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Pseudomonal infections in patients with burns in Al- Mouasat Hospital in Damascus - Syria

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ABSTRACT

Pseudomonas has become an important cause of gram-negative infection, especially when the host defense mechanisms are compromised such as burned patients. It is the most common pathogen isolated from patients when they have been hospitalized longer than 1 week that causes nosocomial infections. It causes complicated infections and can quickly lead to death. The objective of the present study is to identify the prevalence of pseudomonal infections and their changing during hospitalization in 100 burned patients who were admitted to Al-Mouasat Hospital in Damascus- Syria, in addition to identify pseudomonal species and sensitivity to antibiotics during the period from September 2010 until August 2011. Samples were taken in the first 24 hours and during the first and second weeks after admission. Isolation of the bacteria was performed using standard procedure. Out of 100 patients: no bacteria in the 1st culture and 18% in the 2nd culture, and 30% in 3rd one. The results of bacterial sensitivity to antibiotics tests of 48 pseudomonal cases showed significant resistance to antibiotics. However, the highest level of sensitivity to antibiotics is recorded for Imipenem (47.92%), Meropenem (50%), and cefepime (41.67%). The causative species are classified as following: Pseudomonas Aeruginosa (37.5%), Ps. Fluorescens (58.33%), and Ps. Stenotrophomonas maltophilia (4.16%). Pseudomonas comes first among isolated bacteria, the possibility of infection increases as the patient stays in the burn center.

KEY WORDS: Pseudomonal infection, Pseudomonas aeruginosa, burn and wound infection, nosocomial infection.

1. INTRODUCTION

Pseudomonas Aeruginosa is an aerobic bacterium, measuring 0.5-0.8 by 1.5-3.0 µm, according to Gram stain; it is gram-negative rod. It is a member of the family Pseudomonadaceae and its class is gamma proteobacteria (Shyamala R, 2014; Karna S, 2016; Ahmadi K, 2016; Friedrich M, 2015; National Institutes of Health (US), 2014) It is a free-living bacterium, it can live in soil and water (Todar K, 2005). Almost all strains are motile by means of a single polar flagellum. It produces several pigments such as blue–green (pyocyanin) and yellow-green (florescent, pyoverdin). Pseudomonas is often characterized by a sweet odor and it has beta-hemolytic on Blood agar. (Friedrich M, 2015) The standard incubation temperature is 37°c, and PH is 7.4-7.6. It is an opportunistic pathogen; it rarely causes disease in healthy people (Karna S, 2016; CDC, 2014; Botzenhardt K, 1993; Turner KH, 2014). It has many virulence factors which contribute to the pathogenesis of the infection especially the pili and the flagella (Church D, 2006; Ghanbarzadehz, 2015). Adding to its pathogenicity, this bacterium can tolerate the variability of physical conditions. It is resistant to the majority of used antibiotics and high concentrations of salts and dyes (National Institutes of Health (US), 2014) only a few antibiotics are effective against *P. aeruginosa*, such as ciprofloxacin, gentamicin and imipenem (Ghanbarzadehz, 2015). The genus Pseudomonas contains over 100 species but the most medically important species are: *P. aeruginosa, P. fluorescens* and *P. putida* (Public Health England, 2007).

A burn is a type of injury to skin , or\ and mucosa, which caused by heat, electricity, chemicals, friction, or radiation (Khan A, 2007). There are three types of burns: First-degree burns cause minimal damage, they affect the outer layer of skin, second-degree burns; more serious damage, the affect the top layer and the layer underneath, third-degree burns are the most sever, they damage all layers of skin and tissues underneath.(14) (Khan A et al, 2007) Deep and superficial burns are a rich protein area involving tissue necrosis which is a suitable medium for the colonization of microbes and their generations (Meskini M, 2015). The main common cause of morbidity and mortality in burn patients is infection. The diagnosis and management of burn infection are difficult that due to the many physiologic features unique to burn injury. The most common organisms remain Staphylococcus and Pseudomonas (Gauglitz G, 2015). Bacterial infection after severe burns can be most simplistically attributed to massive breaches in the skin barrier. The fact that *P. aeruginosa* occurs widely in the environment makes it extremely probable that an individual suffering severe burns will be threatened with this pathogen before the burns heal (Karna S, 2016; Turner KH, 2014; Church D, 2006; Ghanbarzadehz, 2015; Gauglitz G, 2015; Lipovy B, 2010).

2. MATERIALS AND METHODS

Sample Collection: From September 2010 to August 2011, a total of 100 swab samples were taken from patients who were admitted to burns center in Al-Mouasat hospital in Damascus- Syria. Swabs were taken from various types of superficial burned infections. Age, gender, burn degree, and period of staying in hospital were recorded for each sample.

Ethical committees of the educational hospitals approved the general principles and framework of the present study. Written informed consent was obtained from all patients. Personal information of all patients remains confidential.

Pseudomonas Isolation: The bacteriological isolation was performed in the central microbiology laboratory of Al-Mouasat hospital. The swabs were dipped in Tioglycolate transport medium, then plated on Blood agar (Abtek-UK) and Eosin-methyl thionine blue agar (EMB) (Abtek-UK), and incubated at 37°C for 24 hours. The organisms were identified by using convencional protocol including colony morphology, pyocyanin pigment production, gram staining, oxidase test and Automatic device (BD Phoenix System) was used to confirm the isolates. The API20E strips (BioMèriex. USA) were used to identify the species of pseudomonas.

Antimicrobial Sensitivity Testing of Pseudomonas Isolates: The sensitivity to antibiotics was accomplished by disk diffusion on agar technique (Kirby-Bauer Antibiotic test). The Mueller-Hinton agar medium (Abtek-UK) was used. 10 commonly antibiotics were used, including norfloxacin (10 µg/disk), ampicillin (10 µg/disk), imipenem (30 u/disk), gentamicin (30 µg/disk), ciprofloxacin (30 µg/disk), cefepime (30 µg/disk), cotrimoxazole (30 µg/disk), meropenem (10 µg/disk), amikacin (30 µg/disk), ceftazidime (30 µg/disk) (Abtek-UK).

3. RESULTS

This study included 100 burned patients who were admitted to the hospital, and was meant to show the prevalence of pseudomonal infections in burn center, its species and its sensitivity to antibiotics. The sample included 56 male patients and 44 female patients. There were (74) patients with 2nd degree burns. All samples showed absence of bacterial pathogens in the first 24 hours. During the first week, pseudomonas was found in 18 cultures and in 30 cultures during the second week of admission. The degree of burn was not significantly correlated with the incidence of infection (P ~ 0.73). Table 1 shows the characterization of 100 patients, Table 2 shows the frequency of pseudomonas isolates identified from 100 patients in first hours and other weeks, Table 3 shows total distribution of pseudomonas species according to API20E test, and Table 4 shows antibiotic resistance pattern of pseudomonal isolates.

Table.1.Total distribution of patients according to gender, age and burn degree

Gender	Male	56
	Female	44
Age/years	<10	34
	10-20	14
	21-30	42
	>30	10
Burn degree	2nd	74
	3rd	24

***= values are expressed as numbers**

Table.2.Frequency of pseudomonas isolates identified from 100 patients in first hours and other weeks:

Time of the cultures	*
First 24 hours	0
The end of 1st week	18
The end of 2nd week	30

***= values are expressed as numbers**

Table.3.Total distribution of pseudomonas species according to API20E test

species	*
Pseudomonas aeroginosa	18
Pseudomonas fluorescens	28
Stenotrophomonas maltophilia	2
Total	48

***= values are expressed as numbers**

Table.4.Antibiotic resistance pattern of pseudomonas isolates:

Antibiotic agent	*	**Antibiotic agent**	*
Ampicillin	48(100)	Ceftazidime	29(60.41)
Amikacin	30(62.5)	Imipenem	25(52.08)
Gentamycin	36(75)	Meropenem	24(50)
Ciprofloxacin	38(79.16)	Cefepime	28(58.33)
Norfloxacin	43(89.58)	Cotrimoxazole	48(100)

*= values are expressed as No(%)

DISCUSSION

Nosocomial infections are an important problem for hospitals and other health services in all countries, with important effects on high-risk patients, such as burn patients. Infections of burn areas are very serious problems that can threaten the patient's life. *Pseudomonas aeruginosa* remains one of the most important opportunistic causes of nosocomial infections and it has developed resistance to a range of antimicrobial agents in burn centers. No sufficient research on nosocomial infections in burned patients had been done. Despite several epidemiological investigates accomplished in burn infections in Syria, data is not enough about nosocomial infections.

The results of the present study showed that *P. Fluorescens* has a higher prevalence in various species of pseudomonal infections according to biochemical test with API20E strips. The prevalence of pseudomonal infection in this study was 30% that corresponded with another studies such as Ranjan et al (2010) (27.7%), Bhattacharjee et al. (2006) (32%), Masaadeh and Jaran (2009) (27.78%), Ekrami A and Kulantar E (2007) (37.5%). High prevalence rates of pseudomonal infection have been reported by Ahmadi K. et al (2016) (62.2%), and Azimi L. et al (2011) (55.5%), and Bhatt et al (2015) (54.9%), Naqvi, 2005, (59.6%). In contrast with Qader et al (2010) (18%). The variation of prevalence rates of pseudomonal infection in this study and others may be due to the differences in the number of samples, type of samples (swab or biopsy) or health care managements and sterility procedures in hospitals and their instruments.

The results showed high prevalence rates of pseudomonal infections in male patients due to high hospitalization rates in them caused by their work that contradicts with the study done by Ahmadi K et al (2016), Mulu et al (2012), and Okon et al (2009). High prevalence rates were reported in patients ranging in age between 21-30 years which were different from the study by Ahmadi (2016) which showed that the most infected patients distributed in age groups of 50-70 and >70 years, whereas in Srinivas B et al (2012) study; a high prevalence rate of infection was in children. This study also showed high rates in the 2nd degree burns. According to the period of hospitalization, an increase of pseudomonal infection in the present study that corresponds with Azimi L. et al (2011). Sensitivity to antibiotics results showed the highest levels of resistance against ampicillin and cotrimoxazol (100%), and the resistance against norfloxacin, ciprofloxacin, amikacin and ceftazidime was 89.58%, 79.16%, 62.5%, 60.41% respectively. And relatively lower resistance against imipenem (52.08%) and meropenem (50%), compared with Ahmadi K et al (2016) (100%, 40%, 41.6%, 100%, 96.6%, 43.3%, 5% and 5% respectively). And with M. S. RAZA ET AL. (2013) (amikacin 0%, gentamycin 0%, norfloxacin 0%, ciprofloxacin 50%, and cotrimoxazole 100%), in Hamid Vaez et al investigation (2015); the results showed that the isolates had a high level of resistance to antibiotics against imipenem and meropenem (100%), ciprofloxacin and ceftazidime (90%), whereas the lowest resistance rate (61.6%) was seen against amikacin. This difference between the present study and others may be due to the strains variation.

4. CONCLUSION

Burn centers should routinely determine and track the specific patterns of burn area microbial Colonization especially pseudomonas, time-related changes in the predominant microbial flora of the burn area in individual patients, Sensitivity to antibiotics of bacteria in burn infections during the period of hospitalization, and trends in the nosocomial incidence of them. Coordination among plastic surgeons, infectious disease physicians, and clinical microbiologists is essential to facilitate the managment of burn infections based on an updated microbiological data and outcome analyses.

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