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AEROBIC BACTERIAL ISOLATES FROM INFECTED WOUNDS

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ABSTRACT

Background: Wound infection causes great distress in terms of associated mortality and morbidity, increased length of hospital stay, profound discomfort and significant increased in healthcare cost. Infection in a wound delays healing and may cause wound break down, herniation of the wound and complete wound dehiscence.Therefore the knowledge of the causative agents of wound infection will be helpful in the control of wound infection and selection of empiric antimicrobial therapy as an infection control measure. Methods:A total of 207 wound specimens collected from patients attending the University of Benin Teaching Hospital were used for this study. All specimens were collected using sterile swabs sticks. Specimens were processed using standard microbiological methods. Results:A total of 278 bacterial isolates were obtained from 207 wound specimens processed in this study. Positive growth were observed in 185 (89.4%) of the wound cultures and no bacterial isolates were obtained in 22 (21.1%) of the cultured materials. *Staphylococcus aureus* (26.9%) was the most predominant isolate followed by *Klebsiellapneumoniae* (17.6%), *Pseudomonas aeruginosa* (16.9%) and *Escherichia coli* (12.6%). All isolates were resistant to ampicillin, amoxyillin-clavulanate and tetracycline but show variable susceptibility to other antibacterial used. Majority of the isolates produced beta lactamase. Conclusion: A high proportion of the wounds were infected.The variety of microorganisms observed in this study support the need to obtain culture specimen from infected wounds for microbiological evaluation and antibiotic susceptibility determination, so that adapted chemotherapy can be prescribed.

Key words:wound infection, polymicrobial, immune status, host

INTRODUCTION

A wound is any physical injury involving a break in the skin (1). The exposed subcutaneous tissues provides a favourable substratum for a wide variety of microorganisms to contaminate and colonize, and if the involved tissue is devitalized and the host immune response is compromised, the conditions become optimal for microbial growth (2).This is because the host immune response plays a critical role in determining whether wound infection will arise (3).

Wound infection refers to the deposition and multiplication of bacteria in tissue with an associated host reaction (4). This may be characterized by the classic signs of redness, pain, swelling and fever (5).

The progression of a wound to an infected state is likely to involve a multitude of microbial or host factors including the type, site, and depth of wound, the extent of non viable exogenous contamination, the general health and immune status of the host, the microbial load, and the combined virulence expressed by the types of microorganisms involved (2).

Although the majority of wounds are polymicrobial involving both aerobes and anaerobes, aerobic pathogens such as *Staphylococcus aureus*, *Pseudomonas aeroginosa*, and beta haemolytic *Streptococci* have been most frequently reported as the cause of delay wound healing (6-9, 3). However, Trengrove et al., (10) reported that no single microorganism or group of organisms was more detrimental to wound healing than any other.

The following organisms are commonly associated with wound infection;*Streptococcus pyogenes*,*Staphylococcus aureus*,*Pseudomonas aeruginosa*., *Escherichia coli*,*Klebsiella*species, *Proteus* species,*Clostridium* species and *Bacteroides*fragilis, *Candida* species and *Aspergillus* species (1-3).
Wound infections cause great distress in terms of associated mortality and morbidity; increased length of hospital stay, delayed wound healing, profound discomfort and significant increased in healthcare cost (11).

Numerous reports exist in the literature regarding wound infection (1-3, 12-17). However, a reassessment of the etiology and antimicrobial susceptibility pattern of wound infection is necessary

for current management of this infection. This study focused on determining the spectrum of aerobic bacterial associated with wound infection in Benin City and their susceptibility to various antibacterial agents.

MATERIALS AND METHODS.

Study Population

A total of 207 wound specimens collected from patients attending the University of Benin Teaching Hospital were used for this study.The Ethical Committee of University of Benin Teaching Hospital approved the protocol for this study.

Specimen Collection and Processing

All specimens were collected using sterile swabs sticks. Specimens were processed according to the method previously described (19). Briefly, the swabs were streaked on the surface of Blood agar, MacConkey agar and incubated aerobically at 37^0C for 24hrs. Smears were prepared on slides and stained by Gram technique, and examined using 40x and 100x objectives for pus cells and bacterial. Emergent colonies from culture were identified.

Identification of Isolates

All bacterial isolates were identified according to the criteria described by Cowan and Steel (20). The criteria include colonial appearance, morphological characteristics as seen by staining and biochemical tests.

Antibacterial Susceptibility Test

The disc diffusion susceptibility test was performed according to the modified Bauer-Kirby method (21-22).

Determination of Beta Lactamase Production.

Beta lactamase production were determined using the iodometric tube method previously described (23).

RESULTS.

The results obtained in this study are shown in table 1-4. Table 1 shows the infection rate from processed specimen. Gender has no effect on wound infection rate. 22 (21.1%) of the processed specimen yielded no bacterial isolate.

A total of 278 bacterial isolates were recovered from various infected wounds, majority of the isolates were from males. *S. aureus* seems to be the most common isolate while *E. feacalis* is the least (Table 2).

All isolates were resistant to Ampicillin, Amoxicillin-clavulanate and tetracycline, while they showed variable susceptibility to other antibacterial agents (Table 3).

A total of 258 (92.8%) out of 278 isolates produced beta lactamase. Majority of the bacterial isolates produced beta lactamase (Table 4).

TABLE 1: INFECTION RATE FROM PROCESSED SPECIMEN.

Gender	No. tested (%)	No. with growth (%)	No. with mixed growth (%)	No. without (%) growth
Male	120	107(89.2)	55(51.4)	13(10.08)
Female	87	78(89.1)	38(48.7)	9(10.3)
Total	207	185 (89.4)	93(50.2)	22(21.1)

DISCUSSION

Infection in a wound delays healing and may cause wound break down, herniation of the wound and complete wound dehiscence (24). Therefore the knowledge of the causative agents of wound infection will be helpful in the control of wound infection and selection of empiric antimicrobial therapy as an infection control measure in hospital and community settings. This study was carried out to generate data that will be useful in the formulation of policy that will aid in the aforemention areas.

The results obtained in this study reveal that 185 (89.4%) out of 207 wounds swabs yielded growth with 50.2% being polymicrobic. The prevalence of high rate of wound infection as well as polymicrobic infection had also been reported by Shittu *et al.,* (9). Gender had no effect on wound infection rate.

TABLE 2: BACTERIAL ISOLATES FROM PROCESSED S

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Organisms	Gender		Total (%)
	Male (%)	Female (%)	
Staphylococcus aureus	43 (26.5)	32 (27.5)	75 (26.9)
Coagulase negative *Staphylococci*	10 (6.2)	4 (3.5)	14 (5.0)
Enterococcus feacalis	5 (3.1)	2 (1.8)	7 (2.5)
Escherichia coli	19 (11.7)	16 (13.8)	35 (12.6)
Klebsiellapnuemoniae	31 (19.1)	18 (15.5)	49 (17.6)
Proteus vulgaris	11 (6.8)	9 (7.8)	20 (7.2)
Proteus mirabilis	13 (8.1)	11 (9.5)	24 (8.6)
Providenciarettegeri	4 (2.5)	3 (2.6)	7 (2.5)
Psuedomonasaeruginosa	26 (16.1)	21 (18.1)	47 (16.9)
Total	162 (58.3)	116 (41.7)	278 (100.0)

TABLE 3: SUSCEPTIBILITY PATTERN OF BACTERIAL ISOLATES

Organisms	Amp. 10µg	Amx-cla 30µg	Amx 30µg	Cef 30µg	Tet 10µg	Gen 10µg	Cip 5µg	Ofl. 5µg
Staphylococcus aureus(75)	0	0	0	19(25.3)	0	22 (29.3)	28 (37.3)	47 (62.7)
Coagulase negative *Staphylococci* (14)	0	0	2 (14.2)	3 (21.4)	0	7 (50.0)	9 (64.2)	11 (78.5)
Enterococcus feacalis(7)	0	0	1 (14.2)	2 (28.5)	0	4 (57.1)	6 (85.7)	6 (85.7)
Escherichia coli (35)	0	0	0	9 (25.7)	0	10 (28.5)	22 (62.8)	30 (85.7)
Klebsiellapnuemoniae(49)	0	0	0	14(28.5)	0	18 (36.7)	27 (55.1)	32 (65.3)
Proteus vulgaris (20)	0	0	0	6 (30.0)	0	9 (45.0)	11 (55.0)	13 (65.0)
Proteus mirabilis (24)	0	0	0	7 (29.2)	0	10 (41.6)	12 (50.0)	16 (66.7)
Providenciarettegeri(7)	0	0	0	2 (28.6)	0	31 (42.9)	4 (57.1)	4 (57.1)
Psuedomonasaeruginosa(47)	0	0	0	12 (25.5)	0	14 (39.8)	23 (48.9)	29 (61.7)

Abbrevation: Amp- Ampicillin, Amx- Amoxicillin, Amx-cal – Amoxicillin-clavulanate, Cef- CefuroximeTet- Tetracycline, Gen- Gentamicin, Cip- Ciproloxacin, Ofl- Ofloxacin

A total of 278 clinical isolates were obtained from this study. *S. aureus* (26.9%) was the most predominant isolate in this study. This agrees with the reports of previous investigators (9, 15, 17, 25-26,). But does not agrees with the report of Thanni *et al* (18) who reported *S. aureus* as the second most common organism in their study. The other isolates in decresing order of prevalence were *K. pnuemoniae* (17.6%), *P. aeruginosa* (16.9%), *E. coli* (12.6%), *P. mirabilis* (8.6%), *P. vulgaris* (7.2%), coagulase negative *S. aureus* (5%), *E. feacalis* and *P. rettgeri* (2.5%) respectively. These isolates are common isolates found in wounds (9, 25). These isolates contribute to pathology of the wound infection, for example Streptococcal invasion of wound delays healing as well as results in deterioration of wounds (27). Pseudomonas spp, Enterococci spp, and Proteus spp are responsible for extensive tissue destruction with poor blood circulation to the affected site especially diabetic foot ulcer (28).

TABLE 4: NUMBER AND TYPE OF ISOLATES PRODUCING BETA LACTAMASE

ORGANISMS	NO TESTED	NO POSITIVE (%)
Staphylococcus aureus	75	69 (92.0)
Coagulase negative *Staphylococci*	14	13 (92.8)
Enterococcus feacalis	7	7 (100.0)
Escherichia coli	35	31 (88.6)
Klebsiellapnuemoniae	49	44 (89.8)
Proteus vulgaris	20	18 (90.0)
Proteus mirabilis	24	24 (100.0)
Providenciarettegeri	7	7 (100.0)
Psuedomonasaeruginosa	47	45 (95.8)
TOTAL	278	258 (92.8)

All isolates were resistant to ampicillin, amoxillin-clavulanate and tetracyline. The resistant observed for ampicillin and tetracyline could be due to their long period of use. But that of amoxicillin-clavulanate is surprising as the use of this drug is more recent than ampicillin and tetracycline. Susceptibility pattern of the bacterial isolates to other antibacterial agents varies.

Majority of the bacterial isolates in this study produced beta lactamase. This enzyme is used by microorganism to inactivate beta lactam antibacterials. This may explain the resistance observed for ampicillin, amoxicillina and amoxicillin-clavulanate. The fluroquinolones and gentamicin were more effective in this study. This agrees with the report of Mordi and Momoh, (15). These antibacterial should be use in the management of wound infection.

The variety of microorganisms observed in this study support the need to obtain culture specimen from infected wounds for microbiological evaluation and antibiotic susceptibility determination, so that adapted chemotherapy can be prescribed. This will not only facilitate successful wound management but also assist in the control of anatibiotic usage and hence stem the spread of antibiotic resistant bacterial. Continous dialogue between the microbiology department and wound care practitional is strongly advised.

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