Antibiotic Sensitivity Patterns of Aerobic Bacterial Agents in Post-Surgical Orofacial Infections

Barasa GB[1], Butt F[1], Onyango JF[1], Mutua FM[2], Dimba E[1]

1. School of Dental Sciences, University of Nairobi
2. School of Medicine, University of Nairobi

Correspondence to: Dr. Godfrey Bwire Barasa, P.O Box 66994-00200 Nairobi, Kenya. Email: bwire22@yahoo.com

Abstract

Background: There is no data on the bacteriology of postsurgical orofacial infections at our institutions. Uncontrolled use of antibiotics may lead to development of resistance with the resultant increase in morbidity and cost of treatment. We aimed to determine the aerobic bacterial agents and antibiotic sensitivity of post-surgical infections in the orofacial region. **Methodology**: Patients were evaluated for post-surgical wound infection from the 5th to 30th post-operative day. In cases where a surgical implant was placed the period of review was extended for up twelve months. The specimens were collected using sterile swabs and transported to the microbiology laboratory within two hours of collection. The specimens were then analyzed for bacteriology according to the standard bacteriological techniques. A wide range of antibiotics including those commonly used to treat orofacial infections were tested for sensitivity against the isolates obtained using the disk diffusion test (Kirby-Bauer procedure, using CLSI protocols). **Results**: Staphylococcus aureus accounted for 40% of the isolates followed by Klebsiella species (23%) and the Pseudomonas species (19%). Amoxycillin/clavulinic acid, the 2nd and 3rd generation cephalosporins were effective against most of the bacterial infections from the orofacial region. **Conclusion**: Staphylococcus aureus, Klebsiella, and Pseudomonas species are the commonest isolates from the oral facial region. Antibiotics which showed adequate efficacy against them were the augmented Penicillins and newer generations Cephalosporins.

Key Words: Oro-facial infection, Post-surgical, Antibiotics

Introduction

Although antibiotics have played a major role in the treatment of infection, uncontrolled use has led to alarming rates of development of resistance with a resultant increase in morbidity and cost (1). One area where antibiotic use is often applied is in the control of post-operative wound infection. Although most prescriptions are usually on an empirical basis, regular culture and sensitivity tests should be undertaken to give guidelines on antibiotic regimes reasonable for empirical treatment as well as the updates on the prevailing sensitivity and resistance patterns within specific healthcare institutions. The ideal drug is that which is safe for that specific patient, specific to that infecting microorganism, readily available, affordable and can be given in a reliable and convenient form. The widely used Centre for Disease Control (CDC) and Prevention criteria define surgical site infections (SSIs) as infections related to the operative procedure that occurs at or near the surgical incision within 30 days of an operative procedure or within one year if an implant is left in place. The clinical criteria used to define SSI include any of the following: a purulent exudate draining from a surgical site, a positive fluid culture obtained from a surgical site that was closed primarily, the surgeon's diagnosis of infection and a surgical site that requires reopening (2–4). Wound infection refers to the presence of replicating microorganisms within a wound that cause host injury. Features of an infected wound include increased exudate, swelling, erythema, pain, increased local temperature and peri-wound cellulitis (4). The aim of the study was to determine the aerobic bacterial agents and antibiotic

sensitivity of post-surgical infections in the orofacial region at our institution.

Methods

This was a descriptive cross-sectional study with clinical and laboratory components that was conducted between May and July 2011. The clinical part of the study was conducted at the Kenyatta National Hospital (KNH) in the Dental, Ear, Nose and Throat (ENT) clinics, Maxillofacial and ENT wards and other departments treating patients with orofacial surgical conditions as well as the University of Nairobi Dental Hospital (UNDH). The two institutions are national referral and teaching hospitals in Kenya. The laboratory analysis was conducted at the University of Nairobi (UON) Medical Microbiology laboratory.

We recruited patients undergoing orofacial surgeries and had no infection in the immediate pre-surgical period. Post operatively those who had surgical drains placed were also included. The exclusion criteria were presence of immunosuppression due to HIV-infection characterized by AIDS defining conditions, uncontrolled diabetes mellitus or corticosteroid therapy for more than three months in the period prior to the study and cancer chemotherapy. The sample size of the population was 65 patients, calculated using the prevalence of postsurgical infections following third molar surgery of 4% where the minimum sample size was 59.

Patients were evaluated for wound infection from the 5th post-operative day, when the clinical features were highly likely evident, up to the 30th postoperative day unless a surgical implant was in situ, when the period was extended to up to a year. Infected surgical sites or wounds were those exhibiting pus or any three of the following signs and symptoms increased exudate, swelling, erythema, pain, local temperature and peri–wound cellulitis. The specimens were taken using sterile swabs (Hardwood Properties, USA) and transported to the microbiology laboratory within 2 hours of collection in order to optimize on the yield of cultures, avoid overgrowth of some microorganisms, desiccation of the sample or the death of more fastidious ones. Patient details with special reference to antibiotic history were recorded accordingly.

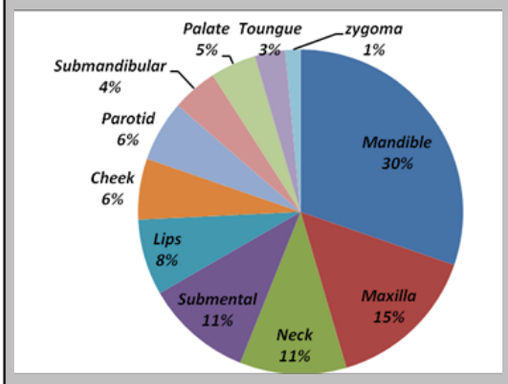
Bacteriological processing was done using the Standard UON/KNH microbiology operating procedures. All cultures were processed by Gram stain, colony morphology and biochemical tests such as catalase, oxidase and coagulase. The specimens were inoculated on MacConkey‟s agar (Oxoid Ltd, England) and Blood agar, prepared from horse blood and Mueller-Hinton agar (Oxoid Ltd, England) on the standard petri-dishes (IsoLab.GmbH).The plates were then examined for selective growth of organisms. The isolated organisms were then stained by Gram's method, identified by colony characteristics such as morphology, pigment production and ß-haemolysis in blood agar. The cultured organisms were then tested for their susceptibility against various commercially prepared antibiotics by disk diffusion methods (Kirby-Bauer procedure). The test was performed by swabbing a standardized inoculum of bacteria onto a Mueller-Hinton agar (Oxoid Ltd, England) plate. The selection of antibiotics for susceptibility testing was determined by the type of isolated organisms and the KNH/UON laboratories formulary based on the Clinical Laboratories Standards Institute (CLSI) guidelines (5). Antibiotic susceptibilities were determined by measuring the diameter of the zone of inhibition in millimeters for each of the different antibiotic disks. These were then converted to susceptible, intermediate or resistant using a table from the CLSI guidelines (2011) and results copied onto appropriate data sheets (5).The performances of the antibiotic disks and all bench procedures were internally quality controlled using Staphylococcus aureus: ATCC25923 and Pseudomonas aeruginosa: ATCC27853 control strains. Some 8% of all cultures were processed in duplicates, in another laboratory (KNH microbiology laboratory) to check the reproducibility of the results.

The study was approved by the Kenyatta National Hospital and University of Nairobi ethics, research and standards committee.

Results

Sixty-five specimens from 65 patients were analysed. Of the 65 study participants 36(55%) were male, while 29(45%) were female among whom 33(51%) were in-patients and 32(49%) out-patients with an age range of 4 to 71years (mean=39yrs). The types of surgical treatments ranged from resection of benign (43%) or malignant (23%) lesions, repair of maxillofacial soft and hard tissues caused by trauma (18.5%) including those involving the dentoalveolar (15.5%) area (Figure 1). The anatomic regions where the specimens were taken for culture and sensitivity were mandibular (30%), maxillary (15%), cervical (11%) and the submental (11%) (Figure 2).

Figure 1. Oral and Maxillofacial areas of specimen collection



Of the specimens cultured, 52(80%) had growth on culture while 13(20%) had no growth. Overall, 43(66%) of the specimens grew pure cultures while 9(14%) grew mixed cultures. There were five different bacterial agents isolated including Staphylococcus aureus which comprised 25(40%) of the isolates, while the Klebsiella and Pseudomonas species formed 14(23%) and 12 (19%) of the isolates respectively. Others were Proteus mirabilis 7(11%), Streptococcus pyogenes 3(5%) and Escherichia coli 1(2%) as indicated in (Table 1). There were nine mixed growths isolated, with the most frequent combination being Staphylococcus aureus with Klebsiella species at 67 %, followed by Pseudomonas species with Proteus mirabilis (22%) and Staphylococcus aureus with Proteus mirabilis (11%).

All the Staphylococcus aureus isolates were susceptible to vancomycin. There was high susceptibility to cefotaxime (90%), cefuroxime (85%) and amoxycillin/clavulinic acid (85%) with the least susceptibility to ampicillin (25%). Oxacillin resistance was noted in 8% of the isolates indicating the presence of methicillin resistant Staphylococcus aureus (MRSA) (Figure 2). Isolates resistant to oxacillin or methicillin were all interpreted as having been resistant to all ß-lactam agents including cephalosporins, as per the CLSI 2011. guidelines. All the 14 Klebsiella species isolated showed highest susceptibility to meropenem (100%), followed by imipenem (93%), levofloxacin and ceftriaxone and gentamicin (86%), cefuroxime (79%)and the least to amoxycillin/clavulanic acid(57%). The isolated Proteus mirabilis (n=7) were all susceptible to imipenem and cefuroxime. There was high susceptibility to meropenem, levofloxacin, ceftriaxone and cefotaxime(86%) and least to gentamicin (43%). There was only one Escherichia coli isolate which was susceptible to all the other antibiotics except ceftriaxone to which it was resistant. Pseudomonas isolates (n=12) were highly resistant to amoxycillin/clavulinic acid with only 7% susceptibility. They were, however, not tested against cefuroxime and ceftriaxone. Their susceptibilities to other tested drugs were as follows: amikacin 75%, cefepime 83%, ceftazidime 92% and ticarcillin/clavulinic acid 83% (Figure 3).

Table 1: Aerobic Bacterial Isolates

Isolates	**Frequency**	**Percentage (n=62)**
Gram negative micro-organisms		
Pseudomonas ssp	12	23%
Klebsiella ssp	14	23%
Proteus mirabilis	7	11%
Escherichia coli	1	2%
Gram –positive micro-organisms		
Staphylococcus aureus	25	40%
Streptococcus pyogenes	3	5%

Figure 2. Antibiotic susceptibility for *Staphylococcus aureus* (n=25)

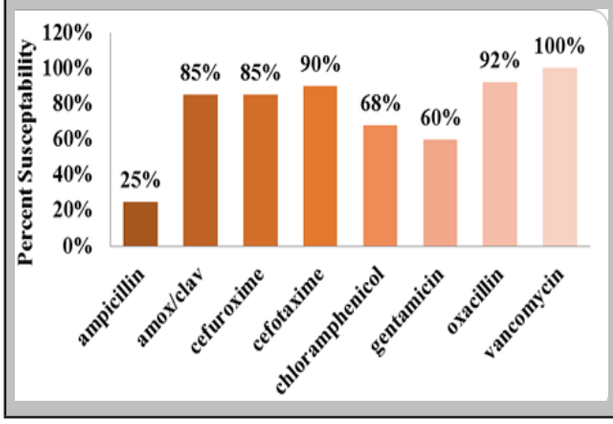
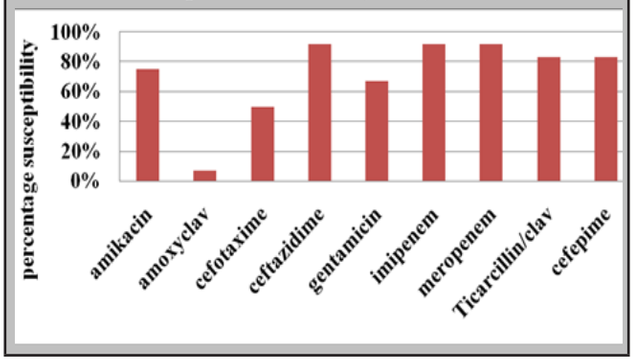


Figure3. Antibiotic susceptibility for the *Pseudomonas species* isolates (n=12)



Discussion

Identification of the organisms causing post-surgical oro-facial infections may be used to optimise patient management, minimise postoperative complications, shorten post-operative hospital stay and consequently reduce the cost of healthcare. The additional information gained about the susceptibility profile of the isolated bacteria may be used to enrich the existing knowledge on antibiotic use. The isolation of bacteria from clinical samples yields useful information that is translated directly into therapeutic strategies for the patients (1). The results of this study demonstrate the polymicrobial nature of post-operative orofacial infections. All hospital in-patients were given a five-day intravenous antibiotic course and upon discharge prescribed an additional five days of oral antibiotics. However, not all these patients left the hospital immediately, with some staying for over a month after discharge due to inability to settle the hospital expenses. The patients had been evaluated as from post-operative day 5 when clinical features of infection were most likely evident. A wound that yields pathogens on microbiological sensitivity with no clinical evidence of infection does not justify antibiotic therapy, supporting the aphorism of "treat the patient, not the microbiology swab" (5). Several studies have shown that surgical site infections (SSI) represent most hospital-acquired infections with the major impact being on average hospital stay and cost of hospitalization (2). A history of antibiotic prophylaxis and surgical drains were not considered reasons for exclusion from the study. Another had found no statistically significant relationship between post-surgical infection and the use of pre-operative antibiotics (6).

In this particular study, the drains had been removed by the 4th post-operative day, hence minimizing the risks of retrograde infection to less than 2% (6–9). In 20% of the specimens, no growth was found. This is much less than Fehr et al, who, in an observational study, in Tanzania, on patients with SSIs, had found that 35% had cultures that yielded no growth or "no clinically significant organism"(5,10). It emerged that most (40%) of the isolates were Staphylococcus aureus, 23% having been Klebsiella species and 19% Pseudomonas species. Despite the difference in the antibiotics used in post-operative care, the study results concurred with those of that the Staphylococcus aureus and Pseudomonas species were the commonest infections of postorofacial procedures with reports of 28% and 12% of the infections having been diagnosed of the two respectively (11). It also partially compared with the Bratzler, et al conclusions that Staphylococcus aureus was consistently the leading cause of nosocomial infections including SSIs and the incidence of methicillin-resistant Staphylococcus aureus (MRSA) strains was rising dramatically. A study done in Tanzania also showed that Staphylococcus aureus was the most common isolate followed by E Coli and the Klebsiella Species (12). In the present study, all the Staphylococcus aureus isolates were susceptible to vancomycin and highly susceptible to cefotaxime (92%) possibly because the use of these antibiotics in KNH is limited. The susceptibility to amoxycillin/clavulinic acid and cefuroxime was still high (85%). In view of the high resistance rates of the isolates to ampicillin, gentamicin and chloramphenicol, empirical treatment of Staphylococcus aureus infections at our hospital with these antibiotics may not be effective. There was 8% oxacillin resistance compared to 5.3% by Askarian et al (13). This poses a major problem in the treatment of Staphylococcus aureus infections because the isolates resistant to oxacillin or methicillin are all interpreted as resistant to all beta-lactam agents including cephalosporins as these drugs are known to be ineffective against the MRSA following therapeutic corrections. Understanding of the genetic basis for methicillin resistance has advanced significantly in the last few years. So far, Staphylococcal Cassette Chromosome mec (SCCmec) elements are the only vectors that have been described for the mecA gene encoding resistance in staphylococci, therefore, polymerase chain reaction (PCR) testing would be necessary to confirm that the MRSA strains isolated in our study were mecA gene-positive (13,14). The antibiotic sensitivity testing revealed that Pseudomonas isolates were highly resistant to amoxycillin/clavulinic acid and gentamicin with 7% and 43% susceptibility respectively. Antipseudomonal antibiotics such amikacin, ticarcillin/clavulinic acid were found to be suitable for routine use with sensitivities of 75% and 83% respectively. Ceftazidime showed 92% susceptibility, close to imipenem and meropenem that showed 93% sensitivity probably due to their limited use that may be attributed to their high cost. With the widespread use of antibiotics, Pseudomonas aeruginosa has become a leading cause of gram negative bacterial infections especially in patients who need prolonged hospitalization

(15). Based upon the current sensitivity patterns at our institution, the use of a third generation cephalosporin or a cabapenem is most likely to give good response if monotherapy is applied. All the Klebsiella species isolated were susceptible to meropenem. The susceptibility to imipenem stood at 93%, to levofloxacin, ceftriaxone and gentamicin at 86%, to cefuroxime at 79%.The least was to amoxiclav at 57%. The isolated Proteus mirabilis were all susceptible to imipenem and cefuroxime. There was high susceptibility to meropenem, levofloxacin, ceftriaxone, and cefotaxime at 86%. It was least susceptible to gentamicin at 43%. There was only one Escherichia coli isolate, and it was susceptible to all the other drugs it was tested gainst except ceftriaxone to which it was resistant. The quinolone, levofloxacin was more effective against the enterobacteriaceae than the penicillins and most cephalosporins. The differences in efficacy between various 2nd and 3rd generation cephalosporins appeared negligible making the choice between a 2nd and 3rd generation cephalosporin to be probably dictated by availability or cost. For many patients, a 2nd or 3rd generation cephalosporin combined with levofloxacin or metronidazole may be adequate. Cephalosporins within the same group generally showed similar sensitivity trends which compared with findings in a prospective comparative trial involving ceftriaxone and ceftazidime (16). When tested against meropenem and imipenem, all the gram negative isolates showed high susceptibility probably because they still retain activity against the chromosomal cephalosporinases and extended-spectrum beta-lactamases found in many gram-negative pathogens. Though, low in our study, the emergence of carbapenem-hydrolyzing betalactamases threaten the clinical utility of this antibiotic class, bringing us a step closer to the challenge of "extreme drug resistance" in gram-negative bacilli. The optimal therapy for treatment of infection due to carbapenemase-producing organisms is not known, and the antibiotic options are limited (17).

Conclusions

Staphylococcus aureus, Klebsiella and Pseudomonas species were the main types of bacteria isolated in the infected surgical site wounds from the oral facial region. The augmented penicillins and newer generations of cephalosporins showed adequate efficacy against them. However, Meropenem and imipenem remained highly effective against various types of bacteria form the Oral facial region.

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