MICROBIAL SPECTRUM OF HEAD AND NECK SPACE INFECTIONS AT THE MAXILLO-FACIAL CLINIC AT LIVINGSTONE HOSPITAL

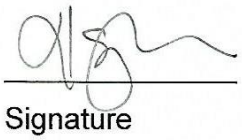
Nerisha Singh

A research report submitted to the Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, in partial fulfilment of the requirements for the degree of Master of Science in Dentistry: Oral Pathology

Port Elizabeth, 2019

Declaration

I, Nerisha Singh, declare that this research report is my own, unaided work. It is being submitted for the Degree of Master of Science in Dentistry in the field of Oral Pathology at the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at any other University.



Signature

27th day of February 2019 in Port Elizabeth

Abstract

Introduction and background

Head and neck space infections remain one of the most commonly encountered conditions at Maxillo-Facial clinics, countrywide. These infections are regularly treated by antibiotics, surgical incision and drainage. Pus aspirates are a vital diagnostic tool, as they permit the micro-organism/s to be identified. This leads to the utilization of appropriate antibiotics, resulting in rapid recovery and shorter hospital stays by the patients.

Objective

The study was a review of the microbial spectrum of the head and neck space infections seen at the Maxillo Facial clinic at Livingstone Hospital in the Eastern Cape. The period of the study was from the 1 January 2012 to 31 December 2016.

Materials and methods

The result sheets of pus aspirates and swabs from patients presenting with head and neck space infection were analysed and information regarding age, gender, aetiology, fascial space involved, microorganisms isolated and antibiotic sensitivity was gathered.

Results

Aspirates and swabs were collected from a hundred and forty five patients. Most patients were males, between the ages of 21 – 40 years. The most common cause of head and neck infections was non-odontogenic in origin, while the fascial space most commonly implicated was the submandibular space. The principal microorganisms identified were gram positive facultative anaerobes.*Bacteroides* species and *Staphylococcus aureus* displayed sensitivity to clindamycin and amoxicillin with clavulanic acid. Viridans streptococci were sensitive to both penicillin and clindamycin.

Conclusion

Bacteroides species were the most commonly isolated bacteria, followed by Viridans streptococci and *Staphylococcus aureus*. Although Clindamycin was found to be effective against the most commonly isolated microorganisms in this study, this antibiotic should be reserved for severe penicillin resistant infections to curtail the increasing resistance reported in literature.

Acknowledgements

My heartfelt thanks are due to:

Dr Sizakele Ngwenya, Head: Department of Oral Pathology, School of Oral Health Sciences, - University of the Witwatersrand for patiently guiding me through every step of the research component.

Dr Julitha Molepo, Head: Department of Oral Biological Sciences, School of Oral Health Sciences, University of the Witwatersrand for her assistance with my research and writing.

Dr Ismael Gopal, Head: Maxillo Facial Department, Livingstone Hospital for graciously allowing me time to conduct my research.

Prof Ellie Grossman for her kind assistance with my research report

The National Health Laboratory, Port Elizabeth for their support in sourcing all the relevant data.

Table of Contents

Contents Page

List of Figures

List of Tables

Abbreviations

ICU: Intensive Care Unit

NHLS: National Health Laboratory Service

HIV: Human Immuno-deficiency Virus

Staph: *Staphylococcus*

Strep: *Streptococcus*

Spp: Species

CHAPTER 1: INTRODUCTION

1.1 Literature Review

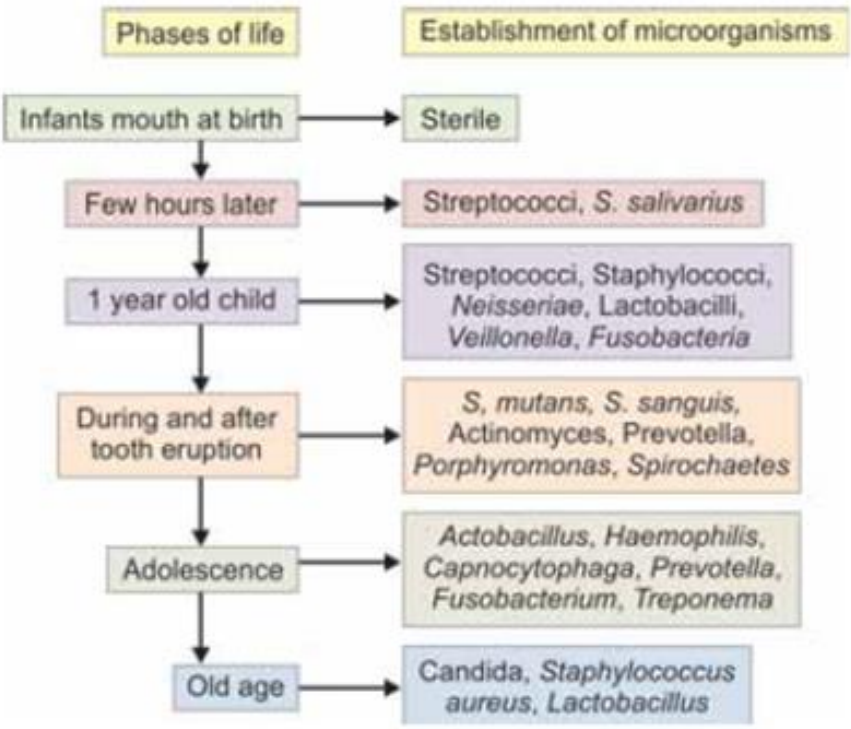
1.1.1 A Review of the normal oral flora

The oral microbiological environment is highly diverse with over 500 species cultivable and many others not cultivable. It comprises bacteria, fungi, viruses, and rarely protozoa; with bacteria being the microbe that is responsible for initiating disease (Vineet, 2015).

According to Sowmya (2016), these micro-organisms vary depending on temperature, pH, nutrients, anaerobiosis, host defences, host genetics and antimicrobial agents. The great population of normal oral flora provides a challenge to invading pathogens, which contend for nutrients and receptor sites. Furthermore, some bacteria produce antimicrobial substances that defend their territory from attack by other organisms.

Acquisition of the normal oral flora

Patil et al (2013) proposed the following regarding the acquisition of normal oral microflora in the phases of life:



Classification of the normal oral microbiology

Patil et al (2013) divided the normal flora into three groups: Indigenous, supplemental and transient flora.

Indigenous flora: Micro-organisms that occur in high numbers in a particular site. They are compatible with the host and do not compromise the host's survival. Examples are *Streptococcus, Actinomyces and Neisseria.*

Supplemental flora: Organisms that are always present, but in low numbers (less than 1% of the viable flora). Environmental changes may lead to these organisms becoming indigenous. An example is *Lactobacillus*, an acid tolerant microbe, which becomes dominant in a carious lesion, where the pH of plaque becomes acidic.

Transient flora: Organisms present in the oral cavity for short periods. They present in food and drinks and quickly vanish. In immunocompromised individuals, this flora may flourish and become opportunistic pathogens.

Sowmya (2016) suggested that, if the conditions permit the normal flora gain access to normally sterile tissues (like the dental pulp or peri-radicular tissues) and become opportunistic pathogens. He reported that if the assault of microbes produces tissue injury, infection is created. Therefore, a sound understanding of the oral microflora may be beneficial in grasping the pathogenesis of numerous oral diseases.

Table 1.1. Working classification of the normal oral flora - Patil (2013)

<table>
<tr><th colspan="6">NORMAL ORAL MICROFLORA</th></tr>
<tr><th colspan="5">ADULT</th><th>NEONATE</th></tr>
<tr><th colspan="4">BACTERIA</th><th>PROTOZOA</th><th>FUNGI</th><th>MOSTLY BACTERIA</th></tr>
<tr><th colspan="2">Gram- positive</th><th colspan="2">Gram –negative</th><td>-Entamoeba gingivalis

-Trichomonas tenax</td><td>-Candida species

-Mycoplasma</td><td rowspan="3">-Mutans streptococci

-Lactobacilli

-Anaerobes</td></tr>
<tr><th>Cocci</th><th>Bacilli</th><th>Cocci</th><th colspan="2">Bacilli</th><td colspan="2" rowspan="2"></td></tr>
<tr><td>- Streptococcus

- Stomatococcus mucilaginosus

- Staphylococcus aureus</td><td>-Actinomyces

-Lactobacillus

-Eubacterium

-Propionibacterium</td><td>-Neisseria
-Veillonella</td><th>Facultative anaerobes</th><th>Obligate anaerobes</th></tr>
<tr><td colspan="2"></td><td colspan="2">-Haemophilus

-Actinobacillus

-Eikenelia

-Capnocytophaga</td><td>-Porphyromonas

-Prevotella

-Fusobacterium

-Leptotrichia

-Wolinella

-Treponema</td><td colspan="2"></td></tr>
</table>

1.1.2. Fascial spaces of the head and neck

Deep neck spaces are interlinked potential spaces separated from one another by different layers of cervical fascia. This allows spread of infection to adjacent spaces with resultant acute infections, some of which can be life-threatening. (Shrivastav, 2014)

Cohen et al (2010) classified fascial spaces into 4 anatomical groups:

i. The mandible and below - the mental, submental, sublingual and submandibular spaces

ii. The cheek and lateral face - the buccal, submasseteric and temporal spaces and the maxillary buccal vestibule

iii. The pharyngeal and cervical areas - the pterygomandibular, para-pharyngeal and cervical spaces

iv. The midface - the palate, base of the upper lip, canine and periorbital spaces

1.1.3. Summary of international studies

In an 18 month review of odontogenic infections, Akinbami et al (2010) found that 11.3% (261) of all oral and maxillofacial infections were odontogenic in origin. There were 146 (59.8%) female and 98 (40.2%) male patients who exclusively presented with dento-alveolar tissue involvement. Patients with infections that had spread into regional soft tissue spaces and beyond constituted 10 (58.8%) males and 7(41.2%) females.

Akinbami et al (2010) also found the submasseteric space to be the most commonly affected space (n = 10; 22.7%), followed by the submandibular space (n = 9; 20.5%) and the sublingual space (n = 6; 13.6%). The most common causative micro-organisms identified in this study were *Klebsiella* and *Streptococcus* spp. Furthermore, the most commonly utilised antibiotics were amoxicillin, amoxicillin with clavulanic acid and metronidazole.

Bahl et al (2014) investigated odontogenic infections with fascial space involvement in 100 patients younger than 60 years. The submandibular space was the most frequently involved fascial space in both single and multiple space infections. Mixed growth (aerobic and anaerobic) was identified in culture smears of 60 patients. Aerobic bacterial growth was observed in 25 patients and anaerobic growth in 15 patients. The

most commonly isolated aerobic bacteria were *Streptococcus viridans* while *Bacteroides* and *Prevotella* appeared to be the most common anaerobic bacteria. In this study, the researchers established that odontogenic infections were caused by both aerobic and anaerobic microorganisms.

Chang et al (2010) examined deep neck infections in a paediatric population. Of 50 patients that presented with deep neck space infections, 29 were adolescents and 21 were children. Nine infections occurred in the retropharyngeal space, 17 in the para-pharyngeal space, and 21 in the peri-tonsillar space while three presented with multiple space involvement. The study also showed that all retropharyngeal abscesses occurred predominantly in children, whilst the majority of peri-tonsillar abscesses (81%) were found in adolescents. The cultures of 32 patients with deep neck infections showed 7 mixed flora, 5 *Streptococcus pyogenes*, 4 *Streptococcus* other than pyogenes, 2 anaerobic bacteria, 1 each *Staphylococcus aureus* and *Klebsiella pneumoniae*, 5 normal flora while 6 showed no growth. The authors concluded that the location of the deep neck space infections differ in different paediatric age categories.

The study by Fating et al (2014) was aimed at investigating the presence of microbial flora and the antibiotic sensitivity patterns present in orofacial space infections, solely of odontogenic origin in order to improve the management of these infections. The results of the study showed that the infections occurred most commonly in males, in the third and fourth decades of life and involved the submandibular space.

The microbial flora most commonly isolated were aerobic alpha haemolytic streptococci followed by anaerobic streptococci and *Bacteroides* sp. With regard to the antibiotic sensitivity, the authors found clindamycin, gentamycin, linezolid and imipenem to be important in the management of these infections as 20 % of the aerobic bacteria were resistant to penicillin. *Streptococcus* species were the most commonly isolated microorganisms in orofacial infections of odontogenic origin. Effective management consisted of the administration of amoxicillin and clavulanic acid and metronidazole, followed by surgical drainage and extraction of offending tooth/teeth.

The 5 year retrospective study by Mathew et al (2012) emphasised the significant contribution of odontogenic infections to maxillo-facial space infections associated with several life-threatening complications. The majority of patients in this study were male (66.4%) and the mean age at presentation was 40 years. Most odontogenic maxillo-

facial infections were attributed to pulpitis (70.8%), involved the lower third molars and occurred in the submandibular space.

In their study on bacteriology and management of orofacial infections in a South African population group, Molomo et al (2016) reported that 57.9% of patients were male, 61.1% were less than 40 years of age, 64.6% had the infection for less than a week, 71.9% had received no antibiotics and 90.4% had no previous history of dento-facial surgery. The submandibular space was the most commonly involved space (57.5%). A total of 122 pathogens were cultured in this study. The predominant microorganisms were aerobic, the four most common being *Streptococcus viridans*, coagulase negative *staphylococcus* and *Staphylococcus aureus*. The antibiotics with the highest efficacy were found to be penicillin, clindamycin and gentamycin. The authors recommended penicillin as the empirical drug of choice, and advised against the indiscriminate use of metronidazole in the management of orofacial infections. In addition, they recommended that second line drugs be utilised in resistant infections.

Rega et al (2004) examined the anatomical spaces, the micro-organisms responsible for deep space head and neck infections and the antibiotic resistance encountered during treatment in 103 patients. The results showed that 54% of patients were male, with a mean age of 33 years. Similarly, the submandibular space was the most common locality for a single space infection (30%), followed by the buccal (27.5%) and the lateral pharyngeal space (12.5%). Sixty-three patients had multiple space involvement and the submandibular space was the predominant space (28.2%) in this subgroup, followed by the submental space (14.8%) and the lateral pharyngeal space (14.0%)

This study yielded 269 bacterial strains from 103 patients. Gram positive microbes were isolated in 63.5% of patients, gram-positive cocci in 57.7% and gram-negative rods in 33% of cultures. Furthermore, 178 aerobes (65.7%) and 91 anaerobes (34.3%) were isolated, the predominant bacteria being Viridans streptococci, *Provetella*, Staphylococci and *Peptostreptococcus*. The cultures displayed greater aerobic growth (65.7%) than anaerobic growth. Viridans streptococci were found to be highly susceptible to ampicillin (98.4%), cefazolin (100%), ciprofloxacin (100%), clindamycin (86.3%), erythromycin (83.4%), levofloxacin (98.6%) and vancomycin (100%). Staphylococci were mildly sensitive to penicillin (27.3%) and ampicillin (41.2%), but

more susceptible to cefazolin (70.0%), ciprofloxacin (95%), clindamycin (89.5%), erythromycin (75%), levofloxacin (84.2%) and vancomycin (100%).

Shah et al (2016) identified the aerobic micro-organisms that were responsible for deep fascial spaces of head and neck infections and evaluated the resistance of antibiotics used in the management of these infections. The results showed that 78% of the sample was male, the mean age was 36 years, the most commonly implicated tooth was the 46 and the submandibular space appeared to be the most commonly affected fascial space. Seventy three percent of the isolates comprised aerobic gram positive bacteria, 18% were aerobic gram negative bacteria and 9% percent displayed no growth. The most common gram positive bacteria were *Streptococcus viridans* (47% of cases) while *Klebsiella pneumoniae* was the predominant gram-negative microorganism (11% of cases).

Amoxicillin eftriaxone, carbenicillin, amikacin, and imipenem displayed high sensitivity levels while amoxicillin showed resistance (48.4%) compared to other antibiotics. All the isolated microbes displayed sensitivity to ceftriaxone and amoxicillin with clavulanic acid at 82.4% and 64.8% respectively. They also advocated the utilisation of third generation cephalosporins as a replacement for amoxicillin. The study also reported 93.4% sensitivity of all microbes against carbenicillin, amikacin, and imipenem.

Singh et al (2014) showed that odontogenic infections were most common in the third and fourth decades of life and predominantly occurred in the submandibular space. The empirical management consisted of extraction followed by incision and drainage. Of the 30 microbial isolates that were acquired, 43 % of the strains were anaerobes, 39 % were aerobes and 18.52 % contained a combination of aerobic and anaerobic bacteria. The most common aerobic bacteria were alpha haemolytic *Streptococcus*; mainly *Streptococcus aureus* and *Peptostreptococcus* and the most common anaerobes were *Bacteroides* and *Prevotella*. This study found that 22% of the aerobic bacteria were resistant to penicillin, while 100% of the strains were sensitive to amoxicillin and clavulanic acid combination. The authors concluded that there was a changing trend where anaerobic dominated aerobic bacteria.

Ye et al (2017) evaluated the prevalence and antimicrobial susceptibility of aerobic and anaerobic strains in 100 patients with single and/or multiple head and neck abscesses. Sixty five percent of the patients were male, 74% had abscesses involving a single

space while 26% had multiple space envelopment. In both single and multiple space involvement, the submandibular space was the most common site, at 76.92% and 84.92% respectively. The authors isolated 114 microbes, of which 48 were aerobic and 65 anaerobic bacteria, with the predominant bacteria being *Klebsiella*, *Clostridium tertium*, *Staphylococcus aureus* and *Actinomyces odontolyticus*. The anaerobic bacteria displayed 100% resistance to amoxicillin and penicillin, but high sensitivity levels were obtained for erythromycin (100%), cefotaxime (98.48%) and clindamycin (96.96%). However, the aerobic strains were resistant to penicillin (47.91%) but sensitive to amoxicillin (79.16%).

Selfars et al (2004) identified infections of oral mucosal, odontogenic and salivary origin; blunt or penetrating trauma and local spread from adjacent spaces as predisposing factors for the development of head and neck space infections. With the advent of antibiotics, odontogenic infections including dental caries, periodontal disease, and pulpitis have become the most common cause of infections in deep neck spaces particularly in adults (Ibeyemi et al., 2014).

Prior to that, upper respiratory infections such as tonsillitis, sinusitis and otitis media were the main cause (Maharaj, 2007). In children however, tonsillitis remains the most common cause (Chang et al., 2010; Poeschl et al., 2010 and Wang et al., 2010). Odontogenic infections are generally caused by abscess forming bacteria. The roots of teeth provide a pathway for infecting bacteria to enter deep tissues. Parotid, submandibular and sublingual gland infections may also involve deep spaces.

The HIV epidemic has resulted in the resurgence of tuberculosis with frequent involvement of the cervical spine resulting in an increase in infections of the prevertebral space (Garg et al., 2011). In many cases of head and neck space infections, the causative agent remains unidentified. Odontogenic causes of infections were identified as the most common source of head and neck infections, with a substantial 46.9% of cases reported by Boscolo-Rizzo et al (2009).

Of all odontogenic infections investigated by George et al (2012), 71%, 17%, 6%, 5% and 1% originated from the pulp, periodontium, extraction sites, pericoronitis and needle tracts, respectively. These authors reported lower third molars (36%) and upper third molars (7%) as the most frequently involved teeth in head and neck space infections with an odontogenic origin. These findings were corroborated by other investigators

(Akinbami et al., 2010; Bahl et al., 2014; Fating et al., 2014). In contrast Singh et al (2014) and Shah et al (2016) reported the first mandibular molar as the most common tooth, followed by the mandibular third molar. The mandible was found to be the most frequently involved jaw (Akinbami et al., 2010; Ibeyemi et al., 2014). Boscolo-Rizzo et al (2009) identified submandibular sialadenitis (14.8 %) as the most common non-odontogenic aetiology of head and neck space infections, followed by pharyngitis (6.2%) and parotitis (3.7%). The authors could not identify the aetiology in 28.4% of the cases.

Studies revealed that the majority of infections occurred in individuals younger than 65 years (Boscolo-Rizzo et al., 2009; George et al., 2012; Singh et al., 2014). Fating et al (2014) and Bahl et al (2014) narrowed the age group most commonly involved to the third and fourth decades of life, while Shah et al (2016) reported the third decade of life as the most common. Head and neck space infections were shown to be more common in males than females (Boscolo-Rizzo et al., 2009; George et al., 2012; Fating et al., 2014; Bahl et al., 2014; Singh et al., 2014; Shah et al., 2016).

1.1.4. Microbial flora in odontogenic infections

According to Hupp et al (2008), indigenous bacteria that form part of the normal oral flora are responsible for eliciting odontogenic infections. These bacteria are primarily aerobic and anaerobic gram positive cocci and anaerobic gram negative rods. Supra-gingivally these bacteria cause caries, gingivitis and periodontitis. When these bacteria gain access to the deeper underlying tissues through a necrotic dental pulp or a deep periodontal pocket, they cause infections in head and neck spaces. As the infection progresses deeper, different members of the existing flora may find better growth conditions and begin to outnumber previously dominant species.

As reported by Hupp et al (2008), aerobic bacteria predominantly found in odontogenic infections are of the *Streptococcus milleri* group, which consists of the three members of the *S. viridans* group: *S. anginosus*, *S. intermedius* and *S. constellatus*. Staphylococci, other streptococci, *Neisseria*, *Corynebacterium* and *Haemophilus* spp. are rarely found. Anaerobic bacteria that play an important pathogenic role include gram-positive cocci such as *Streptococcus* and *Peptostreptococcus* spp. and gram negative rods such as *Prevotella* and *Fusobacterium* spp. The gram-negative cocci and the gram-positive rods

appear to be opportunistic organisms, with little or no role in the initiation of odontogenic infections (Hupp et al., 2008).

1.1.5. Mechanism of spread of infection

According to Brook et al (1991), after initial inoculation into the deeper tissues, the *S. milleri* group synthesises hyaluronidases, which allow the infecting organism to spread through the connective tissues, starting a cellulitis-type of infection. Metabolic by-products cause the release of essential nutrients, lower the pH in the tissues and consume local oxygen supplies. This creates a favourable environment for the growth of anaerobes, which in turn, cause a decrease in the local oxidation-reduction potential. This results in the production of collagenases leading to liquefactive necrosis of tissues and formation of micro-abscesses. Anaerobic bacteria predominate in the abscess stage while aerobic streptococci appear in the early cellulitis stage. Brook et al (1991) further report that anaerobic bacteria account for 50% of odontogenic infections, while aerobic bacteria account for 6% and a combination of both aerobic and anaerobic bacteria are responsible for the remaining 44%.

1.1.6. Anaerobic and aerobic bacteria

In previous studies by Rega et al (2004) and Walia et al (2013), 70% and 65.7% of isolated bacteria were aerobic while 25% and 34.3% were anaerobic, respectively. Shah et al (2016) only isolated aerobic bacteria. *Staphylococcus* species was the most isolated bacteria in the study by Walia et al (2013), while *Streptococcus viridans* was the most isolated in studies by Rega et al (2004) and Shah et al (2016).

In their study on bacteriology and management of orofacial infections in a South African population group, Molomo et al (2016) found *Streptococcus, Staphylococcus* and *Klebsiella* spp. to be the most common organisms. These results echo the findings of a study by Cabral et al (2016) which concluded that *Staphylococcus aureus* and *Streptococcus* spp. are the most common pathogens responsible for head and neck infections. Akinbami et al (2010) reported *Klebsiella* and *Streptococcus* spp. as the most commonly found organisms while Ye et al (2017) demonstrated *Klebsiella*, *Clostridium tertium*, *Staph aureus* and *Actinomyces odontolyticus* as predominant organisms.

1.1.7 Microbial susceptibility

Streptococcus viridans showed susceptibility to erythromycin (83.4%), clindamycin (86.3%), penicillin (87.1%), vancomycin (100%) cefazolin (100%), ciprofloxacin (100%) and levofloxacin (98%) in a study by Rega et al (2004). Walia et al (2013) reported susceptibility to penicillin (75%), erythromycin (75%), gentamycin (75%), ciproflaxin (100%) and cefotaxime (100%). Shah et al (2016) reported maximum resistance to amoxicillin (34%) and a sensitivity of 68.1% to amoxicillin with clavulanic acid. The highest sensitivity rates observed in this study were for carbenicillin, amikacin, imipenem at 100%. Molomo et al (2016) reported *Streptococcus viridans* to be sensitive to penicillin (96.8%), erythromycin (92%), clindamycin (100%), gentamycin (100%) and cefotaxime (100%).

Staphylococci showed minimal resistance (16.7%) to ceftriaxone, carbenicillin, imipenem and moxifloxacin; however maximum resistance was observed for amoxicillin (Shah et al., 2016). In that study ceftriaxone and carbenicillin had the highest sensitivity rate (83.3%). The microorganisms were 71.4% resistant to penicillin and erythromycin but 100% susceptible to gentamycin, ciprofloxacin and cefotaxime in a study by Walia et al (2013). Molomo et al (2016) reported 61.5% penicillin, 82% erythromycin, 93% clindamycin, 60% cloxacillin, 100% gentamycin and metronidazole sensitivity to Staphylococci.

E. coli was most susceptible to amikacin, with 100% sensitivity noted by Walia et al (2013). Shah et al (2016) reported a slightly lower sensitivity of 66.7% to both carbenicillin and moxifloxacin. Shah et al (2016) reported *Klebsiella pneumoniae* to be highly susceptible (100%) to imipenem, ceftriaxone, carbenicillin, amikacin while moxifloxacin and amoxicillin showed resistance of 36.4% and 63.6% respectively. *Pseudomonas* had a 100% resistance to amoxicillin and amoxicillin with clavulanic acid, as reported by Shah et al (2016). Efficacy of amoxicillin with clavulanic acid (64.8-81%) and ceftriaxone (82.4%) was reported for all organisms (Shah et al., 2016).

Patients admitted with deep space head and neck infections tend to have prolonged hospital stays and often require intensive care support. This places financial and logistic

constraints on our health care system. There are also growing concerns, worldwide, regarding antibiotic resistance.

This study focussed on the microbial spectrum of head and neck space infections with the hope to assist in the identification of changes in the microbial spectrum and antibiotic sensitivity patterns in head and neck space infections over the years. It was anticipated that it would serve as an important aid in guiding the prescription of appropriate antibiotics, both before and after microbial culture and sensitivity results thus decreasing the length of hospital stays and contributing to the fight against antimicrobial resistance.

1.2 Aim

The aim of this study was to determine the microbial spectrum of head and neck space infections in patients admitted to the Maxillo-Facial clinic at Livingstone Hospital

1.3 Objectives

- To identify the causative micro-organisms responsible for head and neck infections.
- To determine whether age, gender and source of infection influence the microbial spectrum of head and neck infections.
- To assess the sensitivity patterns of the identified micro-organisms to commonly prescribed antibiotics.

CHAPTER 2: MATERIALS AND METHODS

2.1 Study design

This was a retrospective study which comprised the analysis of patient variables (age, gender and source of infection), microbial spectrum and antibiotic sensitivity patterns in patients presenting with head and neck space infections at the Maxillo-Facial Clinic, Livingstone Hospital, Eastern Cape, South Africa.

The period of the study was 5 years (1 January 2012 to 31 December 2016). The study period was determined in accordance with two previous studies which had a shorter

study period and lower sample sizes of 88 and 93 patients each (Yuvaraj et al, 2012; Cabral et al, 2016).

The study included records of all patients presenting with head and neck space infections and admitted to the Maxillo-Facial clinic. Records with complete demographic, medical, surgical and microbiological data were included and those with 20% or less data available were excluded.

2.2. Sampling method

Cases were selected by a convenience sampling method. The admissions book at the Maxillo-Facial clinic was analysed to identify all patients with head and neck space infections admitted to the clinic at Livingstone Hospital. The study period was from 1st January 2012 to 31st December 2016.The medical records of all identified patients were reviewed. All patients who had microbial culture and sensitivity tests performed were included in the study. Data was extracted from the microbiological result sheets from the archives of the National Health Laboratory Services (NHLS). This study was approved by the Wits Human Research Ethics Committee (Ethical Clearance number: M170719). See Appendix A.

2.3. Data collection

Pus swabs and/or aspirates are routinely taken from all patients who present with suppurative head and neck space infections. These are sent to the NHLS for microbial culture and sensitivity.

The information gathered from the NHLS result sheets included patient age, source of infection, gender, spaces involved, microbial spectrum and antibiotic sensitivities. **(Appendix B)**

2.4 Antibiotics prescribed at the ICU at Livingstone Hospital

Some of the patients with life threatening head and neck space infections are transferred to the Intensive Care Unit at Livingstone Hospital while under treatment.Most of the antibiotics tested for sensitivity and resistance by the NHLS are utilised by the ICU **(Appendix C)**. However, the following antibiotics are not tested for sensitivities by the NHLS but administered in ICU: Acyclovir, Amphotericin B,

Anidulafungin, Azithromycin, Caspofungin, Ceftriaxone, Clarithromycin, Cloxacillin, Cotrimoxazole, Daptomucin, Doripenem, Doxycycline, Fluconazole, Ganciclovir, Metronidazole, Moxifloxacin, Tobramycin and Voriconazole **(Appendix D)**.

2.5. Data analysis

The data was recorded on standardised data collection sheets. Descriptive and inferential statistics were computed for all variables. Bivariate analysis was used for identifying association. P-value based on the Chi-square test was utilised. A p-value of <0.05 was considered to be statistically significant.

The microbial spectrum of head and neck space infections were analysed against patients' variables. Comparative analysis was performed on:-

- Gender
- Age groups
- Odontogenic versus non-odontogenic source of infections
- Single compared to multiple space involvement
- Microbial sensitivity to antibiotics

2.6 Ethical considerations

A letter granting permission to conduct research at Livingstone Hospital was obtained from the Eastern Cape Department of Health. Patients' confidentiality was respected and protected by using patient codes and removing all personal identifiers. A letter of approval was also obtained from the National Health Laboratory Services in order to utilise the microbiology reports of the study population.

Permission to undertake this study was obtained through the Human Research Ethics Committee at the Faculty of Health Sciences at the University of Witwatersrand (ethics clearance certificate number M170719) - **Appendix A**.

CHAPTER 3: RESULTS

3.1. Patient Characteristics

A hundred and forty five records were analysed.

3.1.1. Gender

The majority of patients presenting with head and neck space infections were male (n=97; 67 %), while only 48 (33 %) were female (Figure 3.1).

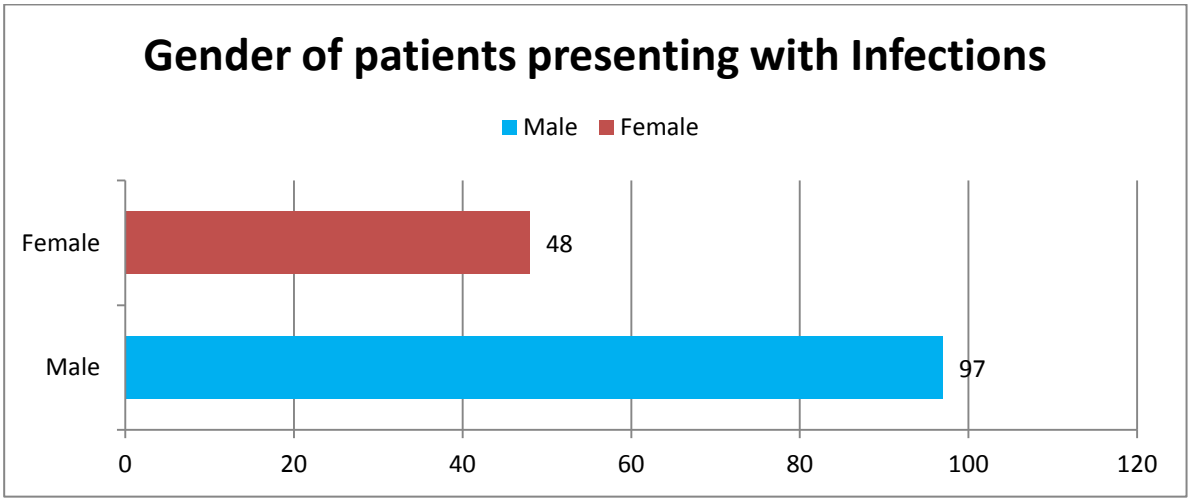


Figure 3.1: Gender distribution of the study

3.1.2. Age Groups

Eighty five patients (59%) were between 21 to 40 years of age. The youngest patient was 13 months and the oldest was 92 years old. Thirty six patients (25%) fell into the 41-60 year age category, while 21 patients (14%) were between 0-20 years old. Head and neck infections appeared to be least common in patients older than 60 years, with only 3 patients (2%) falling into this category. This information is reflected in figure 3.2.

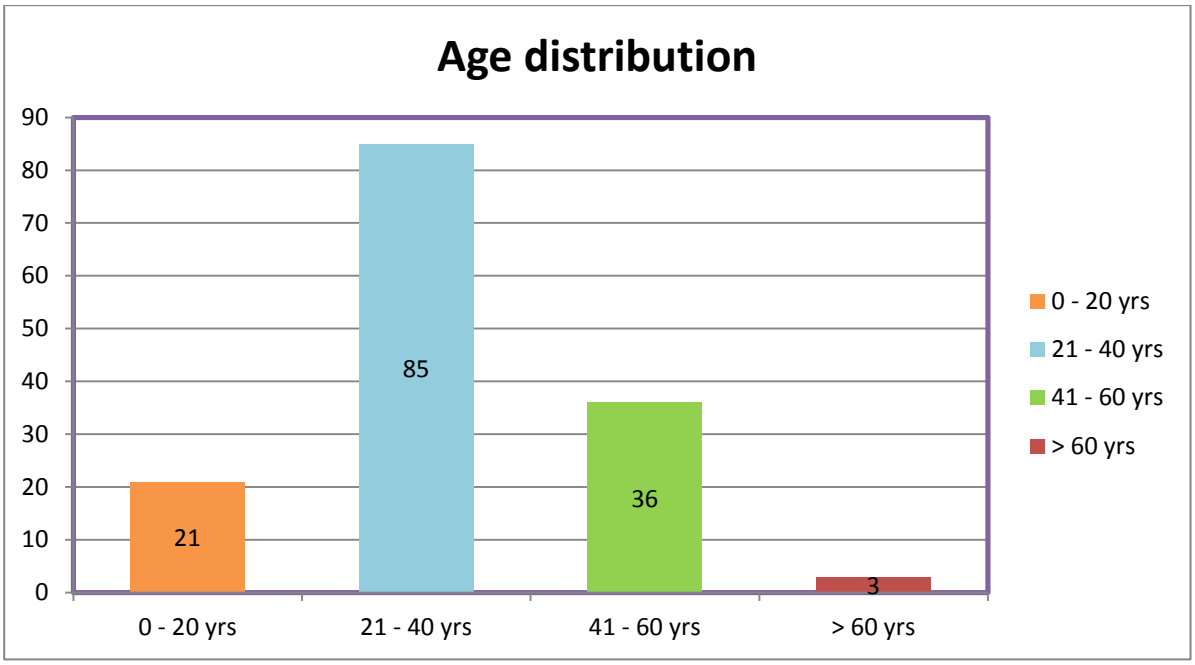


Figure 3.2: Age distribution of the patients with head and neck infections

3.2. Sources of Infection

3.2.1. Odontogenic vs Non-Odontogenic aetiology

Odontogenic origins of infection were classified into peri apical lesions, pericoronitis, periodontitis and alveolar osteitis.

Non-odontogenic causes were identified as trauma, sialadenitis, pharyngitis, Burkitt's lymphoma, immune suppression, presence of a foreign body and unknown aetiology.

Sixty four patients presented with an odontogenic cause of infection, while 81 presented with a non-odontogenic cause. Peri-apical lesions presented in the majority of patients with odontogenic cause of infection (n=32; 50%), followed by alveolar osteitis (n=22; 34%), pericoronitis (n=4; 10%) and periodontitis (n=4; 6%), as illustrated in figure 3.3. Where the aetiology was non-odontogenic, 60 patients (74%) presented with infection of an unknown aetiology, followed by trauma (n=18; 22%) being the second most common cause of non-odontogenic infections. One patient presented with Burkitt's lymphoma, another with immune suppression and yet another with the presence of a foreign body as the cause of infection. (Figure 3.4)

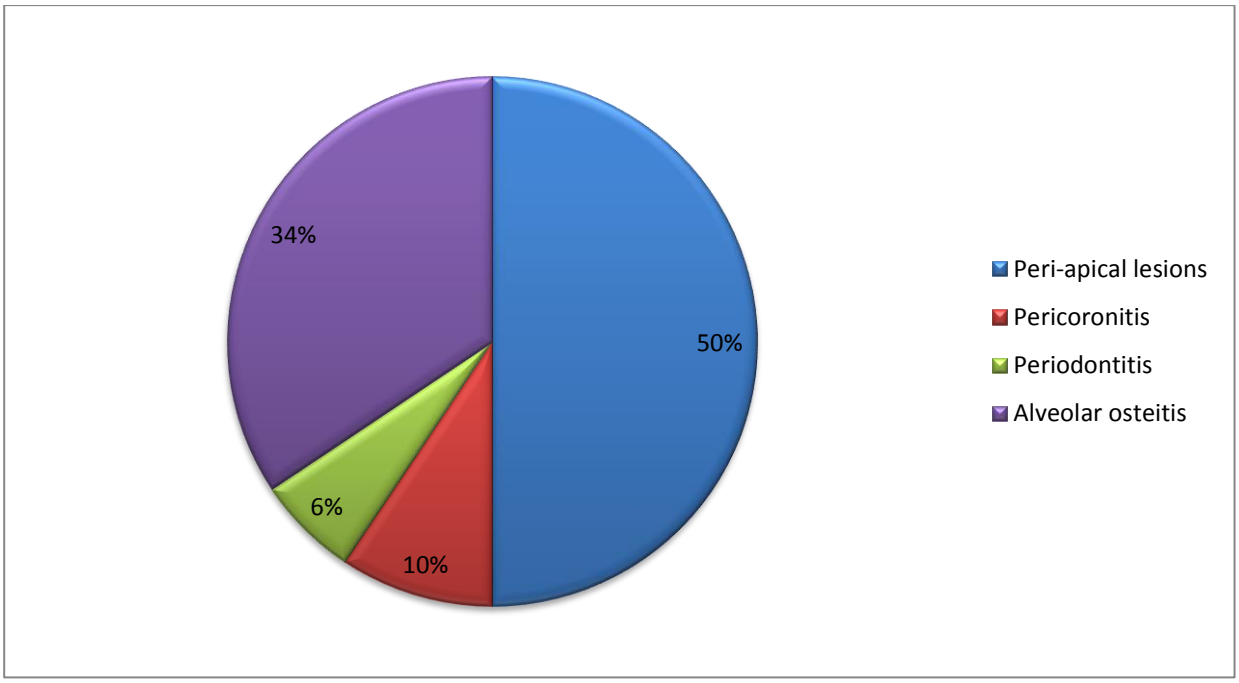


Figure 3.3: Cases of odontogenic aetiology

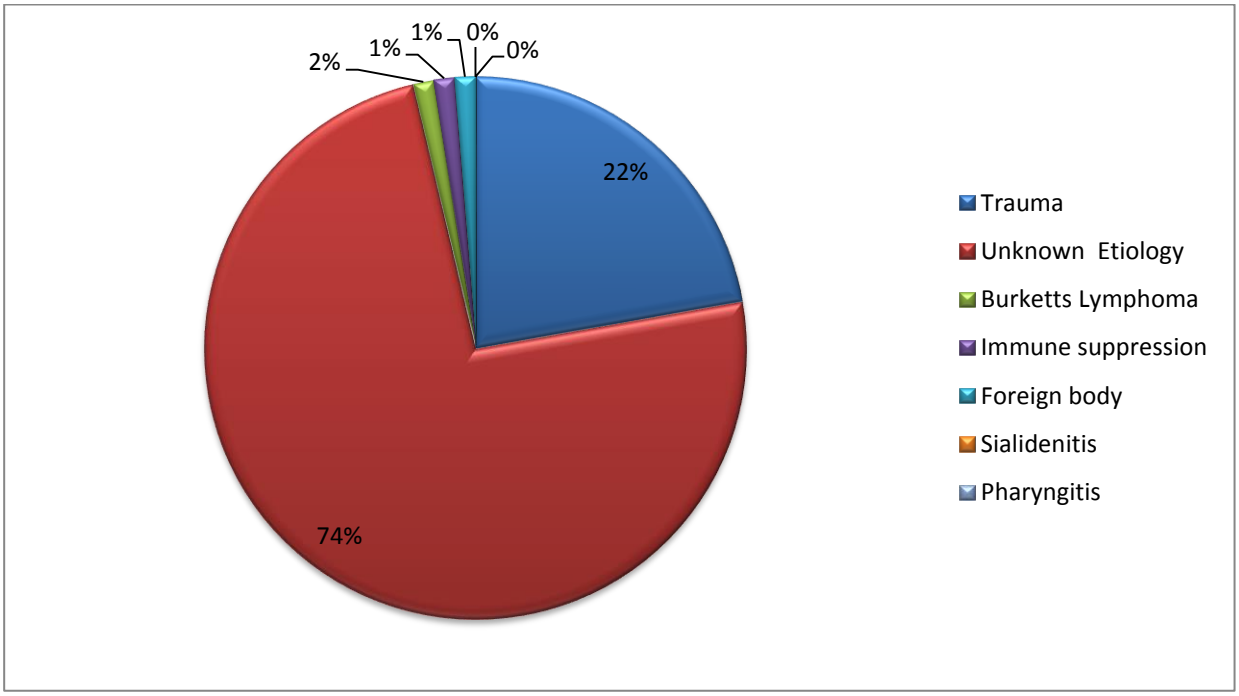


Figure 3.4: Cases of non-odontogenic aetiology

3.3. Fascial Space Involvement

The majority (81) of patients had only a single space involved, while 54 had multiple fascial space involvement. In 10 cases the fascial space remained unidentified.

3.3.1. Single Fascial Space Involvement

The most common single fascial space involved was the submandibular space with a total of 50 patients presenting with infection in this space. Ten cases involved the buccal space (1%), 8 the submental (10%), 6 the peri-orbital (8%) and 3 the zygomatic space (4%), respectively. The submasseteric, para-pharyngeal, superior labial and temporal spaces were less commonly affected and presented with 1 case each. This is represented in figure 3.5.

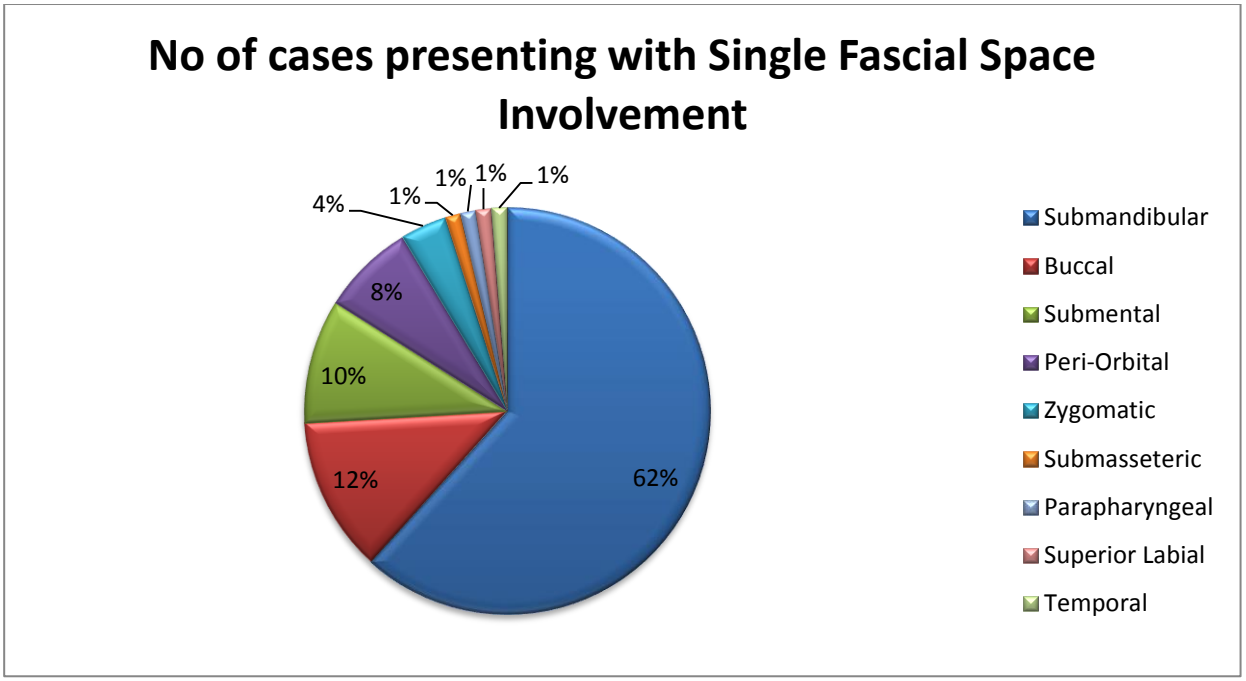


Figure 3.5: Number of cases presenting with single fascial space involvement

3.3.2. Multiple Fascial Space Involvement

Of the 54 patients that presented with multiple space involvement, 31 (57%) had 2 spaces affected, 22 (41%) had 3 spaces affected and only 1 (2%) had 4 fascial spaces being affected, as shown in figure 3.6.

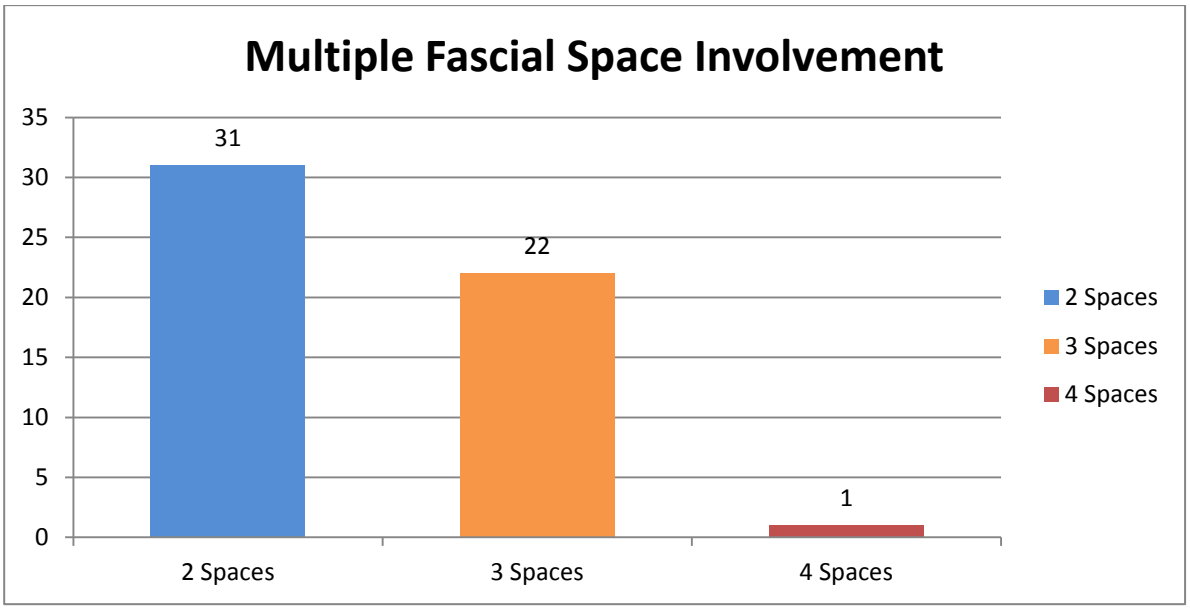


Figure 3.6: Number of cases presenting with multiple fascial space involvement

The most common space in multiple space involvement was the submandibular space (35 cases), followed by the sublingual (15 cases) and submental (14 cases) spaces, respectively.

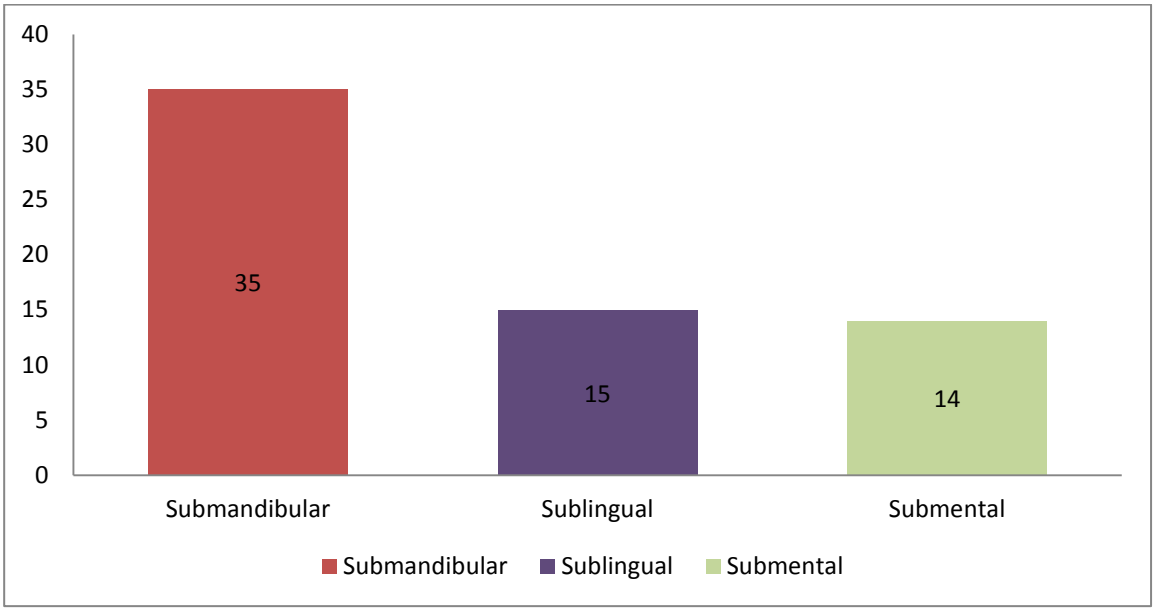


Figure 3.7: Common spaces in multiple fascial space involvement

3.4. Isolated bacteria

Thirty eight different types of micro-organisms were isolated from patients presenting with head and neck space infections. The most commonly isolated bacteria were *Bacteroides* species, appearing in 35 pus swabs (15%). No bacteria were isolated in 27 swabs (11%), while Viridans streptococci were isolated in 25 aspirates (11%) and *Staphylococcus aureus* in 18 (8%).

Less commonly isolated were coagulase negative *Streptococcus* (9 specimens - 4%), *Streptococcus constellatus* (10 specimens - 4%), alpha; beta and non-haemolytic *Streptococcus* (19 specimens - 3%), *Morganella morganii* species (8 specimens - 3%), S*treptococcus anginosus* (8 specimens - 3%) and *Streptococcus mitis/oralis* (8 specimens - 3%).

Table 3.1: Isolated micro-organisms

Group	Micro-organism	No of specimens presenting with isolates
AEROBIC GRAM -	*Acinetobacter baumannii complex*	2
	Burkholderia cepacia	3
	Escherichia coli	1
	Klebsiella oxytoca	1
	Klebsiella pneumoniae	3
	Proteus species	1
	Pseudomonas aeruginosa	3
	Pseudomonas putida	1
AEROBIC GRAM +	*Coagulase negative staphylococcus*	9

ANAEROBIC GRAM -	*Bacteroides*	35
FACULTATIVE GRAM -	*Enterobacter cloacae complex*	4
	Gram negative bacillus	6
	Haemophilus parainfluenzae	1
	Morganella morganii	8
FACULTATIVE GRAM +	*Cornybacterium species*	4
	Gemella morbillorum	1
	Micrococcus species	1
	Staphylococcus species	4
	Staphylococcus aureus	18
FUNGI	*Candida albicans*	3
	Yeast not Candida albicans	3
NORMAL FLORA	*Normal oral flora*	1

FACULTATIVE ANAEROBIC GRAM +	*Staphylococcus epidermidis*	7
	Staphylococcus haemolyticus	3
	Streptococcus anginosus	8
	Streptococcus alpha haemolytic	13
	Streptococcus bovis	4
	Streptococcus constellatus	10

	Streptococcus cristatus	1
	Streptococcus group A	3
	Streptococcus group C	1
	Streptococcus group F	4
	Streptococcus mitis/ oralis	8
	Streptococcus non-haemolytic	6
	Streptococcus pyogenes	1
	Staphylococcus sanguinis	1
	Staphylococcus warneri	1
	Viridans streptococci	25

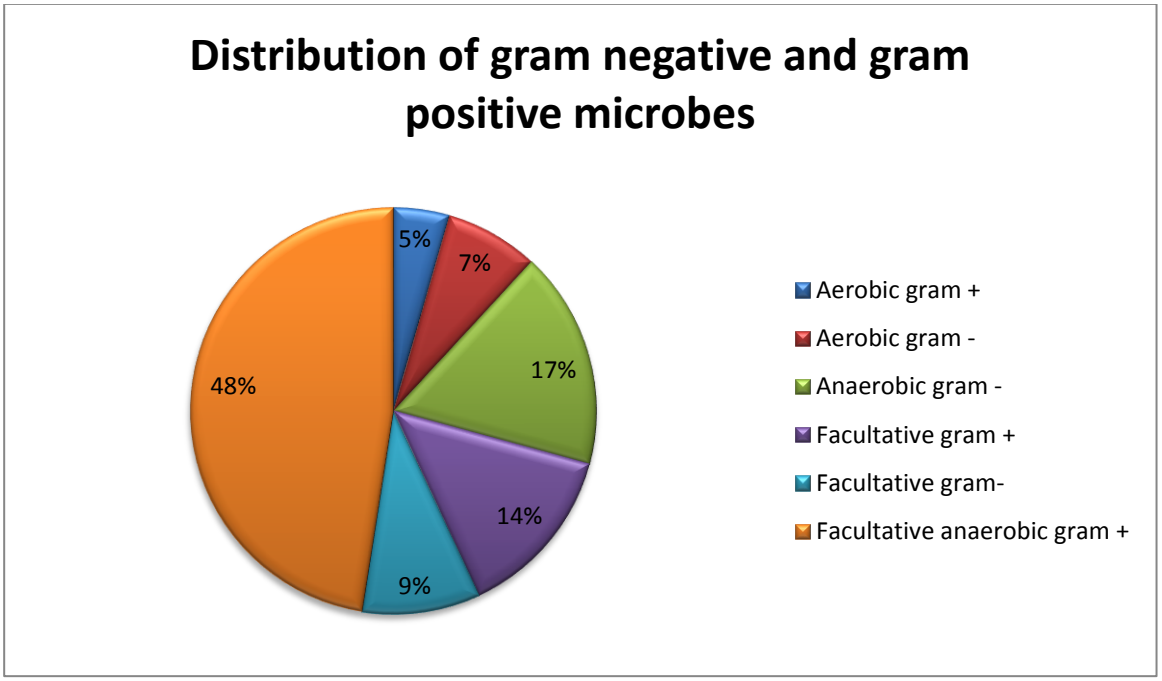


Figure 3.8: Distribution of gram negative and gram positive microbes

The isolated microbes were clustered into 6 categories: aerobic gram positive, aerobic gram negative, anaerobic gram negative, facultative gram positive, facultative gram

negative and facultative anaerobic gram negative. The majority of isolates (n=96; 48%) were facultative anaerobic gram positive flora, followed by anaerobic gram negative (n=35; 17%), facultative gram positive (n=28; 14%), facultative gram negative (n=19; 9%) and aerobic gram negative (n= 15; 7%). The least commonly isolated group were the aerobic gram positive flora (n=9; 5%). This is displayed in figure 3.8.

3.5. Antibiotic Sensitivities

Bacteroides species, found in 35 specimens, were 100% sensitive to metronidazole, carbapenems, piperacillin, clindamycin, cefoxitin, chloramphenicol and amoxicillin with clavulanic acid.

Six of the 25 isolates of Viridans streptococci showed resistance to Erythromycin/azithromycin (24%), while 15 displayed sensitivity (60%).
Four (16%) of the isolates displayed resistance to Clindamycin while 16 (64%) showed sensitivity.
Seven isolates showed sensitivity to penicillin/ampicillin (28%) while only 1 showed resistance (4%). Four isolates were sensitive to cefotaxime/ceftriaxone (16%) while 1 (4%) was resistant. Four isolates were sensitive to vancomycin (16%) and 1 to linezolid (4%).

Staphylococcus aureus were isolated in 18 specimens, and showed 100% resistance to penicillin and 44.4% (8 isolates) resistance to trimethoprim-sulphamethaxazole. Seven isolates (38.9%), however, showed sensitivity to trimethoprim-sulphamethaxazole.
There was 83.3% (15 isolates) sensitivity to cloxacillin, while 11.1% (2 isolates) showed resistance. Clindamycin was effective in 77.8% (14 isolates) but resistance was noted in 16.7% (3 isolates).
Erythromycin/azithromycin was also effective against staphylococci, showing sensitivity rates of 72.2% (13 isolates) and a low resistance of 5.5% (2 isolates).
With regard to tetracycline, 3 isolates (16.7%) were resistant and 38.9% (7 isolates) were found to be sensitive. Only 1 case (5.5%) was sensitive to vancomycin.

Other commonly isolated microbes and their sensitivities patterns are displayed on Table 3.2:

Table 3.2: Antibiotic sensitivities to commonly isolated microbes

	BACTEROIDES		*ST. VIRIDANS*		*STAPH AUREUS*		*ST. CONSTELLATUS*		*COAGULASE- STREP*		*MORGANELLA MORGANII*		*ENT. CLOACAE*		*ST. MITIS/ORALIS*	
	R	S	R	S	R	S	R	S	R	S	R	S	R	S	R	S
Penecillan/ampicillin			4,76%	33,33%	100%			66,66%	50%	25,00%	75%				87,5%	
Amoxicillan with clavulanic acid		100%									62,5%		100%			
Trimethoprim-sulphamethaxazole					44,44%	38,88%					25,0%	50%		100%		
Cefuroxime											100%		100%			
Metronidazole		100%														
Ceftriaxone			4,76%	19,04%												
Cefoxitine		100%											75%			
Cloxacillin					11,11%	83,33%										
Ciprofloxacine												50%		100%		
Tetracycline					16,66%	38,88%										
Cephotaxime								22,22%							50%	12,50%
Clindamycin		100%	19,04%	76,19%	16,66%	77,70%		77,77%	25%	75%					12,5%	75%
Erythromycin/azithromycin			28,5%	71,42%	5,55%	72,22%	11,11%	66,66%	37,5%	62,5%					50%	25%
Vancomycin				19,04%		5,55%		11,11%		25%						50%
Gentamycin												100%		100%		
Imipenem																
Amikacin												50%		100%		12,5%
Linezolid				4,76%												
Ertapenem												87,5%		100%		
Meropenem												87,5%		75%		
Colistin											37,50%			25%		

Streptococcus constellatus was also sensitive to clindamycin, vancomycin, cefotaxime and ceftriaxone.

Streptococcus mitis/oralis displayed sensitivity to clindamycin but resistance to penecillan, erythromycin, azithromycin, cefotaxime and ceftriaxone.

Coagulase negative streptococci were also resistant to penicillan but sensitive to cloxacillin, erythromycin, azithromycin, clindamycin, trimethoprim-sulfamethoxazole and tetracycline.

3.5.1. Sensitivity of isolates to commonly prescribed antibiotics

Twenty two of the isolates were sensitive to amoxicillin with clavulanic acid (15.2%), 28 were sensitive to penicillin (19.3%) and 61 to erythromycin/azithromycin (42.1%). The majority of the isolates (95) were sensitive to clindamycin (65.5%).

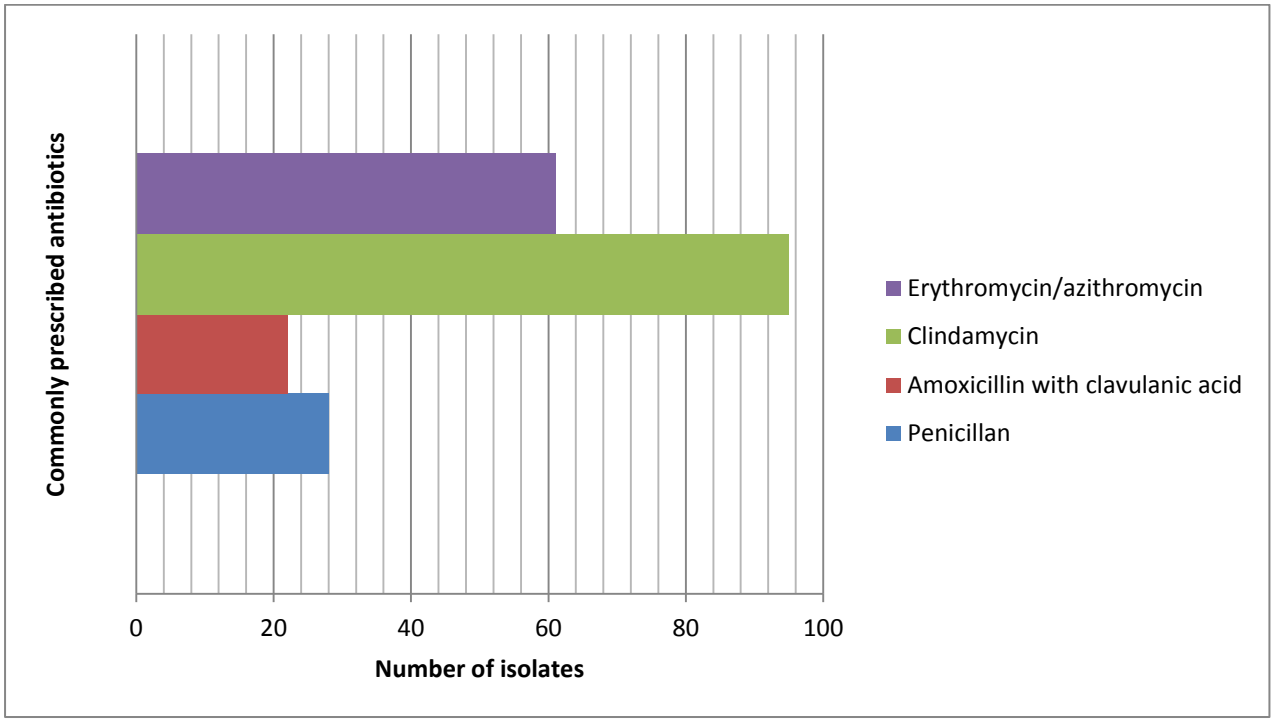


Figure 3.9: Sensitivity of commonly prescribes antibiotics

3.6. Statistical analysis

Descriptive and inferential statistics was computed for all variables. Bivariate analysis was used for identifying association. P-value based on the Chi-square test and Cramer's test was used. A p-value of <0.05 was considered to be statistically significant.

The microbial spectrum of head and neck space infections was analysed against patients' variables. Comparative analysis was performed on:-

- Gender
- Age groups
- Odontogenic versus non-odontogenic source of infections
- Single compared to multiple space involvement
- Microbial sensitivity to antibiotics

3.6.1. Age and cause of disease

Table 3.3: Association between age and cause of disease

		Cause of disease									Total
		Periapical abscess	Periodontal abscess	Pericoronitis	Alveolar osteitis	Trauma induced	Burketts lymphoma	Foreign body	Immune suppression	Unknown	
Age Category	[0 - 20]	6	1	0	0	1	0	0	0	14	22
	[21 - 30]	14	0	1	10	8	0	0	1	11	45
	[31 - 40]	7	0	4	3	8	0	0	0	15	37
	[41 - 50]	6	3	1	8	2	0	1	0	12	33
	[51 - 60]	0	0	0	0	0	1	0	0	3	4
	[> 60]	0	0	0	1	0	0	0	0	2	3
Total		33	4	6	22	19	1	1	1	57	144

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	80.248[a]	40	0,000
Likelihood Ratio	58,819	40	0,028
Linear-by-Linear Association	0,432	1	0,511

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	0,747	0,000
	Cramer's V	0,334	0,000

Interpretation of chi-square test

The p-value associated with the chi-square test is used to determine whether the association between two categorical variables is statistically significant. If the p-value is less than 0.05, then the association between the variables is statistically significant.

As the level of association is significant, the Cramer's V value can be interpreted. The Cramer V value provides the strength of association. The general interpretation is as follows:

V = 0.10 Small effect

V = 0.3 Medium effect

V = 0.50 Large effect

Using these guidelines the association between age category and cause of disease is significant (p = 0.000) with a medium effect size (Cramer's V = 0.334).

This indicates that there is a significant association between age category and cause of disease.

3.6.2. Age and space

Table 3.4: Association between age and fascial space

Age category and space cross tabulation				
		Single/multiple space		
		Single space	Multiple space	Total
Age Category	[0 - 20]	20	2	22
	[21 - 30]	30	15	45
	[31 - 40]	21	17	38
	[41 - 50]	14	19	33
	[51 - 60]	4	0	4
	[> 60]	2	1	3
Total		91	54	145

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	16.898[a]	5	0,005
Likelihood Ratio	19,712	5	0,001
Linear-by-Linear Association	6,573	1	0,010
N of Valid Cases	145		

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	0,341	0,005
	Cramer's V	0,341	0,005

Using the above guidelines the association between age category and fascial space is statistically significant (p = 0.005) with a medium strength of association (Cramer's V = 0.341).

3.6.3 Age and presence of microorganisms

Table 3.5: Association between age and identified microorganisms

Age category and microbiology organisms cross tabulations				
		Presence of microorganisms		
		Yes	No	Total
Age Category	[0 - 20]	19	3	22
	[21 - 30]	33	11	44
	[31 - 40]	33	5	38
	[41 - 50]	27	6	33
	[51 - 60]	2	2	4
	[> 60]	3	0	3
Total		117	27	144

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	5.549[a]	5	0,353
Likelihood Ratio	5,540	5	0,354
Linear-by-Linear Association	0,001	1	0,982
N of Valid Cases	144		

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	0,196	0,353
	Cramer's V	0,196	0,353

Using the above guidelines the association between age and presence of microorganisms is statistically insignificant (p = 0.353). It is therefore not necessary to calculate the Cramer's V value.

3.6.4. Gender and cause of disease

Table 3.6: Association between gender and cause of disease

Sex and cause of disease cross tabulation											
		Cause of disease									
		Periapical abscess	Periodontal abscess	Pericoronitis	Alveolar osteitis	Trauma	Burkitt's lymphoma	Foreign body	Immune suppression	Unknown	Total
Sex	Male	21	2	1	13	12	1	1	1	45	97
	Female	12	2	5	9	7	0	0	0	12	47
Total		33	4	6	22	19	1	1	1	57	144

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	13.541[a]	8	0,095
Likelihood Ratio	14,239	8	0,076
Linear-by-Linear Association	6,179	1	0,013

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	0,307	0,095
	Cramer's V	0,307	0,095

Using the above guidelines the association between sex and cause of disease is statistically insignificant (p = 0.095). It is therefore not necessary to calculate the Cramer's V value.

3.6.5. Gender and space of infection

Table 3.7: Association between gender and fascial space

Sex and space of infection				
		Single/multiple		
		Single space	Multiple space	Total
Sex	Male	58	39	97
	Female	33	15	48
Total		91	54	145

Chi-Square Tests			
	Value	Df	Asymptotic Significance (2-sided)
Pearson Chi-Square	1.102[a]	1	0,294
Continuity Correction[b]	0,752	1	0,386
Likelihood Ratio	1,117	1	0,291
Linear-by-Linear Association	1,094	1	0,295
N of Valid Cases	145		

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	-0,087	0,294
	Cramer's V	0,087	0,294
N of Valid Cases		145	

Using the above guidelines the association between sex and fascial space is insignificant (p = 0.294). It is therefore not necessary to calculate the Cramer's V value.

3.6.6. Gender and microbiology

Table 3.8: Association between gender and presence of microorganisms

Sex and microorganisms cross tabulations				
		Microbiology organisms		
		Yes	No	Total
Sex	Male	72	24	96
	Female	45	3	48
Total		117	27	144

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	7.385[a]	1	0,007
Continuity Correction[b]	6,205	1	0,013
Likelihood Ratio	8,570	1	0,003
Linear-by-Linear Association	7,333	1	0,007

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	-0,226	0,007
	Cramer's V	0,226	0,007

Using the above guidelines the association between the patient's sex and presence of microorganisms is statistically significant (p = 0.007) with a small to medium strength of association (Cramer's V = 0.226).

3.6.7. Microorganisms and antibiotic resistance

Table 3.9: Association between microorganisms and antibiotic resistance

Microorganisms and resistance				
		Resistant		
		No	Yes	Total
Microorganisms	Yes	58	59	117
	No	26	0	26
Total		84	59	143

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	22.320[a]	1	0,000
Continuity Correction[b]	20,288	1	0,000
Likelihood Ratio	31,659	1	0,000
Linear-by-Linear Association	22,164	1	0,000
N of Valid Cases	143		

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	-0,395	0,000
	Cramer's V	0,395	0,000

Using the above guidelines the association between the microorganism and resistance is statistically significant (p = 0.000) with a medium strength of association (Cramer's V = 0.395).

3.6.8. Microorganisms and antibiotic sensitivity

Table 3.10: Association between microorganisms and antibiotic sensitivity

Microorganisms and sensitivity cross tabulations				
		Sensitive		
		No	Yes	Total
Microorganisms	Yes	16	101	117
	No	26	0	26
Total		42	101	143

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	76.418[a]	1	0,000
Continuity Correction[b]	72,313	1	0,000
Likelihood Ratio	79,783	1	0,000
Linear-by-Linear Association	75,884	1	0,000
N of Valid Cases	143		

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	-0,731	0,000
	Cramer's V	0,731	0,000

Using the above guidelines the association between the microorganism and sensitivity is statistically significant (p = 0.000) with a large strength of association (Cramer's V = 0.731).

3.6.9. Fascial space and antibiotic resistance

Table 3.11: Association between fascial space and antibiotic resistance

Fascial space and antibiotic resistance				
		Resistant		
		No	Yes	Total
Single/multiple	Single space	52	38	90
	Multiple space	33	21	54
Total		85	59	144

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	.155[a]	1	0,694
Continuity Correction[b]	0,048	1	0,827
Likelihood Ratio	0,155	1	0,693
Linear-by-Linear Association	0,154	1	0,695

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	-0,033	0,694
	Cramer's V	0,033	0,694

Using the above guidelines the association between fascial space and resistance to antibiotics is statistically insignificant (p = 0.694). It is therefore not necessary to calculate the Cramer's V value as it is insignificant.

3.6.10. Fascial space and antibiotic sensitivity

Table 3.12: Association between fascial space and antibiotic sensitivity

Infection space and sensitivity level crosstabulations				
		Sensitive		
		No	Yes	Total
Single/multiple	Single space	29	61	90
	Multiple space	14	40	54
Total		43	101	144

Chi-Square Tests			
	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	.639[a]	1	0,424
Continuity Correction[b]	0,374	1	0,541
Likelihood Ratio	0,646	1	0,421
Linear-by-Linear Association	0,634	1	0,426

Symmetric Measures			
		Value	Approximate Significance
Nominal by Nominal	Phi	0,067	0,424
	Cramer's V	0,067	0,424

Using the above guidelines the association between fascial space and sensitivity to antibiotics is statistically insignificant (p = 0.424). It is therefore not necessary to calculate the Cramer's V value.

CHAPTER 4: DISCUSSION

Head and neck infections may result in serious morbidity and mortality. Septicaemia, airway obstruction, cavernous sinus thrombosis, necrotizing fasciitis, and mediastinitis are grave complications of infections in the fascial planes of the head and neck (Mathew et al, 2012). Early recognition of infection, as well as a thorough understanding of micro-organisms involved, and their sensitivity patterns are critical in the efficient management of such conditions. In addition to surgical incision and drainage, antibiotic therapy is vital for successful treatment. In order to administer anti-microbial agents effectively to a patient, microbiological data on the infection must be acquired. However, information on the microbiology and antibiotic susceptibility requires time, and subsequently antimicrobials are administered as the principal treatment modality prior to obtaining the aspirate results (Ye et el, 2017). Thus, the selection of the appropriate antibiotics is essential for successful treatment of these infections.

The purpose of this study was to identify the microbial flora present in head and neck space infections and in so doing, provide a better understanding on the management of these infections.

In this study 145 patients who had pus aspirates and swabs taken, were selected as the sample population. The patients that most commonly presented with head and neck space infections ranged from 21 to 40 years. This outcome is similar to the age distributions described by Fating et al (2014) and Bahl et al (2014). Similarly to the findings of previous studies by Singh et al (2014), George et al (2012) and Shah et al (2016), head and neck space infections were more common in males than females.

Single fascial spaces were more commonly involved than multiple spaces. This result corroborates the findings of Ye et al (2017). Conversely, Ibeyemi at al (2014) reported multiple space involvement as being more common. The most common single space involved in head and neck space infections was the submandibular space, followed by the buccal and submental space, a finding that is in agreement with the findings of Singh et al (2014). The spaces most often implicated in multiple fascial space involvement were the submandibular, sublingual and submental spaces, respectively. Rega et al (2004) and Fating et al (2014), however, reported the submandibular space as the most common followed by the submental and lateral pharyngeal spaces.

Most infections in this study were non-odontogenic in origin (56%). This is contrary to the findings by Boscolo-Rizzo et al (2009), who reported odontogenic infection as the most common cause. Odontogenic causes of infection compromised 50 % peri-apical lesions, 34% alveolar osteitis, 10% pericoronitis and 6% periodontitis. In contrast, George et al (2012) reported 71%, 17%, 6%, and 1% of odontogenic causes of infection to be due to pulpitis, periodontitis, alveolar osteitis and needle tracts respectively.

4.1. Microorganisms

The microbiology that was isolated in this study was credited to the primary source of the infection, which is the oral cavity mucosa. This was consistent with the results found by Selfars et al (2004).

In the present study, the microorganisms isolated ranged from gram positive and negative facultative aerobes, gram positive facultative anaerobes, gram negative anaerobes, gram positive and negative aerobes. Similar to a study by Singh et al (2014) which reported anaerobic dominance in isolates, the current study found gram positive facultative anaerobes to be the most commonly isolated, followed by gram negative facultative anaerobes.

Bacteroides spp. was the most commonly isolated micro-organism and the only anaerobe isolated in 17% of all cases. In contrast to the current study *Bacteroides fragilis* and *Bacteroides corrodens* comprised 5% and 2.5% of isolates respectively, in the study by Walia et al (2013).

The most commonly isolated gram positive facultative anaerobe was Viridans streptococci (11%), followed by *Staphylococcus aureus* (8%). This finding is in agreement with those of previous studies which reported Bacteroides spp, Viridans streptococci and *Staphylococcus aureus* as the most commonly isolated micro-organisms in head and neck infections (Cordesmeyer et al., 2017; Farmahan et al., 2014; Rega et al., 2006).

Molomo et al (2016) and Cabral et al (2016) found *Staphylococcus aureus* to be the most common isolate, followed by other *Staphylococcus* spp.

Gram negative aerobes were isolated in 6% of specimens and the distribution was as follows: *Klebsiella* in 4 specimens, *E. coli* in 1 specimen and *Pseudomonas aeruginosa* in 3 specimens. The presence of gram negative bacilli was also reported by Walia et al (2013). In addition, *Prevotella* was the most commonly isolated gram negative bacilli in a study by Singh et al (2014), occurring in 25.81% of the specimens.

One unanticipated finding was the identification of *Enterobacter cloacae* complex and *Morganella morganii* which were isolated in 2% and 3% of cases respectively. *Enterobacter cloacae* is a member of the normal gut flora in many humans and is not usually a primary pathogen. However, some strains have been associated with urinary and respiratory tract infections in immunocompromised individuals (Stock, 2001).

Miller (2015) reported that, although *M. morganii* has a wide distribution, it is considered an uncommon cause of community-acquired infection and is most often encountered in postoperative and other nosocomial infections such as urinary tract infections. He elaborated that patients in whom bacteraemia develops are typically immunocompromised, diabetic, or elderly, or have at least one serious underlying disease. This is consistent with the patient in whom the isolate was obtained. She was HIV positive and the source of infection was chronic sepsis secondary to the placement of a mandibular reconstruction plate.

Fungi were isolated in 6(2%) of specimens. The presence of fungal spores in pus is consistent with the report by Walia et al (2013).

Some of the isolated micro-organisms are part of the normal flora; they may however cause infections should there be a breach in the host defense mechanism. An endogenous source is commonly reported particularly in association with odontogenic infections (Anguzu, 2007).

4.2. Antibiotic sensitivities

In the present study, *Bacteroides* spp was 100% sensitive to metronidazole, carbapenems, piperacillin, clindamycin, cefoxitin, chloramphenicol and amoxicillin with

clavulanic acid. The South American study by Fernadez-Canigia et al (2012) showed similar results with sensitivity rates of 100% to metronidazole and tigecycline, 99% to imipenem and piperacillin-tazobactam, 96% to ampicillin-sulbactam and 91% to moxifloxacin. Lower sensitivity rates, however, were found to cefoxitin (72%) and clindamycin (52%).

Viridans streptococci displayed highest sensitivities to erythromycin/azithromycin (71.43 %) and clindamycin (76.19 %). Shah (2016) also reported high sensitivity levels of 100% to carbenicillin, amikacin and imipenem and 89.4% to ceftriaxone. This differed from the findings by Rega (2004), who presented the highest sensitivity rates to penecillan (87.1%).

Staphylococcus aureus showed sensitivities of 83.33% to cloxacillin, 77.77% to clindamycin and 72.22% sensitivity to erythromycin/azithromycin. The microbe was however, 100% resistant to penicillin and 44.44% resistant to trimethoprim-sulphamethaxazole. Shah (2016) reported a higher sensitivity rate of 100% to amoxicillin with clavulanic acid and a lower resistance of 31.3% to amoxicillin.

Streptococcus constellatus and *Streptococcus mitis/oralis* were also most sensitive to clindamycin, but resistant to penicillan. This was consistent with a report by Rams et al (2014), who found that clindamycin was most effective against *S. constellatus*. Bancescu et al (2004) also reported clindamycin as having the highest sensitivity rates to the S*treptococcus mitis* group.

Coagulase negative streptococci were also resistant to penicillin (50.0%) but highly sensitive to clindamycin (75.0%) and erythromycin/azithromycin (62.50%). This result was consistent with the study by Shah (2016), who found a 100% resistance of this microbe to amoxicillin and amoxicillin with clavulanic acid, but 100% sensitivity to imipenem.

Gentamycin, meropenem and ertapenem were effective against *Morganella morganii* and *Enterobacter-cloacae* complex, characteristic for nosocomial infections. Cefuroxime was ineffective, as well as amoxicillin with clavulanic acid, displaying 100% resistance to *Enterobacter-cloacae*.

Studies by Stock (1998 and 2001), identified the natural antibiotic susceptibility of *Morganella morganii* and *Enterobacter cloacae* complex. The microbes were resistant to penicillin, cefoxitine, lincosamines and fusidic acid among others. Morganella morganii displayed sensitivity to aminoglycosides, piperacillin, third and fourth generation cephalosporins, carbapenems, trimethoprim, cotrimoxazole, and chloramphenicol, while Enterobacter was most susceptible to fosfomycin.

K. Rogers, a pharmacologist, toxicologist and senior editor of biomedical sciences at Encyclopaedia Britannica, reported that drug-resistant *Enterobacter* organisms have complicated treatment regimens particularly in nosocomial settings, where these organisms had become increasingly common. She added that treatment of *Enterobacter* infections comprised single-agent antimicrobial therapy, typically with an aminoglycoside, a fluoroquinolone, a cephalosporin, or imipenem. Rogers also found that *Enterobacter* may produce beta-lactamases, which cleave the central ring structure responsible for the activity of beta-lactam antibiotics, a group that includes imipenem and cephalosporins. Thus, repeated exposure to these drugs selects for beta-lactamase-synthesizing *Enterobacter*, giving rise to drug resistance. She adds that more modern approaches to *Enterobacter* infections have embraced combination therapy regimens involving multiple antibiotics with different core structures, such as an aminoglycoside or a fluoroquinolone in combination with a beta-lactam agent (Rogers 2017).

The three most commonly isolated microorganisms in the current study were sensitive to Clindamycin similar to findings reported by Bahl et al (2014) and Singh et al (2014). Clindamycin has been proven not to be effective as an empirical drug of choice in the treatment of odontogenic infections (Heim et al., 2017). Furthermore this antibiotic should be reserved for severe penicillin resistant infections to curtail the increasing resistance reported in literature (Poeschl et al., 2010; Border and Coke, 2017).

Bacteroides spp, the most commonly isolated bacteria, was found to be 100% sensitive to amoxicillin with clavulanic acid. Several studies have reported penicillin to be the drug of choice in the management of most odontogenic infections (Rega et al., 2006; Farmahan et al., 2014, 2015; Molomo et al., 2016; Shah, 2016; Heim et al., 2017).

Mathew et al (2014) reported penicillin to be highly effective, cheap, and display minimal side-effects. Fating et al (2013) reported that amoxicillin with clavulanic acid administered together with metronidazole, played a crucial role in the management and resolution of infection. It is for this reason that amoxicillin with clavulanic acid may be recommended for use as the empirical drug of choice for head and neck space infections.

Viridans streptococci and *Staphylococcus aureus* show high sensitivity to Erythromycin/azithromycin. Contrary to this finding, Singh et al (2014) reported low sensitivity rates to erythromycin in all isolates (38.89%).

In nosocomial infections the drug of choice would be limited to imipenem, meropenem and gentamycin. Shah (2016) also recommended that imipenem, carbenicillin and ceftriaxone should be cautiously used in more serious hospital borne infections.

4.3. Statistically insignificant associations

The results reveal that age did not influence whether micro-organisms were present in an aspirate, nor did it influence the antibiotic sensitivity or resistance profiles of the associated micro-organisms.

Gender did not play a significant role in the aetiology of the infection, nor the degree of antibiotic resistance. There seemed to be no substantial correlation between the age of the patient and the fascial space involved in the infection.

The results also show that the fascial space, in which the infection occurred, did not significantly influence neither the antibiotic resistance nor sensitivity profiles of the associated micro-organisms.

Furthermore, the aetiologic agent seemed to have no significant impact on the antibiotic resistance profiles of the microbes.

4.4 Statistically significant associations

There was a statistically significant association between age and aetiology.

Older patients in the 51-60 and greater that 60 year groups respectively, presented more commonly with unknown causes of infection. The aetiology could not be identified and could have perhaps been multifactorial or systemic in nature. This may be attributed to the fact that as patients age, the likelihood of a compromised immune system increases, bringing with it a susceptibility to not only oral infections, but pneumonia, intra-abdominal and urinary tract infections and even cancer.

The younger patients in the 0-20 and 21-30 age groups appeared to be more commonly affected with odontogenic causes of infection, such as alveolar osteitis, periodontal and peri-apical abscesses. This finding could be ascribed to the increasing loss of teeth and subsequent reduced risk for odontogenic infections. Bahl et al (2014) reported similar findings.

There was likewise a statistically significant association between the age and fascial space involved with the majority of patients presenting with single space involvement. This correlates with the findings of Walia et al (2013), where single space involvement was significantly more common. Patients younger than 40 years presented more commonly with single space involvement, while the older patients in the 5^{th} decade of life presented more commonly with multiple space involvement.

The association between gender and presence of microorganism was also statistically significant. The majority of specimens were acquired from males. This was consistent with studies by Bahl et al. (2014), Fating et al. (2014) and Walia et al. (2013) where the majority of isolates were obtained from male patients.

However, more microorganisms were identified in females (45 of 48 patients or 94%) than in males (72 of 97 patients or 74%).

There was moreover a statistical significance between the presence of micro-organisms and antibiotic sensitivity and resistance profiles. This is understandable, as the microorganisms isolated would have displayed sensitivity or resistance to an individual antibiotic.

It was similarly noted that gender of the patient was significantly associated to the antibiotic sensitivity profile. The majority of females (81.2%) presented with head and neck space infections caused by micro-organisms that were sensitive to particular antibiotics.

CHAPTER 5: CONCLUSION, LIMITATIONS AND RECOMMENDATIONS

Conclusion

This study clearly indicates that the predominant micro-organisms responsible for head and neck infections are gram positive facultative anaerobes. *Bacteroides* spp were the most commonly isolated bacteria, followed by Viridans streptococci and *Staphylococcus aureus*.

Most infections occurred in the third and fourth decades of life. Patients older than 50 years presented more commonly with non-odontogenic causes of infection, while younger patients more frequently presented with odontogenic causes of infection. Patients younger than 40 years presented more commonly with single space involvement, while the older patients in the 5th decade of life presented more commonly with multiple space involvement. Microorganisms were more frequently isolated in females than males and in patients in the age range of 21-40 years.

The most commonly isolated microorganisms (*Bacteroides* spp, *Staphylococcus aureus*, Viridans streptococci and coagulase negative streptococci) in the current study were sensitive to Clindamycin. *Bacteroides* spp, the most commonly isolated bacteria, was found to be 100% sensitive to amoxicillin with clavulanic acid.

The four most commonly prescribed antibiotics at the Livingstone Hospital Oral and Maxillo-facial Clinic are amoxicillin with clavulanic acid, penicillin, erythromycin and clindamycin.

Limitations of the study include the following:

- Incomplete medical reports
- This was a retrospective study which limits the variables that can be studied and prevents these variables from being modified
- The method of collection of the aspirates may have influenced the microbiology results, especially if the anatomical site of collection was not adequately cleaned with an alcohol swab prior to the procedure.

- Since the Maxillo facial clinic at Livingstone Hospital is a tertiary care centre, it is likely that most patients included in this study would have received antibiotics prior to admission. Therefore the emergence of resistance strains due to administration of these antimicrobial agents may not have been accurately assessed.

Recommendation

A longer study period with a larger sample size evaluating the microbial spectrum and resistance patterns may be necessary to monitor developing trends. Since odontogenic infection was found to be a common cause of head and neck space infections, particularly in the third and fourth decades of life, oral health education should further emphasise the importance of caries prevention, good oral hygiene practices, early presentation and intervention to minimise the complications of pulpitis.

Although Clindamycin was found to be effective against the most commonly identified microorganisms in this study, this antibiotic should be reserved for severe penicillin resistant infections to curtail the increasing resistance reported in literature.

6. REFERENCES

Akinbami, B.O., Akadiri, O. and Gbujie, D.C. 2010. Spread of Odontogenic Infections in Port Harcourt, Nigeria Journal of Oral and Maxillo-facial surgery 68(10): 2472-2477

Anguzu, J.R. and Olila, D. 2007. Drug sensitivity patterns of bacterial isolates from septic post-operative wounds in a regional referral hospital in Uganda African Health Sciences 7(3):148-154

Bahl, R., Sandhu, S., Singh, K., et al. 2014. Odontogenic infections: Microbiology and management Contemporary Clinical Dentistry 5(3): 307-311

Bancescu, G., Dumitriu, S., Bancescu, A., et al. 2004. Susceptibility testing of *Streptococcus mitis* group isolates Indian journal of medical research 119:257-261

Border M, Coke D. 2017 Oct. Increased Incidence of Clindamycin-Resistance in Head and Neck Infections within Oral and Maxillofacial Surgery. Journal of Oral and Maxillofacial Surgery. 75(10): e380

Boscolo-Rizzo, P., and Da Mosto, M. C. 2009. Submandibular space infection: a potentially lethal infection International Journal of infectious diseases 13(3):327-333

Brook, I., Frazier, E.H., and Gher, M.E. 1991. Aerobic and anaerobic microbiology of periapical abscess Oral microbiology and Immunology 6:123-125

Cabral, M., Gowrishankar, S., and Amerally, P. 2016. Investigation of the microbiology and antibiotic sensitivity of skin and soft tissue infections of the head and neck region. British Journal of Oral and Maxillofacial Surgery 54(10):124-125

Chang, L, Chi, H., Chiu N.C., Huang F.Y., et al. 2010. Deep neck infections in different age groups of children. Journal of Microbiology, Immunology and Infection. 2010 43(1):47-52.

Cohen, S., and Hargreaves, K. M. 2010. Cohen's pathways of the pulp-10th edition.

Cordesmeyer R, Kauffmann, Tröltzsch M, et al. 2017. Bacterial and histopathological findings in deep head and neck infections: a retrospective analysis. Oral Surg Oral Med Oral Pathol Oral Radiol. 2017 Jul;124(1):11-15.

Farmahan S, Tuopar D, Ameerally PJ, et al 2014. Microbiological examination and antibiotic sensitivity of infections in the head and neck. Has anything changed? Br J Oral Maxillofac Surg. 2014 Sep;52(7):632-635.

Farmahan S, Tuopar D, Ameerally PJ: A study to investigate changes in the microbiology and antibiotic sensitivity of head and neck space infections. Surgeon. 2015 Dec;13(6):316-20

Fating, N. S., Saikrishna, D., Vijay Kumar, G.S., et al. 2014. Detection of bacterial flora in orofacial space infections and their antibiotic sensitivity profile Journal of Maxillofacial and Oral surgery 13(4): 525-532

Fernadez-Canigia, L., Legaria, M.C., Castello, L., et al. 2012. First national survey of antibiotic susceptibility of the *bacteroides fragilis* group: Emerging resistance to carbapenems in Argentina. Antimicrobial agents and chemotherapy 56(3):1309-1314

Garg, R.K., Somvanshi D.S. 2011. Spinal tuberculosis: A review. The Journal of Spinal Cord Medicine 34(5):441-454

George, M. C., Ranganathan, L. K., Ghandi, S., et al. 2012. Odontogenic maxillofacial space infections at a tertiary care centre in North India: a five year retrospective study International Journal of Infectious diseases 16(4): e296-e302

Heim N, Faron A, Wiedemeyer V, et al. 2017. Microbiology and antibiotic sensitivity of head and neck space infections of odontogenic origin. Differences in inpatient and outpatient management. J Craniomaxillofac Surg. 2017 Oct;45(10):1731-1735.

Hupp, J. R., Ellis, E. and Tucker, M. R. 2008. Contemporary Oral and Maxillofacial Surgery-fifth edition. Missouri: Mosby Elsevier

Ibeyemi, S.T., Okoje –Adesomoju, V.N., Dada-Adegbola, H.O., et al. 2014. Journal of the West African College of Surgeons 4(4): 112-141

Maharaj, S. H. 2007. Microbiology of Submandibular space infections at the University of Witwatersrand. 2017. Research report. http://wiredspace.wits.ac.za/handle/10539/8540 [Accessed 28 June 2017]

Mathew, G. C., Ranganathan, L. K., Ghandi, S., et al. 2012. Odontogenic maxillofacial space infections at a tertiary care centre in North India: a five year retrospective study International Journal of Infectious diseases 16(4): e296-e302

Miller,J. R., 2015. Morganella Infections. Medscape https://emedicine.medscape.com/article/222443-overview#a0101 [Accessed 18 September 2018]

Molomo, E.M., Motloba, D.P., Bouckaert, M.M., et al. 2016. Bacteriology and management of orofacial infections in a Maxillofacial and Oral Surgery Clinic, South Africa. South African Dental Journal, 71(10): 474 - 477

Poeschl, P.W., Spusta, L., Russmueller, G., et al. 2010 Antibiotic susceptibility and resistance of the odontogenic microbiological spectrum and its clinical impact on severe deep space head and neck infections. Oral Surg Oral Medicine Oral Pathology Oral Radiology Endodontics. 110(2):151-6.

Rams, T.E., Feik, D., Mortensen, J.E., et al. 2014. Antibiotic susceptibility of periodontal streptococcus constellatus and streptococcus intermedius cinical isolates Journal of Periodontology. 85(12):1792-1798

Rega, A.J., Aziz, S.R. and Ziccardi, V.B. 2004. Microbiology and antibiotic sensitivities of deep neck space infections Journal of Oral and Maxillofacial surgery 62(1):25-26

Rogers, K. 2017. Enterobacter Bacteria Genus Britannica. Available: https://www.britannica.com/science/Enterobacter[Accessed 15.09.2018]

Selfars, J., Adielsson, A., Ebenfelt, A., et al. 2004. Deep neck infections remain a surgical challenge: A study of 72 patients Acta Otolaryngology 124:1191-1196

Shah, A., Ramola, V., and Nautiyal, V. 2016. Aerobic microbiology and culture sensitivity of head and neck space infection of odontogenic origin National Journal of Maxillofacial Surgery 7(1):56-61

Patil, S., Rao, R. S., Amrutha, N. et al. 2013. Oral microbial flora in health World journal of dentistry 4(4): 262-266

Shrivastav, R. P. 2014. An illustrated textbook Ear, Nose and Throat and Head and Neck surgery-second edition. New Delhi: Jaypee Brothers

Singh, M., Kambalimath, D. H., and Gupta, K. C. 2014. Management of odontogenic space infection with microbiology study Journal of Maxillofacial and Oral surgery 13(2): 133-139

Sowmya, Y. 2016. A review on the human oral microflora Research and reviews: Journal of dental sciences 4(3):1-5

Stock, I.and Weidemann, B. 1998. Identification and natural antibiotic susceptibility of Morganella morganii Diagnostic Microbiology and Infectious disease. 30(3):153-165.

Stock, I., Gruger, T. and Weidemann, B. 2001. Natural antibiotic susceptibility of strains of the Enterobacter cloacae complex International Journal of antimicrobial agents. 18(6):537-545.

Vineet, R.V. Flora of the oral cavity [Internet]. 1st ed. Smashwords; 2015. [cited 2019 Feb 16]; Available from https://www.researchgate.net/publication/291521896_Flora_of_the_oral_cavity

Walia, I.S., Borle, R.M., Mehendiratta, D., et al. 2013. Microbiology and antibiotic sensitivity of head and neck space infections of odontogenic origin Journal of Maxillofacial and Oral Surgery 13(1):16-21

Wang L.F., Tai C.F., Kuo W.R., et al Chien 2010. Predisposing factors of complicated deep neck infections: 12-year experience at a single institution. Journal Otolaryngology Head and Neck Surgery 39(4):335-41.

Ye, L., Liu, Y., Geng, A-L., Fu, H-Y. 2017. Microbiological examination to investigate the differences in microorganisms and antibiotic sensitivity of head and neck space infections. Biomedical Research 28(1): 290-294

Yuvaraj , V., Alexander, M., and Pasupathy, S. 2012. Microflora in Maxillofacial Infections – A Changing Scenario? Journal of Oral and Maxillofacial Surgery 70(1): 119-125

APPENDICES

APPENDIX A: ETHICS CLERANCE CERTIFICATE

R14/49 Dr N Singh

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
CLEARANCE CERTIFICATE NO. M170719

NAME: (Principal Investigator)	Dr N Singh
DEPARTMENT:	School of Oral Health Sciences Department of Oral Pathology Medical School
PROJECT TITLE:	Microbial spectrum of head and neck space infections at the Maxillo-Facial Clinic at Livingstone Hospital
DATE CONSIDERED:	28/07/2017
DECISION:	Approved unconditionally
CONDITIONS:	
SUPERVISOR:	Drs S Ngwenya and J Molepo
APPROVED BY:	Professor PE Cleaton-Jones, Chairperson, HREC (Medical)
DATE OF APPROVAL:	24/11/2017

This clearance certificate is valid for 5 years from date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Research Office Secretary on 3rd floor, Phillip V Tobias Building, Parktown, University of the Witwatersrand, Johannesburg.
I/We fully understand the conditions under which I am/we are authorised to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated from the research protocol as approved, I/we undertake to resubmit to the Committee. **I agree to submit a yearly progress report**. The date for annual re-certification will be one year after the date of convened meeting where the study was initially reviewed. in this case, the study was initially reviewed in **July** and will therefore be due in the month of **July** each year. Unreported changes to the application may invalidate the clearance given by the HREC (Medical).

Principal Investigator Signature

25 · 11 · 2017
Date

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES

APPENDIX B: DATA COLLECTION SHEET

NO	DATE OF PS	AGE				SEX		CAUSE									SPACE	MICROBIOLOGY ORGANISMS	ANTIBIOTIC SENSITIVITY		
		0-20	21-40	41-60	>60	M	F	OD PA	AO	PC	PD	NOD S	T	P	U				RESISTANT	SENSITIVE	INTERMEDIATE
A001	2015-10-09		17			M		PA									submandibular	Staphylococcus aureus	Penicillin / Ampicillin Trimethoprim-sulfamethoxazole	Cloxacillin Clindamycin Tetracycline	
A002	2014-10-15		9			M									U		submandibular	Staphylococcus aureus	Penicillin / Ampicillin	Cloxacillin Erythromycin / Azithromycin Clindamycin Trimethoprim-sulfamethoxazole Tetracycline	
A003	2016-05-10				54		F								U		submandibular	Acinetobacter baumannii		Ciprofloxacin Ceftazidime Cefepime Gentamicin Piperacillin/tazobactam	
A004	2016-03-03			26			F	PA									submandibular	Streptococcus non-haemolytic Streptococcus alpha-haemolytic Staphylococcus species Gram negative bacillus			
A005	2016-04-29			25		M							T				submandibular	No bacteria isolated			
A006	2016-03-07			36		M							T				submandibular sublingual retro-auricular buccal	Streptococcus non-haemolytic Streptococcus alpha-haemolytic Staphylococcus species Gram negative bacillus			
A007	2016-05-04				41	M									U		submandibular sublingual submental	No bacteria isolated			
A008	2016-12-10	7m					F								U		submandibular submental	Proteus species Gram negative bacillus (GNB) Streptococcus non-haemolytic Staphylococcus species			
A009	2015-03-03			26		M		PA									parapharyngeal	Klebsiella pneumoniae Streptococcus bovis Bacteroides species	Erythromycin / Azithromycin Trimethoprim-sulfamethoxazole Ampicillin / Amoxicillin	Clindamycin Ciprofloxacin Cefuroxime (Parenteral) Cefuroxime (Oral) Gentamicin	Amoxicillin-clavulanic acid Piperacillin/tazobactam
A010	2016-06-16			23		M									U		buccal	Bacteroides species		Metronidazole Carbapenems Piperacillin/tazobactam. Clindamycin Cefoxitin Chloramphenicol Amoxicillin-clavulanic acid	

Key:

A: Alveolar osteitis

B: Burkitts lymphoma

F: Foreign body

I: Immune suppression

Pa: Periapical

Pc: Pericoronitis

Pd: Periodontal

Ph: Pharyngitis

Pt: Parotitis

S: Sialedenitis

T: Trauma

APPENDIX C: NATIONAL HEALTH LABORATORY ANTIBIOTIC SENSITIVITY PROFILES

Gram negative bacteria

NHLS Port Elizabeth

bioMerieux Customer:
System #:

Laboratory Report

Printed Jul 17, 2017 15:11 CAT
Printed by: megan

Isolate Group: UB00640067-1

Bench: bld cults

Card Type: GN Testing Instrument: 000007D55B56 (The Damn male)
Card Type: AST-N255 Testing Instrument: 000007D55B56 (The Damn male)

Bionumber: 0407610554526610

Susceptibility Information	Card: AST-N255		Lot Number: 6550308103	Expires: Sep 30, 2018 12:00 CAT	
	Completed: Jul 15, 2017 23:25 CAT		Status: Final	Analysis Time: 9.50 hours	
Antimicrobial	**MIC**	**Interpretation**	**Antimicrobial**	**MIC**	**Interpretation**
Ampicillin	>= 32	R	Imipenem	<= 0.25	S
Amoxicillin/Clavulanic Acid	4	S	Meropenem	<= 0.25	S
Piperacillin/Tazobactam	<= 4	S	Amikacin	<= 2	S
Cefuroxime	4	S	Gentamicin	<= 1	S
Cefuroxime Axetil	4	S	Ciprofloxacin	<= 0.25	S
Cefoxitin	8	S	Tigecycline	<= 0.5	S
Cefotaxime	<= 1	S	Nitrofurantoin	128	R
Ceftazidime	<= 1	S	Colistin	<= 0.5	S
Cefepime	<= 1	S	Trimethoprim/Sulfamethoxazole	>= 320	R
Ertapenem	<= 0.5	S			

+= Deduced drug *= AES modified **= User modified

AES Findings:	Last Modified: Oct 30, 2014 14:07 CAT	Parameter Set: NHLS PE Copy of Global+Phenotypic
Confidence Level:	Consistent	

Installed VITEK 2 Systems Version: 07.01
MIC Interpretation Guideline: Copy of GLOBAL, 2003

Therapeutic Interpretation Guideline: Tshwane Academic Copy of PHENOTYPIC

AES Parameter Set Name: NHLS PE Copy of Global+Phenotypic

AES Parameter Last Modified: Oct 30, 2014 14:07 CAT

Page 2 of 2

Gram negative bacteria

NHLS Port Elizabeth

bioMerieux Customer:
System #:

Laboratory Report

Printed Jul 17, 2017 15:11 CAT
Printed by: megan

Isolate Group: UB00640067-1

Bench: bld cults

Card Type: GN Testing Instrument: 000007D55B56 (The Damn male)
Card Type: AST-N255 Testing Instrument: 000007D55B56 (The Damn male)

Bionumber: 0407610554526610

Susceptibility Information	Card: AST-N255		Lot Number: 6550308103	Expires: Sep 30, 2018 12:00 CAT	
	Completed: Jul 15, 2017 23:25 CAT		Status: Final	Analysis Time: 9.50 hours	
Antimicrobial	**MIC**	**Interpretation**	**Antimicrobial**	**MIC**	**Interpretation**
Ampicillin	>= 32	R	Imipenem	<= 0.25	S
Amoxicillin/Clavulanic Acid	4	S	Meropenem	<= 0.25	S
Piperacillin/Tazobactam	<= 4	S	Amikacin	<= 2	S
Cefuroxime	4	S	Gentamicin	<= 1	S
Cefuroxime Axetil	4	S	Ciprofloxacin	<= 0.25	S
Cefoxitin	8	S	Tigecycline	<= 0.5	S
Cefotaxime	<= 1	S	Nitrofurantoin	128	R
Ceftazidime	<= 1	S	Colistin	<= 0.5	S
Cefepime	<= 1	S	Trimethoprim/Sulfamethoxazole	>= 320	R
Ertapenem	<= 0.5	S			

+= Deduced drug *= AES modified **= User modified

AES Findings:		Last Modified: Oct 30, 2014 14:07 CAT	Parameter Set: NHLS PE Copy of Global+Phenotypic
Confidence Level:	Consistent		

Installed VITEK 2 Systems Version: 07.01
MIC Interpretation Guideline: Copy of GLOBAL, 2003
Therapeutic Interpretation Guideline: Tshwane Academic Copy of PHENOTYPIC
AES Parameter Set Name: NHLS PE Copy of Global+Phenotypic
AES Parameter Last Modified: Oct 30, 2014 14:07 CAT

APPENDIX D: LIVINGSTONE HOSPITAL ICU ANTIBIOTIC DOSING PROTOCOL

Livingstone Hospital Adult Critical Care Protocol

Antibiotic dosing in ICU

In the critical care environment always consider a loading dose of the antibiotic as soon as an infectious aetiology is considered, or as soon as an organism is considered to be a *pathogen*. Also note the doses in this guideline are for the use in the critically ill patient and are often off-label recommendations as accepted and recommended by the national and international critical care physicians.

Please note both CrCl and GFR are used as references as not one entity is used in literature

***To be kept at Temperature < 25°C**

▼To be stored at 2°-8°C

Other Suggestions re combination therapy

- Use Colistin with tigecycline for XDR ESBLs/acinetobacter
- Use Colistin with rifampicin, ceftazidime, imipenem, ciprofloxacin for other organisms
- For KPCs colistin monotherapy has 14% success
  vs. 73% for combinations with tigecycline/aminoglycosides
- Acinetobacter: Tigecycline combinations better clinical/microbial success vs. monotherapy

Daikos Antimicrob Agents Chemother 2009

Hirsch J Antimicrob Chemother 2010

KEY

CAP- community acquired pneumonia	
CAPD-peritoneal dialysis	MRSA-methicillin resistant staphylococcus aureus
CrCl-creatinine clearance	MSSA-methicillin sensitive staphylococcus aureus
CRE-carbepenamase resistant enterobacteraciae	MDR-multiple drug resistance
ESBL-extended spectrum beta lactamase	P.o. - per os
Fx- function	SSTI-skin and soft tissue infections
GFR-glomerular filtration rate	UTI- urinary tract infection
HAP-hospital acquired pneumonia	VAP-ventilator acquired pneumonia
IAI-intra-abdominal infections	VISA-vancomycin intermediate resistant staphylococcus aureus
IBW – ideal body weight	VRSA- vancomycin resistant staphylococcus aureus
ICU-intensive care unit	VRE-vancomycin resistant enterococci
IVI-intravenous	VAP-ventilator acquired pneumonia
MDR- multi drug resistant	VISA-vancomycin intermediate resistant staphylococcus aureus
MIC-minimum inhibitory concentration	VRSA- vancomycin resistant staphylococcus aureus

Drug	Normal renal function	Renal dysfunction:	Haemodialysis	Pitfalls and Pearls
Acyclovir	Herpes simplex encephalitis (14-21days) and other severe varicella-zoster infections (7-14days): 10mg/kg IVI 8hourly	GFR10-50 give 100% of dose12-24hourly GFR<10 give 50% of dose 24 hourly	Dose after dialysis	**Please note** to add Hydrocortisone in **Varicella Pneumonia** at dose of 200mg IVI 6hourlyx48hours then 100mgIVI 6hourlyx24hrs Then 100mg 8hourlyx24hrs, then continue to taper
✓ **Amikacin**	15mg /kg IVI once daily Amikacin* trough level (taken before next dose) <1mg/litre	AVOID		Obese patients should be dosed according to IBW with correction factor = IBW+ [0,4x(actual body weight-ideal body weight)]
✓ **Amoxicillin/ Clavulanic acid**	1.2g IVI 6hrly (with high creat. Clearance may need increased dose)	>30 GFR no adjustment 10-30 GFR 12hrly <10GFR 24hrly	Dose after dialysis	Does not cover ESBLs. Must be given immediately after reconstituting as it begins to degrade after 20 minutes. Does cover anaerobes. Avoid with severe hepatic impairment.
Amphotericin B	0,7mg-1mg/kg IVI for candidaemia given **as continuous infusion** 1,0mg-1,5mg/kg for cryptococcal meningitis or filamentous fungal infections	Pre-existing renal failure does not require adjusted dose but if on therapy renal function deteriorates significantly an echinocandin or azole will need to be considered		Monitor renal function, potassium, magnesium; prolonged therapy may lead to risk of anaemia. Nephrotoxicity is usually reversible.

Drug	Normal renal fx	Renal dysfunction:	Haemodialysis	Pitfalls and Pearls
Ampicillin	2g 4-6hrlyIVI	>50 CrCl: no change 30-50:6-8hrly 10-29:8-12hrly <10:12-24hrs	Dose as for CrCl<10, on dialysis days dose after dialysis, CAPD 250mg 12hrly	For E.faecalis(VSE), leads to E.coli resistance
Anidulafungin▼	Loading dose: 200mg IVI, then 100mg Infusion rate: not to exceed 1,1mg/min	No adjustment	Can be given without regard to haemo-dialysis	Newest echinocandin Do not give IV bolus Not studied for endocarditis, osteomyelitis and meningitis. Monitor LFTs, note for bronchospasm.
Azithromycin	500mg daily p.o.	No adjustment	No adjustment	Due to lower serum levels not associated with QTc prolongation
Caspofungin	70mg loading dose then 50mg daily IVI if > 80kg then 70mg dailyIVI	No adjustment	No adjustment	Mild hepatic insufficiency(Child-Pugh score 5-6) no adjustment; moderate hepatic insufficiency(Child-Pugh7-9) 35mg daily C.parapsilosis may be less susceptible. Not drug of choice for UTI due to Candidaemia(use azole or AmphoB) Echinocandin preferred in patients who have had recent azole exposure, whose illness is severe, or who are at risk to get C. glabrata or C. krusei
Note: Cephalosporins do not have enterococci/ESBL cover. Cephalosporins are an inappropriate choice for hospital acquired infections.				
Cefepime*	2g IVI stat 4-6g daily over 24hrs/ 2g over 3-4 hours, 8hrly infusions	30-49 CrCl:1g over 30min 8hrly <30 CrCl: 1g over 30 min 12hrly <10 CrCl: 1g daily	Dose as for CrCl<10 1g 24hrly, on dialysis day give after dialysis CAPD 1g daily	Anti-pseudomonal cephalosporin active against ceftazidime resistant Pseudomonas. Has activity against MSSA.
Ceftazidime*	2g IVI stat and 6g over 24hrs/ 2g 8hrly	>50 Cr Cl no dose change 30-50CrCl: 1g 12hrly 10-29CrCl: 1g 24hrly <10 CrCl: 1g 48 hrly	Dose as for CrCl <10, on dialysis days supplement 1g after dialysis CAPD 500mg daily	Very little anti- MSSA activity. Does not have streptococcal cover.
Ceftriaxone*	2g IVI 12 hourly (Meningitis 2g 12hourly)	No adjustment	On dialysis days give dose after dialysis	Associated with pseudo-biliary lithiasis. Has activity against MSSA. Caution: co-administration of calcium containing solutions e.g. TPN may lead to intravascular and pulmonary precipitation.

Drug	Normal renal fx	Renal dysfunction:	Haemodialysis	Pitfalls and Pearls
Ciprofloxacin	400mg IV as infusion 8hrly.	≥ 30 no change ≤30 GFR 400mg daily	On dialysis days, dose after dialysis	Lowers seizure threshold. If given via NGT absorption decreased ≥30%. Good cover against Haemophilus, Legionella and gram neg. rods like Pseudomonas. Drug of choice for shigellosis, typhoid and uncomplicated UTIs. Less potent against Streptococcus pneumonia and Enterococcus faecalis May increase QT.
Clarithromycin	500mg IV 12hrly Do not give as bolus	≥ 30 no change <30GFR 500mg Stat then 250mg 12-24hrly	Dose after dialysis On dialysis dose as for GFR<30	Interacts with Amiodarone, may increase QT interval; interacts with carbamazepine increase carbamazepine levels Interacts with cyclosporine increasing levels Interacts with statins increases risk for rhabdomyolysis
Clindamycin	600mg IVI 6hrly	No adjustment	No adjustment	Penetrates staphylococcal biofilms, useful adjunctive therapy when prosthetic devices cannot be removed. Penetrates well into avascular infected bone. Rare but can cause pseudomembranous colitis. Reduce exotoxin production by gram negatives.
Cloxacillin	2g IVI 6 hourly, in septic arthritis or disseminated MSSA we use 3g 6 hourly.	No adjustment	No adjustment	Best agent for staphylococcal clox.-susceptible. But note that Penicillin, amoxicillin, ampicillin, Piperacillin do not cover later organism, but in mixed infections co-amoxiclav. and cephalosporins will cover. Bacteriocidal inhibits bacterial cell wall synthesis Concurrent use of other hepatotoxic medications with cloxacillin ... may increase the potential for hepatotoxicity. Should not be routinely administered with Erythromycin.
Co-Amoxiclav	See Amoxicillin/Clavulanic acid above			
Colistin	See separate protocol for Colistin			
Cotrimoxazole (TMP/SMX) IV 80/400mg/5ml	20mgTMP/kg/day in divided doses 6hrly IV for sepsis (4 amps 6h if 80/400 vial)	CrCl >30 ml/min: no adjustment CrCl 15-30 ml/min: decrease dose by 50% CrCl <15 ml/min: Try to avoid if no alternative use 50% of dose	Dose after dialysis	Used for Stenotrophomonas bacteraemia in critical care. Avoid in respiratory infections as there is an association with MDR S. Pneumonia

Drug	Normal renal function	Renal dysfunction:	Haemodialysis	Pitfalls and Pearls
Daptomycin	6mg/kg for bacteraemia and right sided endocarditis, 4mg/kg for SSTI	If GFR >40 no adjustment If <40 give 48 hourly	Give after haemodialysis If patient on CVVHD larger dose required 8-10mg/kg	Bacteriocidal. Acts on biofilm. No lung penetration. No blood brain barrier penetration. For bacteraemia and right sided endocarditis. For SSTI. Protein binding. Side-effects; eosinophilic pneumonia, elevated CK with prolonged use
Doripenem	1g IVI over 4 hours 8 hrly, NOTE OFF LABEL, and there have been problems with higher dose	>50 CrCl no dose adjustment 30-50 CrCl give 50% of dose 8hrly 10-30 CrCl	Post HD 250mg infusion over 4 hours Post PD 500mg infusion over 4 hours	For nosocomial pneumonia and VAP, complicated intra-abdominal infection, complicated UTI
Doxycycline	**100 mg 12hourly p.o.**			Should be reserved for patients with tick bite fever. But rather use ciprofloxacin if patient intubated and/or shocked. Needs to be taken after meals with 250ml water. Drug interactions with carbamazepine, phenytoin, barbiturates, rifampicin
✓ **Ertapenem**	1g IVI daily, in critically ill 1g 12hrly	≤30 GFR then dose 500mg IVI daily	Dose after dialysis	Does not cover Enterococci, Pseudomonas or Acinetobacter. Covers ESBL enterobacteraciae
✓ **Erythromycin**	125mg IVI 6hrly given as prokinetic. Can cause hepatotoxicity. Can cause diarrhoea.			
Fluconazole	800mg IVI daily in ICU 400mgIVI daily in abdominal surgery prophylaxsis see Pifalls and pearls	>50 no dose adjustment 20-50 50% of dose 24hourly <20 25% of dose daily	200mg after dialysis	No activity against C.krusei Note drug interactions. In this unit there has been resistance to C glabrata as well as resistance shown to C parapsolosis. Not drug of choice if non-albicans species suspected. Recommended as prophylaxis against invasive candidiasis in patients recently operated (abdominal) and now have recurrent perforations or anastomotic leaks.
Ganciclovir	Induction:5mg /kg 12hrly IVI for 14-21days, followed in most cases by maintenance, but GIT CMV in HIV does not routinely require maintenance	>50 GFR 50% dose 12-24hrly; 10-50 GFR 25-50% 24hrly; <10 GFR 25% of dose 3 times/week	Give after dialysis and give dose as for GFR<10	Used for sight or life threatening CMV infection in immunocompromised patients and for prevention of CMV in transplant patients .Neutropenia in 40%, Thrombocytopenia in 20%
✓ **Gentamycin**	DailyIVI: 7 mg/kg: trough <1mg/l (daily level before next dose)	AVOID		TDS in infective endocarditis.

Drug	Normal renal function	Renal dysfunction:	Haemodialysis	Pitfalls and Pearls
Imipenem	1g IVI STAT over 40-60 min then 1g IVI 8 hourly over 3hrs	30-49 CrCl: 500mg over 30 min 8 hrly <30 CrCl: 500mg over 30 min 12 hrly	Dose after dialysis	Cyclosporine (↑cyclosporine levels),ganciclovir(↑risk of seizures), valproic acid(↓seizure threshold)
Linezolid	300mg IVI stat Then 300mg infusion over 12 hours. Then 600mg over 12 hours as infusion every 12 hours	No adjustment in renal insufficiency, CrCl >30ml/min	No dose adjustment but should be administered after haemodialysis in patients receiving haemodialysis	Bacteriostatic For VRE & MRSA. CI in patients on MAOI, pregnant or lactating. Thrombocytopenia has been reported usually related to duration of therapy
Macrolides: literature supports Azithromycin, Clarithromycin and Erythromycin in treatment of biofilm in respiratory Pseudomonal infections as in cystic fibrosis and Staphylococcal infection with devices insitu				
Meropenem	2g IVI STAT over 3-5 min then 2g 8 hourly over 3 hours	30-49 CrCl: 500mg over 30 min 8hrly <30 CrCl: 500mg over 30min 12hrly	Dose after dialysis	Preferred in CNS infections For pneumonia and primary bacteraemia
Metronidazole	400m t.d.s p.o. or 500mg IV 6-8hourly	>= 10:no change <10 500mg 8-12hrly Adjust for hepatic failure	Dose after HD	For C.difficile colitis. Oral may lead to increase VRE prevalence. Please refer to Clostridium difficile protocol for when to use oral vs. IVI
Moxifloxacin	400mg IV daily	No change	None	Respiratory quinolone. Cover same as for other quinolones except not useful in treatment of pseudomonas. Currently at GSH reserved for patients with Penicillin allergy and MDR-TB
Piperacillin–tazobactam*	18g daily as IV infusion	30-49 CrCl:13,5g continuous infusion <30 CrCl: 9g continuous infusion		No ESBL cover. Consider adding Amikacin if you want additional cover.
Rifampicin	10mg/kg IV daily	<10 give 50% of dose		For MDR Acinetobacter and CRE. In TB when only IV can be given In Orthopaedic patients as adjunctive when biofilm on prosthesis suspected.
Teicoplanin	800mg IV 12hrly x2 doses then 400mg 12hrly	400mg IV 12hrly x 1day then daily		Bacteriostaic

	Drug	Normal renal function	Renal dysfunction:	Haemodialysis	Pitfalls and Pearls
✓	**Tigecycline**	200mg IV stat and 100mg-150MG BD in critically ill due to higher volume of distribution	No adjustment		For complicated intra-abdominal and soft tissue infections (SSTI) and secondary bacteraemia. Not to be used in primary bacteraemia or infective endocarditis or UTI. Treatment for Enterococcus faecium (incl.VRE), Staph. Aureus (MRSA, VISA, VRSA), Clostridium diff., Acinetobacter (incl.MDR), enterobacteraciae (ESBL, AmpC, MBL, CRE), anaerobes, atypicals: Legionella, Mycoplasma, Streptococci, Listeria, Corynebacterium, Stenotrophomonas maltophilia NOT active against MORGANELLA,PROTEUS, PSEUDOMONAS and PROVIDENTIA. In combination with polymyxin/ rifampicin in MDR Acinetobacter . Caution in severe liver impairment.
	Tobramycin	Age < 30: 6 mg/kg: trough <1mg/ml (daily level before next dose) Age 30-60: 5mg/kg Age >60: 4mg/kg	AVOID		
✓	**Vancomycin**	15mg/kg IVI stat as 30-60 min infusion then infuse 2g daily as 24 hour infusion **(maintain trough level 15-20mg/L or 10,4-13,86µ mol/L)**	15mg/kg slow IV -repeat levels daily before next dose **(maintain trough level 15-20mg/L or 10,4-13,86µ mol/L)**	Does not need to be given post dialysis	Bacteriocidal, but bacteriostatic if [ ] <4-5 times MIC When to suspect MRSA: Glycopeptide Resistance • h-VISA :(MICs 2mg/l): reduces clinical response in HAP & VAP without impact on mortality • VISA: (MIC 4-8mg/l) modifies cell wall to "trap" glycopeptide: increase mortality • Occurs with vancomycin exposure, esp. inadequate dose • VRSA: (MIC 32-1024mg/l) acquires VRE vanA gene via transposon (Tn1546)
	Voriconazole	Loading dose: 6mg/kg every 12 hours IVI (400mg bd) Maintenance in ICU: 4mg/kg every 12 hours	IV Not recommended if CrCl <50, only oral maintenance		Indicated for invasive candidiasis in non-neutropenic patients with other deep tissue Candida infections (eg, Candida albicans, Candida glabrata, Candida krusei, Candida parapsilosis, Candida tropicalis)

Date: July 2015 Signed: Designation: Head: ICU

1. Antibiotic Essentials 2013
2. Applied Pharmacology in Anaesthesiology and Crit Care
3. Bariran JAC 2003
4. Dhand F1000 Med Rep 2012
5. Doribax Australian approved
6. Drugs on the Go Nov 2012
7. ESCMID Candida Guidelines Dec 2012
8. Gilbert et al
9. Harigaya BMC Infect Dis 2011
10. Khatib JAC 2011
11. Kuti IDSA 2007 abstract 1026
12. Mathew J clin Pharm Ther 1994
13. Mieke Carlier Population pharmacokinetics and dosing simulations of amoxicillin/clavulanic acid in critically ill patients
14. M Mer, G Richards Chest 1998;114;426-431
15. Nannini Curr Opin Pharmacol 2010
16. Park JAC 2012
17. Richardson JAC 1981
18. SAMF 11th Edition
19. Satola J Clin Microbiol 2011
20. Turnidge Infect Dis Clin North Am 2003
21. Van Hal PLoS ONE 2011
22. Van Hal AAC 2011
23. Western Cape Academic Hospitals Antimicrobial Recommendations 2013

APPENDIX E: TURNITIN REPORT

Dr

by Nerisha Singh

Submission date: 08-Oct-2018 01:43PM (UTC+0200)
Submission ID: 1015950518
File name: Nerisha_Singh.docx (1.25M)
Word count: 9916
Character count: 62233

Dr

ORIGINALITY REPORT

11%	6%	9%	6%
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

MATCH ALL SOURCES (ONLY SELECTED SOURCE PRINTED)

3%

★ Submitted to University of Southampton

Student Paper

Exclude quotes	On	Exclude matches	< 1%
Exclude bibliography	On		