

ORIGINAL ARTICLE

Molecular Identification of Different Bacteria isolated from Infected Wounds at Hayatabad Medical Complex Peshawar Pakistan

SYED LUQMAN SHUAIB¹, MOHSINA HAQ^{2*}, BAKHT BILAND KHAN³, FARISHTA HAQ⁴, IHTESHAMUL HAQ⁵, IBRAR AHMAD⁵, WAHID ULLAH⁶, AZHAR NAZIR⁶, MUHAMMAD FAROOQ⁶, HASNAIN ISRAR⁶, SHAHID HUSSAIN⁶, KAMRAN KHAN⁷

¹Department of Microbiology Khyber Medical College Peshawar Pakistan

²Department Microbiology and Pathology Peshawar Medical college, Riphah international University, Warsak road Peshawar Pakistan

³Department of Medicine, Mercy Teaching Hospital, Peshawar Medical College Peshawar Pakistan.

⁴MBBS Final year student Peshawar Medical college Pakistan

⁵Department of Biotechnology and Genetic Engineering Hazara University Mansehra Pakistan

⁶Department of Medical Laboratory Technology, The University of Haripur, Haripur, Pakistan

⁷Medical C Ward Khyber Teaching Hospital Peshawar Pakistan

Corresponding author: Ihteshamul Haq, Email Address: ihteshamulhaq384@gmail.com

Co-Corresponding Author: Mohsina Haq, Email Address: dr.mohsinahaq@gmail.com

ABSTRACT

Wound infections are one of the most common hospital acquired infections and are an important cause of morbidity and account for 70-80% mortality. The Purpose of the current study was to determine the commonest bacteria associated with wound infections at Hayatabad Medical Complex Peshawar January 2019 to June 2020. A total of 100 culture positive samples from patients with mean age of 6.2±0.25 were analyzed. Patient history and clinical findings were collected on a pre-coded form. Pus samples or wound swabs were collected from infected wounds and were analyzed through culturing and biochemical methods for aerobic bacteria. A total of 109 bacteria were isolated from 100 samples with almost same frequency of Gram positive cocci 54(49.54%) and Gram negative bacilli 55(50.45%). Most frequently isolated organism was *S. aureus* 45(41.28%) followed by *Pseudomonas* species 20(18.35%). From this study we conclude that Wound infection remains an ongoing problem which, although, cannot be completely eradicated, however by taking prompt control measures against the most commonly isolated organism and proper care of wound may lead to the minimum of wound infection.

Keywords: Abscess, wound infection, padiatric surgical wounds, arobic bacteria

INTRODUCTION

Skin, the largest organ in the human body, plays a crucial role in the sustenance of life through the regulation of water and electrolyte balance, thermoregulation, and by acting as a barrier to external noxious agents including microorganisms, however, when the epithelial integrity of skin is disrupted, a wound results [1]. Wound infections are one of the most common hospital acquired infections and are an important cause of morbidity and account for 70-80% mortality [2]. Development of such infections represent delayed healing, cause anxiety and discomfort for patient, longer stays at hospitals and add to cost of healthcare services significantly[3]The importance of wound infections, in both economic and human terms, should not be underestimated [4]. In a study, on average, patients with a wound infection stay about 6 - 10 days more than if the wounds heal without infections [5]. This additional stay almost doubles the hospital cost that is equivalent to between £1,168 and £2,398 [6] Wound infections can be caused by different groups of microorganisms like bacteria, fungi and protozoa [7]. However, different microorganisms can exist in polymicrobial communities especially in the margins of wounds and in chronic wounds [8]. The infecting microorganism may belong to aerobic as well as anaerobic group [9]. Most commonly isolated aerobic microorganism include *Staphylococcus aureus* (31%), Coagulase-negative staphylococci (CoNS) (5%), Enterococci (5%), *Escherichia coli* (9%), *Pseudomonas aeruginosa* (14%), *Klebsiella pneumoniae* (3%), Enterobacter species (9%), *Proteus mirabilis* (3.5%), other streptococci (3%), *Candida* (1.3%) with 80% *Candida albicans*, Group D streptococci (2%) and *Acinetobacter* (2%). Other gram-positive aerobes (2%) and anaerobes (2.7%) also cause wound infections

MATERIALS AND METHODS

This observational descriptive study was conducted at Hayatabad Medical Complex Peshawar January 2019 to June 2020. The study population was of children irrespective of sex, either admitted to different wards in hospital or visiting the out-patient department, with mean age of 6.25±0.25 years. A total of 100 culture positive samples, from different types of infected wounds, received at microbiology department during this period, were analyzed. 38% samples were from OPD while 62% were from IPD

(Table-1). The specialty-wise distribution from different wards is given in Table-2

Table 1: Total number of positive cases from out-patient and Inpatient departments (n=100)

Category	=n	%age
OPD	38	38
IPD	62	62

Data were collected on standardized pre-coded forms. Basic clinical information on patient's demographics, way of wound acquisition, underlying disease status, treatment taken, antibiotics usage and previous microbiological analysis of wound, if done, was obtained. Draining/aspirated pus from an infected wound was collected aseptically in a sterilized container. When no pus was discharging, a sample was collected from the infected site by using a sterile cotton swab. Liquid pus samples were 63% while wound swab samples contributed 37% to total. Samples were immediately transported to the laboratory where they were processed promptly.

Table 2: Frequency distribution of positive cases from Different wards (n=87)

Ward	Cases	
	=n	%age
CICU	2	2.78
Emergency	3	4.17
Gastroenterology	2	2.78
GMW-I	1	1.39
GMW-II	3	4.17
GMWI-ICU	2	2.78
General surgical	4	5.56
MICU	1	1.39
NSW	6	8.33
Orthopedic	11	15.28
Private	1	1.39
PSW	23	31.94
Surgical follow-up	2	2.78
Surgical ICU	3	4.17
Surgical recovery	6	8.33
Surgical emergency	1	1.39
Urology	1	1.39

All samples were inoculated on the 5% blood agar as well as on MacConkey agar plates and Gram smear was examined.

Cultures were aerobically incubated at 37°C overnight. Positive cultures were identified using API (Analytical Profile Index) system along with standard diagnostic microbiological laboratory methods like Catalase, Coagulase, D Nase, Oxidase tests [10].

RESULTS

Among different types of infected wounds only surgical and soft tissue wound sample were received with relative frequency of 57% and 43% respectively. Majority of the wounds was infected with a single organism however three wounds were infected with a couple of bacteria while from two specimens 3 organisms were isolated. A total of 109 bacteria were isolated from 100 samples with almost same frequency of Gram positive cocci 54(49.54%) and Gram negative bacilli 55(50.45%). Most frequently isolated organism was *S. aureus* 45(41.28%) followed by *Pseudomonas* species 20(18.35%). Rest of the organisms isolated from infected wounds included *E.coli*, *Klebsiella* species, CoNS, *Proteus* species and *Serratia* species. Their relative frequencies are given in Table-3

Table 3: Frequency of different pathogens isolated from infected wounds (n=109)

Pathogen	Frequency	Relative frequency (%age)
<i>Staphylococcus aureus</i>	45	41.28
Coagulase negative <i>Staphylococci</i> (CoNS)	6	5.50
<i>Streptococcus</i> species	3	2.75
<i>Escherichia coli</i>	15	13.76
<i>Klebsiella</i> spp.	14	12.84
<i>Pseudomonas aeruginosa</i>	6	5.50
<i>Pseudomonas fluorescens</i>	5	4.59
Other <i>Pseudomonas</i> spp.	9	8.26
<i>Proteus</i> spp.	5	4.59
<i>Serratia</i> spp.	1	0.92

DISCUSSION

This study provides the data about the incidence of locally infected wounds in different age and sex groups of children, visiting or admitted in Hayatabad Medical Complex Peshawar. In this study Mean age of the children contracting wound infection was 6.2±0.25 S.E.M. with the highest frequency in the age group 4-6 years (34%). The reported mean age in literature is also in accordance to the results obtained by us i.e. 5.4±3.5 years [11] Rest of the split of the infection in age was 26% in 1–3 years, 18% in 7–9 years, 16% in 10-12 years and 6% in 13–15 years. It indicated that most of the children contracting wound infection, especially undergoing surgery are in the age group range of 1–6 years. The chances for the wound infection go on increasing as the age of the patient increases [12]. due to the weakening of the immune response, similarly children have immune system that is not so much strong to compete all types of infections so chances for infection increase many folds. Over all male patients with wound infection were 20% higher than female. Kaabachi et al (2005)[13], in a study at Tunis has reported a sex ratio of 1.6 boys/1 girl while Jamali et al. (2001)[14] has reported 1.7 boys /1 female in Pakistan. Our overall calculated sex ratio is 1.5 boys /1 girl. It suggests that contracting wound infection has no relation with sex. The slight difference that has been noted is just due to our social behavior where males are given superiority to the female and if get diseased are brought immediately to hospitals in comparison to female for treatment. The incidence of different types of wound infections was found to be the highest of surgical wound infection (57%) followed by acute soft tissue infection (43%). In literature, incidence for postoperative wound infection has been reported to be 31.37%[15]. Thus our SSIs incidence is 25.63% higher than cited in literature. It is due to the poor care of the wounds and dirty surgical procedure, a source for wound contamination, carried out by patients themselves or due to the untrained practitioners in different districts where people initially go for the primitive treatment.[16]. The frequency of Gram positive and negative organisms was found to be almost

equal, 49.54% and 50.45% respectively. However, *S. aureus* was most frequently isolated organism (41.28%) followed by *E. coli* (13.76%), *Klebsiella* spp. (12.84%), *P. aeruginosa* 5.50% and *P. fluorescens* 4.54%, other *Pseudomonas* spp. (8.26%), CoNS (5.50%), *Proteus* spp. (4.59%), *Streptococcus* spp. (2.75%) and *Serratia* spp. (0.92%). Mumtaz et al. (2002)[17] has also found *S. aureus* as the most common pathogen (49%) followed by *E. coli* (25.9%) *Klebsiella* spp. (9.5%), *P. aeruginosa* (8.6%) and *Proteus* spp. (4%). However, Mahmood (2000)[18] has found somewhat different pattern of frequent organism like *S. aureus* (50.32%) followed by *P. aeruginosa* (16.3%), *E. coli* (14.37%), *Klebsiella pneumoniae* (11.76%), miscellaneous gram negative rods (5.88%) and *Streptococcus pyogenes* (1.30%). Gales et al. (2000)[19] have also found the same prevalence pattern of organism in infected wounds. The slight variability that is found to be seen in different studies is due to the different settings and populations, albeit, general pattern for contaminating organisms is almost same.

CONCLUSION

Our Research Conclude that Contracting wound infection remains an ongoing problem. The main culprit for the wound infection are the trivial organisms like *S. aureus*, *E. coli*, *Pseudomonas*, *Klebsiella*, etc. Although complete eradication of wound infections is not possible however by taking the preventive measures and adopting prompt clean surgical procedures and proper care of wounds, the incidence of wound infection may be limited to minimum. Otherwise wound infections may lead to the morbidity and mortality of to a high count especially in children.

REFERENCE

1. Enoch, S. and Price, P. 2004. Cellular, molecular and biochemical differences in the pathophysiology of healing between acute wounds, chronic wounds and wounds in the aged. Available from; URL: <http://www.worldwidewound.com>
2. Gottrup, F., Melling, A. and Hollander, D. 2005. An overview of surgical site infections: aetiology, incidence and risk factors. *E.W.M.A. Journal*; 5(2): 11- 15. Available from: URL: <http://www.worldwidewounds.com>
3. Wilson, A. P. R., Gibbons, C., Reeves, B. C., Hodgson, B., Liu, M. and Plummer, D. 2004. Surgical wound infections as a performance indicator: agreement of common definitions of wound infections in 4773 Patients. *B.M.J.*; 329:720-722.
4. Mohantay, S., Kapil, A., Dhawan, B. and Das, B. K. 2004. Bacteriological and antimicrobial susceptibility profile of soft tissue infections from Northern India. *Indian J. Med. sci.*; 58: 10-15
5. Collier, M. 2004. Recognition and management of wound infections. *Wounds*; Available from: URL: <http://www.worldwidewounds.com>
6. Plowman, R. 2005. The socioeconomic burden of hospital acquired infection. *Euro. Surveill.*; 5(4): 49-50.
7. Zoutman, D., McDonald, S. and Vethanayagan, D. 1998. Total and attributable costs of surgical-wound infections at a Canadian tertiary-care center. *Infect. Control Hosp. Epidemiol.*; 19: 254-259
8. Cooper, R., Kingsley, A. and White, R. 2003. *Wound Infection and Microbiology*. Medical Communications (UK) Ltd for Johnson & Johnson Medical
9. Percevil, S. and Bowler, P. 2004. Understanding the effects of bacterial communities and biofilms on wound healing. Available from: URL: <http://www.worldwidewounds.com>
10. Bowler P G. The anaerobic and aerobic microbiology of wounds: a review. *Wounds* 1998; 10: 170–178.
11. Tayfour, M. A., Al-Ghamdi S. M. and Al-Ghamdi, A. S. 2005. Surgical wound infections in King Fahad Hospital at Al-Baha. *Saudi Med. J.*; 26(8): 1305-1307
12. Cheesbrough, M. 2000. District laboratory practice in tropical countries (2). Cambridge, University Press, United Kingdom
13. Asghar, M., Haq, M., & Saleem, N. (2022). Molecular Identification And Prevalence Rate Of HepatitisC Virus Among Hemodialysis Patients In Peshawar Kp Pakistan. *NVEO-NATURAL VOLATILES & ESSENTIAL OILS Journal* NVEO, 1168-1175
14. Bowler, P.G., Duerden, I. and Armstrong, D. G. 2001. Wound microbiology and associated approaches to wound management. *Clin. Microbiol. Rev.*; 14(2): 244- 269.
15. Asif, A., Asghar, M., Khan, H. U., Haq, I., Shuaib, S. L., Khalid, F., ... & Rehman, N. (2021). Antibiotic susceptibility pattern of clinical isolates of methicillin resistant staphylococcus aureus in

- Peshawar, Pakistan. *Annals of the Romanian Society for Cell Biology*, 25(6), 20116-20131
16. Haq, I., Zahir, F., ur Rehman, A., Ullah, N., Khan, J., Qamar, N., ... & Khan, Y. (2021). Evaluation of change in hematological parameters and epidemiological identification of dengue virus infection at district Peshawar, Khyber Pakhtunkhwa, Pakistan. *International Journal of Mosquito Research*, 8(1, Part A), 11-18.
 17. Anwar, F. (2020). Serological and Epidemiological Evaluation of active HCV Infection in the Volunteer blood Donor at District Swat Khyber Pakhtunkhwa Pakistan. *Bull. Env. Pharmacol. Life Sci*, 9, 08-15 56.
 18. Ahmad, S. U., Khan, M. S., Jan, Z., Khan, N., Ali, A., Rehman, N., ... & Zahir, F. (2021). Genome wide association study and phylogenetic analysis of novel SARS-COV-2 virus among different countries. *Pakistan Journal of Pharmaceutical Sciences*, 34(4)
 19. Bashir, Z., Ahmad, S. U., Kiani, B. H., Jan, Z., Khan, N., Khan, U., ... & Mahmood, T. (2021). Immuno informatics approaches to explore B and T cell epitope-based vaccine designing for SARS-CoV-2 Virus. *Pakistan Journal of Pharmaceutical Sciences*, 34
 20. Haq, I., Muhammad, A., Fazli Zahir, M. K., Anwar, F., Akhtar, M. S., & Ullah, F. (2020). Serological and Epidemiology study of *Helicobacter pylori* infection among Dyspeptic patients in District Peshawar Pakistan. *Adv. Biores*, 11(3), 81-85.
 21. Qamar, Z., Anwar, F., Ahmad, R., Haq, I., Khan, A. M. K., Hussain, R., ... & Khan, J. (2021). Prevalence of Hepatitis C virus and determination of its genotypes in subjects of Tehsil Daggar District Buner, KP, Pakistan. *Clinical Epidemiology and Global Health*, 12, 100809.