زانکۆی سەلاحەدین - هەولێر
Salahaddin University-Erbil

Microbiology of wound infections among Patients in Erbil City

Research Project

Submitted to the Department of Biology in partial fulfilment of the requirements for the degree of BSc. in Biology

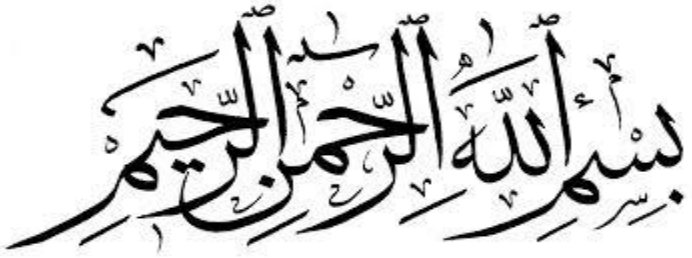
BY:

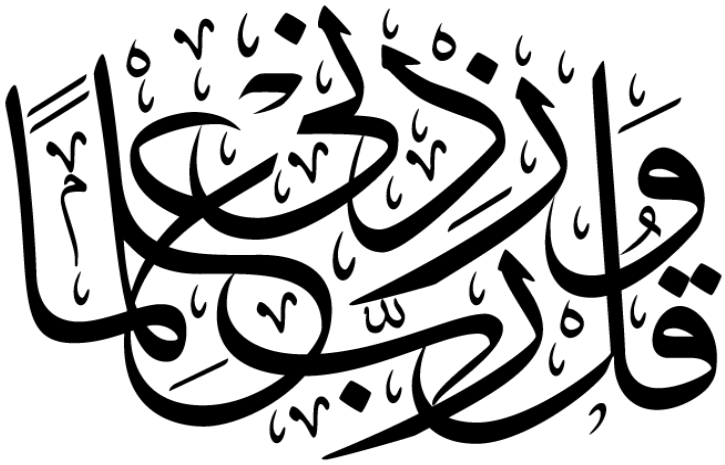
Marwan Ismahel Qadr

Supervised by:

Asst. Prof. Dr. Suhaila Nafee Darogha

May - 2020





صدق الله العظيم

SUPERVISOR CERTIFICATE

This research project has been written under my supervision and has been submitted for the award of the degree of BSc. in Biology with my approval as a supervisor.

Signature**:**

Name**: Dr. Suhaila Nafee Darogha**

Date**: May, 2020**

I confirm that all requirements have been fulfilled

Signature:

Name**: Assist. Prof. Dr. Saeed Omer Ibrahim**

Head of the department of biology

Date**: May, 2020**

DEDICATION

This work is dedicated to

My Parents

My friends

All whom I appreciate

Marwan

ACKNOWLEDGEMENTS

I would like to thank Allah for his blessing virtues to implant the soul of endurance and faith in myself for completing this study. Also, I have no appropriate words that fully express the immense indebtedness and deep gratitude I owe to my worthy learned research supervisor Dr. Suhaila Nafee Darogha for her keen interest, admirable, inspiring behavior and valuable knowledge which she contributed in this work in multitude ways. Whenever I needed she was there near me and always strayed me out from the disaster.

Marwan

SUMMARY

Wound infection plays an important role in the development of chronicity, delaying wound healing. This study aimed to identify causative agents of wound infections. Fifty nine wound swab samples were collected from 59 patients and analyzed for the isolation and identification of microorganisms from June to December 2019. The results showed that only 16 (27.11%) were showed no growth while the remaining wound swabs, 43 (72.88%), were considered infected and one or more microbial species with clinical importance were isolated from them. Rates of isolation of Gram-negative and Gram-positive were 55.81% and 37.20%, respectively. A total of 19 species were isolated from 43 infected wounds. The most common bacterial species detected was *Pseudomonas aeruginosa* and *Escherichia coli* followed by *Klebseilla pneumonia* and *Acinetobacter baumanni*, Dimicrobial infection was found in 4/43 (9.3%) while Monomicrobial isolates represent 39/43 (90.7%) of the samples.

Key words: wound infection, gram positive bacteria, gram negative infection, fungal infection

TABLE OF CONTENTS

Title **Page**

LIST OF TALES

Title **Page**

INTRODUCTION

1. INTRODUCTION

The primary function of intact skin is to control microbial populations that live on the skin surface and to prevent underlying tissue from becoming colonized and invaded by potential pathogens (Ndip *et al*., 2007). Exposure of subcutaneous tissue following a loss of skin integrity (i.e. wound) provides a moist, warm, and nutritious environment that is conducive to microbial colonization and proliferation. Since wound colonization is most frequently poly-microbial, involving numerous microorganisms that are potentially pathogenic, any wound is at some risk of becoming infected (Dai *et al*., 2010).

Wound infection is defined as the presence and growth of microorganism in wound (Bowler *et al*., 2001). Infection of wound results in discharge of pus formed (Collier, 2002). Pus is an exudates, typically white yellowish fluid that forms by the process of necrosis (Bowler *et al*., 2001). Development of wound infection depends on the interplay of a many factors; the breaking of the host protective layer, the skin, and muscles. This disturbance in the protective layer induces many cell types into the wound to initiate host response (Collier, 2002).

Wound infections are one of the most common hospital acquired infections and are an important cause of morbidity and account for 70-80% mortality (Gottrup *et al*., 2005). Wound infections may be endogenous or exogenous. Endogenous

infections or auto Infections are caused by organisms that have been leading a commensal existence elsewhere in the patient s body. In exogenous infection the source of infecting organism is outside the body of patient who becomes infected (Wilson *et al*., 2004).

A number of bacteria may potentially cause wound infection which includes Gram positive bacteria involving *Staphylococcus aureus, Enteococcus faecalis* and other β-hemolytic Streptococci. Gram negative bacteria involving aerobic rods, *Pseudomonas aeruginosa*, and facultative rods, including *Escherichia coli, Enterobacter* spp., *Klebsiella* spp., *Proteus* spp. etc. and some anaerobes like: *Clostridium* spp., *Bacteroides* spp. etc are involved in wound infection (Coller, 2004).

Wound healing needs a good healthy environment so that the normal physiological process will result in a normal healing process with minimal scar formation. One of the most important strategies to keep the process of healing ongoing is to sterilize damaged tissue from any microbial infection (Al-Waili *et al*., 2011).

The aim of the present study was to find out common bacterial pathogens responsible for wound infection.

LITERATURE REVIEW

2. LITERATURE REVIEW

2.1 Wounds

A wound is a break in the integrity of the skin or tissues, which may be associated with disruption of the structure and function. Another way, a wound may be defined as disruption of the normal continuity of bodily structures due to trauma, which may be penetrating or non-penetrating (Al-Tameemi *et al*., 2013). Development of wound infection depends on the many factors including preexisting illness, length of operation, wound class and contamination (Razavi *et al*., 2005). Infection of the wound is the invasion and proliferation by one or more species of microorganisms sometimes resulting in pus formation (Mohammed *et al*., 2013). Wound can be infected by a variety of microorganisms ranging from bacteria to fungus and parasites (Bowler *et al*., 2001).

Wounds are commonly encountered in plastic surgical practice and constitute a significant source of physical, psychological, and economic burden to patients in particular, and the society at large in terms of the huge resources spent in treating such wounds as well as time/days lost at workplaces. Clinically, wound may be categorized as acute or chronic based on the duration of their existence, 6 weeks usually the dividing line (Gurtner and Wong, 2007).

2.2 Wound types

Wounds are broadly classified as being either acute or chronic.

2.2.1 Acute wounds

Acute wounds are caused by external damage to intact skin and include surgical wounds, bites, burns, minor cuts, abrasions, lacerations and those caused by crush or gunshot injuries (Flanagan, 1998). Acute wound infection is initially identified by the presence of clinical signs and symptoms of infection such as: pain, erythema, localized edema, heat and purulence (Stotts and Hunt, 1997). Assuming a relatively healthy host, acute wounds heals within a predictable time frame, progressing through a reparative process of coagulation, inflammation, proliferation or regeneration, maturation, and re-modelling (Enoch and Harding, 2003; Schultz *et al*., 2003; Sibbald *et al*., 2003). The variance in the time frame occurs according to the type, anatomical location and the depth of the wound (Bowler *et al*., 2001).

2.2.2 Chronic wounds

Chronic wounds are usually caused by endogenous mechanisms associated with predisposing conditions that compromise the integrity of the dermis and epidermis. Pathophysiological conditions such as arterial disease, venous disease, malignancy, and diabetes mellitus may predispose an individual to the development of chronic wounds such as leg ulcers, foot ulcers, malignant wounds, and pressure ulcers (Dow, 2003). In chronic wounds clinical signs of covert infection include: serous exudate,

delayed healing, discoloration of granulation tissue, pocketing at the wound base, malodor, and friable granulation tissue (Schultz *et al*., 2003).

2.3 Wound infections

Wound infection is defined as "The deposition and multiplication of bacterial or fungal in tissue with an associated host reaction" Wound infection occurs when there is replication of one or more microorganisms in a wound, which provokes a series of local and systemic host responses that leads to a delay in wound healing (Bowler *et al*., 2001; Sibbald *et al*., 2003). Typical features are cellulitis (spreading erythema) and purulent exudate. The time it takes for wound to convert to colonize to an infected wound is difficult even for the expert clinician to determine at the bedside. A wound can be initially colonized with pathogenic bacteria without inducing host injury, as the bacteria replicate covert infection results (Dai et al., 2010). A covert wound infection may not entail significant tissue invasion, yet will include sufficient injury at the wound-host interface to impair wound healing. Ultimately the opportunistic pathogenic bacteria replicate further with local invasion of the soft tissue (overt wound infection), or systemic dissemination (sepsis) (Dow *et al*., 1999).

Wound infection is directly related to the host's ability to resist infection, the number and virulence of the organisms. Bacterial infections of wounds are among the leading causes of morbidity and mortality throughout the world and are regarded as one of the most common nosocomial infections. Wound infections have been

reported to vary between 3 and 11% in developed countries and estimated to be as high as 40% in developing countries. Wound infections increase with the degree of wound contamination, and it is estimated that 50% of wounds contaminated by bacteria become clinically infected (Ducel *et al*., 2002; Taiwo and Okesina, 2002). Infection in wound constitutes a major barrier to healing and can have an adverse impact on the patient's quality of life as well as on the healing rate of the wound. Infected wounds are likely to be more painful, hypersensitive and odorous, resulting in increased discomfort and inconvenience for the patient (Kotz *et al*., 2009).

The prevalent organisms that have been associated with wound infection include *Staphylococcus aureus (S. aureus)* which from various studies have been found to account for 20-40% and *Pseudomonas aeruginosa (P. aeruginosa)* 5-15% of the nosocomial infection, with infection mainly following surgery and burns. Other pathogens such as Enterococci and members of the Enterobacteriaceae have been implicated, especially in immune compromised patients and following abdominal surgery (Taiwo and Okesina, 2002).

2.4 Pathogenesis of wound infection

Physiological responses to microbial pathogens varies greatly in acute and chronic wounds. In the acute wound environment, the invasion of pathogenic pathogens stimulates the host's immune system and provokes an inflammatory response. This is initiated by the release of cytokines and growth factors. This initial inflammatory reaction produces vasodilatation and an increase in blood flow to the

area of injury (Stotts, 2000). Increased vascular permeability permits the entrance of neutrophils and macrophages to phagocytose the invading organisms. Lymphocytes release lymphokines that recruit neutrophils and monocytes to the area of injury. There is an increase in exudate due to the increase in permeability and number of cells recruited to the area. Antibodies are activated if the individual has had previous exposure to the same organism. Collectively they remove microorganisms, foreign debris, bacterial toxins, and enzymes (Dow *et al*., 1999).

In chronic wounds the continuous presence of virulent microorganisms can lead to host injury by prolonging the inflammatory response. There is a persistent production of inflammatory mediators such as prostaglandin E2 and thromboxane together with a steady influx of neutrophils, which release cytolytic enzymes and free oxygen radicals Vasoconstrictions metabolites and localized thrombosis can lead to further replication of pathogens and tissue destruction. The increase in bacterial numbers can cause depletion of complement and platelets (Laato *et al*, 1988; Kotz *et al*., 2009).

2.5 Diagnosis of wound infection

In acute wounds it is recognized that the invasion of bacteria into the surrounding tissue provokes a sequence of systemic and local host responses. Left untreated sepsis, multi-organ failure and death can occur (Dow *et al*., 1999; Enoch and Harding, 2003). Overt signs of infection include cellulitis (erythema and local heat), pain, swelling, and purulent discharge. Identified additional criteria for

identifying infection in open wounds including; delayed healing, discoloration, friable granulation tissue which bleeds easily, unexpected pain/tenderness, pocketing at base of wound, bridging at base of wound, abnormal smell (Bowler, 2003; Carville *et al*., 2008).

Wound swab culture is the most frequently employed method of confirming wound infection. Properly performed wound swab cultures provide useful data to augment diagnostic and therapeutic decision making (Bonham *et al*., 2009). Many ongoing studies reported a pre-dominance of Gram-negative aerobes over Gram positive bacteria in these specimens. It is of significant importance in health care system because, in the routine practice of management of wound infections, surgeons / physicians mostly prescribe the antibacterial coverage of Gram positive bacteria and the anaerobes (Banashankari *et al*., 2012).

METHODOLOGY

3. METHODOLOGY

A retrospective, record based study was carried out in Rizgary Teaching hospital in Erbil city of Iraqi Kurdistan Region. The study was carried out based on the records of Culture reports of patients admitted to this hospital, during seven months from June to December of the year 2019. The study was conducted on the data of gram negative and positive bacteria and fungi isolated from total of 59 wound swabs taken from 28 male and 31 female. The wound swabs received were processed following standard microbiological procedures. The colonies grown were identified with the help of colony characteristics, Gram staining and biochemical test. Pure colony of each type was identified by VITEK 2 directly (Niranjan *et al*., 2014).

RESULTS AND DISCUSSION

4. RESULTS AND DISCUSSION

Bacterial contamination of wounds is a serious problem in the hospital especially in surgical practice where the site of a sterile operation can become contaminated and subsequently infected (Pondei *et al*., 2013).

Among 59 study subjects, bacterial pathogens were isolated from 43 patients with the isolation rate of 72.88%., while 16 (27.1%) were showed no growth as showen in Table (1). This was higher than the previous study done in Gondar (52%), Bahir Dar (53%), and Addis Ababa (42%), Ethiopia (Mulu *et al*., 2006; Biadglegne, *et al*., 2009; Kibret and Abera, 2011). This high rate of bacterial isolation in the present study may be due to the differences of the quality of wound swab specimens and bacteriological techniques. On the other hand the type of wound pathogens and their rate of isolation in these findings were found to be lower than studies done in India (79%) and Nigeria (94%) (Wariso and Nwachukwu, 2003; Valarmathi *et al*., 2013).

Table 1. Growth pattern in total samples

Growth	Number	Percentage
Growth cultures	43	72.88
No growth cultures	16	27.11
Total	59	99.99

The current findings showed that the rates of isolation of Gram-negative and Gram-positive were 55.81% and 37.20%, respectively. This was in agreement with

studies done in Zaria, Nigeria, 55% and 44%, respectively (Garba *et al*., 2012). However, the present result is different from the previous report from Gondar University Hospital, Ethiopia (29% versus 71%) (Mulue *et al*., 2006).

The present findings show higher rates of isolation of Gram negative wound pathogens from the same area. This high rate of Gram-negative and low rate of Gram-positive isolates from wound in the same area may be due to high number of cases included from inpatients in the present study compared to out patients. This may probably contribute high number of Gram-negatives than Gram-positives (Mohammed *et al*., 2017).

Table 2.Number and percentage of gram positive, gram negative bacteria isolates and Candida spp.

Microorganisms	Number	Percentage
Gram positive bacteria	16	37.20
Gram negative bacteria	24	55.81
Candida spp.	5	11.62

The occurrence of higher number of male patients than female patients for the collection of wound samples may be due to the higher involvement of males in physical outdoor works for earning livelihood as compared to females and more chances of accidents during the activities. A study carried out in three hospitals (Federal Medical Centre, Owerri, Imo State University Teaching Hospital, Orlu and General Hospital, Okigwe) by Ohalete *et al*. (2010) also supported the result of the present study who reported that the males (59.3%) were more prone to wound infection than females (40.7%). The higher growth positive cases in male patients

(58.8%) than in female (44.2%) was observed in this study (Table 3), and was supported by the similar study carried out by Shrestha (2013).

Table 3: Distribution of wound infection according to gender.

Gender	Number	Percentage
Male	24/28	55.8
Female	19/31	44.2
Total	43/59	100

The results of the present study showed that nineteen different microbial species were isolated; 24 (60%) were Gram-negative bacteria, 16 (40%) were Gram-positive bacteria and 5 (11.62) were *Candida* spp. The most predominant gram negative bacteria detected were *Pseudomonas aeruginosa* and *E. coli* 6/45 (15% for each of them), followed by *Klebseilla pneumonia* 5/45 (12.50%), *Acenetobacter baumannii* (10.00%), *Proteus mirabilis, Enterobacter cloacae and Morganella morganii* (2.50% for each one). Among gram positive bacteria, *Staphylococcus intermedius* was the most predominant species with 4 (10.00%) of total isolates followed by *Staph. haemolyticus* (7.50%), *Staph. epidermidis* and *Streptococcus agalacticae* (5.00%),and only one isolates (2.50%) were for *Staph. saprophyticus, Enterococuus faecalis, Enterococcus faecium and Locunia kristinae.* While about *Candida* spp. Four isolates (9.30%) wer related to *Candida albicans* and one isolate (2.32%) was *Candida glabrata* (Table 4 and 5).

Table 4: Distribution of bacterial isolates.

Bacteria isolates	Number	Percentage
Gram positive bacteria		
Staphylococcus intermedius	4	8.9
Staphylococcus haemolyticus	3	6.7
Staphylococcus epidermidis	2	4.4
Staphylococcus saprophyticus	1	2.2
Staphylococcus aureus	1	2.2
Streptococcus agalactiae	2	4.4
Enterococcus faecalis	1	2.2
Enterococcus faecium	1	2.2
Locunia kristinae	1	2.2
Gram negative bacteria		
E. coli	6	13.3
Pseudomonas aeruginosa	6	13.3
Klebseilla pneumoniae	5	11.1
Acinetobacter baumannii	4	8.9
Proteus mirabilis	1	2.2
Enterobacter cloacae	1	2.2
Morganella morganii	1	2.2

Table 5. Distribution of *Candida* spp.

Candida spp.	Number	Percentage
Candida albicans	4	8.9
Candida glabrata	1	2.2

The most common isolate was *Staphylococcus spp.* which was also reported in many other studies to be the predominant microorganism (40–60% of the total microorganisms) isolated from different types of wounds (Bowler and Davies, 1999). *Pseudomonas aeruginosa* and *E. coli* were the Gram-negative more detected, which is also in agreement with other reports (Davies *et al*., 2004: Burnolle *et al*., 2010). It is well documented that bacteria such as *Staphylococcus spp.* and

Pseudomonas aeruginosa produce very destructive virulence factors, responsible for maintaining infection and delay healing in chronic wounds. *S. aureus* causes clinically relevant infections mostly because of its virulence factors such as coagulase, catalase, clumping-factor A and leucocidines (Dissemond, 2009). Similarly, the production of an elastase by *Pseudomonas. aeruginosa* has been associated to its pathogenicity in the wound environment (Schmidtchen, 2003).

A study reported that 129 swabs and pus specimens from various types of surgical sites suspected to be infected on clinical ground were processed and the most common organisms were *Staphylococcus aureus* (50.32%) followed by *Pseudomonas aeruginosa* (16.3%), *Escherichia coli* (14.37%), *Klebsiella pneumoniae* (11.76%), miscellaneous gram negative rods (5.88%), and *Streptococcus pyogenes* (1.30%) (Hanifah, 1990). In another study out of 171 cases of wounds of various etiologies examined and screened bacteriologically *S. aureus* was the mostfrequently isolated (39.9%) single organism and other organisms being *E. coli* (26.1%), *Pseudomonas aeruginosa* (15.4%), *Klebsiella* species (5.8%), *Streptococcus pyogenes* (4.9%), *Proteus* species (4.8%) and coliform organisms (3.1%); however, collectively the gram-negative organisms were the majority among the isolated organisms (Malone *et al*., 2002). A study showed wound infection in the post-operative elective surgeries was 11.3% and the microorganisms found were *Staphylococcus aureus* 70.5% and *Escherichia coli* 29.5% (Wilson *et al*., 2004).

In our study, only 9.30 % of the wounds displayed polymicrobial infections as shown in Table 6. It is known that interspecies interactions consist mostly in bacterial synergy that enhances survival, therefore hampering the infection eradication. Moreover, microorganisms have the ability to establish themselves and proliferate as a biofilm, both in monomicrobial and polymicrobial biofilms, which are often considered to be a further complication that has a significant contribution to the lack of successful antibiotic treatment (Kirketerp *et al*., 2008).

Table 6: Growth pattern of Monomicrobial and dimicrobial isolates in wound swabs.

Types of growth	Numbers	Percentage
Monomicrobial isolates	39	90.7
Dimicrobial isolates	4	9.3
Total	45	100

To prevent or reduce wound infection is a goal shared by health care practitioners in charge of wound management and care; however, when infection is already established, wound management practices should be specifically addressed and become more challenging and demanding. Antibiotic treatment is recommended but, previously, an antibiotic susceptibility test should be performed.

CONCLUSSIONS

5. CONCLUSSION

In the conclusion it can be said that the clinical and microbiological aspect of wound infection are very wide range. Multiple bacteria as well as other organism cause different wound infection. Proper wound management should be implemented to combat this problem.

6. REFERENCES

Al-Tameemi, H.A.A.N and Khalaf, .JM. (2013). Isolation and Identification of Fungi from Wounds and Burns of Human and Farm Animals. The Iraqi Journal of Veterinary medicine. 37(2):251-256.

Al-Waili, N.S.; Salom, K. and Al-Ghamdi, A.A. (2011). Honey for wound healing, ulcers, and burns. The Scientific World Journal, 11:766–787.

Banashankari, G.S.; Rudresh, H.K. and Harsha, A.H. (2012). Prevalence of Gram Negative Bacteria in Diabetic Foot-A Clinico-Microbiological Study. *Al Ameen J Med Sci*. 5 (3) : 224-232.

Biadglegne, F.; Abera, B.; Alem, A.and Anagaw, B. (2009). "Bacterial isolates from wound infection and their antimicrobial susceptibility pattern in Felege Hiwot Referral Hospital, North west Ethiopia," *Ethiopian Journal of Health Sciences*, 19(3): 173–177.

Bonham, P. (2009). Swab Cultures for Diagnosing Wound Infections: A Literature Review and Clinical Guideline, *Journal of Wound, Ostomy & Continence Nursing*. 36 (4) 389-395.

Bowler, P. (2003). Bacterial growth guideline: Reassessing its clinical relevance. *Ostomy Wound Management, 49*(1), 52-53.

Bowler, P.; Duerden, B. and Armstrong, D. (2001). Wound Microbiology and Associated Approaches to Wound Management. Clin Microbiol Rev, 14(2):244-69.

Bowler, P.G. and Davies, B.J. (1999). The microbiology of infected and noninfected leg ulcers. *Int. J. Dermatol.,* **38**:573–578.

Carville, K., Cuddigan, J., Fletcher, J., Fuchs, P., Harding, K., Ishikawa, O. (2008). *Prinicples of best practice: Wound infection in clinical practice. An International Consensus.* London:

Collier M. (2002). "Wound bed management: Key princples for practice". *Journal of Professional Nursing* 18(4) : 221-225.

Dai T, Huang Y-Y, Sharma SK, Hashmi JT, Kurup DB, Hamblin MR: (2010).Topical antimicrobials for burn wound infections. *Recent Pat Antiinfect Drug Discov* ,5(2):124-151.

Davies, C.E.; Hill, K.E.; Wilson, M.J.; Stephens, P.; Hill, C.M.; Harding, K.G. and Thomas, D.W. (2004). Use of 16S ribosomal DNA PCR and denaturing gradient gel electrophoresis for analysis of the microfloras of healing and nonhealing chronic venous leg ulcers. *J. Clin. Microbiol.,* **42**:3549–3557.

Dow, G., Browne, A., & Sibbald, G. (1999). Infection in chronic wounds: Controversies in diagnosis and treatment. *Ostomy Wound Management, 45*(8), 23-40.

Ducel, G.; Fabry, J. and Nicolle, L. (2002). Prevention of hospital-acquired infections. 2nd ed. Geneva: World Health Organization.

Enoch, S., & Harding, K. (2003). Wound bed preparation: The science behind the removal of barriers to healing. *Wounds, 15*(7), 213-229.

Flanagan, M. (1998). *Wound Management*. Edinburgh: Churchill Livingston.

Garba, I.; Lusa, Y.; Bawa, E.; Tijjani, M.; Aliyu, M. and Raji,U. (2012). "Antibiotics susceptibility pattern of *Pseudomonas aeruginosa* isolated from wounds in patients attending Ahmadu Bello University Teaching Hospital, Zaria, Nigeria," *Nigerian Journal of Basic and Applied Science*, 20, 32–34

Gottrup, F.; Melling, A. and Hollander, D. (2005). An overview of surgical site infections: aetiology, incidence and risk factors. E.W.M.A. Journal; 5(2):11-15.

Gurtner, G.C.and Wong, V. (2007). Grabb Plastic Surgery. 6th ed. Philadelphia: Wolters Kluwer-Lippincot William and Wilkins; Wound healing: Normal and abnormal; pp. 15–22.

Hanifah, Y.A. (1990). Post-operative surgical wound infection. Med. J. Malaysia., 45(4):293-297.

Kibret, M. and Abera, B. (2011). "Bacteriology and antibiogram of pathogens from wound infections at Dessie Laboratory, North East Ethiopia," *Tanzania Journal of Health Research*, vol. 13, no. 4, pp. 1–11.

Kirketerp-Møller, K.; Jensen, P.Ø.; Fazli, M.; Madsen, K.G.; Pedersen, J.; Moser, C.; Tolker-Nielsen, T.; Høiby, N.; Givskov, M. and Bjarnsholt ,T. (2008). Distribution, organization, and ecology of bacteria in chronic wounds. *J. Clin. Microbiol.,* **46**:2717–2722.

Kotz P, Fisher J, McCluskey P, Hartwell SD, Dharma H: Use of a new silver barrier dressing, ALLEVYN Ag in exuding chronic wounds. *Int Wound J* 2009, 6: 186-194.

Laato, M., Niinikoski, J. and Lundberg, C. (1988). Inflammatory reaction in blood flow and experimental wounds inoculated with *Staphylococcus aureus European Surgery Research, 20*, 33.

Malone, D.L.; Genuit, T.; Tracy, J.K.; Gannon, C. and Napolitano, L.M. (2002). Surgical site infections: reanalysis of risk factors. Journal of Surgical Research. 103(1):89-95.

Mohammed A, Adeshina GO, Ibrahim YK. Incidence and Antibiotic Susceptibility Pattern of Bacterial Isolates from Wound Infections in a Tertiary Hospital in Nigeria. Tropical Journal of Pharmaceutical Research. 2013;12(4):617-21.

Mohammed, A.; Seid, M.E.; Gebrecherkos, T.; Tiruneh, M. and Moges, F. (2017). Bacterial Isolates and Their Antimicrobial Susceptibility Patterns of Wound Infections among Inpatients and Outpatients Attending the University of Gondar Referral Hospital, Northwest Ethiopia. International Journal of Microbiology Volume 2017, Article ID 8953829, 10 pages

Mulu, A.; Moges, F.; Tessema, B.; and Kassu,A. (2006). "Pattern and multiple drug resistance of bacterial pathogens isolated from wound infection at University of Gondar Teaching Hospital, northwest Ethiopia," *Ethiopian Medical Journal*, 44:(2), 125–131.

Ndip, R.N.; Takang, A.E.M.; Echakachi, C.M.; Malongue, A. and Akoachere, J.F.T.K (2007). In vitro antimicrobial activity of selected honeys on clinical isolates of Helicobacter pylori. Afr. Health Sci., 7(4):228–231.

Niranjan, V. and Malini, A. (2014). Antimicrobial resistance pattern in *Escherichia coli* causing urinary tract infection among inpatients. Indian J. Med. Res. 139: 945-948.

Ohalete,, C.N. (2012). Bacteriology of different wound infections and their antimicrobial susceptibility pattern in IMO state Nigeria. World Journal of Pharmacy and Pharmaceutical Sciences. 1(3): 1155-1172.

Pondei K, Fente BG, Oledapo O. (2013). Current Microbial Isolates from Wound Swabs, Their Culture and Sensitivity Pattern at the Niger Delta University Teaching Hospital, Okolobiri, Nigeria. Trop Med Health. 41(2):49-53.

Razavi SM, Ibrahimpoor M, Kashani AS, Jafarian A. (2004). Abdominal Surgical Site Infections: Incidence and Risk Factors at an Iranian Teaching Hospital.

Schmidtchen, A.; Holst, E.; Tapper, H. and Bjorck, L. (2003). Elastase-producing *Pseudomonas aeruginosa* degrade plasma proteins and extracellular products of human skin and fibroblasts, and inhibit fibroblast growth. *Microb. Pathog.*, **34**:47–55.

Schultz, G., Sibbald, G., Falanga, V., Ayello, E., Dowsett, C., Harding, K.. (2003). Wound bed preparation: A systematic approach to wound management. *Wound Repair and Regeneration, 11*(2), S1- S28.

Shrestha, A. and Sharma, V.K. (2013). Bacteriology Study of Wound Infection and Antibiotic susceptibility Oattern of Isolates. Nepal Journal of Science and Technology 14(2): 143-150.

Shrestha, P. (2013). Bacteriology of Wound Infection and their Antibiotic Susceptibility Pattern. M.Sc. dissertation submitted to the Department of Microbiology, Nepal.

Sibbald, R., Orsted, H., Schultz, G., Coutts, P., and Keast, D. (2003). Preparing the wound bed 2003: Focus on infection and inflammation. *Ostomy Wound Management, 49*(11), 24-51.

Stotts, N. (2000). Wound infection: Diagnosis and management. In R. Bryant (Ed.), *Acute and Chronic Wounds: Nursing management* (2nd ed., pp. 179-188). Sydney: Mosby.

Stotts, N., and Hunt, T. (1997). Managing bacterial colonization and infection. *Clinics in Geriatric Medicine, 13*(565-73).

Taiwo S, Okesina A, Onile B: In vitro antimicrobial susceptibility pattern of bacterial isolates from wound infections in University of Ilorin Teaching Hospital. *Afr J Clin Exp Microbiol* 2002,3(1):6-10.

Valarmathi, S.; Pandian, M.R. and Senthilkumar, B. (2013). "Incidence and screening of wound infection causing microorganisms," *Journal of Academy Industry Research*, 1 : (8), 508–510.

Wariso, B.A. and Nwachukwu,C.O. (2003). "A survey of common pathogens in wound in patients at University of Port Harcourt Teaching Hospital (U.P.T.H.), Port Harcourt," *West African Journal of Medicine*, 22 :(1), 50–54.

Wilson, A. P. R.; Gibbons, C.; Reeves, B. C., Hodgson, B.; Liu, M. and Plummer, D. (2004). Surgical wound infections as a performance indicator: agreement of common definitions of wound infections in 4773 patients. B.M.J., 329:720-722.