Microbial Analysis of Air in a Public Hospital in the City of Fez, Morocco

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ABSTRACT
The hospital environment generally comprises, water, air, and surfaces... which are often contaminated with microorganisms of human and environmental origins. These germs constitute microbial reservoirs that may present a risk of infection for hospitalized patients. In this light we studied the microbiological quality of the air in a public hospital in the city of Fez, in order to prevent nosocomial infections. A prospective study over a period of 4 months was conducted at eight services. The samples were prepared with the sedimentation technique, by exposing the Petri dishes containing nutrient agar, in an area of 1 m² for 15 minutes. The dishes were incubated at 37 ± 1 °C for 48 h. Isolates were identified by conventional biochemical gallery and bioMérieux API gallery. A total of 32 samples were analyzed. Microbial identification showed the presence of 14 bacterial types, with the dominance of coagulase negative Staphylococcus (48%), and followed by Bacillus sp (14%), Staphylococcus aureus and Aeromonas salmonicida (8%) each, Gram negative cocci (7%) and Pseudomonas vesicularis (5%). Other bacteria such as Lactobacillus, Pseudomonas putrificiens, Streptococcus, Aeromonas hydrophila, Serratia liquefaciens, putrificiens Serratia, Serratia rubidaea and Stenotromonas maltophilia were isolated with low proportions. All studied hospital blocks showed a diverse microbial load: trauma (20%), surgery (17%), intensive care (16%), central operating room (14%), neonatology (13%), kitchen (10 %), cardiovascular gastrology (7%) and emergency (4%). These results demonstrated the need to implement systematic measures of hospital surveillance a comprehensive policy for the prevention of nosocomial infections including a systematic air treatment protocol.

Keywords: Hospital, Air Environment, Microbiological analysis, Nosocomial Infections, Fez, Morocco.

INTRODUCTION
The air quality in health facilities has become a matter of increasing concern. It requires special attention to protect patients against nosocomial infections caused by microorganisms. The microorganisms in the air are carried by supports of variable sizes, dust, skin flakes, and saliva droplets or micro-droplets. The later are emitted during talking and remain suspended and can diffuse and penetrate by inhalation to the alveoli of patients. In addition, the hospital can be considered a dynamic environment influenced by seasons, weather, ventilation systems, and moisture intrusion. The presence of visitors, human activities and cleaning in hospitals can also affect the air quality. These factors may be associated with microbial growth conditions, which may cause serious infections. Although the contamination of the air varies qualitatively and quantitatively in time, from one institution to another and within the same institution depending on hospital units, patient and the nature of care and practiced techniques. The air control in health facilities is essential. In fact, it is important to know the types of bacteria present in suspension in an air sample to assess the initial situation and the effectiveness of corrective measures. In addition, microbiological monitoring of the air is a subject of actuality for the prevention of nosocomial infections especially with the media coverage related to recent outbreaks of infections involving air born bacteria.
The aim of this study was to characterize microbial contamination levels in the air of eight units of a provincial public hospital in the city of Fez, Morocco as well as to identify airborne bacteria that may be associated with nosocomial infections for the purpose of improving patients care.

MATERIALS AND METHODS

Type, time and place of study
A prospective study was conducted at the Regional Diagnostic Laboratory of Epidemiological and Environmental Health in the city of Fez, Morocco over a 4-month period spanning from March to June, 2014.

Sampling sites
The samples were collected in eight units in a public hospital in the city of Fez, having a capacity of 172 beds. The units concerned are: neonatology, traumatology, emergency unit, operating room, intensive care, surgery, cardio-gastrology, and the hospital kitchen. The selection of sampling locations was based on the most critical and the most representative sites in each hospital unit.

Sampling techniques
The samples were prepared using the qualitative sedimentation technique based on the exposure of 90 mm diameter petri dishes containing nutrient agar, in an area of 1 m² for 15 minutes. The samples were quickly transported to the laboratory in a 4°C cooler for further treatments.

Samples analysis
In the laboratory, the petri dishes previously exposed to the air in the eight units were incubated at 37 ± 1°C. The results’ reading was done after 48h of incubation. The colonies count, appearance, size and color were scored and the purification of different types of colonies was performed by exhaustion on Plate Count Agar (PCA) medium.

Purified strains were subject to identification by both Gram stain and by classical biochemical strips and BioMérieux API strips.

Data analysis
For data processing and achieving the figures, we have used the Excel 2007 version software.

RESULTS
Air microbial analysis was done for a total number of 32
sites; 31 of which (96.88%) showed positive counts.

A comparison of the number and types of colonies found in each unit showed a level of contamination in the trauma unit (20%) and in the surgery department (17%). The other unit showed 16%, 14%, 13%, 10%, 7% and 4%, respectively for the intensive care, central operating room, neonatal, cooking, cardiovascular gastrology and emergency units (Figure 1).

The frequency of the bacterial species isolated from the studied hospital units showed a predominance of coagulase-negative *Staphylococcus* (48%), followed by *Bacillus* sp (14%), *Staphylococcus aureus* (8%), *Aeromonas salmonicida* (8%), of Gram negative cocci (7%) and *Pseudomonas vesicularis* (5%). Others found germs were found in a small proportions included *Lactobacillus* sp, *Serratia rubidaea* and *Stenotrophomonas maltophilia* (1% each) (Figure 2).

Coagulase-negative *Staphylococcus* were the most frequent in all units (100% at the emergency unit), followed by *Bacillus* sp which were found at five units: trauma (29%), gastroenterology and cardiovascular (22.22%), kitchen (12%), neonatology (9%), and surgery (6.67%). *Aeromonas salmonicida* was present in four units: cardiopulmonary-gastrology (22.22%), trauma (18%), kitchen (13%) and neonatology (9%).

*Staphylococcus aureus* was isolated with a frequency of 38% in the kitchen, 28% in neonatology, and 6% in the trauma unit. Lactobacilli were encountered at the neonatology (9%) and trauma (6%) units. *Aeromonas hydrophila* and *liquifaciens Serratia* were isolated in the kitchen with a frequency of 12% each. *Serratia rubidaea* was encountered only at the trauma unit (6%) while *Serratia putrificiens* was isolated at the cardio-gastrology unit (11.11%). *Pseudomonas vesicularis* was present at the intensive care (21.42%) and the surgery unit (6.67%). *Pseudomonas putrificiens* was isolated in the central surgery unit (16.67%). *Stenotrophomonas maltophilia* was isolated at the intensive care (7.14%).

Other bacteria such as gram-negative cocci were isolated from the air in the following units: cardiovascular gastrology, central surgery, intensive care and surgery with the respective frequencies of 33.34%, 8.33%, 7.14 % and 6.67%.

**DISCUSSION**

Exposure to certain pathogenic microorganisms in hospitals is associated with an increased risk of nosocomial infections. Such infections constitute a major concern for public health because of the increased length of stay of patients and the cost of hospital care they may cause. Microbiological monitoring of the environment in healthcare facilities allows highlighting the effect of saprophytic bacteria in nosocomial infections despite of the cleaning protocols and the ventilation systems in place. The air in the hospital is a ubiquitous contamination vector. It disperses particles carrying different microorganisms that can be inhaled causing
infections such as respiratory and nosocomial preoperative infections in vulnerable patients. The microbiological air monitoring in hospitals is essential because it remains a major priority to address the risk of infection, and allows the identification of pathogens as well as the detection of risk areas in the hospital.

In our study, we noticed that bacterial germs are present indifferent sites analyzed within the studied unit. 100% of the tests were positive for the neonatal unit, traumatology, emergency block, cooking, surgery, gastro-enterology, cardiovascular and intensive care. 83.33% of analyzed sites for the operating room present bacterial germs. Ortu S. et al., 2005 reported that the presence of certain pathogenic microorganisms in the air of the hospital, especially in the operating room could be the cause of severe postoperative infections. The bacterial load found in the air of these units could be the cause of the increased risk of infection in hospitalized patients. Bacteria identification showed the presence of 14 bacterial genera among which Staphylococcus genus was the most predominant. From these results we noticed that hospital air is largely colonized by bacteria belonging to pathogenic species. Our result is consistent with a study conducted in Portugal in 2015 in which Cabo Verde S. et al., 2015 also observed high bacterial load in hospital units’ air system. The presence of these types of germs in hospitals is generally linked to several types of infections. Several examples are described in the literature by different authors. This includes Aeromonas that are responsible for gastroenteritis and wound infections. Staphylococcus aureus which is involved mainly in lung, bone, heart and blood infections. Bacteremia is mainly due to infections by Bacillus sp. The presence of Lactobacillus and Serratia rubidaea in the hospital environment can cause urinary tract infections, respiratory and catheter infections. Stenotrophomonas maltophilia can represent a frequent nosocomial infection risk in immunocompromised patients. On the other hand, the literature reports that airborne microorganisms can come not only from humans (including patients), but can also be caused by various hospital features and environmental sources. According to data and information obtained in this study, as well as other earlier studies, the implementation of a comprehensive policy for the prevention of nosocomial infections especially by establishing a systematic air treatment protocol is necessary. Regular monitoring is essential to assess the effectiveness of the control of air and to detect irregular introduction of airborne particles via patients, visitors and/or medical staff. Moreover, microbiological survey data should be used to clearly define specific guidelines for air quality especially in controlled environments in hospitals. The compliance with good hygiene practices can however also reduce the number of nosocomial infections and their growing resistance to antibiotics remaining always a global concern.

CONCLUSION
Mastering the environment quality in health care facilities is essential to protect both patients and staff. Microbiological analysis of air in eight hospital units in this study showed a significant colonization of air by bacterial species of human and environmental origin that have the potential to infect hospital patients. We identified 14 bacterial types with the predominance of coagulase-negative Staphylococci (48%), followed by Bacillus sp (14%).

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Conflict of interests
The authors declare that they have no conflict of interests.

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