AEROBIC BACTERIAL AGENTS AND ANTIBIOTIC SENSITIVITY OF POST-SURGICAL OROFACIAL INFECTIONS AT THE KENYATTA NATIONAL HOSPITAL AND UNIVERSITY OF NAIROBI DENTAL HOSPITAL

GODFREY BWIRE BARASA.

MDS-OMFS

THIS DISSERTATION IS SUBMITTED IN PARTIAL FULFILMENT FOR THE **MASTER OF DENTAL SURGERY** DEGREE IN **ORAL & MAXILLOFACIAL SURGERY** OF THE **UNIVERSITY OF NAIROBI**

2011

DECLARATION

This is my original work and has not to my knowledge been presented for a degree in any other university.

Signed.....Date.....19/10/2011.....

Dr Godfrey Bwire Barasa

(Candidate)

This dissertation was supervised and has been submitted for examination with my approval.

PROF. J F ONYANGO

Associate professor and Chairman, Department of Oral & Maxillofacial surgery, Oral Medicine &pathology,

University of Nairobi

Signed..... Date...19/10/2011.....

DR FLORENCE M. MUTUA

Lecturer, Department of Medical Microbiology

University of Nairobi

Signed..... Date.....19/10/2011.....

DR ELIZABETH DIMBA

Lecturer, Department of Oral & Maxillofacial surgery, Oral Medicine &pathology,

University of Nairobi

Signed..... Date.....19. 10. 2011.....

DR FAWZIA BUTT

Consultant Oral & Maxillofacial surgeon and Lecturer,

Department of Human Anatomy,

University of Nairobi

Signed..... Date.....19-OCT-2011.....

DEDICATION

This work is dedicated to my wife Makena for her patience and support throughout the study. I also dedicate it to my parents Mr. and Mrs. Barasa and my daughters Nekesa, Karimi and Neema.

ACKNOWLEDGEMENTS

I would like to thank my supervisors, Prof J F Onyango, Dr Florence Mutua, Dr Elizabeth Dimba and Dr Fawzia Butt for continued guidance and assistance during this study. I also appreciate the contributions of Prof Gunturu Revathi, Prof. Omu Anzala and Prof Symon Guthua towards the completion of this study.

Finally, special thanks to Mr Jonathan Oloo and Mr Amos Momanyi for kindly accepting to offer laboratory and statistical services respectively.

LIST OF ABBREVIATIONS

AIDS: Acquired Immunodeficiency Syndrome

ATCC: American Type Culture collection

CDC: Centres for Disease Control

CLSI: Clinical Laboratories Standards Institute

C/S: Culture & Sensitivity

ENT: Ear, Nose & Throat

KNH: Kenyatta National Hospital

MDS: Master of Dental Surgery

MRSA: Methicillin-Resistant Staphylococcus aureus

OMFS: Oral & Maxillofacial Surgery

SPSS: Statistical Method for Social Sciences

SSI: Surgical site infection

TMJ: Temporomandibular Joint

UNDH: University of Nairobi Dental Hospital

UON: University of Nairobi

WHO: World Health Organisation

DEFINITION OF TERMS

Clean contaminated: a wound involving normal but colonized tissue.

Contaminated wound: a wound containing foreign or infected material.

Oro-facial region: region encompassing the face, upper neck, jaws and related structures.

Surgical drain: a tube used to remove pus, blood, or other fluids from a wound.

Surgical implant: a medical device used to replace a missing biological structure, or support a damaged biological structure or enhance an existing biological structure.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ABSTRACT

Main objective: The aim of this study was to determine the aerobic bacterial agents and antibiotic sensitivity of post-surgical infections in the orofacial region.

Methodology: clinical case evaluation and laboratory investigation of microbial sensitivity to antibiotics.

Clinical methods: Patients were evaluated for post-surgical wound infection from the 5th post-operative day up to the 30th post-operative day unless a surgical implant was in situ when the period was extended to up to a year. The specimens were collected using sterile swabs and transported to the microbiology laboratory within 2 hours of collection.

Laboratory methods: The specimens were then analysed 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 (Bauer-Kirby procedure, using CLSI protocols).

Results: Both gram-negative and positive bacteria were isolated. *Staphylococcus aureus* formed 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 bacteria. Enterobacteriaceae showed high susceptibility to levofloxacin and carbapenems. The Pseudomonas species was highly susceptible to the 3rd generation cephalosporins and carbapenems.

Conclusion: The bacteria isolated in the infected surgical site wounds in the oro-facial region are *Staphylococcus aureus,* Klebsiella species, and Pseudomonas species with augmented penicillins and newer generations of cephalosporins still being effective against them.Cabapenems remained highly effective against a large variety of bacteria.

CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Although antibiotics have played a major role in the treatment of infection, uncontrolled use has led to alarming rates of development of resistence with a resultant increase in morbidity and cost.[1,2] The widely used Centres for Disease Control and Prevention (CDC) 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.[3, 4, 5] Wound infection refers to the presence of replicating micro-organisms within a wound that cause host injury. Features of an infected wound include increased exudate, swelling, erythema, pain, local temperature and peri -wound cellulitis. [5] A five-month prospective survey of surgical-site infections (SSI) conducted in the department of general surgery at Kilimanjaro Christian Medical Centre, Tanzania, showed that 77 (19.4%) of the 397 patients studied developed SSI.[6] A surprising 87% of the patients who developed SSI had received antibiotics, the majority having received them for several days. A survey of antibiotics used in the USA showed that dentists wrote an average of 4.45 prescriptions a week. Antibiotics prescribed after treatment primarily were penicillin and its derivatives, with amoxicillin accounting for 72.2% of prophylaxis prescriptions which were mainly for bacterial endocarditis.[7] An assessment of antibiotic prescriptions following oro-dental infections showed that many were for broad spectrum penicillins.Inappropriate use of antibiotics was noted to be widespread in dentistry.[8] When antibiotics are prescribed for the treatment of infections, clinicians should choose them on a case-specific basis and the choice should be based on several factors, such as laboratory data, patient health, age, allergies, drug absorption and distribution ability and plasma levels. Penetration and metabolism of the drug, cost of drug, type or location of the infection and previous use of antibiotics should also be considered.[4, 9, 10, 11] An antibiotic with activity against viridians streptococci and oral anaerobes should be suitable for treatment of dento-alveolar infection, pericoronitis and periodontitis. Cefmetazole, clindamycin and minocycline may be effective against most pathogens, including penicillin-unsusceptible bacteria.[12]

A study in England on patients who had orofacial infections found the most frequently prescribed drugs to be penicillins,cephalosporins,macrolides and quinolones in that order.[13]

The prescribing patterns of general dental practitioners differ significantly with those of general medical practitioners .This was noticed in the reliance of the dental practitioners to prescribe metronidazole as first choice even as an alternative in patients allergic to penicillins.[14] The advantage of using the 2nd or 3rd generation cephalosporins over the 1st generation in minor oral surgery appears marginal and there is no significant evidence to support the practice.[15] In the management of acute dentoaveolar abscesses, the duration of antibiotic therapy can safely be 3 days provided drainage has been established. It is, therefore, not necessary for majority of patients to complete a week's dose. This can greatly reduce the cost of medication in a case where an expensive drug has been prescribed. Besides the unit cost of the antibiotic, the frequency of administration plays an important role in determining the cost of medication provided that the clinician understands the seriousness of a particular kind of the problem.[16] A comparison of ceftriaxone with penicillin for antibiotic prophylaxis for compound mandibular fractures at 1gm/day and 2mu penicillin (every 4 hours for 2 weeks,)found ceftriaxone to have been more cost effective in the long run in the management of compound maxillofacial fractures. Cefuroxime (zinacef) is suitable for preoperative surgery during maxillofacial procedures because of its favourable kinetics and broad spectrum of action.[17, 18] Animal studies have shown that antibiotic prophylaxis is most effective in preventing post-surgical infections when administered before the start of surgery, and pharmacokinetic data suggest administration as near the time of incision as possible. A prospective observational study monitored the timing of antibiotic prophylaxis in 2847 patients in "clean" or "clean contaminated" surgery where it found that preoperative antibiotics within two hours of incision had the lowest rate of infection as compared to antibiotics given after incision or earlier than two hours prior.[19] These findings being supported by the findings of van Kasteren.[20] Controversy exists regarding the use of antibiotic prophylaxis for clean cases. When antibiotic prophylaxis is given, the agent should target Staphylococcus aureus, the most common organism causing SSIs in clean cases. When bone is incised, the use of prophylactic antibiotics is clearly recommended. A good choice in this situation, or for cardiothoracic or vascular surgery, is cefazolin or cefuroxime (or clindamycin or vancomycin for penicillin allergy).For general surgical clean cases, the decision is less clear.[21, 22]

Similarly, a prospective, randomized, double-blind study on the use of prophylactic antibiotics for paediatric surgery in 289 children at a teaching hospital in Nigeria was carried out. Patients were randomly assigned to receive either doses of ampicillin/cloxacillin

(Ampliclox) with vitamin B (Group A, treatment group), or vitamin B only (Group B, placebo group). The doses were begun at induction and continued for five days post-operatively. Patients were then evaluated for wound infection at postoperative day 5 and then again at postoperative day 7 to 10 during suture removal. Wound infection was defined as the presence of erythema, indurations or discharge. Group A had a 4.3% infection rate compared to 5% in group B, a difference that was not statistically significant.[23] For clean-contaminated and contaminated cases, antibiotic prophylaxis is recommended. The most commonly encountered organism in clean-contaminated and contaminated SSIs is still *Staphylococcus aureus,* though other aerobic as well as anaerobic bacteria are also culprits. As such, prophylaxis should be broader than that used for clean cases.[24] Studies have revealed that the most efficacious regimens include coverage against both aerobic and anaerobic organisms such as a 2nd or 3rd generation cephalosporin, or gentamicin in combination with metronidazole.[25] The differences in efficacy between various 2nd and 3rd generation cephalosporins appear negligible and the choice between them can probably be dictated by the availability or cost. For penicillin-allergic patients, clindamycin combined with gentamicin, aztreonam or ciprofloxacin or metronidazole combined with gentamicin or ciprofloxacin have been found to be adequate choices.[26] The rational use of medicines requires that patients receive medications appropriate to their clinical needs, in doses that meet their own requirements, for an adequate time and at the lowest cost to them and their community. Antimicrobial resistance has become a serious worldwide public health problem and global strategies of interventions to slow the emergence and reduce the spread of antimicrobial-resistant micro-organisms have been and continue to be formulated.[1,12,27,28,29] Choosing inappropriate therapy is associated with increased costs, including the cost of the antibiotic and increases in the overall costs of medical care because of treatment failures and adverse events.[30] Murphy et al. (1998) studied 406 post-operative clean wounds for the presence of sepsis and antibiogram of organisms were established.[31] The over-all post-operative sepsis rate was 13% (clinical) and 12% (bacteriological). *Staphylococcus aureus* (32%) and the Pseudomonas species (21%) were the commonest organisms recovered and netilmycin, cephaloridine and norfloxacin were the most effective antibiotics against both gram positive and negative infections.

According to them, this study reflected the change in the pattern of infecting bacterial flora in the case of post-operative wound infections and its antibiogram.[31]

A retrospective review of available records to establish the prevalence of post-surgical infections associated with various periodontal surgical procedures was carried out.[32] Of the 1,053 surgical procedures evaluated in this study, there were 22 infections giving an overall prevalence of 2.09%.Patients who received antibiotics as part of the surgical protocol (pre- and/ or post-surgically) developed eight infections in 281 procedures (2.85%) compared to 14 infections in 772 procedures (1.81%) where antibiotics were not used. Procedures in which chlorhexidine was used during the post-surgical care had a lower infection rate (17 infections in 900 procedures, 1.89%) compared to procedures after which chlorhexidine was not used as part of the post-surgical care (five infections in 153 procedures, 3.27%).

The use of a post-surgical dressing demonstrated a slightly higher rate of infection (eight infections in 300 procedures, 2.67%) than non-use of a dressing (14 infections in 753 procedures, 1.86%). Despite these trends, no statistically significant relationship was found between post-surgical infection and any of the treatment variables examined, including the use of preoperative antibiotics.

Some studies found no statistically significant relationships between post-surgical infection and the use of preoperative antibiotics. At the same time, no study existed correlating wound drain contamination directly with early or late wound infection.[32, 33]

The frequency of anaerobic recovery is highly variable and the bacteriologic patterns depend largely upon the flora at adjacent mucocutaneous sites. Anaerobic infections are suspected when there is putrid discharge, a polymicrobial flora on gram stain, or infection adjacent to a mucosal surface that is normally colonized by anaerobes. Most anaerobic infections are treated empirically, since they are difficult to recover using standard culture techniques.[34]

Most clinical laboratories will not perform susceptibility tests unless they are specifically requested. In addition, many hospitals do not offer this service, those that do often use techniques that are not considered reliable and the results may be obtained after therapeutic decisions have been made. Antimicrobial agents are usually chosen empirically for the treatment of anaerobic infections without the benefit of in vitro susceptibility tests. This is due to inadequate anaerobic culture techniques, poor quality control of in vitro susceptibility results, and difficulty in obtaining test results within a useful time frame.[35]

JUSTIFICATION

The results of the study could help formulate an empirical antibiotic policy in the management of post-surgical oro-facial infections at KNH and UNDH.

OBJECTIVES

Main objective

- To determine the aerobic bacterial causative agents of post-surgical infections in the orofacial region and their antibiotic sensitivity patterns.

Specific objectives

1. To determine the aerobic bacterial agents in the post-surgical orofacial infections.

2. To investigate the bacterial sensitivity to antibiotics based on CLSI guidelines.

HYPOTHESIS

- Post-surgical orofacial infection at the KNH &UNDH is caused by a wide range of micro-organisms.

- Most aerobic bacterial agents causing post-surgical orofacial infections are susceptible to penicillins and cephalosporins.

CHAPTER 2

MATERIAL AND METHOD

STUDY DESIGN

This was a descriptive cross-sectional study with clinical and laboratory components.

STUDY AREA

- 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 and 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 Medical Microbiology laboratory.

STUDY POPULATION

All patients attending KNH and UNDH Maxillofacial and ENT outpatient units and wards as well as other departments treating patients with orofacial surgical conditions who met the inclusion criteria and consented to the study.

INCLUSION CRITERIA

All patients who had surgical procedures in the orofacial region and had no clinically identifiable infection in the immediate pre-surgical period irrespective of antibiotic prophylaxis and presence of surgical drains.

EXCLUSION CRITERIA

1. Patients in the target population who did not consent to the study.
2. Confirmed HIV infected patients presenting with the AIDS defining illnesses.
3. Patients with other uncontrolled immunosuppressive conditions such as diabetes mellitus.
4. Patients on long-term corticosteroid and cancer chemotherapy.

SAMPLE SIZE

Was determined using the Kish and Leslie's formula ,$N=Z^2p(1-p)/d^2$ where Z is the standard normal deviate 1.96 which corresponds to 95% CI, d is the absolute precision 0.05 and p is the proportion of the target population estimated to have the desired characteristics[47], thus Z=1.96,p=0.04,1-p=0.95,d=0.05.

$N= (1.96^2 x0,04x0.95)/0.05^2 =58$. A minimum sample size of 58 patients was used.

SAMPLING METHOD

Consecutive sampling where all the patients satisfying the inclusion criteria were included.

PROCEDURE

Clinical methods

Patients were evaluated for wound infection from the 5th post-operative day 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 /wounds were those exhibiting pus or any three of the following: 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.

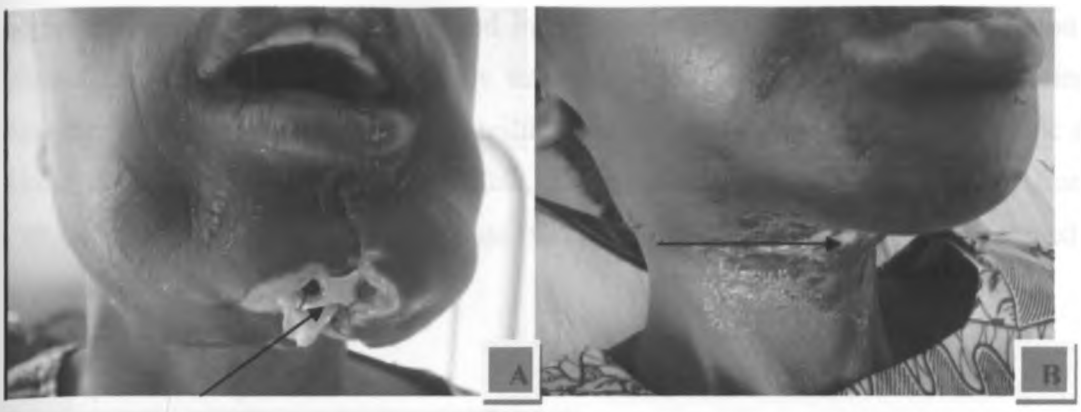

Fig.1. post-operative wound infection.

A: with dehiscence, exposing the titanium plate following resection of an ameloblastoma.

C: starting to show dehiscence following tumuor resection and reconstruction with titanium plate.

Laboratory methods

Bacteriological processing was done using the Standard UON/KNH microbiology operating procedures. All cultures were processed by standard bacteriological techniques including 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 beta haemolysis in blood agar.

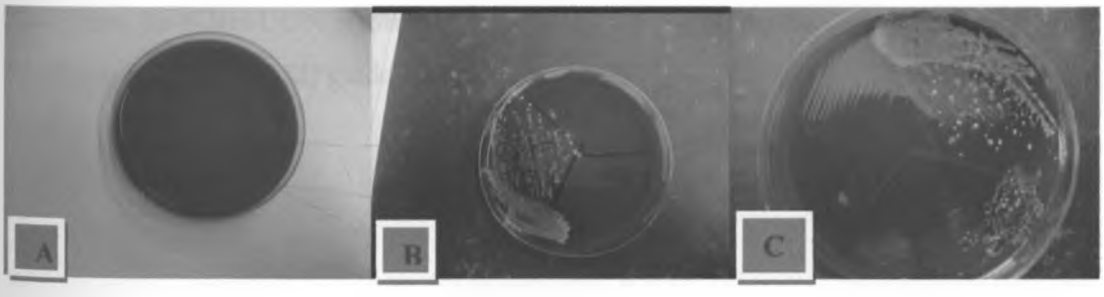

*Fig. 2.*Horse blood agar before inoculation (A), showing pure growth (B), and mixed growth (C).

The cultured organisms were then tested for their susceptibility against various commercially prepared antibiotics by disk diffusion methods (Bauer-Kirby procedure). The test was

performed by swabbing a standardized inoculum of bacteria onto a Mueller-Hinton agar (Oxoid Ltd, England) plate. 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 Clinical Laboratories Standards Institute (CLSI) guidelines (2011) and results copied onto appropriate data sheets.

Fig. 3 Kirby-Bauer disk diffusion test.

Antibiotic susceptibilities are determined by measuring the diameter of the zone of inhibition in millimeters for each of the different antibiotic disks. These are converted to susceptible, intermediate or resistant using a table from the CLSI.

Antibiotic Selection

The selection of antibiotics for susceptibility testing was determined by the type of isolated organisms, the KNH/UON institutional formulary and based on the Clinical Laboratories Standards Institute (CLSI) guidelines (2011).

Plate 1: CLSI Interpretive zone diameters for Staphylococcus aureus.

Antimicrobial	Disc content in micrograms	Zone diameter(mm)		
		Resistant	Intermediate	Sensitive
Cefuroxime	30	≤ 14	15-17	≥ 18
Amoxycillin/clavulinic acid	20/10	≤ 19	-	≥ 20
Chloramphenicol	30	≤ 12	13-17	≥ 18
Cefotaxime	30	≤ 14	15-22	≥ 23
Gentamicin	10	≤ 12	13-14	≥ 15
Oxacillin	1	≤ 10	11-12	≥ 13
Ampicillin	10	≤ 28	-	≥ 29
Vancomycin	15	≤ 14	-	≥ 15

Plate 2: CLSI interpretive zone diameter for *Streptococcus pyogenes*

Antimicrobial	Disc content	Zone diameter(mm)		
		Resistant	Intermediate	Sensitive
Ampicillin	10	≤ 23	-	≥ 24
Amoxycillin/clavulinic acid	20/10	≤ 13	14-17	≥ 18
Cefuroxime	30	≤ 14	15-17	≥ 18
Norfloxacin	5	≤ 12	13-15	≥ 16
Erythromycin	15	≤12	16-20	≥ 21
Chloramphenicol	30	≤ 13	14-20	≥ 21

Plate 3: CLSI interpretive zone diameter for Enterobacteriaceae

Antimicrobial	Disc content in micrograms	Zone diameter(mm)		
		Resistant	Intermediate	Sensitive
Ceftriaxone	30	≤ 13	14-20	≥ 21
Amoxycillin/clavulinic acid	20/10	≤ 13	14-17	≥ 18
Cefuroxime	30	≤ 14	15-17	≥ 18
Levofloxacin	5	≤ 13	14-16	≥ 17
Imipenem	10	≤13	14-15	≥ 16
Ceftriaxone	30	≤ 13	14-20	≥ 21
Cefotaxime	30	≤14	15-22	≥23
Gentamicin	10	≤12	13-14	≥15
Meropenem	10	≤13	14-15	≥16

Plate 4: CLSI interpretive zone diameter for Pseudomonas aeruginosa

Antimicrobial	Disc content	Zone Diameter(mm)		
		Resistant	Intermediate	Sensitive
meropenem	10 Ng	≤ 13	14-15	≥16
imipenem	10 Ng	≤ 13	14-15	≥16
ticarcillin/clavulinic acid	75/10	≤ 14	-	≥15
Amikacin	30	≤ 14	15-16	≥17
ceftazidime	30	≤ 14	15-17	≥18
ceftriaxone	30	≤ 14	14-20	≥21
Gentamicin	10	≤12	13-14	≥15
Cefotaxime	30	≤14	15-22	≥23

Quality Control

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.

Data Analysis

All data were checked for completeness, consistency and accuracy. It was then analysed using the statistical package for social sciences (SPSS) with the help of a statistician. The results are presented in text, graphs, tables and charts.

Ethical Considerations

Consent to conduct this study was sought from the Ethics, Research and Standards Committee of the Kenyatta National Hospital and the University of Nairobi. Only consenting patients were studied. Confidentiality was applied to all the information obtained.

Benefits

Results obtained were used in the management of the patients whose infections were not responding to the empirical treatment they were on.

CHAPTER 3

RESULTS

Demographic Information of the patients

A total of 65 patients were recruited into the study. Sixty-five specimens from the 65 patients were analysed. Of the 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).

Conditions for Which Surgery Was Done

Benign tumours accounted for 43%, malignant lesions 23%, trauma 18.5% and dento-alveolar surgeries 15.5% as shown in figure 4.

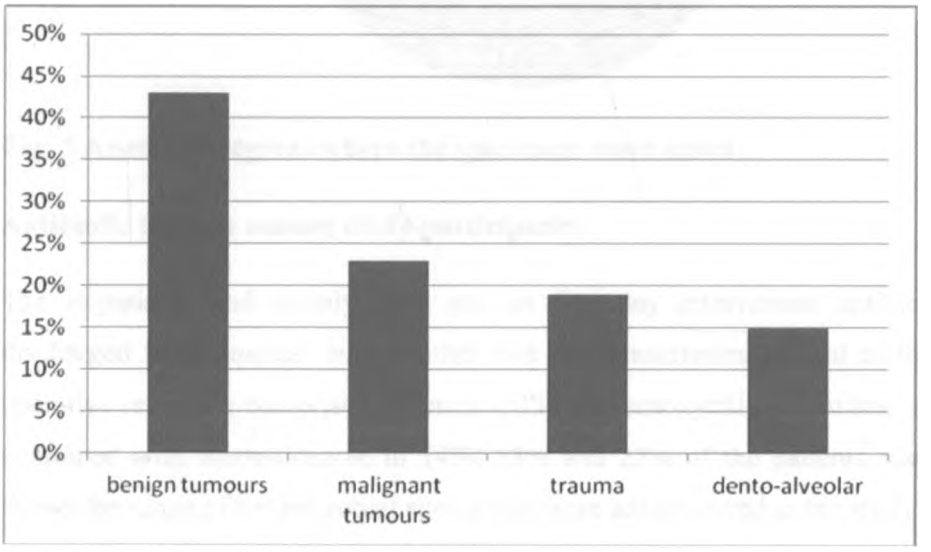

Fig: 4 Summary of Conditions for Which the Surgery Was Done

Anatomic regions where the specimens were taken

The specific anatomic regions from where the specimens were taken included the mandibular region (30%), in the maxillary region (15%) and the neck and Submental areas (11% each). Fig. 5 illustrates all the sites at which the specimens were taken from.

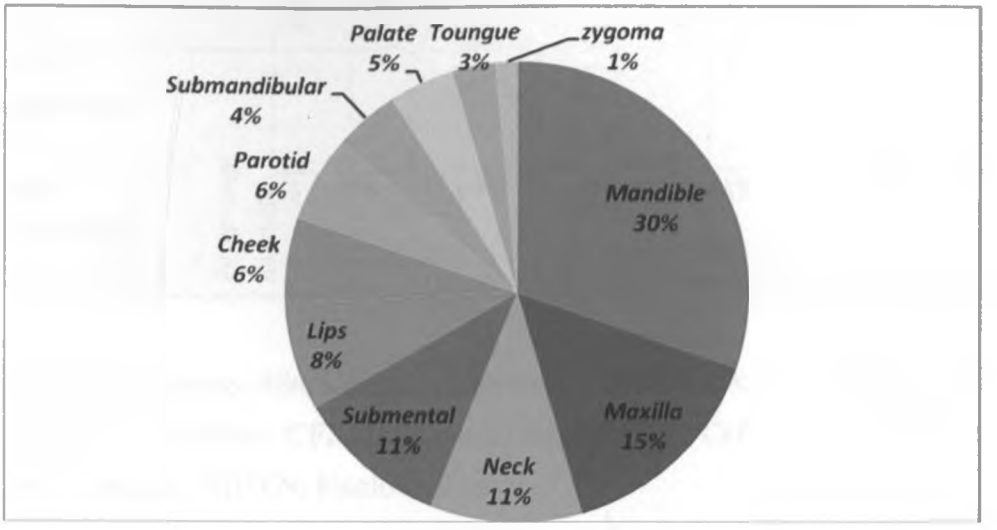

Fig: 5 Anatomic regions where the specimens were taken.

Antibiotic therapy among study participants

The in-patients had mainly been put on five day intravenous antibiotic regimens and discharged from hospital with another five day prescription of oral antibiotics. Most post-operative regimens comprised of amoxycillin and amoxycillin/clavulinic acid or cefuroxime combined with metronidazole in 14%, 15% and 22% of the patients respectively. Table 1 shows the range of antimicrobial agents that were administered to the study participants.

Table 1.Antibiotics administered to the study patients

Antibiotics		AMOXIL	AMX-CLA	CFXME	CFZDM	CFTXN	CLINDA	AMOXIL METRO	AMOX-CLA METRO	CFXME METRO	CFTXN METRO	FLUXN	NO DRUGS
Intra-operatively:	Percentage patients	-	13	29	-	3	-	-	3	3	-	-	49
Post-operatively		6	5	6	3	-	5	14	15	22	5	17	2

AMOXIL: Amoxycillin, **CFTXN:** Ceftriaxone, **AMX-CLA:** Amoxycillin/clavulinic acid, CFZDM: Ceftazidime, **CFXME:** Cefuroxime, **CFTXN:** Ceftriaxone, **METRO:** Metronidazole, **FLUXN:** Flucloxacillin

Laboratory Findings

From the results, 52(80%) of the specimens 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.

Table 2.culture results

Culture Results (n=65)		
Culture results	**Frequency**	**Percentage**
Pure cultures	43	66%
Mixed growth cultures	9	14%
No growth	13	20%
TOTAL		

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%) and12 (19%) of the isolates respectively. Others were *Proteus mirabilis* 7(11%), *Streptococcus pyogenes* 3(5%) and *Escherichia coli* 1(2%) as indicated in Table 3. There were nine mixed growths isolated, with the most frequent combination having been *Staphylococcus aureus* with Klebsiella species at 67 %, followed by Pseudomonas *species* with *Proteus mirabilis* (22%) and *Staphylococcus aureus* with *Proteus mirabilis (11%).*

Table 3. Isolates

Isolates	Frequency	Percentage (n=62)
Gram negative micro-organisms		
Pseudomonas ssp	12	19%
Klebsiella ssp	14	23%
Proteus mirabilis	7	11%
Escherichia coli	1	2%
Gram –positive micro-organisms		
Staphylococcus aureus	25	40%
Streptococcus pyogenes	3	5%

Of the 13 specimens that showed no growth, 77 % were from out-patients whereas 23% were from in-patients. Most of the patients whose samples showed no growths had not had any intra-operative antibiotics. Post-operatively, 4 had been put on flucloxacillin and 3 on cefuroxime while another 4 had been managed with amoxycillin/clavulinic acid.

Table 4.Medications that had been given to the patients whose samples exhibited no growth

INTRA-OPERATIVE MEDICATION				POST-OPERATIVE MEDICATION				
medications	None	cefuroxime	amoxiclav	flucloxacillin	cefuroxime		Amoxiclav	clindamycin
Out-patients	8	0	2	3	0		4	2
In-patients	0	3	0	1	3		0	0

Sensitivity Tests

All the *Staphylococcus aureus* isolates were susceptible to vancomycin. There was high susceptibility to cefotaxime (90%), cefuroxime (85%) and amoxycillin/clavulinic acid (85%).The least susceptibility was to ampicillin (25%).Oxacillin resistance was noted in 8% of the isolates indicating the presence of methicillin resistant *Staphylococcus aureus (MRSA)*. Isolates resistant to oxacillin or methicillin were all interpreted as having been resistant to all beta-lactam agents including cephalosporins, as per the CLSI (2011) guidelines.Fig.6 depicts susceptibility to the other antibiotics.

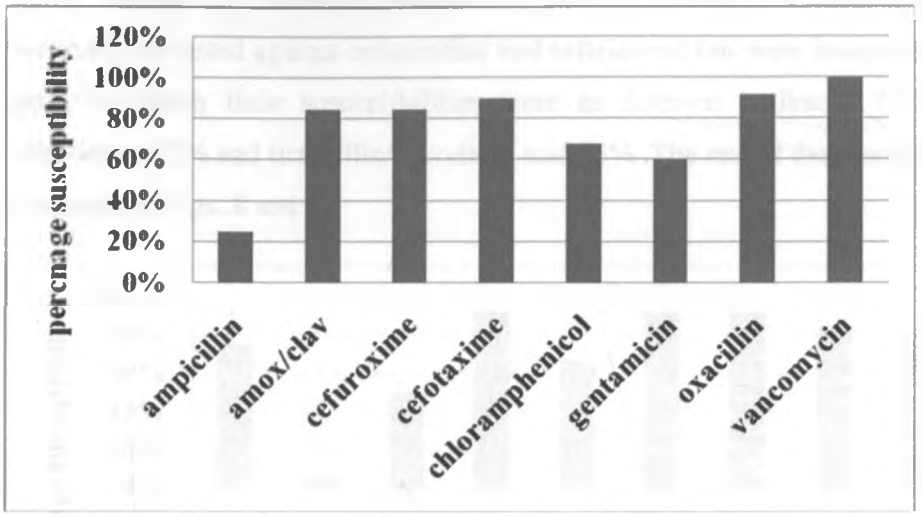

Fig 6.Antibiotic susceptibility for *Staphylococcus aureus*

The susceptibility of the three isolates of *Streptococcus pyogenes* is as shown in figure 7.

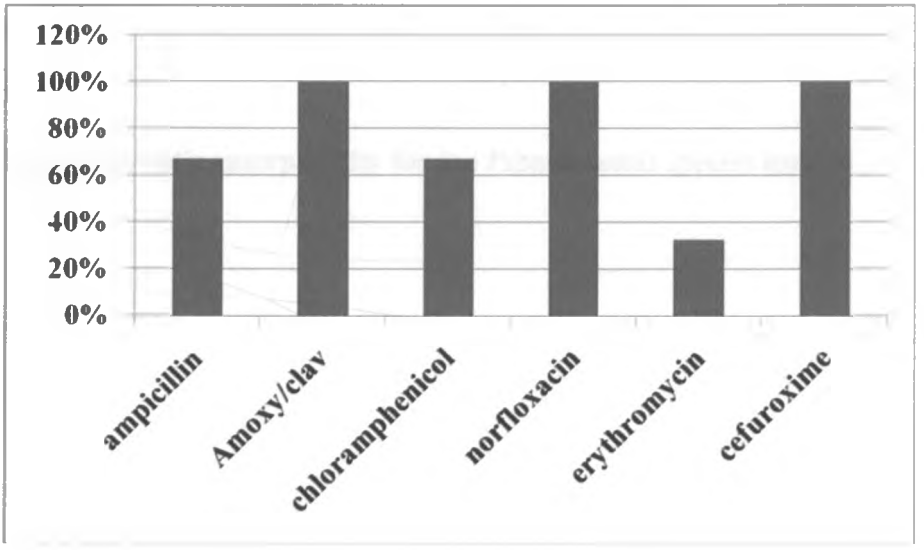

Fig 7.Susceptibility of *Streptococcus pyogenes* (n=3).

The entire Klebsiella species isolated we-re susceptible to meropenem. The susceptibility to imipenem was 93%, to levofloxacin, ceftriaxone and gentamicin at 86%, and 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 which was susceptible to all the other drugs it was tested against except ceftriaxone to which it was resistant. *Pseudomonas* isolates were highly resistant to amoxycillin/clavulinic acid with only 7% susceptibility. They were, however, not tested against cefuroxime and ceftriaxone but were instead tested against other drugs to which their susceptibilities were as follows: amikacin 75%, cefepime 83%, ceftazidime 92% and ticarcillin/clavulinic acid 83% .The rest of the susceptibility patterns are illustrated in Figs. 8 and 9.

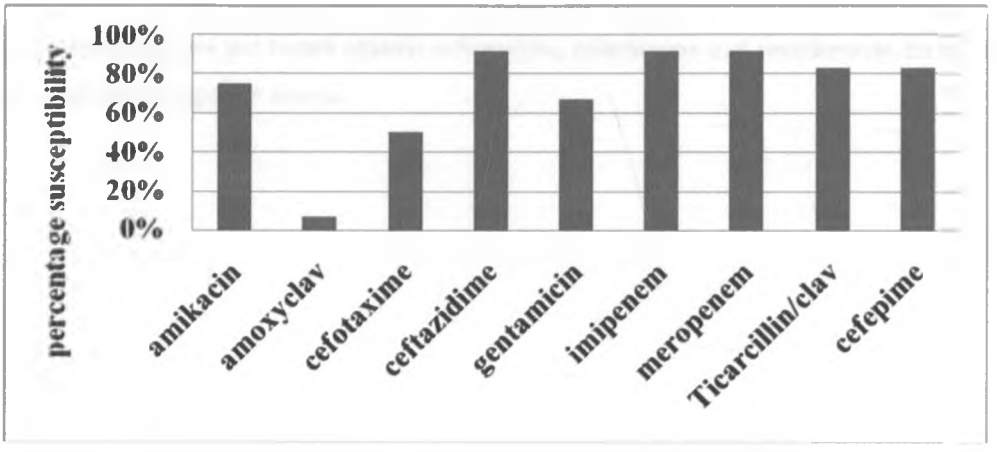

Fig 8. Antibiotic susceptibility for the *Pseudomonas species* isolates

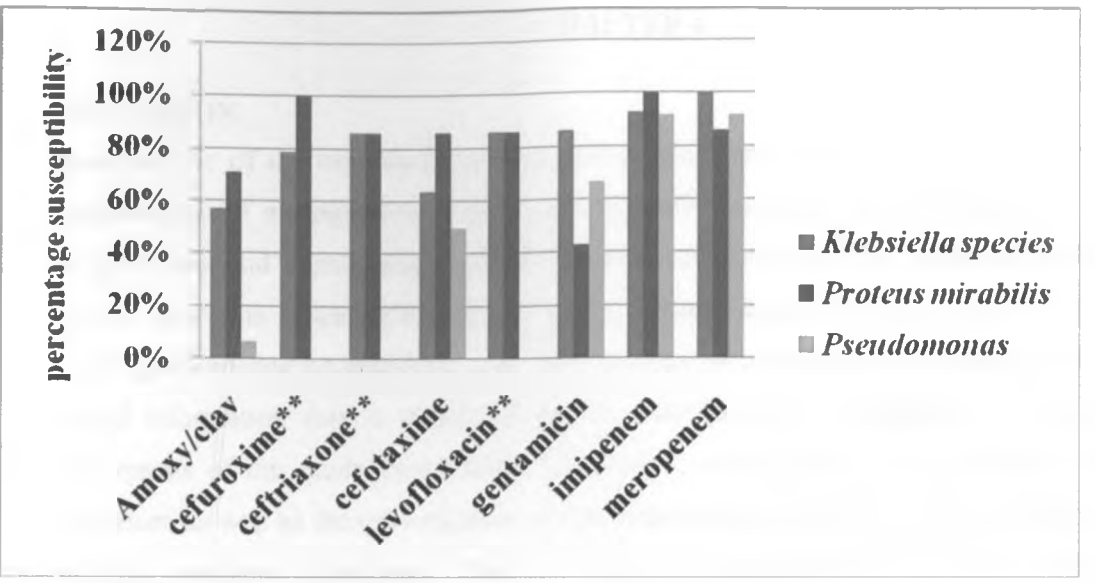

Fig 9. Antibiotic susceptibility for the gram-negative isolates

****Pseudomonas species not tested against cefuroxime, ceftriaxone and levofloxacin.Its full susceptibility pattern is shown in figure 8 above.**

CHAPTER 4

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 as well as the over-reliance of the clinicians at the KNH on a few antibiotics when treating oro-facial infections. This is clearly illustrated by the fact that cefuroxime, amoxycillin/clavulinic acid and metronidazole were the most commonly administered agents to patients intra-operatively. This concurs with results from the study on the prescribing patterns of dental and general medical practitioners which noted the reliance of the dental practitioners on penicillins and cephalosporins and their tendency to prescribe metronidazole as first choice even as an alternative in patients allergic to penicillins.[14]

The in-patients had mainly been put on five-day intravenous antibiotic regimens and discharged from hospital with another five day prescription of oral antibiotics. However, not all these patients left the hospital immediately, with some staying for over a month after discharge, which could have resulted in the majority of the isolates having been from the specimens that had been collected in the 2nd and 4th weeks after surgery. The patients had been evaluated as from post-operative day 5 when clinical features of infection were most likely evident. This was informed by the fact that 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'.[41] 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.[38]

The history of antibiotic prophylaxis and surgical drains were not considered reasons for exclusion from this study because no statistically significant relationship between post-surgical infection and the use of pre-operative antibiotics was anticipated.[32] In this particular study, the drains had been removed by/or on the 4th post-operative day, hence minimising the risks of retrograde infection to less than 2% .[32, 33, 39, 40]

In 20% of the specimens, no growth was found. This is much less than the findings in an observational study in Tanzania on patients with SSIs, in which 35% had cultures that yielded no growth or "no clinically significant organism." [41] 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 another study which showed that the *Staphylococcus aureus* and Pseudomonas species were the commonest infections of post-orofacial procedures with reports of 28% and 12% of the infections having been diagnosed of the two respectively. It also partially compared with the Bratzler, et al (2006) 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. Another Tanzanian study also showed that *Staphylococcus aureus* was the most common isolate followed by *E. coli* and the Klebsiella species.[6]

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 (2009).[43] 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.[44]

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.[45] 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 against 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 Malomo findings in a prospective comparative trial involving ceftriaxone and ceftazidime.[26]

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 beta-lactamases 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.[46]

CONCLUSIONS

1. The bacteria mainly isolated in the infected surgical site wounds in the oro-facial region are Staphylococcus aureus, Klebsiella species, and Pseudomonas species with augmented penicillins and newer generations of cephalosporins being effective against them.

2. Meropenem and imipenem remained highly effective against a large variety of bacteria.

RECOMMENDATIONS

1. Routine culture and sensitivity tests though not feasible for all patients with post-surgical oro-facial infections, should be carried out where response to treatment is poor following intervention and to enable regular and informed updates of antibiotic regime reasonable for empirical treatment.

2. Studies with a larger sample size required to validate the results, in situations that are not time limited.

STUDY LIMITATIONS

Strict time limit of the post-graduate programme.

REFERENCES

1. Bratzler DW, Hunt DR. The Surgical Infection Prevention and Surgical Care Improvement Projects: National initiatives to improve outcomes for patients having surgery. Clin Infect Dis 2006; 43:322.

2. Poulsen KB, Bremmelgaard A, Sorensen AI, et al. Estimated costs of postoperative wound infections. A case-control study of marginal hospital and social security costs. Epidemiol Infect 1994; 113:283.

3. Horan TC, Gaynes RP, Martone WJ, et al. CDC definitions of nosocomial surgical site infections, 1992: A modification of CDC definitions of surgical wound infections. Am J Infect Control 1992; 20:271.

4. Shlaes DM, Gerding DN, John JF Jr, et al. Society for Healthcare Epidemiology of America and Infectious Diseases Society of America Joint Committee on the Prevention of Antimicrobial Resistance: guidelines for the prevention of antimicrobial resistance in hospitals. Infect Control Hosp Epidemiol 1997; 18:275.

5. Carlet J, Ben Ali A, Chalfine A. Epidemiology and control of antibiotic resistance in the intensive care unit. Curr Opin Infect Dis 2004; 17:309-316.

6. Eriksen H, Chugulu S, Kondo S, Lingaas E. Surgical-site infections at Kilimanjaro Christian Medical Center. Journal of Hospital Infection2003; 55:14-20.

7. Epstein JB, Chongs, Le ND'. A survey of Antibiotics used in Dentistry, ADA 2000; 131: 600-609.

8. Kandemir S, Ergul N. Greviances in cases using antibiotics due to orodental problems and assessment of the need of antibiotics. International Dental Journal 2000; 2: 73.

9. Gill Y and Scully C. Orofacial odontogenic infections: review of microbiology and current treatment. Oral Surg Oral Med Oral Pathol 1990; 70: 155–158.

10. Sandor GK, Low DE, Judd PL and Davidson RJ .Antimicrobial treatment options in the management of odontogenic infections. J Can Dent Assoc 1998;64: 508–514.

11. Baker KA and Fotos PG. The management of odontogenic infections. A rationale for appropriate chemotherapy. Dent Clin North Am 1994; 38: 689–706.

12. Tomoari, Kuriyama. Bacteriologic features and antimicrobial susceptibility in isolates from orofacial odontogenic infections. Oral surg Oral Med Oral Pathol, Oral Rad & Endo 2000; 95: 600-608.

13. Tzimis.Antibiotics prescription for indigent patients in primary care. Clinpharm Ther 1997; 23: 227-235.

14. Muthyknishnan A; Wallers H. Douglas PS. An audit of antibiotics prescribing by general practitioners in the initial management of acute dental infection. Dent update1996; 23: 316-318.

15. Thomas DW; Hill CM. An audit of antibiotics prescribing 3rd molar surgery .Br J Maxillofacial surg 1997; 35:126-128.

16. Martin M. LongmanLP; Hardy P. Acute dentoaleveolar infections;-an investigation of dusanun of therapy BR dent J. 1997; 184: 135-137.

17. Helt JM; Stephen's MR.; Jeffords k .A compassion of cefmaxone with penicillin for antibiotics prophylaxis for compound mandibular fracture. Oral surgery, oral Medicine, oral pathology, oral radiology. 1997; 84: 433-436.

18. Schmdt. The resistance spectrum and antibiotics therapy in progressive infection in the mouth jaws and face areas. Swhwelz Munarrsschr Zahnmed .1995; 105: 159-164.

19. Classen D, Evans R, Pestotnik S, et al. The timing of prophylactic administration of antibiotics and the risk of surgical-wound infection. New Engl J of Med 1992; 326:281-286.

20. Van Kasteren ME, Mannien J, Ott A, et al. Antibiotic prophylaxis and the risk of surgical site infections following total hip arthroplasty: Timely administration is the most important factor. Clin Infect Dis 2007; 44:921.

21. Bratzler D, Houck P. Antimicrobial prophylaxis for surgery: An advisory statement from the National Surgical Infection Prevention Project. Clin Infect Dis2004;38:1706.

22. Barie P, Eachempati S. Surgical site infections. Surg Clin of N America2005; 85:1115-1135.

23. Osuigwe A, Ekwunife C, Ihekowba C. Use of prophylactic antibiotics in a paediatric day-case surgery at NAUTH, Nnewi, Nigeria: a randomized double-blinded study. Trop Doc 2006; 36:42-44.

24. Blumetti J, Luu M, Sarosi G, et al. Surgical site infections after colorectal surgery: Do risk factors vary depending on the type of infection considered? J of Surg 2007; 142:704-705.

25. Song F, Glenny A. Antimicrobial prophylaxis in colorectal surgery. A systematic review of randomised controlled trials. Health Techn Asses 1998; 2:110.

26. Malomo A, Adeolu A, Odebode T, et al. Prospective comparative trial of ceftriaxone versus ceftazidime as prophylactic perioperative antimicrobials in neurosurgery. E and C Afric J of Surg 2007; 12:89-92.

27. Kuriyama T, Nakagawa K, Karasawa T, et al. Past administration of β-lactam antibiotics and increase in the emergence of β-lactamase–producing bacteria in patients with orofacial odontogenic infections. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2000; 89: 186–192.

28. Cooper B.S., Stone S.P and Kibbler C. Isolation measures in the hospital management of methicillin resistant Staphylococcus aureus (MRSA): systematic review of the literature, BMJ 2004; 329: 533.

29. Carlet J, Ben A, and Chalfine A. Epidemiology and control of antibiotic resistance in the intensive care unit. Curr Opin Infect Dis 2004; 17:309.

30. Bosso JA. The impact of antibiotic management on resistance. Pharmacotherapy 2004; 24:224.

31. Murthy R, Sengupta S, Maya N, et al. Incidence of post operative wound infection and their antibiogram in a teaching and referral hospital. Indian J Med Sci 1998; 52:553-555.

32. Powell AC, Mealey BL, Dears ED,et al. Post-Surgical Infections: Prevalence Associated With Various Periodontal Surgical Procedures. J Periodontol 2005; 76:329-333.

33. Drinkwater C J, Nell M J. Optimal Timing of Wound Drain Removal Following Total Joint Arthroplasty .The J of Arthrop 1995; 10: 185- 189.

34. Lassmann, B, Gustafson, DR, Wood, CM, et al. Re-emergences of anaerobic bacteremia. Clin Infect Dis 2007; 44:895.

35. Iwata, K, Takahashi, M. Is anaerobic blood culture necessary? If so, who needs it? Am J Med Sci 2008; 336:58

36. Poole, K. Aminoglycoside resistance in Pseudomonas aeruginosa. Antimicrob Agents Chemother 2005; 49:479.

37. Nguyen MH, Yu VL, Morris AJ, et al. Antimicrobial resistance and clinical outcome of Bacteroides bacteremia: Findings of a multicenter prospective observational trial. Clin Infect Dis 2000; 30:870.

38. Horan TC, Culver DH, Gaynes RP, et al. Nosocomial infections in surgical patients in the United States, January 1986-June 1992. Infect Control Hosp Epidemiol 1993; 14:73-80.

39. Schein M. To drain or not to drain? The role of drainage in the contaminated and infected abdomen: An international and personal perspective. World Journal of Surgery2008; 32:312-321.

40. Clegg-Lamptey J, Dakubo J, Hodasi W. Comparison of four-day and ten-day post-mastectomy passive drainage in Accra, Ghana. East African Medical Journal 2007; 84:561-565.

41. Leaper D J. Risk factors for surgical infection Journal of Hospital infection 1995; 30 (Supplement):127-l393.

42. Fehr J, Hatz C, Soka I, et al. Risk factors for surgical site infection in a Tanzanian District Hospital: A challenge for the traditional national nosocomial infections surveillance system index. Infection Control and Hospital Epidemiology2006; 27:1401-1404.

43. Mehrdad A, Alihosein Z, Aziz J, et al. Prevalence of nasal carriage of methicillin-resistant Staphylococcus aureus and its antibiotic susceptibility pattern in healthcare workers at Namazi Hospital, Shiraz, Iran. Int Journal of Infect Dis 2009; 13: 241—247.

44. Hanssen AM, Ericson Sollid JU. SCCmec in staphylococci: genes on the move. FEMS Immunol Med Microbiol 2006; 46:8—20.

45. Shenoy S, Baliga S, Saldanha DR, et al. Antibiotic sensitivity patterns of Pseudomonas aeruginosa strains isolated from various clinical specimens. Indian J Med Sci 2002; 56:427-430.

46. Queenan, AM, Bush, K. Carbapenemases: the versatile beta-lactamases. Clin Microbiol Rev 2007; 20:440

47. Loukota RA. The incidence of infection after third molar removal. Br J Oral Maxillofac Surg 1991; 29:336–337.

APPENDIX I

PATIENT CONSENT INFORMATION

My research involves determination of bacteriology of post-surgical orofacial infections and sensitivity to antibiotics in patients seen at Kenyatta National Hospital University of Nairobi Dental Hospital.

I request you to participate in this study. This participation is entirely voluntary and you have the choice of withdrawing at any stage during the study. As part of the study the information gathered will be used and you will be requested to fill a specially designed chart with questions routinely raised by health care providers. It is entirely dependent on your will to consent to these questions. Should you refrain from the exercise not in any circumstances will this adversely affect your care. A thorough maxillofacial examination will be done using adequate light, clean instruments, disposal gloves and face masks. Once you consent for your participation, we will take a medical and surgical history, examine you and where necessary take some pus from the wound. To ensure confidentiality your personal identity will not be included in the records. Relevant findings from this exercise will be provided to your current health care provider to facilitate your health management.

The results from this study may be used in the future treatment of yourself and those with oro-facial infection.

I Dr Godfrey Bwire Barasa, confirm that I have explained the relevant parts of the study.

Signed _ _ _ _ _ _ _ _

I, the participant, confirm that I have understood the relevant parts of this exercise and do herby give consent to participate.

Signed _ _ _ _ _ _ _ _

Should you wish to contact me over any issues related to the study and your participation please use the following address.

Dr Godfrey Bwire Barasa

P.O. Box 66994-00200 Nairobi, Mobile 0722-615940

FOMU YA MAELEZO KWA MGONJWA NA MAKUBALIANO YA KUSHIRIKI KATIKA UTAFITI

Utafiti wangu unahusu kutambua maambukizi ya vidonda vinavyofuatia upasuaji wa midomo na nyuso za watu katika hospitali kuu Kenyatta na kituo cha utabibu wa meno cha Chuo Kikuu cha Nairobi.

Nakuomba uwe mhusika katika utafiti huu. Uhusika wako ni wa hiari .Unaweza kujiondoa wakati wowote upendao. Kujiondoa kwako hakutadhuru matibabu yako kwa njia yoyote.

Utahitajika kujibu maswali kadha ambayo kwa kawaida huulizwa na wahudumu wa afya. Majibu utakayopeana yatajazwa kwenye fomu maalum na hakuna mtu mwingine atajua ni yako.Kila jambo na hatua itatuunzwa kwa njia ya siri na uhusika wako hautatangazwa kwa uma.

Matokeo ya huu utafiti yatatumika katika kuwatibu wengine watakopatikana na shida kama hii katika siku za usoni.

Mimi Daktari Godfrey Bwire Barasa nadhibitisha kuwa nimetoa maelezo yote yanayofaa kwa njia inayoeleweka.

Sahihi……..

Mimi mhusika ninadhibitisha kuwa nimeelezwa,nikaelewa na nimekubali kuhusika katika utafiti huu.

Sahihi……..

Ukiwa na swali lolote au ukihitaji maelezo kuhusu Utafiti huu,tumia anuani ifuatayo:

Dkt Godfrey B Barasa

S.LP.66994

NAIROBI

Simu ya rununu 0722615940

APPENDIX II

PATIENT PROFORMER

NAME: _____ **STUDY NO: ____**

IPNO: _____ **GENDER: _____**

AGE: _____ **IN-PATIENT/OUT-PATIENT: _____**

POST-OPERATIVE DAY.....

CLINICAL PRESENTATION

Peri-wound Cellulitis----------

Swelling----------

Dehiscence.....

Pus .....

CONDITION FOR WHICH SURGERY WAS PERFORMED

Malignancy.....

Benign lesion.....

Trauma.....

Pulpitis .....

Other.....

ANATOMIC LOCATION WHERE THE SPECIMEN WAS TAKEN

INTRA-OPERATIVE ANTIBIOTICS

1----------

2----------

3----------

POST-OPERATIVE ANTIBIOTICS

1.....

2.....

3.....

4.....

PRESENCE OF SURGICAL DEVICE

Yes ....., type.....

No .....

APPENDIX III

LABORATORY RESULTS

STUDY NUMBER: .....

LAB TEST	RESULT
Gram stain	Positive.....
	Negative.....
Culture results(micro-organisms)	

SENSITIVITY PATTERNS OF THE ISOLATES TO ANTIBIOTICS

Micro-organism						
Antimicrobial	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant

APPENDIX IV

BUDGET

ITEM	COST(KSH)
GLOVES,ETC	1000
STATIONERY	9000
SECRETARIAL SERVICES	15000
LIERATURE SEARCH	5000
DATA ANALYSIS	30000
INVESTIGATIONS,C/S	90000
CONTIGENCY	15000
TOTAL	**165,000**