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J. ANDHOGA, A.G. MACHARIA, I.R. MAIKUMA, Z.S. WANYONYI, B.R. AYUMBA and R. KAKAI

ABSTRACT

Background: The emergence of bacterial antimicrobial resistance associated with acquired infections has made the choice of empirical therapy more difficult and expensive, hence the need for continuous research to determine their sensitivity patterns.
Objectives: To identify the common aerobic pathogenic bacteria in post-operative wounds at Moi Teaching and Referral Hospital (MTRH) and determine their sensitivity patterns to routine antibiotics used.
Design: Cross-sectional study.
Setting: Moi Teaching and Referral Hospital (MTRH) Surgical, Obstetrics and Gynaecology wards.
Patients: Sixty three patients chosen by convenient sampling between May and June 2001 were included in the study. A total of eighty four isolates were obtained from these patients.
Materials and Methods: Isolation of pathogens and their antibiotic sensitivity determination was conducted in the Department of Medical Microbiology Laboratory, Faculty of Health Sciences, Moi University. The bacteria were cultured on blood agar, MacConkey and Nutrient agar followed by identification using biochemical tests (catalase urease, coagulase, triple sugar iron). Disc diffusion method on Muller-Hinton agar was used for sensitivity testing.
Results: Staphylococcus aureus species were the most common pathogenic bacteria isolated from the post-operative wounds. Other organisms included _Proteus, Pseudomonas_ and _Escherichia coli._ There was a multi-drug resistance pattern observed, with minocyline being the most effective drug against _S. aureus._
Conclusion: There is need for review of policies on prescription practice on the use of existing therapeutics choice to limit the spread of resistance. This will ensure reduced patient stay and cost incurred.

INTRODUCTION

Background and rationale. The emergence of bacterial antimicrobial resistance associated with acquired infections has made the choice of empirical therapy more difficult and expensive. Bacteria have a remarkable ability to develop resistance to many of the antimicrobial agents but appropriate antibiotics will delay and in many cases prevent the emergence of resistance(1). This has remained a global problem that calls for continuing research to identify these strains and hence proper drug use to combat the infections. Like any other wound, post-operative wounds have remained a conducive environment for growth of different bacteria, most of which are acquired during the post-operative period. Knowledge of the common pathogen and their resistance patterns at any facility are vital for both nursing and medical purpose. Though post-operative wounds are expected to be clean, this is not the case in our set-up. The wounds becomes infected making patients stay in the wards longer than expected. This could be due to poor nursing care or wrong antibiotics prescription. Furthermore a range of antibiotics are used to fight the infection without knowing the specific drug(s) that are effective against the incumbent organisms. With the use of wide range of antimicrobials, the chances of bacteria developing resistance are increased. The wounds could therefore become septic as they fail to respond to the treatment. Though the methods of resistance to antimicrobials vary, the ultimate result is ineffective use of drugs without achieving the desired results.

This study aimed at identifying the common aerobic pathogenic bacteria infecting post-operative wounds at Moi Teaching and Refferal Hospital and ultimately determines their antibiotic sensitivity patterns. It has not only increased the knowledge base on the pathogens to anticipate in post-operative wounds, but will aid clinicians in choosing appropriate prophylactic and therapeutic antibiotics. Major reasons for conducting antibiotic sensitivity testing are to improve the quality of patient care, to limit the emergence of resistance and to contain costs(2). The study will thus form a basis for further research into the issue with the aim of formulating appropriate prescription policies at the facility.

MATERIALS AND METHODS

Study area: The study was carried out at the Moi Teaching and Referral Hospital (MTRH) Surgical, Obstetrics and Gynaecology wards.

Study population: The study included all postoperative patients in the surgical, obstetrics and gynaecology wards at Moi Teaching and Referral Hospital.

Study design: This was a cross sectional study.

Inclusion criteria: All post operative patients in the surgical, obstetrics and gynaecology wards. Permission to carry out the study was obtained from the management of the MTRH and the Institution Research and Ethics Committee of the Faculty of Health Sciences, Moi University. Verbal informed consent was obtained from the patient before specimens could be collected from him or her and all information collected on each individual was held in confidence.

Sampling and sample size: Convenient sampling was used due to time limitation and the nursing care being given for each individual patient. Samples were only obtained from the patient at the time of dressing wounds. Every week new patients were enrolled and those previously sampled were excluded. Sixty three patients were sampled and a total of 84 isolates obtained (some patients had more than one type of organism isolated from the wounds).

Variables and measurements: Interviews and hospital records were used to obtain bio data, information on existing chronic disease, previous and current drug use and past medical history. Physical examination of the patients was used to determine the location of the wound and its level of contamination (Table 1), while the body-mass index was used to determine the nutritional status (Table 2). Antibiotic sensitivity patterns were determined using disc diffusion method on Mueller-Hinton agar.

Laboratory procedures: Culturing for colony characteristics followed by Gram staining and biochemical tests was used to identify the pathogenic bacteria. The culture media used were Blood agar, MacConkey agar, Nutrient agar and Triple Sugar Iron (TSI) agar. Culture media was prepared by reconstituting commercial powder in distilled water and sterilizing at 121°C for 15 minutes in an autoclave as per the manufactures instructions. Pus swabs were collected by use of sterile cotton wool swabs, then transported to the laboratory immediately. They were then inoculated on appropriate culture media and incubated aerobically at 37°C for 24 hours. After 24 hours, another gram stain from discrete colonies growing on media was made. Biochemical tests were performed to identify the

Table 1

Operational definitions of level of contamination of post-operative wounds

Level of contamination	Description
Clean	No physical features of inflammation of tissues involved. Wound is dry with no damage.
Clean-contaminated	At least three physical features of inflammation of tissues present involved. Draining serous fluid but no pus.
Heavily contaminated	Draining pus but no secondary colour changes of tissues and the skin, no foul smell and no involvement of surrounding tissues such as bone, muscle.
Septic	Draining pus, secondary changes of tissues and the skin (for example tissue green in colour), foul smelling and invoving the surrounding tissues such as bone, muscle

Table 2

Distribution of patients by age (years), nutritional status and location of wound

Age in years	*Frequency (%)*
0-2	6(9.5)
3-12	7(11.1)
13-24	25(39.7)
25-59	21(33.3)
60 and above	4(6.4)
Total	63(100)
Nutritional status	*Frequency(%)*
Poor	1(1.6)
Fair	28(44.4)
Well nourished	(34(54.0)
Total	63(100)
Location of wound	*Frequency(%)*
Back	2(3.2)
Thorax	4(6.3)
Perineum	7(11.1)
Abdomen	13(20.6)
Head and neck	15(23.8)
Limbs	22(34.9)
Total	63(100)

Well nourished (BMI = 18.5–25), Fair (BMI = 16–18.5 or 25–39), Poor (BMI = below 16 or over 30). % = percent, BMI = Body Mass Index specific bacteria. The tests included catalase and coagualase test for gram positive cocci to identify *S. aureus*. Gram-negative bacilli were inoculated in TSI agar and incubated for a further 24 hours. Isolates were identified based on characteristic colonial morphology, gram stain and biochemical reactions.

After identifying the bacteria, they were inoculated onto Mueller-Hinton agar spread evenly using a sterile cotton wool swab. Sensitivity discs for appropriate drugs were placed onto the media and incubated at 37°C for 24 hours. Commercial antibiotic discs by Himeda laboratories Ltd with different antibiotic regimen of varying concentrations were used both for gram positive (Combi 30) and gram negative (Combi 35) bacteria (Table 3 and 4). After incubation, the zone of inhibition diameter was measured using a transparent ruler and compared with that of a control organism. The zones were interpreted according to their diameters as their sensitive or resistant.

Table 3

Sensitivity pattern of gram negative bacteria to various antibiotics

Antibiotic (mcg)	*E. coli* n = 2 (% sensitive)	*Pseudomonas* n = 10 (% sensitive)	Proteus n = 13 (% sensitive)
Gentamicin(10)	2(100)	6(60)	8(61.5)
Ampicilin (25)	0	0	2(15.3)
Tetracycline(100)	1(50)	8(80)	9(69.2)
Co-trimoxazole(25)	0	0	2(15.3)
Sulphamethoxazole(200)	1(50)	5(50)	8(61.5)
Nalidixic acid(30)	2(100)	4(40)	9(69.2)
Nitrofurantoin(200)	1(50)	5(50)	8(61.5)

Octodics by Himedia Laboratories Ltd (Combi 35)

mcg = micrograms, % = percent, n = number

Table 4

Sensitivity pattern of Staphylococcus aureus to various antibiotics

Antibiotic (mcg)	*S. aureus* n = 46 (% sensitive)
Ampicillin (10)	10(21.7)
Chloramphenicol (30)	7(15.2)
Minocycline(30)	32(69.6)
Erythromycin (15)	9(19.6)
Penicillin (1 IU)	2 (4.3)
Lincomycin (2)	7(15.2)
Methicillin (5)	9(19.6)
Cotrimoxazole(25)	7(15.2)

Octodiscs by Himedia Laboratories Ltd (Combi 35)

mcg = micrograms, % = percent, n = number

Data collection, management and analysis: The bio data, past medical history and prior drug use were obtained from patient's hospital records. The site of the wound, level of contamination of the wound, the duration of the wound and the nutritional status of the patient were obtained by physical examination of the patients (Table 2). Using gram staining techniques and biochemical test the bacteria were identified and broadly classified. Due to limited budgetary allocations, serotyping of the isolated bacteria to specific strains was not performed. The different identified bacteria were then tested for sentivity to various antibiotics such as penicillin, streptomycin and gentamicin. A data collection form was used to record the variables and labaratory results of each patient. Data was entered into computerised data base using the computer software statistical package for Social Sciences (SPSS Version 10.0 for windows). Data was anlysed by Chi-square test. Further analysis involved frequencies and cross tabulations with their associations.

RESULTS

A total of 63 swabs were obtained from the post-operative patients. Table 2 shows the distribution of the patient by their age in years. Most of the patients were aged between 13-24 years (39.7%). The patients were either well or fairly nourished using the body mass index (BMI). Only one patient was poorly nourished.

The presence of chronic debilitating disease was sought, and up to 78.6% of the patients had no history of diabetes mellitus, hypertension, malignancies or any other disease that could clinically be classified as chronic. However, 13 patients (20.6%) had malignancies and three patients (4.8%) were diabetics. Specimens were drawn from post-operative wounds located on various anatomical parts of the body (Table 2). Of the 84 isolates, 22(26.2%) were from heavily contaminated wounds, 21 (25%) from septic, while 16 (19%) and 25 (29.2%) were from slightly contaminated and clean wounds respectively.

The mean duration of the patients in the ward before surgery was 2.5 days and the average length of stay in the hospital after surgery was 7.8 days. Out of 84 isolates, 54.7% were *S. aureus* ($P<0.05$), while *proteus Pseudomonas* and *E.coli* were 15.5%, 11.95 and 2.3% respectively. Fifteen percent (15.5%) of specimens did not have any growth.

The association between the level of contamination and the pathogens was thus; of the clean wounds 48% of isolates had no growth obtained while 44% were *S. aureus* and 8% were *Proteus*; of clean contaminated 75% were *S. aureus, Pseudomonas* 21.5% and Proteus 6.3% while no organisms were found in 6.2% of the wounds in category; of the heavily contaminated 59% of the isolates were *S. aureus*, 22.5% were *Proteus,* 13.6% were *Pseudomonas* and 4.5% were *E.coli*; of septic wounds 47.6% were *S. aures, Pseudomanas* and

Proteus were 23.8% each while *E.coli* was 4.8%. Most patients could not recall previous use of antibiotics and no proper records were kept to this effect. This variable was therefore not available or analysis. The drugs commonly used as prophylaxis for post-operative patients at MTRH were Ampiclox (ampicillin and cloxacillin), augmentin (amoxicillin and clavulanic acid), gentamicin, ampicillin and metronidazole. The sensitivity pattern to various antibiotics was established (Table 3 and 4). *S. aureus* was sensitive to cotrimoxazole (15.2%), chloramphenicol (15.2%), methicillin (19.6%), ampicillin (21.7%) and minocycline (69.6%). Most *Pseudomonas* strains were sensitive to tetracycline (80%) and gentamicin (60%), but there was 100% resistance to ampicillin and contrimoxazole. All *E. coli* isolates were sensitive to gentamicin and nalidixic acid but resistant to ampicillin and cotrimoxazole.

DISCUSSION

This study demonstrates the pattern of post-operative wound infection from a referral hospital in a developing country, which is similar to patterns seen elsewhere(3). From the study the most predominant organisms isolated were *S. aureus* (54.7%), *Proteus* (15.5%), *Pseudomonas* (11.9%) and *E.coli* (2.3%). This confirms that *S. aureus* is the most prevalent gram-positive aerobic bacteria infecting post-operative wounds at MTRH(1,3). Thirteen specimens had no growth obtained indicating either lack of infection or they need special techniques to identify the pathogens. The sensitivity patterns of both *S.aureus* and gram-negative showed multiple resistance to commonly used antimicrobials. *S.aureus* was most sensitive to minocycline (69.6%) followed by ampicillin (21.7%). methicillin (19.6%), cotrimoxazole and chloramphenicol 15.2%. The resistance could be due to indiscriminate use of antibiotics both before admission to the hospital and while in the wards(4). Findings that 80.4% of *S. aureus* were resistant to methicillin (MRSA) is disturbing. This implies that the strain will be resistant to penicillins and cephalosporins. This type of resistance is also conferred to other beta-lactams such as imipenem(5-7). *Pseudomonas* was susceptible to most drugs except ampicillin and cotrimoxazole. *Proteus* displayed a mixed pattern of sensitivity, being least sensitive to ampicillin (15.3%) and contrimoxazole (15.3%). All *E. coli* isolates were sensitive to gentamicin and nalidixic acid but resistant to ampicillin and cotrimozaxole. Resistance to penicillins could be due to beta lactamase production as shown in a study done at Kenyatta National Hospital, Nairobi(3). The commonly used drugs at MTRH, which were available on our test kit, were gentamicin, ampicillin, tetracycline and amoxicillin. Of these drugs, gentamicin showed activity against most strains of *E. coli, Pseudomonas* and *Proteus*. Ampicillin was not effective against most organisms, with sensitivity at 21.7% for *S. aureus*. Cotrimoxazole, penicillin and chloramphenicol also showed low sensitivity patterns.

The results of this study should be interpreted with caution. Poor response to drug may indicate that the drug is not reaching the organism or a large dose of it is required. This is evidenced by the fact that most of the patients enrolled in this study eventually recovered and were discharged despite the levels of *in-vitro* resistance to drugs displayed. It would therefore be premature to generally conclude that the regimes used at MTRH are ineffective. As such the need for more sensitive tests such as the determination of minimum inhibitory concentrations (MIC) should be advocated, to establish the effective dosages required to arrest bacteria growth. Similarly, although this information is useful in guiding the choice of drug, the response of the patient to the drug is the ultimate test of effectiveness of the drug by the clinician(8). Nevertheless the best drug should be selected according to its characteristics and search for a better regimen to treat the infection is mandatory(9). Successful treatment of *S. aureus* will definitely require knowledge of its antimicrobial resistance(10).

In conclusion while appreciating the limitations of the study, there is compelling evidence suggestive of multiple resistance in post-operative wound infections. This could be indication of widespread antibacterial use resulting into selective resistance to certain strains of bacteria(11). Emergence of resistant strains has an impact on the population as it increases morbidity and mortality rates, reducing desired treatment outcomes and prolong hospital stay, while increasing the cost of patient care(12). Urgent need to review policies on prescription practices at any facility should then be put in place, as is the need to develop effective policies on the use of existing therapeutic choices in the most appropriate manner(8,13). A combination of antimicrobials to address multiple resistance is invaluable in current practice.

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ANNOUNCEMENT

KENYA MEDICAL ASSOCIATION 31ST ANNUAL SCIENTIFIC CONFERENCE AND ANNUAL GENERAL MEETING

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– Mombasa Division

Venue: Travellers Beach Hotel, Mombasa

Date: 23rd – 27th April, 2003

Theme: Current Trends in Treatment and Prevention of HIV/AIDS and other Diseases in sub-Saharan Africa

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