakening diagnostic and therapeutic methodology, such as patients affected by hematological illnesses, in particular those subjected to a marrow bone transplant, or the ones that are cured in Intensive Care Units.

In those last years many environmental infection-control guidelines [7-9], were developed to review and reaffirm strategies for the prevention of environmentally-mediated infections, particularly about the procedures among health-care workers and immunocompromised patients: all personnel must follow behavioural rules codified by appropriate operational protocols (filter zone, sterilized clothing, overshoes, masks) [22]. Besides, infection-control strategies involved structural elements of health-care facilities [23], specifying the areas that require special environment such as the rooms used by high-risk, immunocompromised patients [24]. Those areas must fulfill particular environmental requirements such as low concentration of airborne particles, the proper ventilation standards for specialized care environments and the right conditions of temperature, humidity, air pressure and filtering (HEPA filters)

[25], although the exact configurations and specifications might differ among hospitals.

For those reasons, we believe useful, in our work, determining the values of airborne environment contamination in some of the department "at high risk" inside the main hospital of our region.

From the observation of the results regarding bacterial and aspergillar charges at the air-flow emission openings it can be outlined that in more than a half of the samplings there are indeed Colony Forming Units (CFU), and that sometimes maximum values reach levels decisively high.

This induces to think, that even if patients hospitalized in those departments may face a severe risk, the attention turned to this problem is less significant than in operating rooms.

If we take into account artificial ventilation systems equipped with absolute filters HEPA located respectively centrally or peripherally (at the openings of ducts), we can highlight that the air quality from the point of view of microbic and aspergillar contamination turns out to be, with certainty, better in systems with peripheral filters.

This can happen because of a chance that a system equipped with central filters may suffer, as time goes by, from a contamination by different kinds of particles inside the duct after the filter that, eventually, develops in an unfiltered bacterial growth.

Peripheral filters turn out to be particularly efficient also in regards of airborne aspergillar charge as outlined in the Hematology department A where all the readings taken at the air flow emission are negative.

From the compared analysis of the three departments of Hematology we can gather that bacterial charge in the middle of rooms where patients are indeed located, can be anyhow optimized, even if the air introduced is not so good, wherever procedures to limit air contamination are strictly followed by doctors, nurses and visitors as in the Bone Marrow Transplant Centre. On the other hand, it seems to be self-evident that a presence of an artificial ventilation system (in Hematology A and the Bone Marrow Transplant Centre) can lower the airborne bacterial charge as compared to a department with natural ventilation (Hematology G). As far as the aspergillar charge is concerned, it can be safely hypothesize that sources of contamination are indeed present in all three departments, as it is for all hospital departments, still, yet in this case, only the presence of ventilation systems can lead to charge values in the middle of the room decisively more limited. Talking about the influence of seasons in airborne bacterial and fungal growth, it can be noted how hot seasons favor a raise in both, since the high temperature and raise in humidity possibly cause a higher proliferation of microbic and micotic species squandered in the air.

.....

In conclusion, our study drives us to underline the usefulness of air samplings, not only in operating rooms

References

- Bennett JV, Brachman PS. *The inanimate environment*. In: Rhame FS, Eds. *Hospital Infections*. Philadelphia: Lippincott-Raven 1998, pp. 299-324.
- [2] Matsumoto T. The role of health care workers in nosocomial infection transmission. Nippon Rinsho 2002;60:2103-6.
- [3] Nogueras M, Marinsalta N, Roussell M, Notario R. Importance of hand germ contamination in health-care workers as possible carriers of nosocomial infections. Rev Inst Med Trop Sao Paulo 2001;43:149-52.
- [4] Greene VW. *Microbiological contamination control in hospitals*. Hospitals JAHA 1969;43:78-88.
- [5] Weber DJ, Rutala WA. Environmental issues and nosocomial infections. In: Wenzel RP, ed. Prevention and control of nosocomial infections. 3rd Eds. Baltimore: Williams & Wilkins 1997, pp. 491-514.
- [6] Schierholz JM, Beuth J. Implant infections: a haven for opportunistic bacteria. J Hosp Infect 2001;49:87-93.
- [7] Garner JS. Guideline for isolation precautions in hospitals. Part I. Evolution of isolation practices. Am J Infect Control 1996;24:24-31.
- [8] Garner JS, Favero MS. Guideline for handwashing and hospital environmental control. Centers for Disease Control. Atlanta: Department of Health and Human Services, Public Health Service 1985.
- [9] Larson EL. APIC guideline for handwashing and hand antisepsis in health care settings. Am J Infect Control 1995;23:251-69.
- [10] Dharan S, Pittet D. Environmental controls in operating theaters. J Hosp Infect 2002;51:79-84.
- [11] Walsh TJ, Dixon DM. Nosocomial aspergillosis: environmental microbiology, hospital epidemiology, diagnosis and treatment. Eur J Epidemiol 1989;5:131-42.
- [12] AA.VV. SAS-SUPER 100. Milan: International p.b.i. S.p.a. 1999.
- [13] NHS Estates. Ventilation in Healthcare Premises. In: London HMSO, Health Tecnical Memorandum 2025. 1994.
- [14] Andersen BM, Roed RT, Solheim N. Air quality and microbio-
- Received on January 12, 2006. Accepted on April, 4, 2006.
- Acknowledgements: the Authors are grateful to Dr. Giorgio Calochira for assistance with English translation.
- Correspondence: Prof. Paolo Crimi, Department of Health Sciences, via Pastore 1, 16132 Genoa, Italy - Tel. +39 010 5553583 - Fax +39 010 5556684 - E-mail: paolo.crimi@hsanmartino.liguria.it

but also in stay departments, where patients immunedepressed, and therefore at risk of nosocomial infections are hospitalized. As a matter of fact, often departments of such sort are not equipped with properly designed artificial ventilation systems and are not properly surveyed.

Moreover, from our studies, it turns out that the location of absolute filters HEPA directly in the vicinity of the emission openings is important as well, to obtain a total filtering prior to air emission into stay rooms.

Besides, the seasoning of airborne contamination suggests us to raise our surveillance during spring and summer time.

logic contamination in operating theatres. Tidsskr Nor Laegeforen 1998;118:3148-51.

- [15] Kodama AM, Mc Gee RI. Airborne microbial contaminants in indoor environment. Naturally ventilated and airconditioned homes. Arch Environ Hlth 1986;41:306-11.
- [16] Petrova NA, Kliasova GA, Funygina LP. Prevalence of mycelial fungi in hematological hospital. Ter Arkh 2003;75:58-63.
- [17] Rainer J, Peintner U, Poder R. Biodiversity and concentration of airborne fungi in a hospital environment. Mycopathologia 2001;149:87-97.
- [18] Sessa R, Di PM, Schiavoni G. *Microbiological indoor air quality in healthy buildings*. New Microbiol 2002;25:51-6.
- [19] Kistemann T, Huneburg H, Exner M, Vacata V, Engelhart S. Role of increased environmental Aspergillus exposure for patients with chronic obstructive pulmonary disease (COPD) treated with corticosteroids in an intensive care unit. Int J Hyg Environ Health 2002;204:347-51.
- [20] Pannuti CS, Gingrich RD, Pfaller MA, Wenzel RP. Nosocomial pneumonia in adult patients undergoing bone marrow transplantation: a 9-year study. J Clin Oncol 1991;9:77-84.
- [21] Dimopoulos G, Piagnerelli M, Berre J, Eddafali B, Salmon I, Vincent JL. Disseminated aspergillosis in intensive care unit patients: an autopsy study. J Chemother 2003;15:71-5.
- [22] Garner JS. Guideline for isolation precautions in hospitals. Infect Control Hosp Epidemiol 1996;17:53-80.
- [23] American Institute of Architects. Guidelines for design and construction of hospital and health care facilities. Washington, DC: American Institute of Architects Press 2001.
- [24] Hansen W. The need for an integrated indoor air quality program. In: Hansen W, ed. A guide to managing indoor air quality in health care organizations. Oakbrook Terrace: Joint Commission on Accreditation of Healthcare Organizations Publications 1997, pp. 13-18.
- [25] Hahn T, Cummings KM, Michalek AM, Lipman BJ, Segal BH, McCarthy PL. Efficacy of high-efficiency particulate air filtration in preventing aspergillosis in immunocompromised patients with hematologic malignancies. Infect Control Hosp Epidemiol 2002;23:525-31.

.....