REVIEW OF LITERATURE

There is documentary evidence that the historical background of wound infection may be traced to as far back as 1\textsuperscript{st} century AD when a Roman physician, Cornelius Celsus described the four principal signs of inflammation and used ‘antiseptic’ solutions. Claudius Galen (130-200 AD), another Roman physician had such an influence on the management of wounds that he is still thought of by many today as the ‘father of surgery’

The 19\textsuperscript{th} century witnessed the acceptance of the germ theory and introduction of antisepsis through Semmelweis (1818-1865), Pasteur (1822-1895) and Lister (1827-1912). The current terminologies like wound contamination, colonization and infection are defined by a Nursing officer, Mary Ayton. Vincent Falanga, (1994) identified the concept of ‘critical colonization’ with fresh insights into chronic wound healing and non-healing wounds. These current terms are:

- Wound contamination- The presence of bacteria within a wound without any host reaction.
- Wound colonization - The presence of bacteria within the wound which do multiply or initiate a host reaction.
- Critical colonization– Multiplication of bacteria causing a delay in wound healing usually associated with an exacerbation of pain not previously reported but still with no obvious host reaction.
- Wound infection- The deposition and multiplication of bacteria in tissue with an associated host reaction.

The presence of a microorganism within the margins of a wound does not indicate that wound infection is predictable. Some bacteria produce proteins that kill or inhibit other bacteria while in some other cases, bacteria produce a variety of metabolites that inhibit the multiplication of other microorganisms (Kingsley, 2001). This is called protective colonization. The development of an infection will be influenced largely by the virulence of the organism and immunological status of the patient. Virulence describes both the pathogenicity and invasiveness of the relevant microorganism. When microorganisms are present to a degree of $10^5$ per gram of tissue, an infection is likely to be present. Quantitatively, wounds harboring bacteria that exceed $10^5$ colony forming units per gram are considered infected.
Histologically, a wound that is infected shows microorganisms in viable tissue. This produces tissue reactions evidenced in the classical features of inflammation like pain, heat, swelling and purulent discharge. This causes delay in healing. When, a wound that was assessed as healing starts to develop strips of granulation tissue in the base as opposed to a uniform spread of granulation tissue across the whole of the wound.

Skin and gut normally harbor certain bacteria in a commensal relationship with the host that serves to limit invasive, pathogenic microorganisms. When epithelial barrier is breached, the normal host response is a series of concerted, physiologic cascades that result in local inflammation (Patel et al., 2000). Inflammation ultimately protects the host and initiates healing. However, if the initial injury is extensive, infection may develop as organisms that previously colonized skin and gut may now invade tissues. In addition, such an environment may allow secondary invaders to cause infection. These organisms or their liberated toxins may overcome the local protective environment with resultant systemic sepsis.

Most surgical wounds are categorized as acute wounds, healing without complication in an expected time frame (Bale and Jones, 1997). However, like all wounds, healing is affected by intrinsic and extrinsic factors that may result in complications. All wounds follow a specific cellular and biochemical sequence of healing. Immediately after wounding occurs, mast cells degranulate and release inflammatory mediators, which allow local blood vessels to dilate. Neutrophils enter the wounded area to digest bacteria, followed by macrophages, which flood the wound bed and release growth factors and prostaglandins to influence the healing process (Nathan, 1987). This inflammatory phase is often characterized by redness and swelling around the wound, accompanied by heat and pain (Tortora and Grabowski, 1996). In clean surgical wounds, this stage can last for three to seven days.

In the reconstructive, or proliferation stage the growth of new vessels and tissue takes place. Fibroblasts move into the wound and collagen synthesis occurs alongside new vessel growth to fill the wound with granulation tissue (Eckersley and Dudley, 1988). As the defect fills, the wound contracts and epithelial tissue forms at the edges. This stage ends when the wound is fully closed. In the final stage of healing - maturation - the wound regains its tensile strength and, as the collagen fibers reorganize, the scar loses some of its red pigmentation and lies flatter to the surface of
the skin. This phase can take up to 18 months to complete (Nguyen et al., 2009; Clark, 1988).

2.1 Microbiology

Brook et al., (1995) studied bacterial post-operative wound infections and concluded that these are caused mainly by aerobic and anaerobic bacteria. Aerobic bacteria mainly constitute species of *Staphylococcus*, *Streptococcus*, *Pseudomonas* and members of family *Enterobacteriaceae*, while anaerobic bacteria mainly contain species of *Clostridium*, *Spherophorus*, *Peptostreptococcus*, *Fusobacterium*, *Pseudomonas* and *Diptheroids*.

The sources of organisms are found in the patient’s own endogenous normal flora and from exogenous sources from the environment and healthcare personnel. Exogenous organisms from the hospital environment are generally more resistant to antimicrobial agents than endogenous organisms. Organisms associated with post-operative wound infections patients include Gram positive bacteria, Gram negative bacteria, yeast and fungi. The distribution of organism’s changes over time in the individual patient and such changes can be initially colonized predominantly with Gram positive organisms which are quite quickly replaced by antibiotic susceptible Gram negative organisms, usually within week of the infection. If wound closure is delayed and the patient becomes infected requiring treatment with broad spectrum antibiotics, this flora may be replaced by yeasts, fungi and antibiotic resistance bacteria.

Gram positive organisms of particular concern include Methicillin resistant *Staphylococcus aureus* (MRSA), *Enterococci*, Group A β-hemolytic *Streptococci* and coagulase negative *Staphylococci*. MRSA were first observed in the United States in the late 1960s and has become an endemic organism in many hospitals. It has been argued that no extra-ordinary efforts are made to control its speed; however this view has been increasingly challenged in the era of Vancomycin Resistant *Enterococcus* (VRE). With the increasing incidence of VRE in hospitals, the risk associated with infection with this organism is increasing. Risk factors identified in patients colonized with VRE include prior vancomycin use, prior use of third generation cephalosporins and antibiotic active against anaerobes, a critically ill patient with severe underlying disease or immunosuppression, and a prolonged hospital stay (Arabshahi et al., 2006).

A study conducted among patients underwent surgery from small community hospital in the United States of America reported *S. aureus* as the commonest isolate (25.8%), followed by *Enterobacteriaceae* (12.4%), *Streptococcus* spp (11.2%),
Cogulase negative *Staphylococcus aureus* (CoNS) (10.1%), *Enterococcus* sps (7.9%) and *Pseudomonas aeruginosa* (6.7%), but MRSA were isolated from only 4.5% as compared with SSIs (Cantlon *et al.,* 2006). Another recent study in USA among patients who underwent operation for hollow viscous injury (Schnuriger *et al.,* 2010) documented *E. coli* as the most commonly isolated microorganism (64.7%) followed by *Enterococcus* sps (41.2%) and Bacteroides (29.4%). Findings from these two studies suggest that the aetiologic agents of SSIs depend on where the procedures are performed and whether skin was incised or gastrointestinal tract was opened. When incisions are made near the perineum or groin, organisms usually include aerobic Gram positive cocci and fecal flora (anaerobic bacteria and Gram negative aerobes).

Findings from a study carried out at a University hospital in Nigeria (Adegoke *et al.,* 2010) showed that the commonly isolated bacteria were *Staphylococcus aureus* (25%), *Pseudomonas aeruginosa* (20%), *Escherichia coli* (15%), *Klebsiella oxytoca* (10%) and *Proteus mirabilis* (10%). Similarly, in a prospective survey done in Central African Republic, among orthopedic surgical patients, it was found that methicillin-susceptible *S. aureus* was the most frequent species isolated followed by *Enterobacteriaceae* sps and *P. aeruginosa*. A strain of *E. cloacae* harboring extended spectrum β-lactamase (ESBLs) was also isolated (Bercion *et al.,* 2007). Frequent isolation of *S. aureus* (28.8%) and *Escherichia coli* (27.1%) have also been reported among patients with abdominal surgical wounds in Ethiopia (Kotisso *et al.,* 1998).

A recent study at a University hospital in Iran, reported *S. aureus* to be the commonest bacteria pathogen with rate of (43%), followed by *E.coli* (21%), *Klebsiella* sps (13%), *Pseudomonas* sps (10%) and CoNS (5%) among surgical patients (Khorvash *et al.,* 2008). In the past study MRSA accounted for a high rate of 78.9% of all *S. aureus* isolates. Another study in the same region (Le *et al.,* 2006) among patients who underwent orthopedic and neurosurgery reported that the three most frequently isolated pathogens were *P. aeruginosa* (29.5%), *S. aureus* (11.5%) and *E. coli* (10.3%). Although 90% of *S. aureus* isolates were MRSA, Gram negative organisms were the most common causative pathogens in contrast to what has been reported in other studies in which Gram positive organisms predominate.

A study conducted in East Africa, one cross-sectional survey among 63 surgical patients at University teaching hospital in Kenya (Koigi-Kamau *et al.,* 2005), reported that *S. aureus* were the most frequently isolated pathogens (54.7%) while *Proteus* sps, *Pseudomonas* sps and *Escherichia coli* were 15.5%, 11.9% and 2.3%, respectively.
The previous study investigated that patients of all age groups and no attempt were made to characterize the bacteria isolated from SSIs. Similar finding was also reported in a study at referral hospital in Uganda, which documented the *S. aureus* as a commonest isolate (45.1%) followed by coliforms (16.9%) and *Proteus mirabilis* (11.3%). MRSA accounted of 25% among isolated *S. aureus* and the majority of surgical patients underwent caesarian and herniorraphy procedures (Anguzu et al., 2007).

Studies have shown an increase in the trend of SSIs, attributable to antimicrobial resistant pathogens such as MRSA. In the data collected between 2003 and 2007 reported that the proportion of MRSA significantly increased (from 16.1% to 20.6%) among culture positive SSIs patients readmitted in 97 US hospitals (Weigelt *et al.*, 2010). Another study reported that MRSA were the most frequent pathogen recovered and the prevalence rate of MRSA SSIs almost doubled during the study period increasing from 0.12 infections per 100 procedures to 0.23 infections per 100 procedures (Maida *et al.*, 2010; Anderson *et al.*, 2007).

A cross sectional study conducted among patients with wound infection visiting Jimma University Specialized Hospital, South-West Ethiopia reported the infection rate of *S. aureus* (32.4%), *E. coli* (20%), *Proteus* sps (16%), Coagulase negative *Staphylococci* (14.5%), *Klebsiella pneumoniae* (10%) and *Pseudomonas aeruginosa* (8%) were the predominant organisms isolated from wound infections (Mohammedaman *et al.*, 2014; Taye, 2005). The high rate of *S.aureus* infection may be because it is an endogenous source of infection. Infection with *S. aureus* organism may also be due to contamination from the environment e.g. contamination of surgical instruments. With the disruption of natural skin barrier *S.aureus*, which is a common bacterium on surfaces, easily find their way into wounds (Shingray *et al.*, 2013).

Findings from a study carried out at tertiary care hospital in India by Mehta *et al.*, 2013 and Shanthi *et al.*, 2012 reported that, out of the 158 isolates, the most common isolates were *Staphylococcus aureus* (27.8 %), followed by *Escherichia coli* (24.05 %), *Klebsiella pneumoniae* (13.29 %), *Pseudomonas aeruginosa* (6.32%), *Klebsiella oxytoca* (5%), *Enterococcus* sps (5.6%) and other various Gram negative rods (9.4%) and *Streptococcus pyogenes* (1.3%).

A hospital based cross-sectional study by Lopiso *et al.*, (2014) in Ethiopia showed that, out of total 177 aerobic bacteria isolates; 105 (59.3 %) were Gram negative and 72 (40.7%) were Gram positive organisms. *S. aureus* were the most
frequent isolate (37.3%); followed by *E. coli* (25.4%), *Klebsiella* sps (13.6%), *Proteus* sps (10.2%), *P. aeruginosa* (10.2%) and CoNS (3.4%) respectively as reported by Shriyan *et al.*, 2010.

Out of 247 samples, *Staphylococcus aureus* (37.12%) were the most predominant isolate followed by *Klebsiella* sps (20.25%). Overall prevalence of MRSA was 23.47% (Nazeer *et al.*, 2014; Rao *et al.*, 2013; Etok *et al.*, 2012). All Gram positive cocci were susceptible to vancomycin and linezolid including MRSA. All Gram negative bacilli showed positive sensitivity towards Amikacin, Piperacillin-Tazobactum and Imipenem in a range of 70-100%. *Staphylococcus aureus* is the most prevalent organism among wound infections. The prevalence of MRSA was 23.47% and more numbers of MRSA isolates were multidrug resistant as compared with the MSSA isolates. Vancomycin and linezolid continue to remain the stronghold for treatment for MRSA infections (Singh *et al.*, 2010).

A study conducted from November 2010 to June 2011 in Hawassa University Referral Teaching Hospital (Guta *et al.*, 2014) reported that out of 177 bacterial isolates identified in the study, the most dominant isolates were *Staphylococcus aureus*, *Klebsiella* sps, *Escherichia coli* and coagulase negative *staphylococci* (CoNS) accounting for 45 (25.4%), 32 (18.1%), 30 (16.9%) and 26 (14.7%) of the isolates respectively. Other bacteria isolated include *Pseudomonas aeruginosa* (9.0%), *Proteus* sps (6.8%), *Streptococcus* sps (5.1%), *Citrobacter* sps (2.3%) and *Enterobacter* sps (1.7%).

### 2.2 Pathogenesis

Microbial contamination of the surgical site is a necessary precursor of SSI. Following contamination the risk of development of SSIs will depend on several factors, the most important ones being the dose and virulence of the pathogens, and host defense mechanisms. Virulence of bacteria depends on the ability to produce toxins and other substances that increase their ability to invade the host, produce tissue damage or survive within the host cells. For example Gram negative bacteria contain endotoxin or Lipopolysaccharide (LPS) which is the most potent microbial mediator implicated in the pathogenesis of sepsis and septic shock. LPS triggers the release of pro-coagulant factors and inflammatory mediators such as cytokine which may initiate systemic inflammatory response syndrome and cause multiple systemic organ failure (Opal, 2010). Some bacteria produce polysaccharide capsule, which inhibit phagocytosis which is a critical host immune response following bacterial contamination (ManGram
et al., 1991). When incision is made invariably it impairs first line of defenses between the environmental microbes and internal host environment, therefore the exposed tissues are at risk of contamination with endogenous patient’s flora. Exogenous contamination may also occur from operating room environments, surgical teams and instruments. The risk of SSI can be conceptualized according to the following relationship, exogenous and endogenous sources.

**Endogenic Sources**

Microorganisms may contain or produce toxins and other substances that increase their ability to invade a host, produce damage within the host, or survive on or in host tissue. For example, any Gram negative bacteria produce endotoxin, which stimulates cytokine production. In turn, cytokines can trigger the systemic inflammatory response syndrome that sometimes leads to multiple system organ failure. Certain strains of Clostridia and Streptococci produce potent exotoxins that disrupt cell membranes or alter cellular metabolism (ManGram et al., 1999).

For most SSIs, the source of pathogens is the endogenous flora of the patient’s skin, mucous membranes, or hollow viscera. When mucous membranes or skin is incised, the exposed tissues are at risk for contamination with endogenous flora. These organisms are usually aerobic Gram positive cocci (e.g. Staphylococci), but may include fecal flora (e.g. anaerobic bacteria and Gram negative aerobes) when incisions are made near the perineum or groin. When a gastrointestinal organ is opened during an operation and is the source of pathogens, Gram negative bacilli (e.g. E. coli), Gram positive organisms (e.g. Enterococci), and sometimes anaerobes (e.g. Bacillus fragilis) are the typical SSI isolates (ManGram et al., 1999).

**Exogenic Sources**

These SSIs pathogens include surgical personnel (especially members of the surgical team), the operating room environment (including air), and all tools, instruments, and materials brought to the sterile field during an operation. Exogenous flora is primarily aerobes, especially Gram positive organisms (e.g. Staphylococci and Streptococci) (ManGram et al., 1999).

**2.3 Risk factors**

Anyone who has got surgical operation can develop a wound or infection (Hunt and Hopt, 1997). The potential for the development of wound infection depends on a number of patient variables such as the state of hydration, nutrition and existing medical
conditions as well as other extrinsic factors, for example factors related to pre, intra, and post-operative cares if the patient has undergone surgery (Heinzelmann et al., 2002).

Microbial factors that will influence the establishment of a wound infection include the bacterial inoculums, virulence, and the effect of the microenvironment. When these microbial factors are favorable, impaired host defense set the stage for enacting the chain of events that produce wound infection (Krizek and Robson, 1975). Oxygen tensions of between 5 and 20 mm Hg have been recorded in non-healing wounds (Sheffield, 1988) and oxygen tension values of less than 30 mm Hg have been recorded in infected and traumatized tissue (Morykwas and Argenta, 1997; Hohn et al., 1976); which correlates it with a reported pO$_2$ requirement of approximately 30 mm Hg for active cell division (Hunt and Hopt, 1997).

Factors that have a proven or probable influence on the frequency of wound infections are the use of antibiotic prophylaxis; the duration of surgery, the defense mechanisms of the host; the use of ultraclean air in the operating room, the patient’s temperature in the operating room, the use of supplemental oxygen, the presence of hypovolemia, diabetes mellitus, or adiposity in the patient, the patient's nutritional status, the use of blood transfusion and pain control (Nwachukwu et al., 2009; Gardlund et al., 2002; Gottrup, 2000; Lee et al., 1997).

Optimal antimicrobial in the appropriate dose, time and duration, which has been selected on the basis of the antimicrobial susceptibility pattern of the most common isolates in the hospital, would ensure a decreased rate of post-operative wound infections. Therefore the surveillance of nosocomial infections with an emphasis on antimicrobial audit will reduce the risk of post-operative wound infections (Amrita et al., 2010; Saxer et al., 2009).

Patient variables, Pre-operative preparation, Operative procedure and Post-operative care are four main factors which influence the infection rates in surgical wounds (Patel, 2012).

**Patient variables**

Diseases like Diabetes, Cancer of liver and kidney or Lung conditions that may slow the healing process. Medical condition, such as low blood protein may also affect healing. The immune system is the part of the body that fights infection. For some type of operation, severe protein-calorie malnutrition is crudely associated with post-operative nosocomial infection, impaired wound (Garg, 2000). The immune system may be weakened by radiation, poor nutrition, certain medications (anti-cancer...
medicines or steroids). Weight and age may also decrease the ability to respond to injury (Beaver, 2008).

**Pre-operative care**

Prolonged pre-operative hospital stay is frequently suggested as a patient characteristic associated with increased SSI risk. However, the length of pre-operative stay is a likely surrogate for severity of illness conditions requiring inpatient work-up and therapy before the surgery.

Manigram *et al.*, (1999) described that, pre-operative transfusion of leukocyte containing allogeneic blood components is an apparent risk factor for the development of post-operative bacterial infection including SSI. However, there is currently no scientific basis for withholding necessary blood transfusion from surgical patients as a means of either incisional or organ / space SSI risk reduction.

**Operative procedure**

The type of surgery, site of surgery and duration of surgery are important factors in the post-operative wound infection. An emergency surgery on traumatic injuries and over 3 hour’s surgery also increases the risks of SSI. It may also include surgeries also done on certain body organs, such as the stomach or intestines (bowels). The risk may be greater if an object pierced through the skin and into an organ. SSI is likely to occur after an open surgery than a laparoscopy surgery. Drains and blood transfusion may increase the chance of bacteria reaching the wound causing infection (Eron, 1999).

Patients involved in an accident, usually some foreign objects, such as glass or metal or dead tissues present in the wound may delay wound healing. It is also possible to have SSI if there is an infection on another part of the body or a skin disease (Donald, 2007).

**Post-operative Care**

When the surgical incision is left open for a few days before it is closed (delayed primary closure), it is likely for the site to be infected or patients condition may prevent primary closure. When a surgical incision is left open to heal by second intention, it is packed with sterile moist gauzes and covered with sterile dressing. It is also recommended that when changing the dressings, it is appropriate to use sterile gloves to reduce the chances of infection.

Nicotine use delays the primary wound healing and also increases the risk of SSI. At the same time patients receiving steroids or other immunosuppressive drugs pre-operatively may be predisposed to developing SSI.
There are several aseptic agents available for pre-operative preparation of the skin at the incision site (Veeraya Paocharoen, 2009). Alcohol is considered to be the most available, inexpensive and the most rapid-acting skin antiseptic. Before the skin is prepared, it should be free of contaminations (*i.e.* soil or dirt). The skin is prepared by applying an antiseptic in concentric circles. The prepared area should be large enough to extend the incision or create new incisions or drain site if necessary.

Mobile phones have been the source of communication within the hospital. According to a recent research by Ulger *et al.*, (2009) hospital operating rooms (OR) and intensive care units (ICU) are the workplaces that need highest standard of hygiene, also the same requirements for the personnel working there and the equipment used by them. They did not do a direct comparison of transmission rate of bacteria from surface to hands. Risk of infection involved in using mobile phones in the OR has not yet been determined as there are no cleaning guidelines available that meet hospital standards. However, mobile phones are used routinely all day long but not cleaned properly as healthcare workers may/do not wash their hands as often as they should. They found out that healthcare workers hands and their mobile phones were contaminated with various types of microorganisms. Mobile phones used by healthcare workers may be the source of nosocomial infections in hospitals.

As known, several factors contribute to the problem of the emergence of MDR bacteria:

1. Device and fomite associated nosocomial infections are frequent in hospital settings everywhere and always (Crnich and Drinka, 2012), with grimy settings.

2. The drug resistance character has ramifications: bacteria may be resistant to representative, frequently used antibiotics of several classes; paradigmatically, the production of extended spectrum β-lactamase has rendered resistance to antibiotics of penicillin/cephalosporin group (Shahcheraghi *et al.*, 2009); carbapenemase production affords resistance in Gram negative bacteria to carbapenems (imipenem, meropenem and ertapenem) in use.

3. Mutation rates in bacteria are faster. One mutant cell in $10^6$ to $10^8$ bacterial cells in the presence of an antibiotic-stress is known to be drug resistant (Gillespie *et al.*, 2005).

4. To avoid host toxicity, antibiotics doses are mostly fixed at some lower concentration that is often below the mutant preventive concentration (Ferrari *et al.*, 2011) giving ways to the development of mutants.
5. Genetic recombination mechanisms (bacterial transformation and conjugation) are operational in natural systems, such as hospital sewage, facilitating the creation of a pool of drug resistant characters in camaraderie MDR strains (Perron et al., 2012).

6. Absence of a stringent antibiotic policy triggers the emergence of resistant bacteria as both clinicians and patients use antibiotics belonging to higher generations without often being warranted.

7. Patients, particularly, often do not complete the course of prescribed dose of an antibiotic, because of the blithesome effect of the control of infection from the start of the course. Thus, bacterial resistance to antibiotics is a consistently complex and dynamic affair, involving major genetic and biochemical mechanisms, bacterial transformations, interchange of integrons, hypermutabilty, plasmids mediated improvements in resistance factors, ending with drug efflux mechanism and gaining of characters to synthesize indigenous and exogenous antibiotic degrading enzymes.

2.4 Clinical features and complications

There are a number of indicators of wound infection; these include the classic signs related to the inflammatory. The classic signs of post-surgical wound infection include localized erythema, localized pain, localized heat, cellulites and oedema. Further signs include abscess, purulent discharge, delayed healing, discoloration of tissues both within and at the wound margins, abnormal smell and wound breakdown associated with wound pocketing/bridging at base of wound (Cutting and Harding, 1994). Common post-operative complications include post-operative fever, atelectasis, wound infection, embolism and deep vein thrombosis. The highest incidence of post-operative complications is between 1 and 3 days after the surgery. However, specific complications occur in the following distinct temporal patterns: early post-operative, several days after the operation, throughout the post-operative period, and in the late post-operative period (Boni et al., 2006; Thompson et al., 2003).

2.5 Antimicrobial susceptibility pattern

Antimicrobial resistance has been a problem in the field of surgery, as advances in control of infections have not completely eradicated the problem of drug resistance (Nwachukwu, 2009). The widespread uses of antibiotics, together with the length of time over which they have been available have led to major problems of resistant pathogens contributing to morbidity, and mortality. The sensitivity pattern of SSI isolates is changing due to increasing emergence of antimicrobial resistant pathogenic bacterial strains like MRSA (Anderson, 2007), making the choice of empirical
treatment more difficult and expensive. The magnitude of antimicrobial resistance among bacteria globally is unknown and more so in developing countries where few data are available.

Matthew et al., (2009) worked on the susceptibility of bacteria and found that susceptibility of bacteria to antimicrobial agents cannot be predicted, testing of individual pathogens against antimicrobial agent is often necessary.

Andhoga et al., (2002) in a study conducted at a University hospital in Kenya among the bacteria isolated from SSI reported that S. aureus were resistant to Ampicillin (78.3%), Chloramphenicol (84.8%), Methicillin (79.4%) and Cotrimoxazole (84.8%). All Pseudomonas sps and Escherichia coli strains were completely resistant to Ampicillin and Cotrimoxazole. Similar observation were also reported in the same region, in Uganda, it has showed that most of the Gram negative bacteria isolated from SSI were highly resistant to first line antibiotics namely Ampicillin (90.6%), Amoxycillin (96.9%) and Chloramphenicol (100%). S.aureus isolates were highly resistant to Ampicillin (97%) and Erythromycin (56.2%), but sensitive to Gentamycin (87.5%), Ciprofloxacin (68.7%) and Methicillin (75%). Pseudomonas sps were sensitive to Gentamycin (87.5%) and Ceftazidine but resistant to Ciprofloxacin as 57.2% (Anguzu, 2007).

Studies conducted in Tanzania have reported variable antimicrobial susceptibility pattern of bacterial pathogens isolated from SSIs (Mawalla et al., 2011). Study done among surgical patients at Muhimbili University of Health and Allied Sciences (Manyahi, 2012; Wayi, 2000) demonstrated that, S. aureus were 100% resistant to commonly used Penicillin G but it has showed 100% sensitive to Methicillin. Klebsiella sps showed susceptibility to Ceftriaxone (93.3%) and Gentamycin (64.7%) but were resistant to Ampicillin (77.8%). But another data from the same study setting revealed that microorganisms isolated from SSIs were frequently resistant to Ampicillin, Chloramphenicol, Tetracycline, Cotrimoxazole and Gentamycin. CoNS were resistant to all antibiotics used in this study (Eriksen, 2003).

Among all bacterial pathogens isolated from SSI at Ifakara hospital, Tanzania, 60% were resistant to antimicrobial agents commonly administered (Fehr et al., 2006). In the same study, 33% and 50% of S. aureus strains isolated from SSI were resistant to Chloramphenicol and Penicillin respectively. Interestingly MRSA were rather uncommon comprising only one of 114 isolate, 40% were fully resistant to the two drugs. However, S. aureus showed increased resistant (60%) to Erythromycin.
The susceptibility testing of the Gram negative organisms showed that; *E. coli*, *Proteus mirabilis* and *P. aeruginosa* were highly resistant to Ampicillin and Ceftriaxone (β-lactam antibiotics) (Pondei *et al.*, 2013). The reported high resistance of organisms to β-lactam is not surprising, as these antibiotics are the most abused drugs in Eastern Nigeria (Amoran, 2013; Sani *et al.*, 2012). Vendors are seen selling them in Motor parks, and Streets without prescription by doctors. Similarly, Fluit and Colleagues (2000) in Europe reported the high resistance of *E. coli, P. aeruginosa* isolated from surgical wounds. Resistance to Aminoglycosides by *P. aeruginosa* isolates was also observed. Out of 16 bacterial isolates studied, 8 (50%) were resistant to Gentamycin, while 7 (43.7%) were resistant to Streptomycin. Ciprofloxacin was highly active against all Gram negative organisms. The susceptibility testing of *Staphylococcus aureus* showed that out of 20 isolates of *S. aureus* tested, 16 (80%) were sensitive to Ciprofloxacin, while 4 (20%) were resistant. Also, 10 (50%) out of the 20 isolates of *S. aureus* were sensitive to Norfloxacin (Moorhouse *et al.*, 1996).

A cross sectional study conducted by Mohammedaman *et al.*, (2014) among patients with wound infection visiting Jimama University Specialized Hospital, South-West Ethiopia reported that 100% of the *E. coli* isolates were resistant to cephaplatin, Ampicillin (96.6%), Tetracycline (79%), Chloramphenicol (65.5%), Ceftriaxone (62%), Sulphamethoxazole trimethoprim (55%) and Gentamycin (51.7%) respectively. *K. pneumoniae* has showed 100% resistance to Ampicillin, 85.7% to Chloramphenicol, Sulphamethoxazole trimethoprim and Cephalothin, (71%) in Ceftriaxone however; it indicates low resistance to Ciprofloxacin (35.7%) and Doxycycline. These were in consistence with the study done in Ethiopia (Fantahun *et al.*, 2009). *Proteus* sps were resistance to Ampicillin (91%), Cephalothin (87%), Tetracycline (73.9%) and Ceftriaxone (65%). The isolates were sensitive to Ciprofloxacin (83%) and Gentamycin (74%). Most of the Gram negative bacteria isolated were resistant to Ampicillin, Cephalothin, Tetracycline and Chloramphenicol. This may be due to the antibiotics having been in use for much longer time and their oral route of administration that affects their rate of absorption into blood stream. Some of them were used as prophylaxis therefore increasing their use in patients (Mulu *et al.*, 2012; Sanjay *et al.*, 2010).

Over use of antibiotics contributes to developing resistance in organisms. *P. aeruginosa* showed reduced sensitivity to commonly used antibiotics like Ampicillin, doxycycline, nalidixic acid and tetracycline, except Ciprofloxacin, Norfloxacin (100%),
and Gentamycin (82%). Ciprofloxacin and Norfloxacin have been stated to be the most potent oral drug available for the treatment of *P. aeruginosa* infections. This report is in agreement with the result of other study in which Ciprofloxacin recorded the least resistance (6.2 to 24%) to *P. aeruginosa* isolates from wound infection; it is undoubtable that at the present time, the oral drug Ciprofloxacin and injection Gentamycin are the most effective antibiotics against *P. aeruginosa* involved in wound infection relative to most other commonly used drugs. *Pseudomonas* spp resistant to third generation Cephalosporins (Ceftriaxone 63.6%) is real treat. In fact, the ridiculous and improper use of antibiotics is responsible for the development of resistance of *Pseudomonas* spp to antibiotic monotherapy. The incidence of *P. aeruginosa* in wound infection among admitted patient is becoming more serious in developing countries because of lack of general hygienic conditions, production of low quality antiseptics and medicinal solutions for treatment (Agwunglefah *et al.*, 2014).

The study conducted by Sepehri *et al.* (2004) in Iran on antibiotic susceptibility of isolates that cause SSI showed that the prevalence of different bacterial types were 51.8, 12.7, 10, 8.2, 6.4, 6.4 and 6.4% for *Staphylococcus aureus*, β-hemolytic *Streptococcus* spp, *Pseudomonas* spp, *Klebsiella* spp, *E. coli*, CoNS and *Enterobacter* spp respectively. The overall antibiotic resistance of these isolates to penicillin G was 94%; in case of Amoxicillin it has showed 83.3% and 88.1% for Ampicillin.

The study conducted by Alireza Sharif (2014) at Kashan University of Medical Sciences, Kashan, Iran, reported that the overall bacterial susceptibility of the 98 isolates, only 8 (8.1%) of them were sensitive to penicillin G, 6 (6.1%) were intermediate and 84 (85.8%) were resistant. There results regarding Amoxicillin were 18 (18.3%), 22 (22.4%) and 58 (59.3%) for sensitive, intermediate and resistant species respectively. Antibiotic susceptibility pattern of Ampicillin was 14 (14.2%), 23 (23.4%) and 61 (62.4%) for sensitive, intermediate and resistant species respectively (Mohammad *et al.*, 2013).

The study conducted to examine Antibiograms of a group of suppurative bacteria isolated from wound swabs from hospitalized patients of all economic groups of a typical Indian teaching hospital showed that, out of 819 bacterial isolates, there were 52 strains of *E. faecalis*, 251 strains of *S. aureus*, 210 strains of *S. pyogenes*, 48 strains of *A. baumannii*, 39 strains of *E. aerogenes*, 62 strains of *E. coli*, 53 strains of *K. pneumoniae*, 24 strains of *P. mirabilis*, 21 strains of *P. vulgaris* and 59 strains of *P. aeruginosa*. Thus, *S. aureus* were the maximally isolated suppurative infection causing
bacterium, followed by *S. pyogenes, E. coli, P. aeruginosa, K. pneumoniae, E. faecalis, A. baumannii, E. aerogenes, P. mirabilis* and *P. vulgaris* (Arun et al., 2013; Nabakishore et al., 2014).

*S. aureus* were the most common isolate of which 52% isolates were MRSA; all sensitive to Vancomycin and linezolid followed by Chloramphenicol, Gentamycin, Ciprofloxacin (Huda, 2005; Mehata et al., 2013). *Enterococcus* sps were 100% sensitive to Vancomycin and Linezolid. Piperacillin-Tazobactum, Ceftazidime, Tobramycin and Gentamycin are the common antimicrobials used for surgical prophylaxis and also for empirical therapy of SSIs. Gram negative bacilli isolated in the study were highly sensitive to these antibiotics. ESBL producers included *Klebsiella* sps (50%); *E. coli* (20%), and *Pseudomonas* sps (30%). *Pseudomonas* sps were mostly sensitive to piperacillin-Tazobactum, Meropenem, Imipenem and Amikacin. Most of the Gram negative bacilli were resistant to Cefamandole, Cefixime and Cotrimoxazole.

*S. aureus* isolates showed (Lopiso et al., 2014; Gautam et al., 2013; Bibi et al., 2012) the highest resistance to Penicillin (100%), Ampicillin (95.5%) and Ceftriaxone (81.8%) while their resistance rate to Amoxicillin-clavulenic acid as showed 30.3%. Isolates of CoNS showed 100% resistance to Vancomycin, Ceftriaxone, Ampicillin and Penicillin; but sensitive to Chloramphenicol. All isolates of *E. coli, Proteus* sps and *P. aeruginosa* were resistant to Ampicillin. Also, no isolates of *Klebsiella* sps and *P. aeruginosa* were sensitive to Gentamycin and Chloramphenicol, respectively. While, *Proteus* sps and *P. aeruginosa* isolates were susceptible to Ciprofloxacin. Single and multiple antimicrobial resistances were observed in 6.8% and 93.2 % of the isolates, respectively. No bacterial isolates were found to be sensitive to all antibiotics tested, and three isolates of *S. aureus* (4.5%) were shown to be resistant to all antibiotics tested.

The study of Post-operative wound infections and their antimicrobial susceptibility pattern in a tertiary care hospital in Salem, by Lalithambigai et al., (2014) showed that the Gram positive cocci were 100% sensitive to Vancomycin, Teicoplanin and Linezolid (Verma et al., 2012). *Staphylococcus aureus* showed resistant to Penicillin, Cefazolin (72.73%) followed by Cefoxitin (55.56%). The Gram negative bacilli showed 100% sensitivity to Imipenem, Pipercillin-azobactum whereas *Pseudomonas aeruginosa* showed 85.71% sensitivity to Pipercillin-Tazobactum. All Gram negative isolates showed more than 85% sensitivity to Levofloxacin, Amikacin and more than 60% sensitivity to Amoxicillin-clavulenic acid, Cefotaxime, Cefepime.
A majority of the isolates showed more than 70% resistant to Ampicillin and cefazolin (Goswami et al., 2011).

### 2.6 Antimicrobial resistance

Before the discovery of antibiotics in the 1928, death was often an inevitable outcome of infection. Heralded by the discovery sulphonamides in the mid-1930s the introduction of penicillin in 1939 was considered a miracle in the battle against the life threatening infections. Penicillin the first $\beta$-lactam employed to treat infectious diseases, inhibited bacteria that were resistant to the sulfonamides and produced fewer side effects. It was unique in its ability to penetrate into dying tissues yet retaining its activity. Uncountable lives were therefore saved from the infectious diseases, especially in World War II. Unfortunately the clinical value of penicillin was short lived. The first incidence of antibiotic resistance to penicillin soon brought novel challenges in the treatment of infection. Although the development of new antibiotics has occurred at an extraordinary pace in recent years, it was paralleled by the appearance of resistance to antibiotics (Leung-Kei, 2002).

There are number of reasons why bacterial resistance should be a concern for physicians. First resistant bacteria, particularly *Staphylococci*, *Enterococci*, *Klebsiella* and *Pseudomonas* spp are becoming common place health care instructions (Jones, 2002; Edmend, 1999). Bacterial resistance often results in treatment failure, which can have serious consequences, especially in critically ill patients. Inadequate empiric antibacterial therapy, defined as the initial use of an antibacterial agent to which the causative pathogen was not susceptible, has been associated with increased mortality rates in patients with infections due to *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Enterobacter* spp, coagulase negative *Staphylococci* and *Enterococci*. Prolonged therapy with antimicrobial agents such as vancomycin or linezolid, may also lead to the development of low level resistance that compromises therapy, but that may not be detected by routine susceptibility testing methods used in hospital laboratories (Tenover, 2004).

Resistant bacteria may also spread and become broader infection control problem, not only within healthcare institutions, but in communities as well. Clinically important bacteria such as Methicillin resistant *Staphylococcus aureus* (MRSA) and extended spectrum $\beta$-lactamase (ESBL) producing *E.coli* are increasingly observed in the community. Infected individuals, including children, often lack identifiable risk factors for MRSA, and appear to have acquired their infections in a variety of
community settings. Community associated MRSA strains are typically less resistant to antimicrobial agents than health care associated MRSA, but are more likely to produce toxins (Francis, 2005). The spread of resistant bacteria within the community poses obvious additional problems for infection control, not just in long term care facilities but also among sport teams, military recruits, and even children attending day care centers, a task that is complicated by the increased morbidity of our population. Finally with respect to the cost containment pressures of today’s health care environment, antibacterial drug resistance places an added burden on healthcare costs, although its full economic impact remains to be determined (McGowan, 2001).

Since the first introduction of Sulfa drugs and Penicillin into clinical use, large number of antibiotics have been developed and used for treating various infections in human. But extensive use of antimicrobials has raised a serious public health problem due to multi-drug resistant bacterial pathogens that immediately develops resistance to every new drug used in clinic. Antibiotics, literally “Against Life” are chemical compounds produced by microorganisms that kills or inhibits the other microorganisms. Antibiotics exerts two effects on bacteria i.e. either bacteriostatic or bactericidal (Hiroshi, 2006).

Antibiotics resistance among bacteria is a worldwide problem. The situation in developing countries like Ethiopia is particularly serious. Since the presence of drug resistant bacteria in the environment is threat to the public, up-to-date information on local pathogens and drug sensitivity pattern is very crucial to manage patients (Mulu et al., 2006).

Resistant organisms pose a great challenge in the treatment of bacterial infections often leading to treatment failure, prolonged duration of illness and great risk of death. Bacteria have ability of undergoing mutation or acquiring a resistance gene when antimicrobial agents are inappropriately used (Davis, 1994; Cohen, 1992). There has been an increase in the number of multidrug resistant organisms isolated from patients in hospitals worldwide. Infection with antibiotic resistant bacteria also increases the likelihood that the patients will receive inadequate therapy.

A case control study in Australia, reported hospitalization within the preceding six months and residence in long care facility as being associated with higher risk of MRSA bacteremia (HO, 2009). In a retrospective study at a university hospital in Malaysia, duration of hospitalization, previous antibiotic use, and bedside invasive
procedures were significantly associated with MRSA than MSSA (Al-Talib et al., 2010).

Prior exposure to antibiotics as a risk factor for emergence of drug resistant bacterial strains has also been reported in studies done in Thailand (Apisaranthanarak, 2007). Use of ventilator and catheter as well as days of stay in hospital wards has also been significantly associated with acquisition of antibiotic resistant isolates (Lyamuya et al., 2011; Mansouri et al., 2011). Multivariate analysis of data from a study done in Madagascar reported that diabetes and use of an invasive procedure were independent risk factors for resistance to third-generation cephalosporins among ESBL-producing Enterobacter isolates from surgical wards and intensive care unit (Randrianirina et al., 2011).

In a case control study conducted in Denmark identifying possible risk factors for MRSA and Methicillin susceptible Staphylococcus aureus (MSSA), prior hospitalization for more than 7 days within the previous six months tended to be associated with MRSA (Bocher, 2008). Furthermore a study done in USA, antimicrobial use 1-6 months to culture, history of boil and having a household member who was a smoker were associated with MRSA compared to MSSA (Como-Sabetti et al., 2010).

Most of antimicrobial resistance is mainly due to the extensive use and misuse of antimicrobial drugs, result in emergence and survival of resistant strains. Bacteria become resistant to antimicrobial agents by a number of mechanisms, the commonest being: production of enzymes which inactivate or modify antibiotics, changes in the bacterial cell membrane, preventing the uptake of an antimicrobial, modification of the target so that it no longer interacts with the antimicrobial and development of altered metabolic pathways by bacteria (Cheesbrough, 2006). Resistance in antimicrobial drugs in bacteria can result from two mutually non exclusive phenomenons: mutations in structural or regulatory genes and the horizontal acquisition of foreign genetic information and the rapid spread of antimicrobial resistance genes on mobile genetic elements such as plasmids and transposons (Courvalin and Trieu-Cuot, 2001).

Antibiotics are most frequently prescribed for acute respiratory tract infections, acute watery diarrhoea, acute trauma and gastrointestinal symptoms (Rashid et al., 2007). The most frequently prescribed antibiotics are ceftriaxone (30.19%) followed by Cefixime (18.87%), and Amoxycillin (16.98%). It has reported that cephalosporins accounted for more than 55% of the total antibiotics used, where the highest uses were
of Ceftriaxone, Cefixime, and Cefuroxime. This probably explains why ceftriaxone and Cefixime have abnormally high resistance. A study suggested that *Pseudomonas aeruginosa* responsible for wound, urine, ear, throat and other infections were more than 50% resistant to commonly used antibiotics in Bangladesh (Paul, 2004), including Ciprofloxacin, Gentamycin, Ceftriaxone, Cefixime and Azithromycin. Azithromycin was 100% ineffective in wound and urine infections, while Ceftriaxone and Cefixime were 100% ineffective in tracheal infections. Another study also reports that *Escherichia coli* were resistant in 40% of cases to commonly used antibiotics Ceftriaxone, Levofloxacin, Ciprofloxacin, Amoxicillin and Ampicillin and 95% resistant to Azithromycin. *Klebsiella pneumoniae* also showed similar patterns (Lina *et al*., 2007). It has reported that 43.2% and 39.5% of isolated *E. coli* and *K. pneumoniae* respectively had ESBL phenotypes. This rate is higher than in countries of the Western Pacific Region, North America or Europe and some South American nations (Darouiche *et al*., 2004). *Enterobacter* isolates which are resistant to expanded spectrum Cephlosporin is becoming a matter of concern for the probability of transmitting antimicrobial resistance from one microorganism to another worldwide (Gebre-Selassie, 2007).

The study of pattern of antimicrobial susceptibility in Ayder Teaching and Referral Hospital, Mekelle, Ethiopia (Zeamanuel *et al*., 2009) reported that Drug resistance of isolated Gram negative bacteria, irrespective of species/genus, were 92.3% to Ampicillin, 92.3% to Tetracycline, 92.3% to Amoxicillin, 81.5% to Ceftriazone, 69.2% to Amoxicillin-Clavulanic acid, 46.2% for Ciprofloxacin, 26.2% to Erythromycin and 16.9% for Gentamycin respectively. *Klebsiella* sps showed 100%, 93.1%, 89.7% and 86.2% resistance for Amoxicillin, Tetracycline and Ceftriazone, respectively. *P. aeruginosa* isolates were 100% resistant for Ceftriazone, Amoxicillin, Amoxicillin-clavulanic acid and Tetracycline. All *P. aeruginosa* isolates were; however, 100% sensitive to Gentamycin. All *Proteus* sps were resistant to Amoxicillin and Tetracycline whereas, 80% were sensitive to Gentamycin. Isolated *E.coli* showed 100% resistance to Amoxicillin-clavulanic acid, Tetracycline and Ampicillin, whereas, all of them were sensitive for Gentamycin. Isolated Citrobacter species were 100% sensitive to Gentamycin, while all (100%) of them were resistant to Ampicillin. *S.aureus* isolates have showed resistance of 90% to Tetracycline, Ceftriazone and Ampicillin, and in case of Cloxacillin antibiotic it has showed 85% resistance. All of the isolates *S.aureus* (100%) were sensitive for Vancomycin. High resistance rate of CoNS were observed
for Amoxicillin, Amoxicillin-clavulanic acid, Ampicillin and Tetracycline, 88.9%, 77.8%, 77.8% and 77.8%, respectively. All isolates of CoNS (100%) were however sensitive for Vancomycin (Mengesha et al., 2014; Godebo et al., 2013).

A study conducted from November 2010 to June 2011 in Hawassa University Referral Teaching Hospital showed that, of the 177 isolates, 173 (97.7%) were resistant to at least 1 antimicrobial, while 164 (92.7%) were resistant to ≥ 2 antimicrobials. The isolates showed resistance as 76.3% to Amoxicillin, 71.2% to Penicillin, 56.9% to Vancomycin, 39.5% to Ceftriaxone, 39.5% to Norfloxacin and 31.1% to Gentamycin. The *S. aureus* as showed susceptibility of 64.4% for Gentamycin antibiotic but it was 100% resistant to Amoxicillin. All isolates of *P. aeruginosa* were resistant to Penicillin and Amoxicillin. The rate of resistance of *S. aureus* to 2 or more antimicrobials was 97.8% and that of *P. aeruginosa* was 100%. This study confirms that the bacteria commonly implicated in post-operative wound infections: *S. aureus*, *Klebsiella*, *E. coli*, CoNS, and *P. aeruginosa* continued to dominate and have developed high level of drug resistance to some important antibiotics. Periodic surveillance of the species of bacteria involved in post-operative wound infection and determination of their antimicrobial resistance is recommended for empirical treatment (Guta et al., 2014).

### 2.7 Treatment and prophylaxis

At any given time, about 25 to 35 percent of hospital patients are under antibiotic treatment for active infections or to prevent potential infections. The large amount of antibiotic use exerts enormous selective pressure for emergence and spread of antibiotic resistant bacteria. Therefore, untreatable bacteria such as stains of VRE and hard to treat bacteria are much more common in hospitals than community at large.

The reporting by the microbiology laboratory of specific microorganisms isolated from a wound and the associated antibiograms may be interpreted by the practitioner as a diagnosis of wound infection that requires antimicrobial treatment (Burke, 2003; Bowler et al., 2001). Although the primary purpose of antibiotics is to treat infection, prophylaxis associated with surgical practice accounts for up to half of all antibiotics prescribed (Periti et al., 1998). The choice of prophylactic antibiotics should cover both facultative and anaerobic intestinal bacteria. The aim is to achieve high concentrations at the time of surgery and throughout the surgical procedure. Particularly in contaminated surgery, where excessive populations of Gram negative bacteria are likely to be present, careful selection of antibiotics is required since some are known to influence endotoxin liberation and hence septic shock (Periti et al., 1998).
Antimicrobials may be used to prevent or treat established surgical infection. Antibiotics do not replace surgical drainage of infection. The use of antibiotics for the treatment of established surgical infection ideally requires recognition and determination of sensitivities of the causative organisms. Antibiotic therapy should not be held back if they are indicated, the choice being empirical and may later be modified depending on microbiological findings. However, once antibiotics have been administered, the clinical picture may become confused and, if a patient’s condition does not rapidly improve, the opportunity to make a precise diagnosis may have been lost. It is unusual to have to treat SSIs with antibiotics, unless there is evidence of spreading infection, bacteraemia or systemic complications. The appropriate treatment of localized SSI is interventional radiological drainage of pus or open drainage and debridement (Williams et al., 2008).

**Approaches to Antimicrobial Therapy**

1. **A narrow-spectrum antibiotic**: These may be used to treat a known sensitive infection; for example, vancomycin for the treatment of MRSA.

2. **Combination of broad spectrum antibiotics**: These can be used when the organism is not known or when it is suspected that several bacteria, acting in synergy, may be responsible for the infection. For example during and after emergency surgery requiring opening of perforated or ischaemic bowel, any of the gut organisms may be responsible for subsequent peritoneal or bacteraemic infection. In this case, a triple therapy combination of broad spectrum penicillin, an aminoglycoside and Metronidazole, may be used pre and post-operatively to support the patient’s own body defense (Williams et al., 2008).

**Judicious use of antibiotics**

Use of antibiotics should be judicious. These should be used only when there is evidence of clinical infection or as part of a policy regarding pre-operative prophylaxis. Choice of antibiotic should be rational. If in doubt, consultation with the microbiologist earlier is better than later. Overuse of antibiotics encourages development of resistance in exposed organisms. It also destroys patient’s normal flora so they are more susceptible to colonization with hospital organisms. Furthermore, it predisposes to infection with *Clostridium difficile*, which can lead to pseudomembranous colitis; third-generation cephalosporins are notorious for this (Kirk and Ribbons, 2004). It has been shown that, for many contaminated and clean-contaminated procedures, post-operative...
infection can be avoided by using appropriate prophylactic antibiotics given prior to surgery (Kirk and Ribbans, 2004).

**The general principles of antibiotic prophylaxis:**

1. Antibiotic prophylaxis should be used only when wound contamination is expected or when operations on a contaminated site may lead to bacteraemia. It is not required for clean-wound procedures except when an implant or vascular graft has been inserted, in valvular heart disease to prevent infective endocarditis, during emergency surgery in a patient with pre-existing or recently active infection, if an infection would be very severe or have life threatening consequences (Tiberi *et al*., 2010; Kirk and Ribbans, 2004).

2. There is no evidence that prolonged prophylaxis has any advantage over short course of 24 hours. Prolonged administration may lead to super-infection. Normally in a clean operation one dose is sufficient. In contaminated operations three doses are often given (Kirk and Ribbans, 2004).

3. Antibiotic should be administered parenterally, immediately prior to operation to achieve effective tissue levels. If they are given soon afterwards they do not prevent infection. If the procedure continues for more than 3-4 hours, or if there is excessive blood loss, a further dose should be given in theatre (Kirk and Ribbans, 2004).

4. Antibiotics should be selected to cover relevant organisms after discussion with the microbiologist regarding likely contaminants and local resistance patterns. Working together with the microbiologist to develop standard policies for the unit is very important, and they should be followed strictly when they are in place (Kirk and Ribbans, 2004). The use of the newer, broad-spectrum antibiotics for prophylaxis should be avoided (Williams *et al*., 2008).

**The Decisive Period**

If antibiotics are given empirically, they should be used when local wound defenses are not established (the decisive period). Ideally, maximal blood and tissue levels should be present at the time of making the first incision (Williams *et al*., 2008).

Flucloxacillin is the best choice prophylactic antibiotic for the control of staphylococcal infection in orthopedic surgery (Kirk and Ribbans, 2004). Lower limb amputation should be covered against *C. perfringens* using 1.2 g of Benzyl Penicillin intravenously at induction of anaesthesia and 6-hourly thereafter for 48 hours (Williams *et al*., 2008). Single doses of broad-spectrum Penicillin, for example Amoxicillin, orally
or intravenously administered, are sufficient for dental surgery (Williams et al., 2008). A second-generation Cephalosporin, such as Cefuroxime is sufficient.

Reduction of infection rate is not possible by concentrating attention in a single area. Control of resistant organisms in all areas within the hospital is mandatory. Aseptic and Antiseptic principles should be obeyed strictly. The highest standard of surgical technique should be practiced. Use of prophylactic antibiotics should be logical. Audit of the results will play a major role to maintain and improve standards (Williams et al., 2008).

Antibiotics have been in used for more than 50 years and many organisms are now resistant to the older agents. For example, in many hospitals more than 50% of isolates of *Escherichia coli* are resistant to Ampicillin. The most obvious example is Methicillin Resistant *Staphylococcus aureus* (MRSA). It is also resistant to flucloxacillin and has to be treated with drugs such as the glycopeptides, Vancomycin and Teicoplanin. Even more worrying is the reported emergence of Vancomycin intermediate *S. aureus* with reduced susceptibility to Vancomycin. It will be dangerous in future, if it become impossible to treat *S. aureus* infection. *Enterococci* are also posing major problems with resistance; glycopeptide-resistant *Enterococci* are now found in many hospitals and they may cause life-threatening infections in immunocompromised patients. Gram negative organisms such as *Pseudomonas aeruginosa* may also be multi-resistant. The increasing use of third-generation Cephalosporins appears to be encouraging the emergence of Gram negative bacilli such as *Klebsiella pneumoniae* and *Enterobacter cloacae* resistant to these and other β-lactams (Jadhav et al., 2012).

### 2.8 Correlation of plasmid with drug resistance

According to the infectious diseases society of America in the January 2009 highlighted the impact of the ESKAPE pathogens (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Enterococcus species*) as group of particularly troublesome bacteria having the ability to escape the effect of current antimicrobial agents (Boucher et al., 2008). Drug resistance property in bacteria is usually born in R-plasmids, which can be disseminated to diverse population and regions causing worldwide problems. R-plasmids from resistant strains may transfer to a sensitive counterpart, which can show the same drug resistance in the donor Strain.
A high rate of spread of resistant gene has been suspected as the cause of increased antibiotic resistance cases in it. Plasmids carry genes that could be spread by replication. Intrinsic and acquired antibiotic resistance makes *P. aeruginosa* one of the most difficult organisms to treat. The high intrinsic antibiotic resistance of *P. aeruginosa* is due to several mechanisms: low outer membrane permeability, the production of an AmpC-lactamase and the presence of numerous genes coding for different multidrug resistance efflux pumps (Akingbade *et al*., 2006).

New stains of *E. coli* and other related bacteria evolve through natural biological process of mutation. They possess ability to transfer DNA via transformation, conjugation and transduction, which allow genetic material to spread horizontally through an existing population (Brussow *et al*., 2004). Normal intestinal flora is a reservoir of resistant genes, the prevalence of resistance in commensal *E. coli* is useful indicator antibiotic resistance in bacteria in community. Oral administration of antibiotics can also influence the normal intestinal micro flora and can lead to an overgrowth of resistant. The correlation between antibiotic resistance and plasmid profile indicate that the genetic information is plasmid borne (Myaing *et al*., 2005).

### 2.9 Cost of Antibiotic resistant bacterial diseases

Over 27 million patients undergo surgery in the United States annually. Despite the use of prophylactic systemic antibiotics and other routine interventions, post-operative surgical site infection (also called surgical wound infection) is a significant complication. It is estimated that over 7, 00,000 surgical site infections occur annually in the US with these infections accounting for $10 billion in costs annually.

Itobi *et al*., (2006) have described that post-operative wound infection delays recovery and often increase length of stay and may produce long lasting sequel. It requires extra resources for investigations, management and nursing care (Wood and Haber, 1994).

Occupational therapy assistant office of technology assessment (OTA) estimates the in-hospital costs of hospital-acquired (nosocomial) infections caused by six common kinds of antibiotic resistant bacteria to be a minimum of $1.3 billion. The estimate ignores the costs of infections caused by other kinds of antibiotic resistant bacteria, costs of lost work days and costs for post-hospital care. If these factors were considered, the total cost to society would be at least several billion dollars per year. Further, these costs can be expected to increase rapidly as the numbers of antibiotic
resistant bacteria increase. Antibiotic-resistant bacteria are found all over the world and are spread among countries as people and goods are transported internationally.

Hospital-acquired infections aid to functional disability and emotional stress of the patient and in some cases, lead to disabling conditions that reduce the quality of life. Nosocomial infections are also one of the leading causes of death. The economic costs are considerable. The increased length of stay for infected patients is the greatest contributor to cost. One study showed that the overall increase in the duration of hospitalization for patients with surgical wound infections has required 8.2 days, ranging from 3 days for gynecology to 9.9 for general surgery and 19.8 for orthopedic surgery. Prolonged stay not only increases direct costs to patients or payers but also indirect costs due to lost work. The increased use of drugs, the need for isolation, and the use of additional laboratory and other diagnostic studies also contribute to costs. Hospital-acquired infections add to the imbalance between resource allocation for primary and secondary health care by diverting scarce funds to the management of potentially preventable conditions.

Because of the costs involved in controlling and monitoring the spread of antibiotic-resistant bacteria, it would be useful to know how much would be saved by reducing the impacts of antibiotic-resistant bacteria. Calculation of the costs imposed by antibiotic-resistant bacteria can include such factors as the direct cost of time in a hospital, the costs of extra physicians’ visits when antibiotics are ineffective, the extra hospitalizations due to community-acquired resistant infections, and the costs of newer antibiotics to replace antibiotics to which bacteria have become resistant. Other costs include lost workdays and deaths, if they occur. Only one such study has been published, and it included the estimate that the cost of antibiotic-resistant bacteria nationwide was between $100 million and $30 billion annually, with different values attached to the cost of a life accounting for most of the wide range of the estimate. A medical society subsequently estimated the costs of such diseases at $4 billion.