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Prevalence of Potential Nosocomial Pathogens Isolated from Environments of Regional Hospital of Korca, Albania

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Abstract

Background: Nosocomial infections make one of the most important issues regarding the surity of the patients at the healthcare institutions. Every hospitalized patient may acquire one nosocomial infection. The microorganisms that cause these infections may be bacteria, viruses and pathogen fungus. Nosocomial infections are the 5th causes of death in hospitals. A third of nosocomial infections are preventable.

Objective: The main purpose of this study was the identification of prevalence of potential nosocomial pathogens isolated from environments of Regional Hospital of Korca, Albania.

Material and methods: A total of 393 bacteria were isolated and identified from the hospital. The microbial identification was done with microscopy after Gram staining, colonies morphology and biochemistry.

Results: The study revealed that the prevalence of microrganisms isolated was as following:

Staphylococcus aureus 237 (60.3%) of isolates, *E. coli* 124 (31.6%), *Klebsiella* spp. 1 (0.3%), *Pseudomonas* spp. 13 (3.3%), *Proteus* spp. 1 (0.3%), *Staphylococcus epidermidis* 4 (1.0%), Saprophytes 13 (3.3%).

Conclusion: *Staphylococcus aureus* was the most prevalent pathogen isolated in the hospitals while *E. coli* was the most frequent gram negative bacteria isolated.

Keywords Nosocomial infection; hospital environment; *Staphylococcus aureus*; *E. coli*

Background

Nosocomial infections make one of the most important issues regarding the surity of the patients at the healthcare institutions. Every hospitalized patient may acquire one nosocomial infection. Hospital-acquired infections are a major challenge to patient safety. It is estimated that in 2002, a total of 1.7 million hospital-acquired infections occurred (4.5 per 100 admissions), and almost 99,000 deaths resulted from or were associated with a hospital-acquired infection [1], making hospital-acquired infections the sixth leading cause of death in the United States [2]; similar data have been reported from Europe [3]. The estimated costs to the U.S health care budget are $5 billion to $10 billion annually [4]. Approximately one third or more of hospital-acquired infections are preventable [5].

Hospital-acquired infections are the sixth leading cause of death in the United States and similar data have been reported from Europe [6], being an increasing problem in intensive care units (ICU), where the patients are more susceptible to colonization and the organisms are often more resistant to antimicrobial agents than in other environments. Since the 80's, infectious disease specialists have recognized that ICU patients acquire nosocomial infections at a much higher rate than patients elsewhere in the hospital [7]. In these confined areas, lower respiratory tract and bloodstream infections happen to be the most lethal; however, urinary tract infections are the most common. Infections caused by gram-negative bacteria have features that are of particular concern since these organisms are highly efficient at up-regulating or acquiring genes that code for mechanisms of antibiotic drug resistance, especially in the presence of antibiotic selection pressure. Most common nosocomial pathogens may well survive or persist on surfaces for a long time, what could explain their survival in ICU wards [8]. Gram-positive bacteria, such as *Enterococcus* spp. (including VRE), *Staphylococcus aureus* (including MRSA), *Clostridium difficile* or *Streptococcus pyogenes*, and many gram-negative bacteria, such as *Acinetobacter* spp., *Klebsiella* spp., *Pseudomonas aeruginosa*, *Serratia marcescens* or *Shigella* spp., may indeed survive for months on dry surfaces. A few others, such as *Bordetella pertussis*, *Haemophilus influenzae*, *Proteus vulgaris* or *Vibrio cholerae*, however, persist only for days [9,10].

Objective

The main purpose of this study was the identification of prevalence of potential nosocomial pathogens isolated from environments of Regional Hospital of Korca, Albania.

Materials and Methods

393 microbial isolates were obtained from various samples in the wards of the Regional Hospital of Korca. The items from which the samples were collected included sterilized materials, laundries, healthcare workers, surfaces, air and the systems of aspiration-

intubation-oxygen. A swab soaked in nutrient broth was used to collect samples from the surfaces, laundries, health care workers and systems of aspiration-intubation-oxygen. All the samples were labeled properly and transported immediately to the microbiology laboratory of Korca for processing. The swab samples were inoculated in Blood agar, Sabouraud agar and incubated at 37°C for 24 h.

Air sampling was performed with settle plates methods. Petri dishes containing Blood and Sabouraud agar were placed at chosen places in the hospital environments at about 1 m above the ground and exposed for 15 min. After this exposure, the plates were covered with their lids and taken to laboratory in sealed plastic bags and incubated at 37°C for 24 h.

Identification of bacteria

After incubation bacterial isolates were identified according to the morphological appearance, cultural characteristics and biochemical reactions.

Bacterial colonies were viewed under a microscope after Gram staining. The procedure was as following: A slide of cell sample was made to be stained. Heat fixed the sample to the slide. Crystal violet was added to the slide and incubated for 1 min. The slide was rinsed with a gentle stream of water for a maximum of 5 s. Gram's iodine was added for 1 min, than the slide was rinsed again with water. The slide was rinsed with alcohol for ~3 s and with water. The sondary stain, safranin, was added to the slide and incubated for 1 min. The slide was rinsed with gentle stream of water for a maximum of 5 s.

Gram positive bacteria retained the prime stain (Crystal violet) and appeared violet under the microscope. Gram negative, lost the primary stain and took the sondary stain, causing it to appear red when viewed under a microscope.

Biochemical reactions were carried out in the isolates as follows

Catalase test

2-3 ml of hydrogen peroxide was purred in a test tube. A colony of test organism was taken with sterile wooden or glass rod and immersed it into hydrogen peroxide solution. Generation of bubbles indicated oxygen production. If bubbles were produced, the organism was catalase positive, if bubbles were not produced, the organism was catalase negative.

Oxidase test

A piece of filter paper was pleased in petri dish and 3 drops of freshly prepared oxidase reagent were added. Using a sterile glass rod, a colony of test organisms was removed from a culture plate and smeared on the filter paper.

Oxidase positive organisms gave blue color within 5-10 s, and in oxidase negative organisms, color did not change.

Citrate test

A bacterial colony was inoculated in simmons citrate agar and incubated at 35°C to 37°C for 18 to 24 h. Than development of blue color was observed.

Citrate positive

Growth was visible on the slant surface and the medium became an intense Prussian blue.

Citrate negative

Trace or no growth was visible and no color change occurred.

Indole test

Test bacterial colony were inoculated in tryptophan broth and incubated at 37°C for 24-28 h. Then 0.5 ml of Kovac's reagent was added.

Positive test

Pink colored rink was observed after addition of reagent. Negative test: no color change after reagent addition.

Results

The study revealed that the prevalence of microorganisms isolated was as following: *Staphylococcus aureus* 237 (60.3%) of isolates, *E. coli* 124 (31.6%), *Klebsiella* spp. 1 (0.3%), *Pseudomonas* spp. 13 (3.3%), *Proteus* spp. 1 (0.3%), *Staphylococcus epidermidis* 4 (1.0%), Saprophytes 13 (3.3%) (Figure 1).

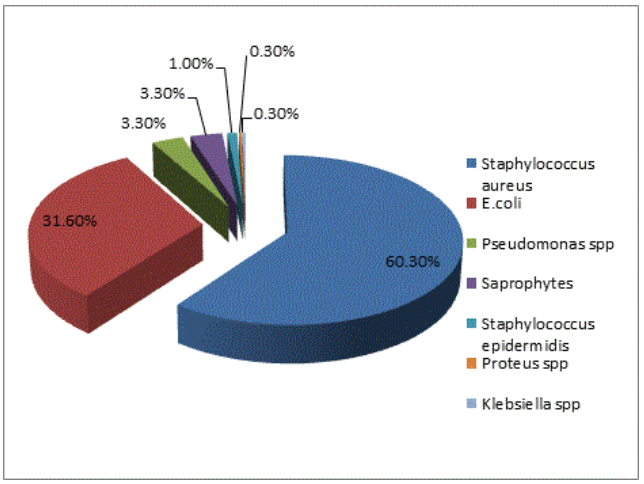

Figure 1: Prevalence of isolates.

The distribution of *Staphylococcus aureus isolates* was as following:

8% of isolates were obtained from the sterilized materials, 14.8% from the laundries, 35.9% from the surfaces, 28.3% from healthcare workers, 2.1% from air and 11.0% from the systems of aspiration-intubation-oxygen samples.

The distribution of *E. coli* isolates was as following:

8.1% of isolates were obtained from the sterilized materials, 16.1% from the laundries, 60.5% from the surfaces, 4.8% from healthcare workers, 2.4% from air and 8.1% from the systems of aspiration-intubation-oxygen samples.

The distribution of *Pseudomonas* spp. isolates was as following:

7.7% of isolates were obtained from the sterilized materials, 15.4% from the surfaces and 76.9% from the systems of aspiration-intubation-oxygen samples.

The distribution of Saprophytes isolates was as following:

7.7% from the laundries and 92.3% from the surfaces samples.

Staphylococcus epidermidis, *Proteus* spp. and *Klebsiella* spp. were isolated only from the healthcare workers samples (Table 1 and Figure 2).

Sample	Microorganisms							
	S. aureus n (%)	*E. coli* n (%)	*Klebsiella* sp. n (%)	*Pseudomonas* sp. n (%)	*Proteus* sp. n (%)	*St.epidermidis* n (%)	Saprophytes n (%)	Total n (%)
Sterilized materials	19 (8.0)	10 (8.1)		1 (7.7)				30 (7.6)
Laundries	35 (14.8)	20 (16.1)					1 (7.7)	56 (14.2)
Healthcare workers	67 (28.3)	6 (4.8)	1 (100.0)		1 (100.0)	4 (100.0)		79 (20.1)
Surfaces	85 (35.9)	75 (60.5)		2 (15.4)			12 (92.3)	174 (44.3)
Air	5 (2.1)	3 (2.4)						8 (2.0)
Systems of aspiration-intubation-oxygen	26 (11.0)	10 (8.1)		10 (76.9)				46 (11.7)
Total n	237	124	1	13	1	4	13	393

Table 1: Distribution of microorganisms.

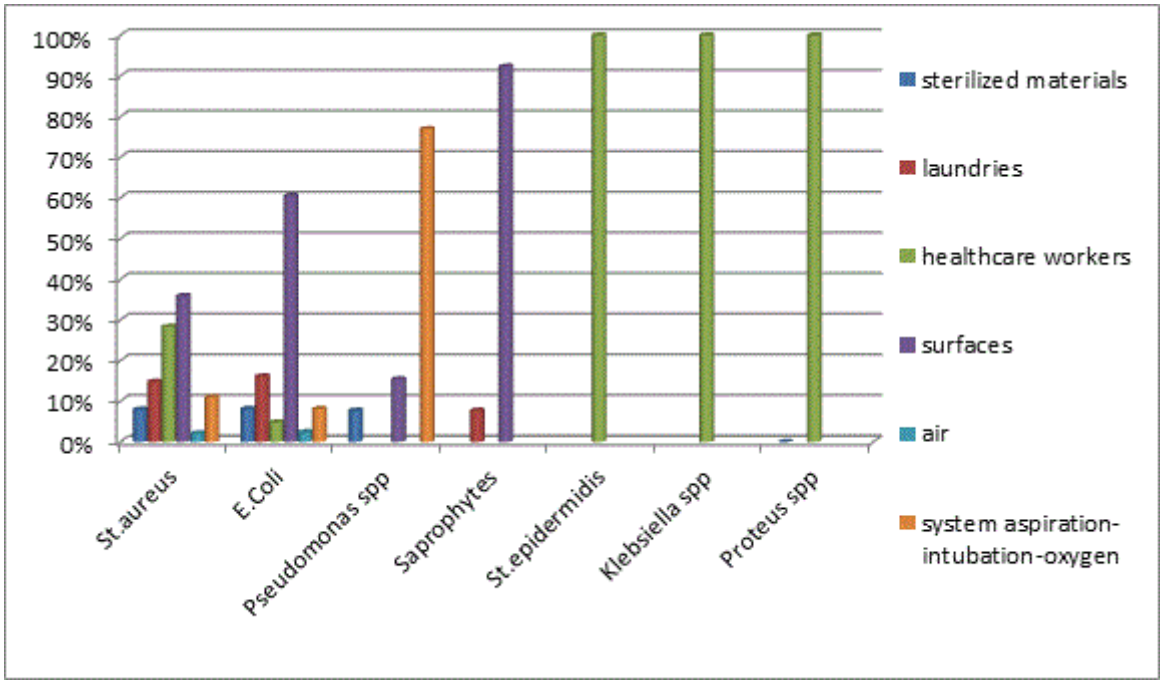

Figure 2: Distribution of microorganisms.

Discussion

Identification of microorganisms of clinical importance in the environments of the Regional Hospital of Korca confirmed the presence of pathogens which are potentially related to nosocomial infections.

From the total of 393 isolates, the microorganism with the higher prevalence was *Staphylococcus aureus* 237 (60.3%) followed by *E. coli* 124 (31.6%), *Klebsiella* spp. 1 (0.3%), *Pseudomonas* spp. 13 (3.3%), *Proteus* spp. 1 (0.3%), *Staphylococcus epidermidis* 4 (1.0%), Saprophytes 13 (3.3%).

This result is similar to other studies that has reported *St.aureus* the most frequent bacteria in hospitals environments (60.30%) [11,12].

Staphylococcus aureus was isolated from all the types of samples, with a higher prevalence in the samples from the surfaces (35.9%).

E. coli was the most frequent gram negative bacteria isolated in hospital environments (31.60%) [13]. *E. coli* was isolated from all the types of samples, predominating in the samples from the surfaces (60.5%).

Pseudomonas spp. was the sond most frequent gram negative bacteria isolated in hospital environments (3.3%). Predomination of *Pseudomonas* spp. in the samples of the systems of aspiration-intubation-oxygen (76.9%) indicates high risk for nosocomial pneumonia [14,15].

The prevalence of Saprophytes isolated from the hospital environments was 3.3%. They were mostly found in the samples from the surfaces (92.3%) [16-18].

Staphylococcus epidermidis, *Klebsiella* spp. and *Proteus* spp. were isolated with a low prevalence, respectively 1.0%, 0.3%, 0.3%. They were isolated only in the samples from the healthcare workers [19].

Conclusion

Hospital environment serve as a potencial reservoir of microorganisms that can cause nosocomial infections. The results of this study revealed that *Staphylococcus aureus* was the most prevalent pathogen isolated in the hospitals while *E. coli* was the most frequent gram negative bacteria isolated. The high prevalence of pathogens in this study might be due to poor hygienic practice in the hospital and lack of proper surveillance system for the control of potential nosocomial pathogens. Therefore, based on this finding it is recommended that management of the hospital should establish new politics which will help to minimize the spread of potential pathogens in the hospital environments.

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