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COLLEGE OF HEALTH SCIENCES

DEPARTMENT OF CLINICAL MEDICINE AND THERAPEUTICS

THE PREVALENCE AND ANTIMICROBIAL SUSCEPTIBILITY PATTERNS OF BACTERIA THAT CAUSE CHRONIC WOUND INFECTIONS AMONG PATIENTS AT KENYATTA NATIONAL HOSPITAL

BY

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STUDENT'S DECLARATION

I hereby declare that this research proposal is my original work carried out with guidance from my supervisors, and references made to work done by others have been indicated and has not been presented for the award of degree in any other university.

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DEDICATION

I dedicate this work to my family, whose support has made it possible so far.

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LIST OF ABBREVIATIONS

ADA	American Diabetes Association
CHF	Congestive Heart Failure
CKD	Chronic Kidney Disease
CLSI	Clinical Laboratory Standards Institute
CoNS	Coagulase Negative Staphylococci
DFI	Diabetic Foot Infection
DM	Diabetes Mellitus
ECM	Extracellular Matrix
ESBL	Extended-Spectrum Beta Lactamase
HIV	Human Immunodeficiency Virus
IDSA	Infectious Disease Society of America
KNH	Kenyatta National Hospital
MCS	Microscopy Culture and Sensitivity
MDROs	Multidrug-Resistant Organisms
MRSA	Methicillin-Resistant *Staphylococcus aureus*
PAD	Peripheral Arterial Disease
ROS	Reactive Oxygen Species

RTA	Road Traffic Accident
SLE	Systemic Lupus Erythematosus
SOPC	Surgical Outpatient Clinic
Spp	Species
TGFβ	Transforming Growth Factor Beta
UON	University of Nairobi

ABSTRACT

Background: Wound infection is a common complication of chronic wounds and a public health issue with significant morbidity and mortality. There is a paucity of data on the prevalence, bacterial causes of wound infections and antimicrobial susceptibility patterns in Kenya. This study was carried out at Kenyatta National Hospital (KNH) to bridge this gap.

Study objectives: To determine the prevalence of chronic wound infection; identify the causative bacteria and their antimicrobial susceptibility patterns in chronic wound infections among patients in medical wards and clinics.

Study design and setting: A descriptive cross-sectional study was conducted for two months in 2020 at KNH medical wards, Surgical Outpatient and Diabetes clinics. Socio-demographic characteristics, clinical data and a wound punch biopsy for microscopy culture and sensitivity were obtained from 106 participants. Data derived was coded into IBM SPSS version 21.0 for descriptive analysis.

Results: The study population was predominantly male (60.4%) with a mean age of 47 years. The most common comorbidities were diabetes mellitus and hypertension. The prevalence of wound infections was 85.8%, with gram-negative bacilli/ rods being the most common (73.6%). The most common organisms were *Proteus mirabilis*, *Proteus aeruginosa* and *S. aureus* at 17.6%, 13.9% and 12.0% respectively.

Conclusion: There were high rates of antimicrobial resistance among gram-negative and gram-positive organisms. Meropenem, Amikacin and Piperacillin/Tazobactam had the best sensitivity rate against the gram-negative organisms. Tigecycline, Teicoplanin and Linezolid had the best

activity against gram-positive organisms and to avoid resistance, antibiotics should only be used when specified.

Recommendations: Tissue culture should be incorporated in the management of infected wounds as per standard practice. Antibiotic use in the management of infected wounds should be guided by sensitivity results after adequate source control/debridement has been achieved. This study supports the current KNH Antibiotic guidelines recommendations on empiric antimicrobial therapy in wound infections

CHAPTER ONE: INTRODUCTION AND PROBLEM STATEMENT

1.0 Introduction

Wounds are a significant cause of morbidity and mortality. Chronic wounds are a potential epidemic that affects a large proportion of the world population, posing a major threat to health. A meta-analysis carried out in 2018 showed that ulcers of different etiologies had a total prevalence of 2.21 per 1,000 population, with leg ulcers alone being estimated to be 1.51 per 1,000 population (1). Only three of the nine studies included in the meta-analysis were from developing countries indicating gap in data in these regions (1). This could be due to financial limitations, poor policy making or poor leadership and management (2). Comparison of data in wound infections across regions is a challenge due to multiple causes and diversity of wounds (3).

Additionally, wounds are often associated with multiple co-morbidities. This can be attributed to the increase in the incidence of predisposing conditions such as diabetes, obesity and generally the rise in health care costs(4). Debridement, appropriate dressing and antibiotics if indicated, is the cornerstone of wound infections management. In the management of antibiotics, antimicrobial susceptibility testing is necessary due to the emergence of resistance patterns (4) (5). Global evidence in recent years has shown progressively reducing antibiotic effectiveness, as resistance to antibiotics continues to increase. Antibiotic resistance is certainly on the rise. Although there is a paucity of data on antibiotic resistance in Africa, there still exists a few local studies that have been done (6). Recent AMR data is not available for more than 40% of the countries despite the level of resistance to commonly prescribed antibiotics was significant (7).

CHAPTER TWO: LITERATURE REVIEW

2.1 Background

A wound is a breach in the integrity of the skin and is often associated with disruption of the skin's normal structure and function. A wound can be superficial affecting the epidermis to form an erosion; or deep into the dermis and underlying tissues causing an ulcer (4) (8). Wounds can be classified based on duration, aetiology, or degree of contamination (9).

Based on the duration, a wound can either be acute or chronic. An acute wound progresses through the normal stages of wound healing within the anticipated time, usually a few weeks (9). A chronic wound, on the other hand, can be defined as a wound that has failed to heal in an orderly and timely manner, to restore the anatomic and functional integrity of the skin. It can also be defined as a wound that undergoes the repair process without establishing a normal anatomic and functional outcome. There is no agreed specific cut-off time that differentiates an acute from a chronic wound. Literature suggests that if a wound surface area fails to reduce by approximately 15% over a week or 50% over one month indicates a chronic state (9)(10). The duration used to define chronic wounds ranges between 3 weeks to 3 months (11). This difference in duration poses a huge challenge in establishing the epidemiology of chronic wounds and comparison of data worldwide.

The Wound Healing Society has classified chronic wounds based on aetiology into four main categories. These include diabetic ulcers, venous ulcers, arterial ulcers and pressure ulcers (3)(12). Other important known causes of wounds include malignancy, radiation, sickle cell disease and lymphoedema. Traumatic causes of wounds include burns, interpersonal conflict injuries, road traffic accidents, postoperative wounds, cellulitis and bites (13). Dermatological conditions that

can lead to chronic wounds include pyoderma gangrenosum, systemic lupus erythematosus, among others (14).

Based on the degree of contamination, a wound can be classified as clean, dirty, contaminated or infected (4). Common causes of chronic wound infection include *Staphylococcus aureus, Escherichia coli, Klebsiella Pneumonia, Streptococcus Pyogenes, Pseudomonas Aeruginosa, Proteus species, Streptococcus species,* and *Enterococcus species*. When wound infection is suspected, tissue culture should be obtained. This is the gold standard. Alternative tests include wound swab and pus aspiration. Other tests that can be carried out include a complete blood count and a metabolic panel.

2.2 Pathophysiology of Chronic Wounds

Wound healing occurs through cellular response to skin or mucosal injury, involving activation of keratinocytes, endothelial cells, fibroblasts, macrophages, and platelets. There are organized cell migration and angiogenesis through activation of endothelial cells. Several growth factors and cytokines are released by the different cell types to coordinate and maintain wound healing. In acute wounds, restoration of skin integrity is usually complete within two to three weeks (14).

Wound healing involves several stages: inflammation, epithelialization, fibroplasia, and maturation (Figure 1 below).

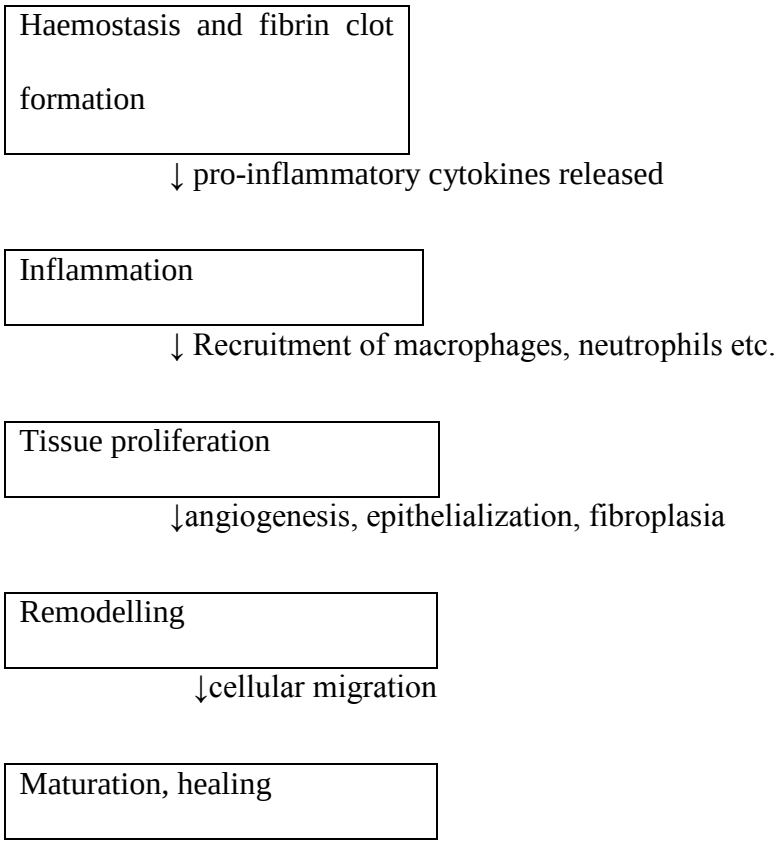

FIGURE 1: ACUTE WOUND

Irrespective of the aetiology, chronic wounds have common characteristics at the molecular level such as excessive levels of pro-inflammatory cytokines, reactive oxygen species (ROS), proteases, and senescent cells. The repeated tissue injury leads to the release of platelet-derived factors like extracellular matrix (ECM) and transforming growth factor-ß (TGF-ß) which stimulate immune reaction (14).

The pro-inflammatory cytokine cascade in chronic wounds becomes amplified and continued for a prolonged time, leading to an increase in serum proteases. In chronic wounds, healing usually stalls at the inflammation stage of healing. When healing progresses to the proliferative phase in a chronic wound, it is usually disordered. The chronic inflammation and disordered proliferative phase lead to non-healing ulcers. Diabetes mellitus has an important role in the development of

non-healing ulcers due to peripheral neuropathy and peripheral artery disease secondary to hyperglycemia (15). Other conditions contributing to the development of chronic wounds include peripheral vascular diseases, autoimmune conditions such as vasculitides and neurological conditions leading to paralysis and resultant pressure ulcers. This data is relevant for risk stratification and well holistic management of patients (9).

Infection is an important contributor to the chronicity of wounds. Biofilm formation contributes to antimicrobial resistance in wound infection due to reduced antibiotic penetration and the development of tolerance to antibiotics. Infection results in chronic inflammation and disruption of proliferation thus an impaired healing process (14) (12). Figure 2 below shows the effects of biofilm on wound healing and its role in the chronicity of wounds.

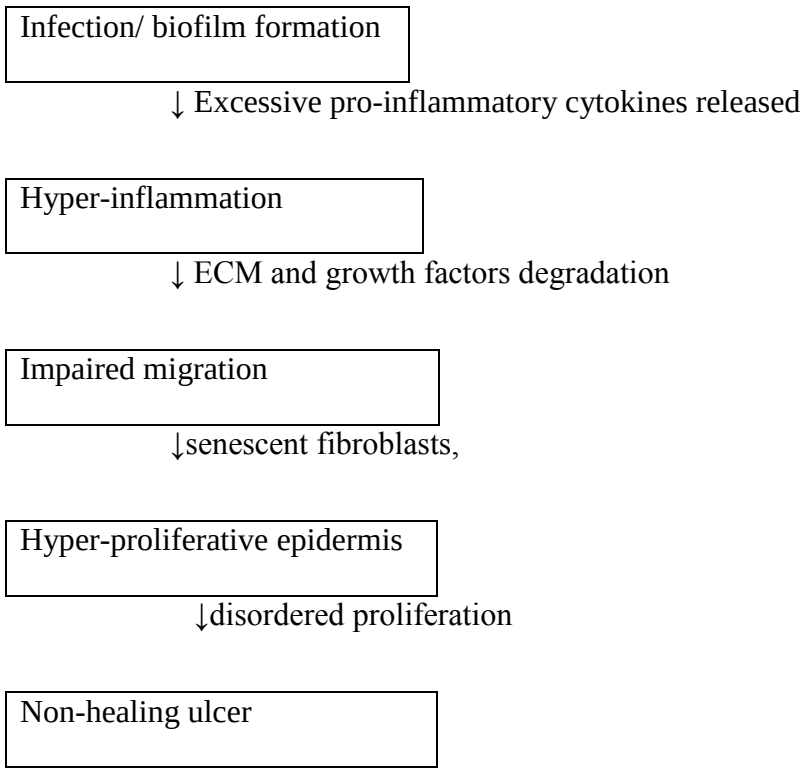

FIGURE 2: CHRONIC WOUNDS

2.3 Prevalence of chronic wounds

It is anticipated that 1-2 % of the population in developed countries will develop a chronic wound in their lifetime(3). A retrospective study in 2018 identified about 8.2 million people with chronic wounds globally (16). A study done in the United States to determine the burden of chronic wounds reported that about 6.5 million people (3% of the population above 65 years of age) have chronic wounds (3). However, a report from Wales in 2016 estimated a higher (6%) prevalence of chronic wounds, indicating an increase in the numbers of chronic wounds in the recent past (16).

In Africa, only a few countries have some data on chronic wounds. A five-year study done in a tertiary hospital in Nigeria among 509 patients, with chronic wounds, found the proportion of wound infection of 70.1% (5). This study included both in-patient and out-patient. However, the prevalence of chronic wounds was not determined. A relatively high proportion of wounds is managed in outpatient set-up hence this study may not have depicted the prevalence in the general population.

A point prevalence study carried out in another tertiary hospital in Nigeria, found 65 (31.5%) of the 206 in-patients were being managed for wounds (17). This study was however done on a single day and hence may differ greatly with period prevalence or incidence. It was also a single centre study that combined both acute and chronic wounds, with only 14 out of the 65 being chronic wounds.

In Kenya, there are a few studies on chronic wounds. A study done at KNH found 82 of 1788 patients with diabetes mellitus to have foot ulcers putting the prevalence of diabetic foot ulcers (DFU) at 4.6%. A study on DFUs at KNH in September 2017 showed a 94% culture-positivity rate; 29% were Gram-positive and 65% were Gram-negative. The main organisms isolated were *S.*

aureus (16%), *Escherichia coli* (15%), *Proteus mirabilis* (11%), *Klebsiella pneumoniae* (7%) and *Pseudomonas aeruginosa* (7%) (15). Another study on fungal cultures on DFU at KNH found 23.7%(n=152) Candida isolates. Among these 46% were drug-resistant, 11% multidrug-resistant, 3% pan-drug resistant and 40% susceptible to all the antifungal agents tested. Candida albicans was the most common species isolated with a low incidence of resistance to echinocandins (26%) and triazoles (26%) (18).

Another study carried out at KNH and the National Spinal Injury Hospital (NSIH) on bed sores, found that 66 out of 1175 in-patients had pressure sores, with a prevalence of 5.5%. Paraplegia was the commonest associated medical condition accounting for up to 35.4% of the pressure sore(19). Only one aetiology of chronic wounds was explored and the study was carried out over one week (20). A study by Ratemo et al found 78.4% (406) positive culture. *S. aureus* was the most frequent isolate (29.9%) (21). Another study by Dinda et al in Aga Khan Hospital on surgical site infection (SSI), found an incidence rate of 7.0%, and *S. aureus* which was the most prevalent bacterial isolate (22).

2.4 Morbidity and Mortality from Wounds

Wounds are associated with significant morbidity and mortality. Thus, prompt management of wound infections is paramount to curbing complications.

Immediate complications of wounds include infection such as infective venous eczema, cellulitis, and osteomyelitis. Wounds can also result in sinus formation, haemorrhage and lower-extremity gangrene, with some requiring amputation. Life-threatening conditions such as skin failure syndrome and shock can occur in large wounds (20).

Long term complications include hypertrophic scars, contractures, glycoprotein metabolic dysfunction such as secondary amyloidosis, and loss of cosmesis. In rare cases, non-healing wounds can undergo malignant transformation into Marjolin's ulcer (22).

Most chronic ulcers last about 12-13 months and can recur in about 60%-70% of the cases. This leads to a negative impact on the quality of life of the patients such as the inability to lead active social lives and depression. The poor quality of life further aggravates healing outcomes (16).

A retrospective study carried out in the United States in 2011 found a mortality of 28% among patients with chronic wounds. Two-thirds of the patients had co-morbidities such as cardiovascular diseases and diabetes (22). An all cause-mortality study carried out in Denmark in 2015 reported a mortality rate of 9.4%. The wounds associated with a higher mortality rate included cancer wounds and pressure ulcers. The mortalities were attributed to the underlying co-morbidities (16) (23).

The mortality rate among patients with chronic leg ulcers drastically increases after the first amputation from 20 to 50% in the first 3 years, with a 5-year mortality rate at 70% (4).

A study in Europe showed the risk of death at 5 years for a patient with a diabetic foot ulcer to be 2.5 times as high as the risk for a patient with diabetes who does not have a foot ulcer. More than half of diabetic ulcers become infected. Approximately 20% of moderate or severe diabetic foot infections lead to some level of amputation. Peripheral artery disease independently increases the risk of non-healing ulcers, infection, and amputation. Mortality after diabetes-related amputation exceeds 70% at 5 years for all patients with diabetes (24).

According to the Centre for Disease Control (CDC), approximately 700,000 individuals lose their lives because of drug-resistant infections each year showing higher mortality rates due to AMR by

2050 in different regions of the world. In the United States, 2 million people are affected every year by AMR and about 23,000 deaths occur as a result. This number is roughly the same as the European Union which has an annual mortality rate of 25,000 (25) (26).

Infections and sepsis are the leading cause of death in non-cardiac Intensive Care Units (ICUs) and account for 40 per cent of all ICU expenditure. They also contribute to prolonged ICU stay (27).

2.5 Cost of Managing Wounds

The cost of treating acute and chronic wounds worldwide in 2018 was estimated to be $28.1-$96.8 billion. Surgical wounds had the highest cost of management followed by diabetic foot ulcers (12). Almost $25 billion is spent on treating wounds and wound-related complications per year in the U.S.A (3). A report in 2016 showed that 5.5% of the National Health Service fund in Wales was consumed in the management of chronic wounds. This also affects individual resources and quality of life (16).

The management of chronic wounds has become advanced, incorporating new technologies, this has improved outcomes while increasing wound care costs especially in developed countries (3) (12).

2.6 Wound Infections

2.6.1 Risk Factors for Wound Infections

Wounds are vulnerable to colonization by bacteria as they provide a conducive environment for microbial invasion, proliferation, and resultant infection. Factors that increase the risk of wound infection include (4):

I. Patient factors that cause debilitation, impaired immune function and vasculopathy. Examples include (4):

   i. Old age

   ii. Co-morbidities – Poorly controlled diabetes mellitus, rheumatoid arthritis, obesity, malnutrition

   iii. Therapy-cytotoxic agents or radiation therapy, prior surgery, corticosteroids and immunosuppressant therapy.

   iv. Psychosocial factors – hospitalization or institutionalization, poor hygiene.

   v. Hypoxic or ischaemic conditions such as anaemia, hypothermia, cardiac or respiratory disease, vascular diseases and sickle cell disease.

   vi. Disorders that impair immune function, e.g., Human Immunodeficiency Virus, acquired immune deficiency syndrome (HIV/AIDS), malignancies.

   vii. Inappropriate antibiotic use, especially in acute wounds

   viii. Alcohol, smoking and drug abuse

   ix. Prolonged hospital stay (4).

II. Wound factors include (4):

   i. Increased tissue debris

   ii. Foreign bodies including catheters

   iii. Ischaemia

   iv. Hematoma

   v. Large in size and/or deep wound

   vi. Anatomical location near a site of potential contamination.

   vii. Increased exudate

viii. Repetitive trauma

ix. Contamination during surgical procedures (4).

2.6.2 Stages of Wound Infections

Wound infections can be caused by multiple microbes including bacteria, fungi, protozoa, and viruses. Bacteria are by far the commonest cause of wound infection (4).

Wound infection is a continuous process including the following stages (4):

I. **Wound contamination**–Wound contains microbes that do not proliferate thus no immune activation. In contamination, the wound has no signs of inflammation (4).

II. **Wound colonization** -Microbes in the wound undergo limited proliferation which does not invoke host immune reaction (4).

III. **Local wound infection**- In this stage, microbes proliferate at a high rate triggering host reaction. The microbes invade deeper wound tissue causing local inflammation. The infection is however not severe enough to spread to other structures or organs.

IV. **Spreading Infection**-This occurs when the wound infection progresses to invade the surrounding tissue. The tissues include muscle, fascia, organs and/or serosa.

V. **Systemic Infection**-Systemic infection occurs when microbe from a wound spread through vascular or lymphatic systems to distant organs. Invasion of the bloodstream can lead to sepsis. Signs and symptoms of systemic infection include fever, and end-organ dysfunction (4).

International Wound Society revised the stages of wound infection in the 2016 guidelines. The term *Critical colonization* was excluded from the stages of infection because it would require

microbe quantification tests. Most laboratories don't carry out quantification tests worldwide. Further, no cut-off value has been set to differentiate critical colonization from contamination (4).

Worldwide, there is a growing understanding of biofilm as a factor in wound infection. Several studies have shown that biofilms develop in wounds using scanning electron microscopy. A study in 2008 found that 60% of chronic wounds had biofilm compared to 6% of acute wounds indicating that biofilm has a role in the chronicity of wounds. Disruption of biofilm is important in the management of chronic wounds. Advanced dressings have been impregnated with topical antibiotics to control and disrupt biofilm (4) (16).

2.6.3 Bacterial Causes of Wound Infection and Antimicrobial Susceptibility Patterns

The most common bacterial causes of wound infection are *Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Streptococcus pyogenes, Pseudomonas aeruginosa, Proteus species, Streptococcus species*, and *Enterococcus species*. Wound infections are often polymicrobial. Pyogenic infections can be endogenous or exogenous (28). *Staphylococcus aureus* is by far the most common pathogen responsible for wound infections (29).

Various studies across the globe have in the past shown similar bacterial profiles in wound infections. This is important for the determination of empirical antibiotic treatment where indicated, while laboratory cultures reports are awaited. In the United States of America, more than 2.8 million antibiotic-resistant infections occur each year, and more than 35,000 people die as a result according to the report by the Centre for Disease Control (CDC). Some of these infections are attributable to wound infections (30). World Health Organization (WHO) reported rising global antimicrobial resistance with most regions recording more than 50% on *Klebsiella pneumonia* and *E. coli* resistance to third-generation cephalosporins (31).

A review article published in 2013 on diabetic foot infections (DFIs) in developed countries, showed that most mild community-acquired infections in antibiotic in-experienced patients are caused by aerobic Gram-positive cocci, especially *Staphylococcus aureus*, followed by *β-streptococci* and coagulase-negative *staphylococci* (32).

In patients with prior antibiotic use, wound infections are mostly caused by multiple microbes, including aerobic Gram-negative and/or anaerobic bacteria. Recent epidemiological studies from developing countries including India have reported a lower prevalence of *S. aureus* wound infections compared to reports from developed nations (30% compared to 75%). There was a higher prevalence of Gram-negative rods instead, especially *P. aeruginosa* noted in these countries(32). The reasons for this geographical difference may be related to differences in weather and the environment, specimen types, prior antibiotic use, regulation of over-the-counter antibiotics or reporting bias but this has not been established yet. It might as well be that the microbiology of DFIs is evolving slowly towards more Gram-negative microorganisms in some regions. Some regions in the southwest of the USA have reported *P. aeruginosa* as more frequent in nosocomial DFIs, and relatively low isolation of *S. aureus* in DFIs (28).

Traumatic deep wounds with moderate to severe infections, especially in patients with prior antibiotic use, were found to be polymicrobial with organisms such as *Escherichia coli*, *Proteus*, *Klebsiella*, *P. aeruginosa* and *Bacteroides*. Severe lower limb wound infections especially in patients whose feet are frequently exposed to water grew *P. aeruginosa* mostly. Fungal, parasitic or mycobacterial DFIs have not been well studied or documented (28).

A study on 114 pus swabs in a South Indian tertiary hospital found 89.47% of the swabs to have a positive culture with 95 % of the culture-positive pus samples growing pure bacterial isolates and only 5% showed mixed infection. *Staphylococcus* aureus (24.29%) was the most common isolate,

followed by *Pseudomonas aeruginosa* (21.49%) and *Escherichia coli* (14.02%). Less common organisms included *Klebsiella pneumoniae* at12.15%, *Streptococcus pyogenes at* 11.23%, *Staphylococcus epidermidis* 9.35% and *Proteus species* at 7.47% (33). Susceptibility of *Staphylococcus aureus* to vancomycin was 100% while that of Levofloxacin and Oxacillin were 76.92% and 73.07% respectively. Gram Negative Bacilli were susceptible to Imipenem (80%), Aztreonam (80%), Piperacillin/Tazobactam (80%), Levofloxacin (80%) (33).

A similar study in Raipur, India also showed that *Staphylococcus aureus* was the commonest microorganism (40%). The second was *Klebsiella species* (33%), then *Pseudomonas species* (18%), *Escherichia coli* (16%), and *Proteus species* (7%). Both studies used the swab technique. This shows variations in the type of bacteria in different zones of the same country (34).

A study on 503 pus samples in Nepal in 2017 yielded 43.7% bacterial growth. Out of the total isolates, 71.82% were Gram-negative and 28.18% Gram-positive bacteria. *Pseudomonas species* cultures were the most common (34.55%) followed by *Staphylococcus aureus*(21.36%), *Escherichia coli* (11.82%), and lastly *Acinetobacter baumannii* (11.36%). Most Gram-negative bacteria were susceptible to amikacin (63.9%) while most Gram-positive bacteria (93.5%) were susceptible to chloramphenicol. The most ineffective antibiotic was ceftazidime with a 94.7% resistance rate. *A. baumannii* isolates showed 100% sensitivity to colistin. The *A. baumannii* isolates, however, showed 100% resistance to ceftriaxone and 96% resistance to ciprofloxacin, ofloxacin, gentamicin, cefoperazone/ sulbactam and piperacillin/ tazobactam. Out of the 220 bacterial isolates, 138 were found to be MDR, with a higher MDR pattern in Gram-negative isolates (48.18%). The target population was similar to those in other studies elsewhere, showing there could be major regional variations in both causative agents as well as resistance patterns (35).

A study on culture and sensitivity patterns in diabetic foot ulcers in a hospital in Gujarat predominantly yielded *Pseudomonas aeruginosa* (27%) (34). The predominance of gram-negative bacilli in diabetic pus was also reported in another study in India in 2009 (36).

A meta-analysis of 21 studies in Ethiopia with over four thousand wound samples in 2019 had about 70% positive wound cultures with a few cultures showing polymicrobial isolates. The pooled culture positivity was found to be 70.0%. *S. aureus* isolate was highest (36%) from which almost half were methicillin-resistant strains. *E. coli* isolate was second at 13%, followed by *P. aeruginosa*, 9%, *K. pneumoniae*, 9% and *P. mirabilis*, 8% (5). *S. aureus* exhibited relatively lower resistance against ciprofloxacin at 12%, and gentamicin at 13%. *E. coli* isolates had the highest resistance towards ampicillin (84%). Poly-microbial cultures had the highest sensitivity to a combination of ciprofloxacin and gentamicin (5). The antimicrobial resistance patterns vary significantly between countries and regions for various bacterial isolates and evolve over time (37). The emergence of multidrug-resistant organisms (MDROs) in wound infections poses a huge challenge in the management of severe wound infections. The most common isolated resistant pathogen has been methicillin-resistant *S. aureus* (MRSA) (22) (34). RSA seems to be declining in most countries as reported in recent studies. Recently, Gram-negative organisms that produce extended-spectrum β-lactamases-(ESBLs) or carbapenemases are of great concern. Isolation of MDROs from a diabetic foot infection has been on the rise over the past ten years (28).

Resistance to ampicillin, penicillin and amoxicillin has been increasing over the last several years (38). A study done in Ethiopia demonstrated that isolates of *P. aeruginosa* were 100% resistant to tetracycline, ceftriaxone, ampicillin and penicillin but 100% sensitive to gentamicin. Coagulase Negative *S. aureus* (CoNS) showed 100% sensitivity to vancomycin. Every bacterial isolate

showed resistance to at least one antibiotic of those tested in the study (38). A similar study in a tertiary Indian hospital in 2014 on 144 aerobic isolates from pus samples showed *Staphylococcus aureus* isolates were resistant to ampicillin with only 33% sensitive to methicillin and among the CoNS, only 58.3% were sensitive to methicillin (33).

Aerobic bacterial isolates from wound infections were found to be the most susceptible to aminoglycosides and quinolones (37) (39) (40). In Ethiopia, a resistance rate of above 50% in *Aureus* isolates was observed. Of these, penicillin had the highest resistance at 95.5%, followed by methicillin and chloramphenicol at 77.3% and 51.5% respectively(23).

Acinetobacter isolates studied in Palestine in 2016, showed almost complete resistance to cephalosporins (>95%) and gentamicin (81.3%). Lower rates of resistance against amikacin 68.3% and ciprofloxacin 69.7% were shown. The most effective antimicrobial drug was doxycycline with the lowest resistance rate of 22.1% (41). In a study in Gujarat in 2014, most gram-negative isolates in diabetic foot ulcers were resistant to amikacin, piperacillin/tazobactam, gentamicin, ampicillin-sulbactam and gatifloxacin. The gram-negative bacilli were highly sensitive to imipenem and polymyxin. Sixty per cent of the wounds grew methicillin-resistant *S. aureus* which was sensitive to vancomycin and linezolid (28). A study done by Karimi et al in 2007 showed a high prevalence of *Pseudomonas spp* (42.6%) followed by *Proteus spp*. (33.9%) and *Staphylococcus aureus* (33%) (42).

Studies carried out in Nigeria, Uganda and Kenya showed similar isolates. *Staphylococcus aureus* showed the highest number of isolates, followed by *Pseudomonas aeruginosa*. Although the gram-negative bacterial infection has increased significantly, *Staphylococcus aureus* is still a major

etiological agent of pyogenic infections according to studies done by Elamenya, Kagwa and Mutonga et al (36) (43) (44) (see Table 1 below):

TABLE 1: TABLE SHOWING COMMONEST ORGANISMS FROM DIFFERENT STUDIES

AUTHOR/ Year	REGION	STUDY POPULATION	Culture positivity/ prevalence %	Commonest organisms	Tissue used
H Al-Azawi et al, 2013	Iran	Inpatient, surgical units	88%	*S. Aureus 34%, E. coli 28% Pseudomonas 24%*	swab
KC, R., Shrestha, 2014	Nepal	Inpatient	60.20%	*S. aureus* 72%	swab
A. Mohammed, 2017	Ethiopia	inpatient and outpatient	83.9%	*S. Aureus 34%*	swab
Kanaga et al 2014	Kenya	paediatric surgical patients	82%	*S. Aureus* 52.7%	swab
Kagwa et al,2016	Kenya	Diabetic foot ulcers	90.6 %	*S. aureus* 37.3%, *Proteus* 21.3%	swab
Mutonga D. et al 2019	Kenya	Diabetic foot ulcers	94%	*S. aureus* 16% *E. coli* 15%	Swab

A study on diabetic foot infections at KNH in 2016 among 75 participants found *Staphylococcus aureus* to be the commonest isolate at 37.3%. Of these isolates, 39.3% were methicillin-resistant. *Proteus spp* was second commonest at 21.3% of positive cultures followed by *Klebsiella* spp at 14.7%. All the bacteria were highly sensitive to imipenem. *S. Aureus* showed 92.9% sensitivity to imipenem Most of the *E. faecalis* isolates were sensitive to imipenem (77.8%), ciprofloxacin (66.7%) and amoxicillin-clavulanate (55.6%) but were all resistant to cefuroxime, ceftriaxone, ceftazidime, clindamycin and vancomycin (44). This study only explored one type of chronic ulcer; hence generalization to other types of wounds may not be applicable. There is overwhelming evidence of increasing resistance which calls for careful selection of antibiotics when indicated, to prevent the development of resistance. A study done by Wangai et al in 2016 in KNH demonstrated significant rates of antimicrobial resistance to most antibiotics including carbapenems, and third-

generation cephalosporins (45). Methicillin-resistant *S. aureus* was high at 50% in a similar study (46). The Department of Pathology at Aga Khan University Hospital released an updated 'Antibiotic Susceptibility Report 2015' for data through 2014 showing 49% *E. coli* resistance to third-generation cephalosporins among hospital inpatients (22).

2.7 Specimen Collection Method

To get accurate and pure bacterial culture, proper specimen collection is mandatory. In most studies, a pus swab is used. The gold standard, however, is a biopsy. The most accepted swab technique across the literature is Levine's Method: After wound cleansing and/or debridement, a sterile swab is rotated over a $1cm^2$ area of the wound while applying pressure to express fluid from the wound tissue. The areas of the wound with the greatest clinical suspicion for infection should be selected for the swab (13).

A multi-centre study in the Netherlands compared bacterial culture yield between swab and biopsy when collected at the same site. Swabs correctly identified 131 of 180 (72.8%) microorganisms cultured from biopsies in wounds. The most frequent organisms cultured were *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *β-Haemolytic Streptococci species* in the two methods of sample collection. The study demonstrated that swabs and biopsies yield similar culture results when taken from the same site, hence no need for invasive biopsy in clinical practice (41). This is further supported by the three-specimen collection technique comparison study in 2011 in Denmark where swab, biopsy and filter paper pad methods were used. The swab method had similar yields compared to biopsy (47).

The use of biopsy and needle aspirate is however the gold standard as recommended by the Infectious Disease Society of America (IDSA) and is widely used in research work and will be

used in this study(48) (48). Adopting the gold standard will not only help with reliability and quality control but comparison with other studies as well.

2.8 Treatment of Wound Infections

Optimal management of wound infection requires a thorough assessment of the patient as a whole. In case of sepsis, immediate resuscitation with fluids, oxygen and antibiotics is required while observing the patient in critical care set up (4).

When antibiotics are indicated, the choice should be based on available local antibiogram which our study could contribute towards.

The KNH empiric cover guideline published in 2018 recommends incision, drainage and debridement as the first line of management in wound infections. Use of systemic antibiotics for five to seven days in chronic wounds is recommended only if there is cellulitis and/or sepsis. No antibiotics are required for small abscesses (<5cm). For anaerobic infection, clindamycin or metronidazole should be used. Tissue culture for infected wounds is mandatory. Tigecycline should not be used in infected diabetic foot ulcers(49). (See table 2 below).

TABLE 2: THE KNH AMS GUIDELINES 2018 ON SOFT TISSUE INFECTIONS

	Category 1	Category 2	Category 3
Description	No contact with health care system No prior antibiotic treatment Patient young with no co-morbidities No organ failure	Recent hospital admission, dialysis etc. Recent antibiotic therapy Patient old with co-morbidities Single organ failure	Long hospitalization Recent and multiple antibiotic therapies Advanced immunodeficiency and Neutropenia, Multiple organ failure
Common Pathogens	Staph.aureus , streptococcus spp.	Staphylococcus spp. Enterobacteriaceae	Pseudomonas, enterobacteriaceae
Empiric Therapy	Flucloxacillin or Amoxicillin/clavulinic acid or Clindamycin or Doxycycline	Clindamycin or Cefriaxone or Tigecycline[6]	Piperacillin / Tazobactam + amikacin or Cefepime +amikacin

Other newer methods of chronic wounds include the use of stem cells such as platelet-rich plasma and fat-derived stem cells (40) (42). Surgical management includes debridement followed by skin grafts and flaps (43).

Table 3 below summarizes the general principles of wound infection management.

TABLE 3: SUMMARY OF APPROACH TO WOUND MANAGEMENT

Optimize host response	Reduce microbial load	General measures
Manage co-morbidities Reduce risk factors for wound infection Nutritional support Hydration Identify and manage other infections Relieve symptoms such as fever Psychosocial support	Adopt aseptic technique Hygiene Timely debridement Appropriate dressings Appropriate topical antiseptics/ antibiotics Antibiotics, where indicated	Clean environment. Correct supplies Wound care education Good general support

2.9 Problem Statement

This study aims to determine the proportion of infected chronic wounds, their bacterial causes and antimicrobial sensitivity patterns among patients at KNH. The outcome of this study will help in understanding the burden of chronic wounds infections. Currently, there are local guidelines formulated in 2018 for empiric antibiotic choice at Kenyatta National Hospital (KNH). There is however need for regular surveillance of antimicrobial resistance (AMR) due to the constant change in susceptibility patterns. There is also a need to strengthen infection prevention and control especially when handling wounds (49).

The results of this study will provide important information on the burden of chronic wound infections locally. The outcome of this study will contribute to informing of appropriate antibiotic cover for chronic wound infections and thus facilitate better outcomes among these patients, leading to improved quality of life.

2.10 Study Significance

The results of this study will provide important information on the burden of chronic wound infections locally. The outcome of this study will contribute to informing of appropriate antibiotic cover for chronic wound infections and thus facilitate better outcomes among these patients, leading to improved quality of life.

2.11 Scope of the Study

This study determined the prevalence and antimicrobial sensitivity of chronic wound infections among patients in medical wards, diabetes, and surgical outpatient clinics at KNH.

The socio-demographic and clinical characteristics of patients with wound infection were established after which a biopsy of the wound bed was collected from each patient and sent to the KNH microbiology laboratory for culture, identification and antimicrobial sensitivity testing.

Prior antibiotic use among the participants was assessed, and the susceptibility patterns of the wound infections were evaluated.

2.12 Research Question:

What is the burden of chronic wound infections among patients admitted to medical wards at the Kenyatta National Hospital?

2.13 Objectives

2.13.1 Main Objective:

To determine the prevalence of chronic wound infection, identify the causative bacterial isolates and describe their antimicrobial susceptibility patterns among patients admitted to medical wards at Kenyatta National Hospital (KNH).

2.13.2 Specific Objectives:

1. To determine the prevalence of infected chronic wounds among patients in medical wards and medical clinics at KNH.
2. To identify the bacteria that cause chronic wound infections, using tissue culture, among patients in medical wards and medical clinics at KNH.
3. To document the antimicrobial susceptibility patterns of the isolated bacteria.

CHAPTER THREE: METHODOLOGY

3.1 Study Design

This was a hospital-based descriptive cross-sectional study.

3.2 Study Setting

The study was carried out in all medical wards and clinics at Kenyatta National Hospital (KNH). KNH is a teaching and referral hospital situated in Nairobi- Kenya, with 1800 beds, making it the largest hospital in East and Central Africa. The study was carried out in the medical wards, Surgical Outpatient clinic, Diabetes Clinic at KNH. The two clinics are the main areas where patients with all types of wounds are dressed at the facility, thus giving a wide representation of the study population. Patients with wounds were identified by the principal investigator with the aid of the staff in the various locations.

3.3 Study Population

Medical patients with chronic wounds presenting at KNH outpatient clinics and those admitted in the medical wards.

3.3.1 Case Definition

A chronic wound was defined as a wound that has lasted for at least 3 weeks. *Wound infection* was defined as a wound with one or more symptoms and signs of infection below (Table 4):

TABLE 4: SIGNS AND SYMPTOMS OF INFECTION

Local covert subtle signs	Local overt /classic signs	Spreading infection	Systemic infection
Excessive granulation tissue Bleeding, friable tissue Excessive pocketing of granulation tissue Enlarging wound Worsening pain Increasing mal-odour	Erythema Local warmth Swelling Purulent discharge New or increasing pain Increasing mal-odour	Extending induration +/- erythema Lymphangitis Crepitus Wound breakdown/ dehiscence +/- satellite lesions General malaise/ lethargy poor appetite Inflammation, swelling	Sepsis Septic shock End-organ failure NB: must not be attributable to other infections.

3.3.2 Inclusion Criteria:

1. Patients aged 18 years and above
2. Patients who gave written and informed consent
3. Patients with a diagnosis of chronic wound

3.3.3 Exclusion Criteria

1. Patients with post-operative wound infections
2. Patients with burn wounds

3.4 Sampling

3.4.1 Sample Size:

The following simple formula (Daniel, 1999) was used to calculate the sample size

$$n = \frac{Z^2 \, x \, P(1-P)}{d^2}$$

Where, n = sample size,

Z = Z statistic for a level of confidence,

P = expected prevalence: S. Sudhaharanet al carried out a study on aerobic wound infections among diabetes patients at an Indian tertiary care hospital in 2017. The study yielded positively cultured in 93.2% of the samples. This percentage was used to estimate the sample size

d = precision (in proportion of one); if 5%, d = 0.05.

Z statistic (Z): For a confidence level of 95%, which is conventional, Z value is 1.96

3.4.2 Sample Size Calculation:

$$n = \frac{1.96^2 \, x \, 0.932(1-0.932)}{0.05^2}$$ **= 97**

The minimum sample size required was 97

3.4.3 Compensation for dropout or data errors

10 % of 97 was added

97+ (10%×97) = **106**

The sample size target was 106

3.5 Sampling Technique

During the study period, participants who met the inclusion criteria in different wards and clinics were recruited consecutively to participate in the study until a sample size of 106 was achieved.

3.6 Recruitment Procedure

Patients who met the eligibility criteria were identified by the principal investigator with the aid of the staff in the various wards/clinics. Recruitment of patients was done in the wards and waiting for bays for the clinics. Given the high number of patients reviewed each day at the facility, research assistants were deployed to help in the recruitment process, to effectively utilize time during the study.

All the eligible patients were then taken through the consent process which included giving the patient general information about the study benefits and risks, confidentiality and their freedom to choose to participate in the study. The patients who gave written and informed consent were then be recruited (Appendix 3).

3.7 Data Collection

3.7.1 Data Collection Procedure

After signing a consent form (Appendix 3), the patient's socio-demographic data and co-morbidities were captured into a proforma (Appendix 4). Antibiotic use in the previous 3 months, if any, was documented. Targeted history and examination were conducted to identify those with symptoms and signs of wound infection. The principal investigator supervised the conduct of the study to ensure standardized data collection.

3.7.2 Specimen Collection and Processing

The wound was cleaned with saline to get to the wound base. Under local anaesthesia, a 4mm deep punch biopsy was taken, put in a sterile container with Stuart Transport Media to preserve fastidious organisms. The samples were taken to the KNH microbiology laboratory for culture, identification and sensitivity testing. Precautions were taken to avoid cross-contamination at all stages. All samples were obtained by the principal investigator (PI).

In the laboratory, the specimens were handled by a qualified microbiology technologist with the guidance of a microbiologist. Gram staining was done to differentiate gram-positive and gram-negative cocci/bacilli. The biopsy was put in soya broth for inoculation. The colonies grown were transferred to aerobic bacterial cultures such as Sheep Blood agar, Cysteine-Lactose-Electrolyte-Deficient Agar (CLED) and MacConkey media were used based on the gram-stain results. The culture media were prepared, poured in Petri dishes and allowed to cool. The inoculums were applied to a small area then spread using a sterile loop of wire to provide for single colonies. All inoculated plates were labelled and incubated at 37°C for 24 hours for the organisms to grow. No anaerobic cultures were carried out because Macintosh anaerobic jar and Robertson media were not available at the time of the study. The Vitek 2 machine was used for bacterial identification and antibiotic susceptibility testing. This is a highly advanced, automated machine used for identification and antimicrobial sensitivity testing of routine clinical isolates, thus reducing human error (50).

All *staphylococcus* isolates were subjected to coagulase testing to differentiate them from Coagulase Negative *staphylococcus* (CoNS). Sensitivity testing was done using antibiotic-impregnated cards and the Vitek machine, which has both gram-positive and gram-negative bacteria identity cards(50).

The gram-positive cards contained: amoxicillin-clavulanate, cefuroxime, ceftriaxone, ciprofloxacin, cefoxitin, cloxacillin vancomycin and ceftazidime benzyl penicillin ampicillin, amoxicillin Dicloxacillin, oxacillin, erythromycin, Clarithromycin, azithromycin, sulfonamide/trimethoprim, clindamycin tetracyclines chloramphenicol, gatifloxacin, moxifloxacin, levofloxacin, imipenem, vancomycin and linezolid.

The gram-negative aerobic cards contained: Amoxicillin–clavulanic acid, ampicillin, piperacillin-tazobactam, cefuroxime, cefuroxime axetil, ceftriaxone, cefoperazone-sulbactam, cefepime, ciprofloxacin, ertapenem, imipenem, meropenem, gentamicin, amikacin, nalidixic acid, nitrofurantoin, and trimethoprim-sulfamethoxazole

3.7.3 Study Flow

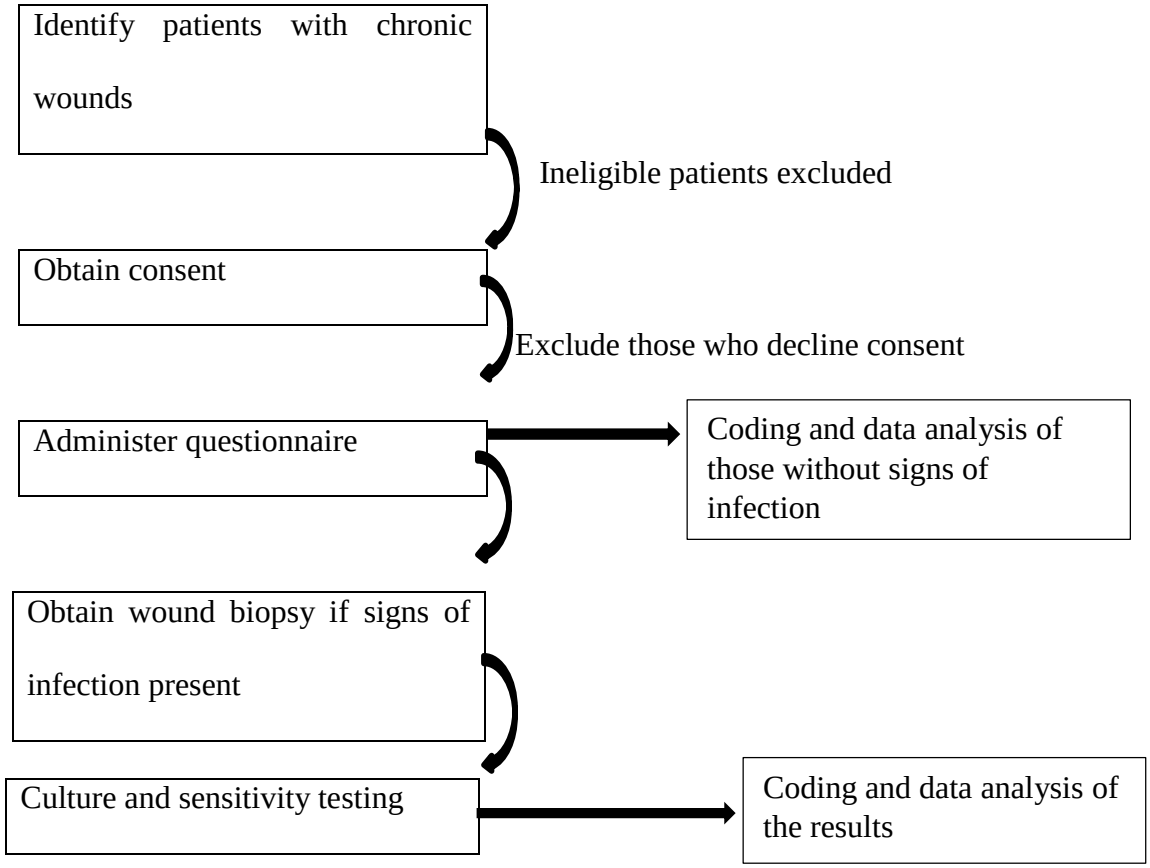

FIGURE 3: A FLOW CHART OF SUBJECT RECRUITMENT INTO THE STUDY

3.8 Study Instruments and Tools

3.8.1 Instruments

1. A study proforma was used to collect socio-demographic and clinical data e.g., sex, age, employment status, marital status, level of education, duration and cause of the wound, any prior use of antibiotics.

2. Culture identification and sensitivity tables

3.8.2 Tools

1. Skin biopsy punch

2. Stuart transport media

3. Vitek AST machines with the antimicrobial cards

3.9 Definition of Clinical Variables

3.9.1 Dependent Variables

1. Wound outcome- infected or not infected based on clinical and culture results.

2. Bacterial profile- different types of bacteria isolated from the wound.

3. Antibiotic sensitivity- reported as per cent susceptible.

3.9.2 Independent Variables

1. Age: in years

2. Sex: male or female

3. Antibiotic use within last 3 months: indicated as yes or no.

3.10 Quality Assurance

Data collection was carried out by the principal investigator for quality control purposes. Standard operating procedures were observed at all times when obtaining the biopsies. In the laboratory, the cultures were carried out using the updated Clinical Laboratory Standard Institute (CLSI) recommendations by a qualified microbiology technologist (51).

Bacteria identification and sensitivity testing of the specimens was done using the Vitek machine which is semi-automated to reduce human error. The machine is validated for clinical use and frequently calibrated to ensure reliability and reproducibility. The KNH microbiology laboratory is ISO 15189:2012 certified laboratory. A qualified and experienced microbiologist was involved during the whole process to guide on best practices and procedures. The KNH Microbiology laboratory has existing in-built controls and external quality checks through the World Health Organization National Institute for Communicable Diseases, South Africa (WHO/NICD) and United Kingdom National External Quality Assurance Service (NEQAS). The laboratory uses the VITEK 2 system which currently conforms to the international recommendations as outlined in the M100Ed30 Performance Standards for Antimicrobial Susceptibility Testing; The laboratory usesVitek-2 machine which conforms to international recommendations outlined in the M100Ed30 *Performance Standard for Antimicrobial and Susceptibility Testing* (52). This document was developed through the CLSI consensus process and provides includes an update to the antimicrobial susceptibility testing standards M02, M07, and M11(51). The Vitek machine is also frequently calibrated to ensure reliability and reproducibility. The KNH microbiology laboratory is ISO 15189:2012 certified laboratory.

In addition, the laboratory applied specific Standard Operating Procedures (SOPs) below to enhance the quality of specimen processing and minimise pre-analytical, analytical and post-analytical errors.

Pre-analytical processes

To minimize pre-analytical errors, the laboratory applied the standard operating procedures entitled 'Collection, Handling and Transportation of Microbiological specimens' (KNH/LABM MED/MICROB/022P). This document entails details of proper specimen collected by trained clinicians, as well as prompt transport of specimens to the laboratory as soon as possible after collection. Once received in the laboratory, scrutiny of the specimens was done, with rejection criteria applied to those which were deemed unfit for processing, such as mislabeled or contaminated specimen. After sorting, proper storage of specimens was ensured before processing, including refrigeration of certain specimens such as urine.

Analytical processes

Quality control during specimen analysis was performed as per the Standard Operating Procedure: 'Media Preparation and Quality Control' (KNH/LAB MED/MICRO/003F1). Standard ATCC (American Type Culture Collection) reference micro-organisms were used to check the performance of culture media. It is important to note that contaminated media and inoculation of old cultures can lead to false results and analytical errors. Adequate bacterial cultures grown were processed by the VITEK-2 machine, according to the Standard Operating Procedure quality control document: 'Operation of VITEK-2 Compact' (KNH/LAB MED/MICRO/057P). Verification of VITEK-2 results was done and the inter-method comparison was performed with offline manual methods such as Kirby-Bauer disk diffusion techniques. External quality assurance

and inter-lab comparisons are usually performed on a quarterly basis to check all stages of processing from culture to VITEK reporting, using external references laboratories. All the above processes were done to ensure that the VITEK results reported were valid.

Post-analytical

In the analytical phase, the Standard Operating Procedure: 'Results Reporting Format' (KNH/LAB MED/MICRO/065P) was applied. The machine print-outs were interpreted by both the machine and by the microbiology laboratory technologist. All results were subjected to a second verification and counter signing by a senior laboratory microbiologist. Any contaminants or commensals reported were flagged and reported to the clinician. The clinician was then advised to request for a second specimen to be collected and tested in pathogenic isolates.

Antibiotics susceptibility test results and breakpoints were violated and interpreted as per the latest CLSI M100-S24 Performance Standard for Antimicrobial Susceptibility Testing; Twenty-Fourth Informational Supplement. Periodic internal quality checks are performed regularly to interrogate culture and sensitivity testing results and locate any discrepancies. In case of any clarifications needed, previous results are retrieved from the manual backups and VITEK storage archives.

3.11 Data Management

Each study proforma was assigned a unique serial code to prevent data duplication. All filled assessment tools were kept under lock and key only accessible to the primary investigator. Data collected were entered into Microsoft Excel Sheet and secured by protected passwords. Upon completion of data entry, the Principal Investigator (PI) cross-checked all entries with the hard copies for any inconsistencies.

3.12 Data Analysis

The results were coded and entered into Microsoft Excel Sheet, and data were analysed using the statistical software, SPSS version 21 (Chicago, Illinois). The study population was described using clinical and socio-demographic characteristics. Categorical variables e.g., sex, employment status, marital status, level of education have been presented as frequencies. Age was presented as mean, mode median.

Prevalence of chronic wound infections was calculated with numerator as several samples with a positive culture and denominator as a total sample number. The prevalence has been presented as a percentage with its 95% confidence interval.

Bacterial isolates and susceptibility patterns from all wounds were tabulated and presented as percentages. Group differences were analyzed using the Independent T-test. Results were presented using tables, figures, pie charts and bar graphs where appropriate.

3.13 Ethical Consideration

Data was collected after the approval of the Kenyatta National Hospital/University of Nairobi Ethics Review Committee (KNH/UoN- ERC); reference number KNH-ERC/A/356. All procedures conformed to the World Medical Association Declaration of Helsinki. The biopsy procedures did not cause any harm, and local anaesthesia was used to alleviate pain. Informed consent was sought from all the study participants (Appendix 3).

All patients got standard care. Other laboratory tests including histology if needed were prescribed and carried out by the primary care physician. Non-participation did not affect patient's care in the health facility. There were no incentives given to the study participants. The cost of conducting

the study was met solely by the Principal Investigator. Results from the study were communicated to the primary care physician and appropriate management was initiated.

All patients' identifiable data e.g., the name was not included in the data collection tool. The patients were identified by study numbers. The data sheet was retained by the researcher and was treated with the utmost confidentiality. Electronic data generated was encrypted with a password only availed to the research team. All hard copy research data was kept in a safely locked cabinet only accessed by the research team.

CHAPTER 4: RESULTS

4.1 Recruitment

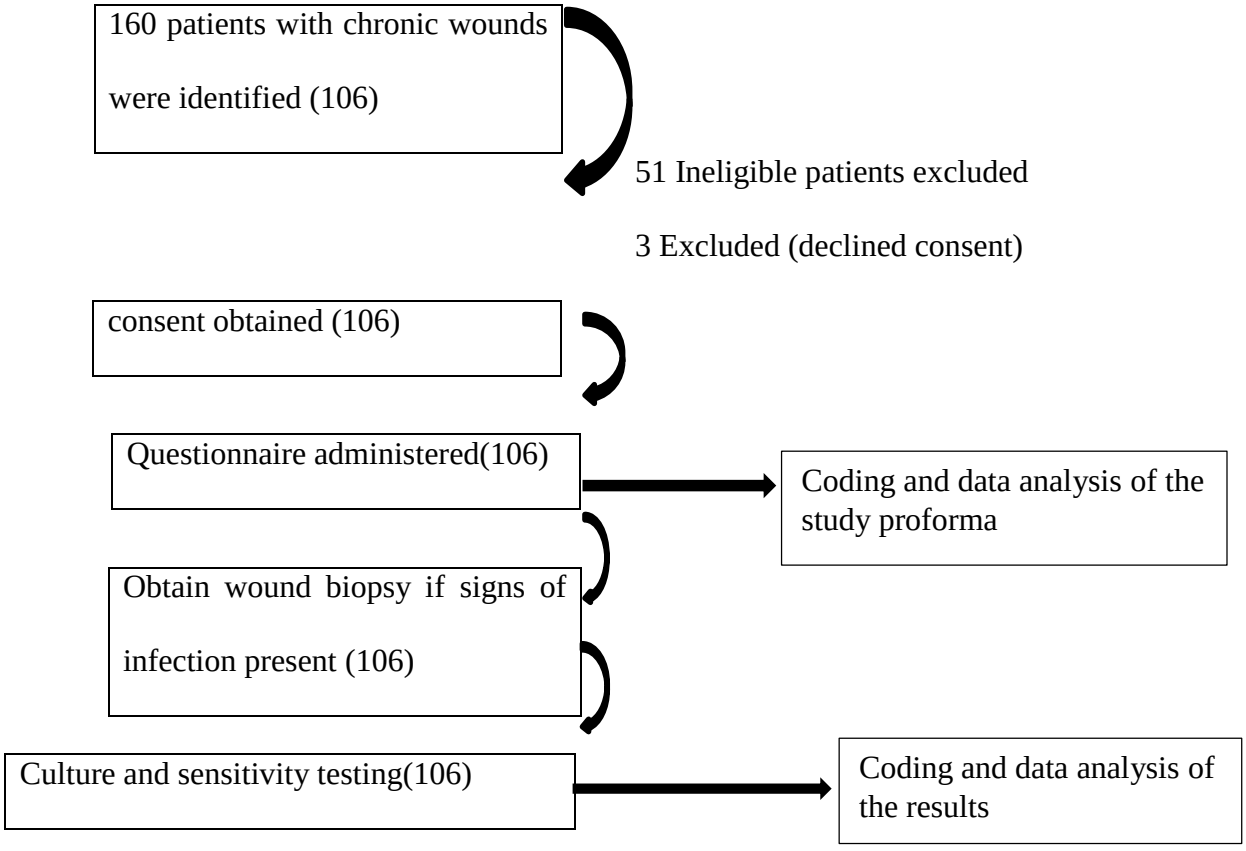

FIGURE 4: A FLOW CHART OF SUBJECT RECRUITMENT INTO THE STUDY

51 patients were excluded from the study due to the following reasons:

1. 18 had post-surgical wounds
2. 13 were below 18 years of age
3. 12 had burn wounds
4. 5 wounds had been confirmed to be malignant
5. 3 wounds were contaminated with fecal material

4.2 Sociodemographic and clinical profiles/characteristics of participants

4.2.1 Sociodemographic characteristics of participants

Most (22.6%) of the study patients were aged between 41 and 50 years, the mean age was 47.4 years with an SD of 16.06, the median age was 47.50 years.

See other sociodemographic characteristics in Table 5 below.

TABLE 5: SOCIO-DEMOGRAPHIC CHARACTERISTICS OF STUDY PARTICIPANTS

Social demographic characteristic		Frequency(n=106)	Percentage
Age (mean=47.48, SD 16.06)			
• <=20		5	4.7
• 21-30		12	11.3
• 31-40		19	17.9
• 41-50		24	**22.6**
• 51-60		19	17.9
• 61-70		20	18.9
• >=71		7	6.6
Gender:	Male	64	**60.4**
	Female	42	39.6
Level of education:	Primary	32	30.2
	Secondary	62	**58.5**
	College	12	11.3
Occupation:	Employed	26	24.5
	Unemployed	80	**75.5**
Marital status:	Single	27	25.5
	Married	76	**71.7**
	Separated	3	2.8
Use of alcohol, cigarettes or bang		25	23.6

4.2.2 Clinical characteristics of participants

Most patients (64.1% had one or more comorbidities. Diabetes was the most common co-morbidity, present in (31.1%) of the study patients. Some patients had more than one co-morbidity as shown in the table below:

TABLE 6: TABLE SHOWING THE DISTRIBUTION OF CO-MORBIDITIES

Co-morbidity	**Frequency**	**Percentage**
Total	**68**	**64.1**
Diabetes Mellitus	35	33.0
Hypertension	19	17.9
Spinal injury	8	7.5
Malignancy	6	5.7
HIV*	4	3.7
CKD*	3	2.8
COPD*	2	1.9
CHF*	2	1.9
SLE*	1	1.2

*HIV- Human immunodeficiency virus, CKD- chronic kidney disease, COPD- chronic obstructive pulmonary disease, CHF- congestive heart failure, SLE- Systemic Lupus Erythematosus.

4.2.3 Causes of Wounds

Diabetes was the most common cause of the wound, accounting for 31.1% of the wounds. Spontaneous wounds were 23.6 % of the total. Other causes of wounds included trivial or major trauma, peripheral arterial disease and venous ulcers (see Table 7 below).

TABLE 7: TABLE SHOWING FREQUENCIES OF THE DIFFERENT CAUSES OF WOUNDS

Cause of wound	**Frequency (n= 106)**	**%**
Diabetes	**35**	**33.1**
Trauma (all)	33	31.1
RTA	15	14.1
Other	18	17
Spontaneous	25	23.6
Bedsore	13	12.3
Bites (all)	3	2.8
Human	2	1.9
Snake	1	0.9
Peripheral arterial disease	1	0.9
Venus ulcer	1	0.9

4.2.4 Signs of wound infection

Based on clinical assessment 96 out of 106 (90.6%) of wounds had signs of infection. These included an increase in discharge or mal-odour from the wound, excess granulation tissue and erythema around the wound.

Eleven (10.3%) patients had signs of spreading infection.

Only 2 patients (1.9%) had signs of systemic infection. See figure 5 below.

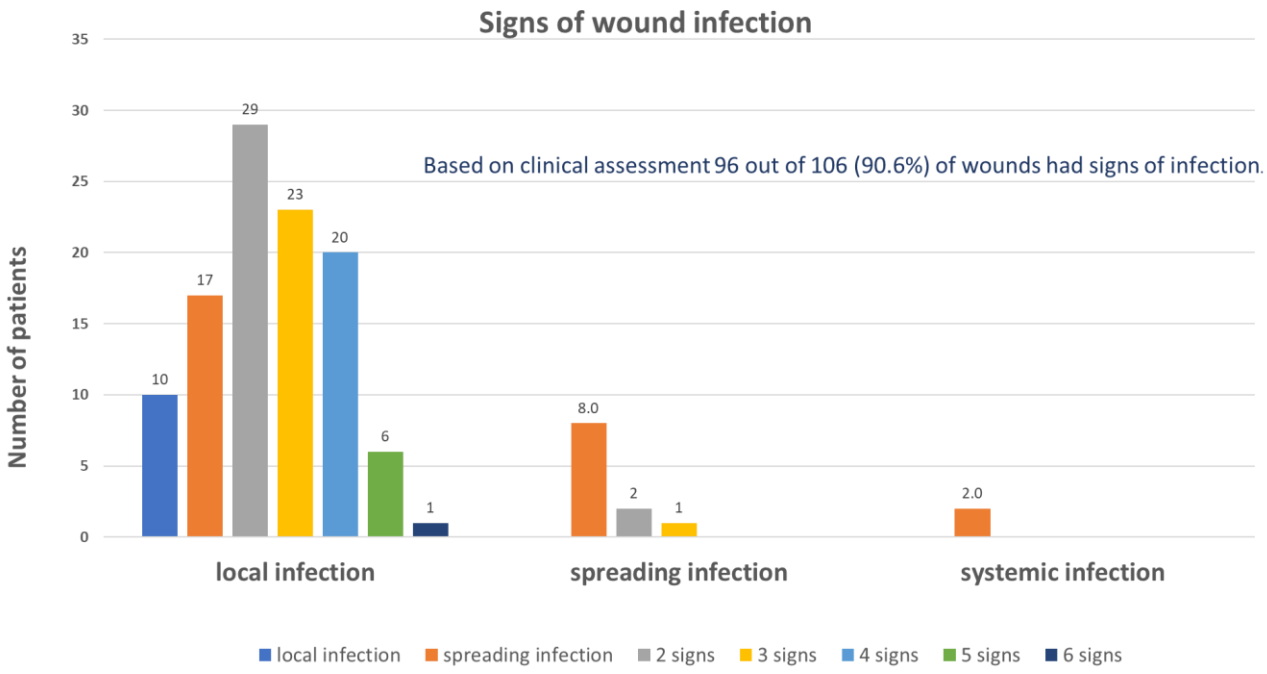

FIGURE 5: FIGURE SHOWING DISTRIBUTION OF SIGNS OF WOUND INFECTION

Table 8 below shows the total number of patients for each sign of wound infection.

TABLE 8: SIGNS OF WOUND INFECTION

Local covert subtle signs	Local overt /classic signs	Spreading infection	Systemic infection
Excessive granulation tissue: 2 Bleeding, friable tissue: 54 Excessive pocketing of granulation tissue: 21 Enlarging wound: 33	Erythema: 23 Local warmth: 18 Swelling: 51 Purulent discharge: 40 New or increasing pain: 37 Increasing mal-odour: 30	Extending induration +/- erythema: 5 Lymphangitis: 0 Crepitus: 0 Wound breakdown/ dehiscence +/- satellite lesions: 7 General malaise/ lethargy poor appetite: 2 Inflammation, swelling: 3	Sepsis: 2 Septic shock: 0 End-organ failure: 0 NB: must not be attributable to other infections.

4.2.5 Duration of Wounds and Hospital Stay

Duration of wounds varied widely, with a range of 3 weeks up to seven years, a median of 695 days (about 23 months) and an interquartile range of 305 days (about 10 months).

The mean hospital stay for the inpatient participants was 76 days.

4.2.6 Antibiotic Use

52 (49.1%) reported antibiotic use within 3 months of the study; of whom, 30% flucloxacillin, 25% ceftriaxone, 15% clindamycin and 13.5% didn't know the antibiotic used.

4.3 Distribution of patients in different settings

53.8% of the study participants were from the clinics, outpatient group, with Surgical Outpatient Clinic (SOPC) contributing 34.9% (see Table 9 below):

TABLE 9: DISTRIBUTION OF PATIENTS BASED ON INPATIENT/OUTPATIENT STATUS

Ward/clinic	(N= 106)	%
Clinic/ outpatient	**57**	**53.8**
SOPC	37	34.9
Diabetes mellitus clinic	20	18.9
Medical wards/ Inpatient	**49**	**46.2**
Total	106	100.0

4.4 Prevalence of wound infections

A significant number (91) of the study patients had wound infection making the prevalence rate 85.8%.

Two of the cultures grew mixed culture (2 isolates each), making the total culture positivity rate 87.7%

(93/106)

Based on clinical assessment, 96 out of 106 (90.6%) of wounds had signs of infection.

4.5 Patterns of wound infections

4.5.1 Gram stain results

Gram-negative bacilli/rods were the most prevalent at 73.6%, followed by gram-positive cocci at 20.8%. Mixed gram stain organisms were 2.8%.

4.5.2 Distribution of organisms identified

Proteus mirabilis was the most prevalent organism at 17.6%. Others include (see Table 7 below):

TABLE 10: TABLE ILLUSTRATING THE DISTRIBUTION OF IDENTIFIED BACTERIA

Identified bacteria	Frequency	Per cent
None	15	13.9
Proteus mirabilis	19	17.6
Pseudomonas aeruginosa	15	13.9
Staphylococcus aureus	13	12.0
Escherichia coli	8	7.4
Morganella morganii *ssp morganii* *ssp sibonii*	8 7 1	7.4
Proteus vulgaris	6	5.6
Klebsiella pneumoniae ssp pneumoniae	6	5.6
Acinetobacter baumannii	3	2.8
Providencia stuartii	3	2.8
Providencia rettgeri	2	1.9
Proteus penneri	2	1.9
Serratia liquefaciens	1	.9
Enterobacter aerogenes	1	.9
Serratia fonticola	1	.9
Serratia marcescens	1	.9
Pantoea agglomerans	1	.9
Proteus hauseri	1	.9
Klebsiella oxytoca	1	.9
Citrobacter koseri	1	.9
Total	108	100.0

4.5.3 Data comparing culture positivity rate between medical wards and outpatient

Culture positivity rate was higher in outpatient (46.3%; 34 from SOPC, and 16 from DM Clinic) compared to inpatient set-up (39.8%, n=43). There was no statistically significant difference noted in the types of isolates among the 3 groups (p-value = 0.618). See figure 6 and 7 below.

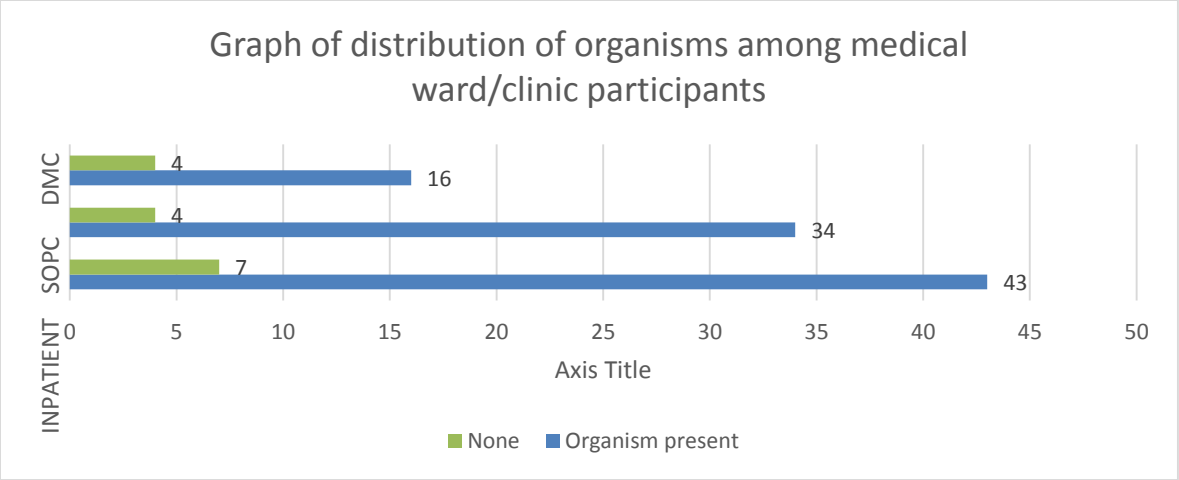

FIGURE 6: DISTRIBUTION OF ISOLATES BASED ON INPATIENT/OUTPATIENT STATUS

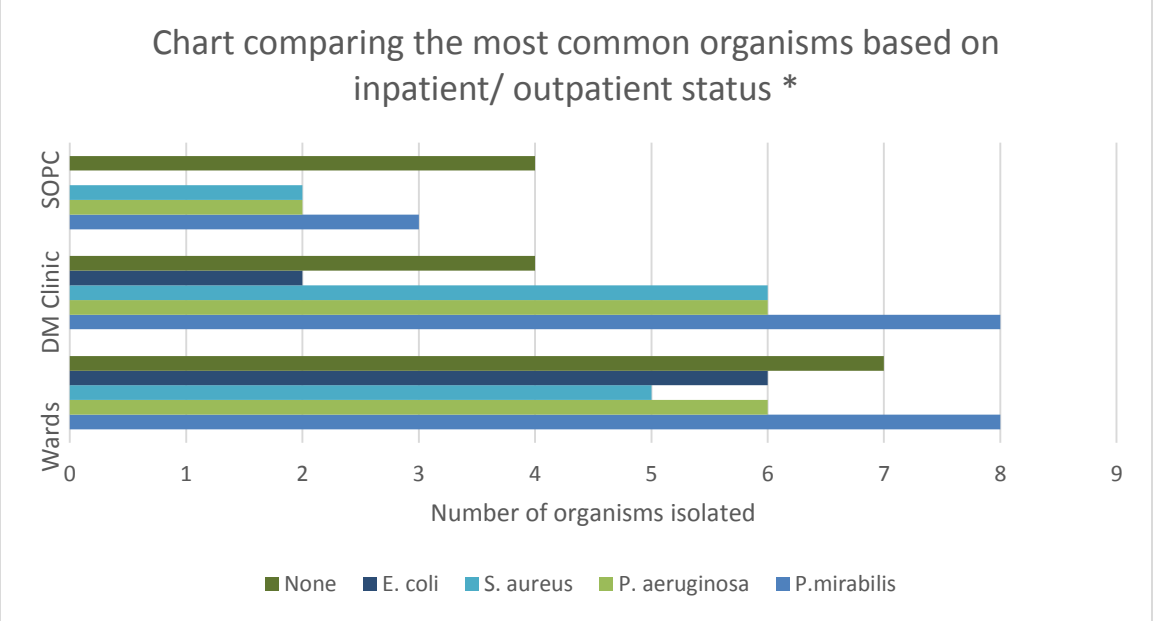

FIGURE 7: DISTRIBUTION OF ORGANISMS AMONG MEDICAL WARD, DIABETES CLINIC AND SURGICAL CLINIC PATIENTS

*Less than 30 isolates, the standard recommendation for reporting

4.6 Sensitivity Pattern of Organisms

4.6.1 Gram-Negative Organisms

P. mirabilis had poor sensitivity to Trimethoprim/Sulfamethoxazole, *S. aureus* to ampicillin-sulbactam and erythromycin (46.1%). It showed moderate sensitivity to some beta-lactamase penicillins, third and fourth generation cephalosporins, gentamicin and ciprofloxacin.

P. aeruginosa was moderately susceptible to cephalosporins and gentamycin (63.2%). One isolate demonstrated extended beta-lactamase activity. This was a sample taken in an outpatient set-up, and the participant had no known comorbidities or recent antibiotic use. It however had an excellent response to piperacillin-tazobactam, amikacin and meropenem.

E. *coli* was poorly sensitive to Ampicillin-salbactum and Trimethoprim/Sulfamethoxazole (25%), moderately sensitive to third and fourth generation cephalosporins, amoxicillin sulbactam and trimethoprim- sulfamethoxazole. It showed a great response to amikacin.

M. morganii had a poor response to cotrimoxazole and third-generation cephalosporins and *K. pneumoniae* showed to amoxicillin-clavulanate, Trimethoprim/Sulfamethoxazole. (See Table 11 and figure 8 below adopted from CLSI M39A4E) (53).

TABLE 11: TABLE SHOWING SENSITIVITY PATTERNS OF GRAM-NEGATIVE BACTERIA

Gram negative organism	No. of strains	PERCENT SUSCEPTIBLE (%S)*												
		Amoxicillin-Clavulanic acid	Ampicillin-Sulbactam	Piperacillin-Tazobactam	Cefuroxime	Cefotaxime	Ceftriaxone	Ceftazidime	Cefepime	Amikacin	Gentamicin	Ciprofloxacin	Meropenem	Trimethoprim-Sulfamethoxazole
Proteus mirabilis	**19**	84.2	84.2	100	63.2	63.2	63.2	63.2	63.2	100	63.2	84.2	100	31.6
Pseudomonas aeruginosa	**15**	-	-	73.3	-	-	-	73.3	80	93.3	80	86.7	80	-
Escherichia coli	**8**	50	25	87.5	50	50	50	50	50	100	75	87.5	100	25
Morganella morganii	**8**	-	37.5	87.5	-	75	75	87.5	87.5	100	87.5	62.5	100	62.5
Proteus vulgaris	**6**	100	100	100	16.7	100	100	100	100	100	83	100	100	100
Klebsiella pneumoniae	**6**	33.3	0	83.3	16.7	33.3	33.3	33.3	33.3	100	66.7	33.3	100	33.3
TOTAL	**62**													

FOOTNOTES

(-) Drug not tested or expected intrinsic resistance ***Calculated from fewer than the standard recommendation of 30 isolates**

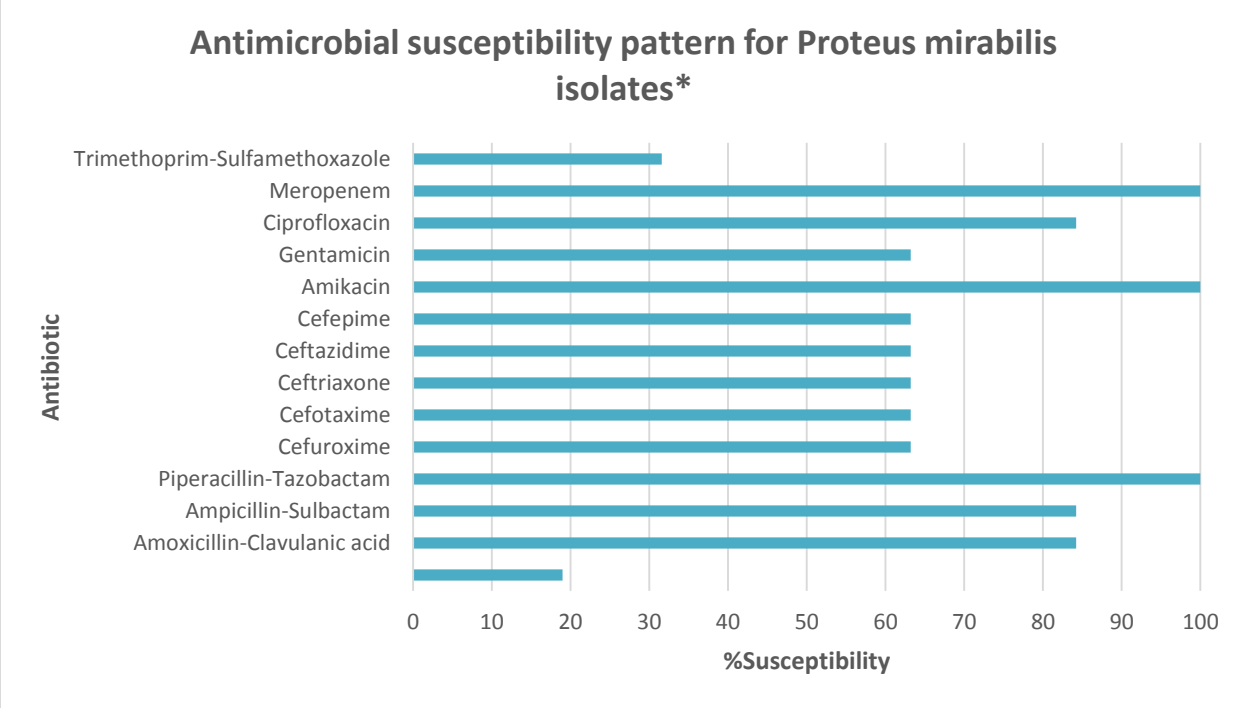

FIGURE 8: ANTIMICROBIAL SUSCEPTIBILITY PATTERNS OF *PROTEUS MIRABILIS*

**** Calculated from fewer than the standard recommendation of 30 isolates**

4.6.2 Gram-Positive Organisms

S. aureus had the lowest susceptibility to penicillin and highest to quinolones and teicoplanin methicillin susceptibility was at 53.8%. (see Table 12 below adopted from CLSI M39A4E) (53).

TABLE 12: TABLE SHOWING SUSCEPTIBILITY PATTERNS OF GRAM-POSITIVE BACTERIA

Gram positive organism	No. of strains (n)	PERCENT SUSCEPTIBLE (%S)*											
		Ampicillin-Sulbactam	Oxacillin	Penicillin G	Levofloxacin	Moxifloxacin	Gentamicin	Tetracycline	Clindamycin	Erythromycin	Linezolid	Trimethoprim-Sulfamethoxazole	Teicoplanin
Staphylococcus aureus	**13**	46.1	63.2	-	**100**	100	92.3	58.3	**76.9**	46.2	**100**	46.2	**100**
TOTAL													

*** Calculated from fewer than the standard recommendation of 30 isolates**

4.7 Multidrug resistance (MDR)

Four of the total isolates (4.4%) showed Methicillin-Resistant S*taphylococcus aureus* (MRSA), making it 30.8% of all *S. aureus* isolates. Two of the MRSA isolates produced mixed infection (*S. aureus* with *P. aeruginosa* and *P. mirabilis* as respectively). 2 of the 4 isolates were from medical patients who had used antibiotics in the preceding 3 months. Over 50% of *S. aureus* demonstrated MDR against amoxicillin-sulbactam, trimethoprim-sulfamethoxazole and erythromycin. *Klebsiella pneumoniae* had multidrug resistance to all third and fourth generation cephalosporins, trimethoprim and amoxicillin-clavulanate. About 50% of E. *coli* had MDR to third and fourth generation cephalosporins, amoxicillin sulbactam and trimethoprim- sulfamethoxazole.

CHAPTER 5: DISCUSSION

Wound infections are among the major causes of morbidity and mortality. They are often associated with multiple socio-demographic factors as well as co-morbidities such as diabetes, obesity. The study aimed to determine the prevalence of chronic wound infection, identify the bacterial isolates and their antimicrobial susceptibility patterns among medical patients at KNH. The objectives were fully met by our study.

The sociodemographic characteristics, the prevalence of chronic wound infections, bacterial profiles and antibiotic susceptibility patterns will be discussed here.

The findings of the study showed that the majority of the patients were from the clinics. This was similar to a review by Sen et al, who also noted that in the majority of studies, the majority (57.8%) of patients with wounds were from outpatient settings(5).

As concerns age, the mean age at presentation in our study was 47.4 years, which was in sharp contrast to the review done by Sen et al (2019) who reported that most chronic wounds are noted in the elderly population(53) (5). Similarly, a study by Jockenhofer et al revealed a mean age of 69.9 years (54), and that by Raeder et al was 85 years (55). In China, the mean age was noted to be 60 years (56) (30) while in Brazil it was 59 years (52). This difference in age of presentation of chronic wounds might be due to proper early wound care in the developed as compared to developing countries or differences in regional epidemiology and population dynamics (56).

The most common affected gender in our setting were males (60.4%) as compared to females. This is in keeping with several studies including one done in Germany where the most commonly affected gender was noted to be males (57) as well as China (54). Our findings were however different from that in Brazil where females (62%) were mostly affected by chronic wounds (58).

The male gender has been confirmed as a risk factor in the meta analyses of Zhang, Lu and Huang, Li with different psychological and physiological states and anatomical structures, healthy behaviour, environmental experiences, reactions to stressful events, and differences in risk behaviour being linked as a cause of these differences (56). The male preponderance in our study could be due to poor health-seeking behaviour in the male population leading to chronic wounds (56).

Most of the patients in our setting had attained secondary education (58.5%), were unemployed (80%) and married (76%). Similar findings were noted in China where most were married (90.95%) and unemployed (79.21%) (54). The high rate of unemployment, financial constraints and limited access to national insurance schemes are likely to contribute to delayed health-seeking behaviour, which can result in progression to a chronic wound.

The most common comorbidity associated with chronic wounds was diabetes and hypertension as compared to the rest, which is an established risk factor for the development of non-healing wounds. This was similar to a review done by Jockenhofer et al (2014) who noted diabetes (46.4%) as the commonest comorbidity followed by hypertension (25.5%) (55). This association could be owing to combined pathological mechanisms in diabetes and hypertension resulting in peripheral arterial disease (PAD) and chronic inflammatory process. Other mechanisms in diabetes include uncontrolled hyperglycemia, diabetic neuropathy, and a higher risk of infection due to immunosuppression (59). Spontaneous wounds comprised 23% (n=25) which is concerning for possible undiagnosed peripheral vascular diseases (55).

The prevalence of chronic wound infections in our study was 85.8%. Specifically, *P. mirabilis* and *P. aeruginosa* were the commonest at 17.6% and 13.9% respectively. Various studies across the globe have in the past shown similar bacterial profiles in wound infections. The higher rate of

gram-negative organisms might be due to antibiotic use and misuse of gram-positive cover which was the case in our study. There was no statistically significant difference between outpatient and inpatient positivity rates. This could be since both categories had frequent contact with our health facility.

These bacterial profiles were comparatively different from the known patterns of the commonest bacterial causes of wound infection: *Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Streptococcus pyogenes, Pseudomonas aeruginosa, Proteus species, Streptococcus species,* and *Enterococcus species*. A review article published in 2013 on diabetic foot infections (DFIs) in developed countries, showed that most mild community-acquired infections in antibiotic in-experienced patients are caused by aerobic Gram-positive cocci, especially *Staphylococcus aureus*, followed by *β-Streptococci* and coagulase-negative *Staphylococci* (3).

Our findings were similar to those done in patients with prior antibiotic use. In these, wound infections were mostly caused by multiple microbes, including aerobic Gram-negative and/or anaerobic bacteria. Recent epidemiological studies from developing countries including India have reported a lower prevalence of *S. aureus* wound infections compared to reports from developed nations (30% compared to 75%). There was a higher prevalence of Gram-negative rods instead, especially *P. aeruginosa* noted in these countries (3).

The reasons for this geographical difference may be related to differences in weather and the environment, specimen types, prior antibiotic use, regulation of over-the-counter antibiotics or reporting bias but this has not been established yet. It might as well be that the microbiology of DFIs is evolving slowly towards more Gram-negative microorganisms in some regions. Some regions in the southwest of the USA have reported *P. aeruginosa* as more frequent in nosocomial DFIs, and relatively low isolation of *S. aureus* in DFIs (23). A similar study in Raipur, India also

showed that *Staphylococcus aureus* was the commonest microorganism (40%). The second was *Klebsiella species* (33%), then *Pseudomonas species* (18%), *Escherichia coli* (16%), and *Proteus species* (7%). This shows variations in the type of bacteria in different zones of the same country (32).

In Kenya, a study by Kagwa et al on diabetic foot infections showed that the most commonly isolated microorganisms were *S. aureus* (37.3%), *Proteus spp* (21.3%), *Klebsiella spp* (14.7%) and *E. coli* (13.3%) (44). *Proteus spp* was the highest isolated gram-negative isolate, which is similar to our study. This could be due to the high number of diabetes comorbidity in our study. In practice, however, *Proteus mirabilis* is more implicated in urinary tract infections (UTI).

Strict anaerobes were not studied due to a lack of laboratory capacity at the time the study was conducted. These by definition, are bacteria that grow in the absence of oxygen and fail to show surface growth in cannot tolerate 0.5 per cent oxygen. Examples include *Clostridium perfringens* and *Bacteroides fragillis*. Obligate anaerobes tend to cause deep-seated acute infections, and rarely cause chronic wound infection, thus this was not a limitation in our study.

Antibacterial susceptibility in our study showed significant rates of resistance. *P. mirabilis* had the poorest sensitivity to Trimethoprim/Sulfamethoxazole, *S. aureus* to ampicillin-salbactum and erythromycin (46.1%), E. *coli* to Ampicillin-salbactum and Trimethoprim/Sulfamethoxazole (25%), *M. morganii* to cotrimoxazole and third-generation cephalosporins. *K. pneumoniae* showed high resistance to amoxicillin-clavulanate, Trimethoprim/Sulfamethoxazole. *P. aeruginosa* was moderately susceptible to cephalosporins and gentamycin (63.2%). The antimicrobial resistance patterns vary significantly between countries and regions for various bacterial isolates and evolve, due to multiple factors such as antibiotic use, various comorbidities such as diabetes mellitus, rate of hospitalization, socioeconomic factors and health-seeking behaviour (20).

Resistance to ampicillin, penicillin and amoxicillin has been noted to increase over the last several years (60). A study done in Ethiopia demonstrated that isolates of *P. aeruginosa* were 100% resistant to ceftriaxone, but 100% sensitive to gentamicin. *S. aureus* showed 100% sensitivity to vancomycin. Every bacterial isolate showed high resistance to at least one antibiotic of those tested in the study (60). A similar study in a tertiary Indian hospital in 2014 on 144 aerobic isolates from pus samples showed *Staphylococcus aureus* isolates were resistant to ampicillin (33% sensitivity) and only 58.3% were sensitive methicillin (28). In Ethiopia, a resistance rate of above 50% in *S. aureus* isolates was observed. Of these, penicillin had the highest resistance at 95.5%, followed by methicillin and chloramphenicol at 77.3% and 51.5% respectively (37). *Pseudomonas aeruginosa, Klebsiella pneumonia* and *Proteus vulgaris pyogenes* were highly susceptible to Ciprofloxacin and Gentamycin(about 78.5%), in a study done in Baqubah, Iraq which is similar to our study (38).

In a study in Gujarat in 2014, most gram-negative isolates in diabetic foot ulcers were resistant to amikacin, piperacillin/tazobactam, gentamicin, ampicillin-sulbactam and gatifloxacin. The gram-negative bacilli were highly sensitive to imipenem and polymyxin. Sixty per cent of the wounds grew methicillin resistant *S. aureus* which was sensitive to vancomycin and linezolid (28). The KNH guide to empiric antimicrobial therapy published in 2018 recommends the use of antibiotics only in cellulitis, sepsis or abscesses larger than 5cm diameter. The Mainstay of wound infection is incision, drainage and debridement. Clindamycin or add metronidazole can be used where the anaerobic infection is suspected. The duration of treatment should be 5-7 days. Tissue culture should be obtained for infected wounds(49).

Understanding these antibiotic patterns is crucial since the emergence of multidrug-resistant organisms (MDROs) in wound infections poses a huge challenge in the management of severe

wound infections. Infection prevention and control remain key to better outcomes in wound management. Some of the measures include hygiene hand washing, aseptic techniques in the hospital, removal of unnecessary catheters and intravenous cannula, antibiotic stewards and patient education (6) (61).

Study strength and limitations

The study utilized tissue culture which is more reliable in differentiating infection from colonization. This is a great strength to the study.

All organisms cultured had less than 30 isolates, which is below the recommended minimum number for antibiogram as per CLSI guidelines (53). This resulted from the small sample size which was time-limited. Being a single centre study, it cannot be generalized to other facilities and regions.

CHAPTER SIX: CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The bacterial wound infection rate was high (85.6%). *Proteus mirabilis, Pseudomonas aeruginosa and S. aureus* were the commonest causes of infections.

Meropenem, Amikacin and Piperacillin/Tazobactam had the best sensitivity rate against the gram-negative organisms. Ciprofloxacin & Amoxicillin/clavulanate oral antibiotics had the best sensitivity. Tigecycline, Teicoplanin and Linezolid had the best activity against gram-positive organisms. Levofloxacin followed by clindamycin, were the oral antibiotics with the highest sensitivity rates. Antibiotics should only be used when indicated to avoid resistance

6.2 Recommendations

1. Tissue culture for microscopy, culture and sensitivity (M/C/S) should be incorporated in the management of infected wounds instead of pus swabs as per local and international guidelines.

2. The use of antibiotics in the management of infected wounds should be guided by sensitivity results after adequate source control/debridement has been achieved.

3. This study supports the current KNH Antibiotic guidelines and IDSA recommendations in the choice of antimicrobial therapy in skin & soft tissue infections.

4. Clindamycin can be used empirically to treat infected wounds where gram-positive organisms are suspected pending culture and sensitivity results.

5. Ceftriaxone alone should not be used for the empirical treatment of infected wounds.

6. Vancomycin should not be used for empirical treatment of assumed MRSA unless confirmed by sensitivity studies (low MRSA rates of 4.4%).

7. Treatment guidelines for use of antibiotics should be followed and reviewed regularly to ensure rational use of antibiotics.

8. There is a need to strengthen antimicrobial stewardship and surveillance to prevent resistance.

9. The laboratory capacity should be built to allow strict anaerobe culture for the completeness of future studies.

10. A multicenter study with a large sample size is recommended to inform guidelines and practice.

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APPENDIX 1: SCREENING PROFORMA

Study No.: …….

Age: …….

Date of Birth: ……

Gender: Female ☐ Male ☐

Are you willing to participate in the study on wound infections at patients at Kenyatta National Hospital?

YES ☐ NO ☐

APPENDIX 2: PATIENT INFORMATION FORM

Introduction

My name is Dr. Kisoi Stella, a post-graduate student in Internal Medicine and Therapeautics at the University of Nairobi. I am carrying out a research to establish the types of bacteria that cause wound infection in Kenyatta National Hospital, their susceptibility to different antibiotics. Wound infections vary from local to severe infections such as sepsis. This will help inform current practice on antibiotic choice in wound infections and hence improved outcomes.

You are free to participate or decline participation in this study and that will not change your current management and treatment that is routinely offered in this hospital for your particular condition. You have a right to refuse or withdraw from this study at any point.

A brief history physical exam to determine signs and symptoms of wound infection will be carried out. The wound will then be washed. Under local anaesthesia, a small piece of tissue will be collected and taken to laboratory for analysis. This will take about 10-20 minutes of your time.

The information obtained will be treated with utmost confidentiality and only be available to the principal investigator and his research team. Your name will not be used in the proforma. We will not be sharing the identity of anyone participating in this research.

The knowledge that we get from this study will be shared with the policy makers in the Ministry of Health, University of Nairobi, KNH and doctors through publications, conferences, journals and presentations. Confidential information will not be shared with any third party.

Thank you for taking time to read this and if you have any questions please don't hesitate to ask.

In case of any clarifications, you may contact the following:

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KNH/UON Ethics and review committee

Tel 2726300 Ext 44102

KIAMBATISHO CHA PILI: FOMU YA HABARI ZA WAGONJWA

Jina langu ni daktari Stella Kisoi. Mimi ni mwanafunzi wa chuo kikuu cha Nairobi katika somo la udaktari. Madhumuni ya kauli hii ni kukujulisha kuhusu utafiti ninaoufanya kuhusu viini vinavyoadhiri vidonda, katika hosipitali kuu ya Kenyatta. Mapendekezo kutokana na utafiti huu yanaweza kutumika na wasimamizi katika kuboresha utoaji wa huduma.

Kushiriki kwako katika utafiti huu ni kwa hiari yako. Iwapo utakubali kushiriki,haya ndiyo yanahusika na utafiti huu:

Kupata takwimu za kijamii kama vile umri, kauli ya ajira, kiwango cha juu cha elimu, muda ambao umeugua kidonda. Jina lako na nambari yako ya usajili hospitalini hazitatajwa katika profoma ya utafiti.

Mtihani mfupi wa kimwili wa kupima ambukizo la viini kutoka kwa kidonda.

Kidonda kitaoshwa na maji yaliyona chumvi, na kugandishwa kisha sampuli itachukuliwa kwenye kidonda na kupelekwa kwamaabara kufanya utafiti ili kujumuisha viini vilivyoko.

Haya yote yatachukua muda wa dakika kama kumi/ishirini (10-20).

Habari utakazozitoa zitakua ni siri.Daktari wako wa kwanza atajulishwa matokeo ya utafiti yanayohusika na matibabu yako.Utahitajika kutia sahihi kwenye fomu ya idhini iwapo utakubali kushiriki kwenye utafiti.Iwapo utataka kujitoa kwenye utafiti huu,unaruhusiwa kufanya hivyo katika hatua yoyote na bila adhabu yoyote.

Usipokubali kushiriki katika utafiti huu vile vile hakuna adhabu yoyote itakupata na matibabu yako yataendelea kama kawaida.

Asante kwa kuchukua muda wako kusoma habari hii na iwapo una maswali yoyote, tafadhali usikose kuuliza.

Kwa maelezo zaidi unaweza wasiliana na;

Daktari.Stella Kisoi

Nambari ya simu: 0725246271

Sanduku la Posta 1387-00100

Nairobi.

Profesa. Munyao

Mwenyekiti,

Idara ya Clinical Medicine and Therapeutics

Chuo Kikuu cha Nairobi

APPENDIX 3: CONSENT FORM

Introduction

My name is Dr. Kisoi Stella, a post graduate student in Internal Medicine and Therapeautics at the University of Nairobi. I am carrying out a research to establish the types of bacteria that cause wound infection in Kenyatta National Hospital, their susceptibility to different antibiotics. Wound infections vary from local to severe infections such as sepsis. This will help inform current practice on antibiotic choice in wound infections and hence improved outcomes.

Procedure

A brief history physical exam to determine signs and symptoms of wound infection. The wound will then be washed and under local anaesthesia to numb pain, a small piece of tissue will be collected and taken to laboratory for analysis. This will take about 10-20 minutes of your time.

Confidentiality

The information obtained will be treated with utmost confidentiality and only be available to the principal investigator and his research team. Your name will not be used in the proforma. We will not be sharing the identity of anyone participating in this research.

The knowledge that we get from this study will be shared with the policy makers in the Ministry of Health, University of Nairobi, KNH and doctors through publications, conferences, journals and presentations. Confidential information will not be shared with any third party.

Benefits

There are no direct benefits to the participants; however, knowledge from the study findings could help improve future care of patients with wound infection. The findings will be communicated to your primary physician and appropriate management implemented. Participants shall not receive any monetary compensation to take part in the study.

Risk

The risks in this study are those associated with punch biopsy only.

Cost and compensation

There will be no extra cost incurred for participating in this study.

Participation

You are free to participate or decline participation in this study and that will not change your current management and treatment that is routinely offered in this hospital for your particular condition. You have a right to refuse or withdraw from this study at any point

Questions about the research

If you have any questions on the study, kindly contact me on this telephone number 0725246271

I ……. Hereby consent to take part in this study to establish the types of bacteria that cause wound infection in Kenyatta National Hospital, their susceptibility to different antibiotics. The nature of the study has been explained to me and I have been assured my participation is voluntary and shall not impact my health negatively.

Signature/ Thumb print: …….

Date: …….

Investigator statement

I explained the purpose and implications of the study to the participant.

Signature: ……...

Date: ……

Researcher's signature..... Date: .....

Name (PRINT): ..... Designation: .....

KIAMBATISHO CHA TATU: FOMU YA IDHINI

Utangulizi

Huu ni utafiti unaofanywa na daktari Stella Kisoi kuhusu viini vinavyoadhiri vidonda, katika hosipitali kuu ya Kenyatta. Mapendekezo kutokana na utafiti huu yanaweza kutumika na wasimamizi katika kuboresha utoaji wa huduma.

Kushiriki

Kushiriki kwako katika utafiti huu ni kwa hiari yako. Iwapo utakubali kushiriki, haya ndiyo yanahusika na utafiti huu:

Utaratibu

Kupata takwimu za kijamii kama vile umri, kauli ya ajira, kiwango cha juu cha elimu, muda ambao umeugua kidonda.

Jina lako na nambari yako ya usajili hospitalini hazitatajwa katika profoma ya utafiti.

Mtihani mfupi wa kimwili wa kupima ambukizo la viini kutoka kwa kidonda.

Kidonda kitaoshwa na maji yaliyona chumvi,na kugandishwa kisha sampuli itachukuliwa kwenye kidonda na kupelekwa kwamaabara kufanya utafiti ili kujumuisha viini vilivyoko.

Haya yote yatachukua muda wa dakika kama kumi/ishirini (10-20).

Faida

Hakuna faida za moja kwa moja kwa washiriki,ila maarifa yatakayotokana na utafitit huu yanaweza kuboresha matibabu ya wagonjwa siku zijazo.Matokeo yatawasilishwa kwa daktari

wako na rufaa mwafaka itafanyika iwapo kuna haja.Washiriki hawatapata fidia yoyote ya kifedha kwa kushiriki katika utafiti huu.

Hatari

Ushiriki wako katika utafiti huu una hatari chache.Utaweza kuhisi kwamba unasumbuliwa utakapokua unajibu maswali kuhusu maisha yako ya kibinafsi**.**

Usiri

Habari utakazozitoa zitakua ni siri.Daktari wako wa kwanza atajulishwa matokeo ya utafiti yanayohusika na matibabu yako.Utahitajika kutia sahihi kwenye fomu ya idhini iwapo utakubali kushiriki kwenye utafiti.Iwapo utataka kujitoa kwenye utafiti huu, unaruhusiwa kufanya hivyo katika hatua yoyote na bila adhabu yoyote.

Usipokubali kushiriki katika utafiti huu vile vile hakuna adhabu yoyote itakupata na matibabu yako yataendelea kama kawaida.

Asante kwa kuchukua muda wako kusoma habari hii na iwapo una maswali yoyote, tafadhali usikose kuuliza.

Maswali kuhusu utafiti

Kwa maelezo zaidi unaweza wasiliana nami kwa nambari 0725246271

Mimi…… nakubali kwamba nitashiriki katika utafiti kuhusu viini vinavyoadhiri vidonda, katika hosipitali Kuu ya Kenyatta.

Nimeelezwa asili ya utafiti huu na kuakikishiwa kwamba kushiriki kwangu ni kwa hiari na kwamba hakutakua na athari mbaya kwa afya yangu.

Sahihi/alama ya kidole: ……

Tarehe: ……

Kauli ya Mtafiti

Nilieleza madhumuni na maana ya utafiti kwa mshiriki.

Sahihi: …….

Tarehe: ……

APPENDIX 4: STUDY PROFORMA

(Fill out the form and Tick in the applicable/appropriate box **clearly**)

Date……

Study No…….

Age (years): ……

1. Gender: Male □ Female □
2. Level of education none□ primary□ secondary□ …...college □
3. Occupation: employed □ unemployed □
4. Marital status: single□ married□ divorced□ separated □
5. Comorbidities: Hypertension□ Diabetes□ Malignancy□ other……
6. Do you use alcohol/cigarettes/bhang/any drugs? Yes □ no □
7. Ward/clinic……**…**
8. Hospital stay if in-patient (from D.O.A) ……
9. Duration of wound……
10. What is the cause of the wound? (tick appropriately)

Diabetes □ Bedsore □ Bites (insect, animal /snake/human)□ spontaneous □Malignancy □

Others (specify)……

11. Have you used any antibiotics used last three months? Yes □ No □

If yes which ones……

TABLE 13: SIGNS OF INFECTION FOUND

(TICK APPROPRIATELY)

Local covert subtle signs	Local overt /classic signs	Spreading infection	Systemic infection
• Excessive granulation tissue • Bleeding, friable tissue • Epithelial bridging and pocketing in granulation tissue • Wound breakdown and enlargement • New or increasing pain • Increasing mal-odour	• Erythema • Local warmth • Swelling • Purulent discharge • New or increasing pain • Increasing mal-odour	• Extending induration +/- erythema • Lymphangitis • Crepitus • Wound breakdown/ dehiscence +/- satellite lesions • General malaise/ lethargy poor appetite • Inflammation, swelling	• Sepsis • Septic shock • End-organ failure NB: must not be attributable to other infections.

12. Duration of symptoms…….

13. M/C/S REPORT collection tool: Identified micro-organisms

(a)Gram positive bacteria isolate (*tick where applicable*)

Staphylococcus spp……

Streptococcus pyogenes……

Enterococcus……

Other…….

(b)Gram negative bacteria isolate (tick where applicable)

Pseudomonas aeruginosa…

Escherichia coli……

Klebsiella spp……

Proteus spp……

Other……

Antibiotic sensitivity ……

TABLE 14: GRAM NEGATIVE BACTERIA

ANTIBIOTIC	SENSITIVE	INTERMEDIATE	RESISTANT

TABLE 15: GRAM POSITIVE BACTERIA

ANTIBIOTIC	SENSITIVE	INTERMEDIATE	RESISTANT