



Research Article

A STUDY ON NECROTIZING FASCIITIS AND OTHER WOUND INFECTIONS

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Received December 28, 2016; Accepted January 25, 2016; Published January 31, 2017;

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Cite This Article: Usharani, P., Vijayalakshmi, P.(2017). A study on necrotizing fasciitis and other wound infections. BMR Microbiology, 3(1).1-7

ABSTRACT

Purpose

A wound is commonly infected because there is loss of integrity of skin. Infected wound delays wound healing due to the lack of defense mechanism, blood supply and lack of oxygen. The present study was focused on the incidence of various pathogens in different types of wound infections and the variations in their antibiotic susceptibility have been analyzed.

Methods

A total of 50 samples were collected randomly from wound infected areas like cellulitis, diabetic foot lesions, post-operative wound infections, early stages of necrotizing fasciitis, burn wounds etc. The samples were analyzed by various microbiological techniques like microscopy, culture methods and Biochemical methods. The antibiotic sensitivity pattern of isolates was tested by using Kirby Bauer's disc diffusion method.

Results

Out of 50 cases, 35 were males and 15 were a female which shows the male preponderance. Wound infection showed a high incidence in the age group of 41-60 which is followed by 21-40. The most predominant bacteria in positive samples are *Pseudomonas aeruginosa* with 44% which is followed by *Staphylococcus aureus* 26%, *Klebsiella pneumoniae* 16%, *Proteus mirabilis* 8% and *Escherichia coli* 6%. Nearly 21 major strains of *Pseudomonas aeruginosa* was highly sensitive to Amikacin (95.4%) and 15 strains were sensitive to Amoxycillin/clavulanate (68.1%). *Staphylococcus aureus* was isolated from two cases of necrotizing fasciitis and the fungi *Aspergillus niger* was isolated from one burn wound infection.

Conclusion

The most common organisms isolated were *Pseudomonas aeruginosa* followed by *Staphylococcus aureus* which showed sensitivity to Amikacin. The percentage of resistance was high among Penicillin, Cephalexin, Ampicillin, Tetracycline and Erythromycin.

KEY WORDS: Wound infection, Microorganisms, Microbiological analysis, sensitivity, resistance.



INTRODUCTION

Wound infections have posed considerable challenges to mankind in the terms that a variety of pathogenic microorganisms have colonized wounds. A wound is inflicted to the human body by an injury. Wound infections may be Endogenous or Exogenous. Endogenous infections or auto-infections are caused by organisms that have been leading a commensal existence elsewhere in the patient's body. In exogenous infections, the source of the infecting organism is out with the body of the patient who becomes infected. The most common effects to the body by invading organisms include inflammation, warmth, redness around the wound, discharge of pus; fever with or without chills and loss of function of that part [1]. Pus exudates are generally used as a sample for identifying the organism and diagnosing the disease. Wound infections are of various types which include Cellulitis, Osteomyelitis, Diabetic foot lesions, Post-operative wounds, Traumatic ulcers, Necrotizing fasciitis, Pyogenic arthritis, cellulitis etc. A diverse range of bacteria are known to colonize wounds. Both gram Positive bacteria and gram negative bacteria are isolated from various wounds. As such there is no specificity regarding the site specific infections of organisms. The possible pathogens in wound infections are *Staphylococcus aureus*, *Streptococcus pyogenes*, *Enterococcus*, *Clostridium perfringens*, *Pseudomonas aeruginosa*, *Proteus*, *Escherichia coli*, *Klebsiella* species etc.

Cellulitis is an acute, spreading pyogenic inflammation of the dermis and subcutaneous tissue, habitually complicating a wound ulcer or dermatosis. The area usually on the leg is swollen, tender, warm, and erythematous. Erysipelas is a superficial cellulitis with conspicuous lymphatic involvement, offering with an indurated "pean orange" appearance with a raised border that is demarcated from normal skin [2]. Bacteria like *Staphylococcus aureus*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa* etc. gain entry into the body in many ways and cause cellulitis [3].

Diabetic foot problems like ulcerations, gangrene are the most common cause of hospitalization among diabetic patients. The critical triad most commonly seen in patients with diabetic foot ulcers includes peripheral sensory neuropathy, deformity, trauma, edema, calluses and peripheral vascular disease [4]. The most commonly encountered pathogens include Methicillin resistant *Staphylococcus aureus*, *Streptococci*, *Enterobacteriaceae*, *Pseudomonas aeruginosa*, *Enterococci* [5].

Post-operative wound infection is considered by the presence of pus at the incision site. It is generally due to increased hospital stay and direct cost of hospitalization. The surgical wounds are inoculated with bacteria at operation. Many factors are responsible for sepsis includes invasive capacity of the organism, the degree of excellence in surgical or antiseptic technique, resistance of the host, the presence of physiologic derangements, the type of tissue involved and climatic or seasonal variation in bacterial flora [6]. The most frequently found organisms

Necrotizing fasciitis is one of the rare infectious diseases caused by "flesh-eating bacteria". It may be fatal if not treated properly. It spreads quickly and aggressively in an infected person. It causes tissue death at the site of infection and beyond. The pathogens generally enter into the body through minor cuts, insect bites, abrasions surgery or any injury. Group 'A' *Streptococci* and *Staphylococci* generally causing necrotizing fasciitis. The symptoms involve severe pain at skin opening area, redness, warmth around the wound and flu like symptoms. After five days more advanced symptoms were identified like swelling, blisters filled with fowl smelling fluid, discoloration, peeling, flakiness, gangrene, drop in blood pressure and toxic shock.

Burn wound infection is a major public health problem and the most devastating form of trauma worldwide [7]. The most risk factors in burn infection include the age of burns, total burn size, body surface area, full thickness burns, prolonged hospital stay, late surgical excision, open dressing, hyper glycaemic episodes, immune suppression, fungal wound colonization etc. Bacteria commonly cause burn wound infections followed by fungi and viruses. Though bacterial infections are decreased worldwide due to better care of burn patients and availability of antibiotics, the fungal infections of wound showed an increasing trend [8, 9]. Non-*candida albicans* group, *Aspergillus*, *Fusarium*, *Mucor* is more commonly isolated from infected area [10, 11].

The present study was aimed to isolate different pathogenic organisms from different types of wound infections specifically diabetic foot lesions, post-operative wound infections, cellulitis, necrotizing fasciitis, burn wound infections etc. To study also evaluate the antibiotic sensitivity pattern of the isolates against different antibiotics.

MATERIALS AND METHODS

A Hospital based cross-sectional study was conducted on 50 wound samples taken from consecutive patients suffered with cellulitis, diabetic foot lesions, post-operative wound infections, necrotizing fasciitis at early stage and burn wound infections. The swab specimens were collected aseptically after cleansing the wounds with normal saline solution using Levine's technique [12]. Both the bacteriological and mycological studies were performed by using standard microbiological techniques [13]. The specimens are examined to detect, isolate and identify the pathogens by using Microscopy (Grams staining, Lactophenol cotton blue staining for fungi), Culture methods using blood agar, MacConkey agar, Mannitol salt agar, Eosin methylene blue agar for bacteria and Sabourauds dextrose agar for fungi and Biochemical methods like catalase, coagulase, oxidase, sugar fermentation tests, IMViC tests. All the plates were incubated at 37°C for 24-48h. However the fungal plates were observed for 1 week. The antibiotic susceptibility pattern of bacteria was tested by Kirby-Bauer's disc diffusion method using Muller-Hinton agar medium and

antifungal susceptibility can be performed by E-Strip method [13].

diabetic foot lesions, post-operative wounds, necrotizing fasciitis, burn wound infections and others etc. (Figure 1) (Table 1).

RESULTS AND DISCUSSION

In the present study a total of 50 samples were isolated from various types of wounds such as Cellulitis of leg,

Table 1: Patients infected with different types of Wound Infections

S. No	Name of the disease	No of patients	Percentage
1	Cellulitis	20	40
2	Diabetic foot lesions	14	28
3	Post-operative wound	9	18
4	Necrotizing fasciitis	2	4
5	Burn wound infection	2	2
6	Others	3	6
Total No. of patients		50	100

Figure 2 showed that out of 50 cases, 35 were males and 15 were females indicate that men are frequently infected. Wound infection showed a high incidence in the age group of 41-60 which is followed by 21-40. Out of 50 samples, the most predominant bacteria in 22 positive samples showed the growth of gram negative bacteria *Pseudomonas aeruginosa* with 44% which is followed by the growth of gram positive bacteria *Staphylococcus aureus* (26%) in 13 samples, *Klebsiella pneumoniae* (16%) in 8 positive samples, *Proteus mirabilis* (8%) in 4 samples and *Escherichia coli* (6%) in 3 samples (Table 2).

Table 2: Percentage of organism Isolated In Wound Infections

S.No	Name of the organism	Positive samples	Percentage
1.	<i>Pseudomonas aeruginosa</i>	22	44
2.	<i>Staphylococcus aureus</i>	13	26
3.	<i>Klebsiella pneumoniae</i>	8	16
4.	<i>Proteus mirabilis</i>	4	8
5.	<i>Escherichia Coli</i>	3	6

Coagulase positive *Staphylococcus aureus* growth was found in the plates of Mannitol salt agar inoculated with the pus swabs collected from the two cases of necrotizing fasciitis. *Staphylococcus aureus* is most commonly isolated from necrotizing fasciitis cases [14, 15]. The fungal growth of *Aspergillus niger* was found in one of the burn wound sample (Figure 3). However the bacterial infection is common in burn patients but now-a-days fungal infections showed increased tendency may be due to improper aseptic conditions [10, 11]. One study had isolated gram positive pathogens from wound infections and found that *Staphylococci* was the major causative agent with 72% positive cases [16]. By analyzing 79 swab specimens obtained from various categories of wounds. The twin genera of *Staphylococci* and *Pseudomonas* contributed equally to burn infections, but *Staphylococcus* species were predominant in post-operative wound infections. Higher incidence of *Escherichia coli* and *Pseudomonas aeruginosa* in ulcers was also noticed [17]. Among the various categories of wound infections surgical infections or post-operative wound infections are always on the rise. Certain lapse in the maintenance of strict aseptic conditions during surgery is the prime reason [18]. The magnitude of bacterial infection of lesions of the diabetic foot in a study carried for about 2 years. Among 107 patients 62 suffered with ulcer showed mixed

aerobes, 26 patients were found to have cellulitis and 12 with gangrene had more than 5 types of aerobes and anaerobes like *Escherichia coli*, *Klebsiella*, *Pseudomonas*, and *Proteus* [19].

Antibiotic sensitivity test was carried out against all the isolates in the present study. Variations in the susceptibilities and resistance patterns of the organism to certain drugs have been noticed (Table 3). Out of 22 strains, 21 strains of *Pseudomonas aeruginosa* were sensitive to Amikacin (95.4%), 15 strains were sensitive to Amoxicillin/Clavulanate (68.1%), 12 strains were sensitive to Gentamycin (54.5%), 10 strains were sensitive to Ofloxacin (45.4%), 9 strains were sensitive to Ciprofloxacin (40.9%), 6 strains were sensitive to Cephalexin (27.2%), 6 strains were sensitive to Erythromycin (27.2%), 3 strains were sensitive to Ampicillin (13.6%), 1 strain was sensitive to Penicillin (4.5%). The drug resistance of amikacin was plasmid mediated and 100% resistance was observed to Amikacin by Shahid et al. [20].

Table 3: Antibiotic Sensitivity Pattern of the Isolates

Name of the organism	Sensitivity (%)									
	P	CP	A	Ak	T	E	G	Am/C	Cf	Of
<i>Pseudomonas aeruginosa</i>	4.5	27.2	13.6	95.4	22.7	27.2	54.5	68.1	40.9	45.4
<i>Staphylococcus aureus</i>	-	46.1	15.3	84.6	30.7	53.8	69.2	38.4	46.1	84.6
<i>Klebsiella pneumoniae</i>	-	25	12.5	62.5	-	37.5	37.5	37.5	50	50
<i>Proteus mirabilis</i>	-	-	-	100	25	50	25	50	75	50
<i>Escherichia coli</i>	66.6	-	-	100	33.3	-	-	-	-	-

Drugs evaluated: P: Penicillin, Cp: Cephalexin, A: Ampicillin, Am/C: Amoxicillin/clavulanate, T: Tetracycline, E: Erythromycin, G: Gentamicin, AK: Amikacin, Cf: Ciprofloxacin, Of: Ofloxacin

However Bharathi and Jogalakshmi were reported 100% sensitivity of *Pseudomonas aeruginosa* to the drug Amikacin [21] and the results were well agreed with the present study 95.4% strains were sensitive to Amikacin which showed lower resistance. According to Shahid et al. 80% strains were sensitive to Gentamycin [20] and 89.4% sensitivity to Gentamycin was reported by Bharathi and Jogalakshmi [21]. In the present study 54.5% strains of *Pseudomonas aeruginosa* were sensitive to Gentamycin which showed higher resistance. Algun et al. analyzed the sensitivity of 136 *Pseudomonas aeruginosa* strains Isolated from clinical materials. The resistant rates with Ciprofloxacin was 12.5% and 19.9% of strains showed resistance to Ofloxacin which showed lower resistant rates [18] than present study in which 59.1% strains showed resistance to Ciprofloxacin and 54.6% strains were resistant to Ofloxacin. Out of 13 strains of *Staphylococcus aureus*, 11 strains were sensitive to Amikacin (84.6%) and Ofloxacin (84.6%), 9 strains were sensitive to Gentamycin (69.2%), 7 strains were sensitive to Erythromycin (53.8%), 6 strains were sensitive to Ciprofloxacin (46.1%) and Cephalexin (46.1%), 5 strains were sensitive to Amoxicillin/Clavulanate (38.4%), 4 strains were sensitive to tetracycline (30.7%), 2 strains were sensitive to Ampicillin (15.3%). Another similar study had found that *Staphylococcus aureus* isolated from various categories of wounds showed moderate sensitivity to Ampicillin and tetracycline [17]. Bharathi and Jogalakshmi had concluded that, *Staphylococcus aureus* strains showed 95.2% sensitivity to the drug Amikacin and resistant to Penicillin and Cephalexin [21]. The current study also determined that many strains of *Staphylococcus aureus* showed more sensitivity to Amikacin (84.6%) and are completely resistant to Penicillin but exhibited 46.1% sensitivity to Cephalexin. Among 8 strains of *Klebsiella pneumoniae*, 5 strains were sensitive to Amikacin (62.5%), 4 strains were sensitive to Ciprofloxacin (50%) and Ofloxacin (50%), 3 strains were sensitive to Erythromycin (37.5%), Gentamycin (37.5%) and Amoxicillin/Clavulanate (37.5%), 2 strains were sensitive to Cephalexin (25%), 1 strain was sensitive to

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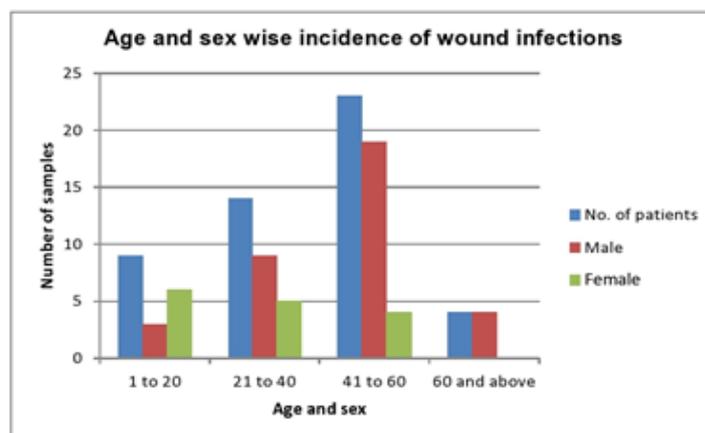
Ampicillin (12.5%). Shen et al. identified that, 7% of *Klebsiella pneumoniae* strains showing sensitivity to Ampicillin isolated from surgical infections [22], where as in the present study it is the slightly higher i.e., (12.5%). Among 4 strains of *Proteus mirabilis*, all showed 100% sensitivity to Amikacin, 3 strains were sensitive to Ciprofloxacin (75%), 2 strains were sensitive to Amoxicillin/ Clavulanate (50%), Gentamycin (50%) and Ofloxacin (50%), 1 strain was sensitive to Tetracycline (25%) and Erythromycin (25%). The similar results were reported by Bharathi and Jogalakshmi but the organisms showed little more resistance to ciprofloxacin and Gentamicin and sensitive to tetracycline [21]. All the 3 strains of *Escherichia coli* showed 100% susceptibility to Amikacin, 2 strains were sensitive to Penicillin (66.6%) and 1 strain was sensitive to Tetracycline (33.3%) and exhibit complete resistance to all other antibiotics. The investigation report of Mahmood on surgical site infection had revealed that, the antibiotic sensitivity pattern showed that more than 60% of the isolates of *Escherichia coli* were found to be resistant to Gentamicin [23] but the organism isolated in the current study showed complete resistance to Gentamicin. The isolated strain of *Aspergillus niger* showed maximum sensitivity to voriconazole followed by terbinafine and ketokonazole, intermediate sensitivity to amphotericin-B, clotrimazole and nystatin with E-test gradient strips of concentration ranging from 1-32µg/ml. Similar results were reported by the previous study of Malini et al. [24].

There is a frightening increase of infections caused by antibiotic resistant bacteria. Lack of standardized antibiotic policy and random use of antibiotics may lead to emergence of antibiotic resistant bacterial strains. In addition, usual antimicrobial susceptibility surveillance is necessary for area-wise monitoring of the resistance pattern. An efficient national and state level antibiotic policy and draft guidelines should be initiated to safeguard the effectiveness of antibiotics and for better patient management. The fight for entire elimination of wound infections will carry on with passable surveillance and with good co-ordination of microbiologists, this fight would definitely be won [25].

Figure 1: Infected wound (Necrosis)



Figure 2: Age and sex wise incidence of wound infections





CONCLUSION

Pus samples were collected from 50 cases of different etiologies. The common organisms isolated were *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Proteus mirabilis* and *Escherichia coli*. *Pseudomonas aeruginosa* was the major pathogen isolated in 22 cases (44%) followed by *Staphylococcus aureus* in 13 cases. All organisms were highly sensitive to Amikacin. The percentage of resistance was high among Penicillin, Cephalexin, Ampicillin, Tetracycline and Erythromycin.

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