

AIM OF WORK

INTRODUCTION

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The development of infection in surgical wounds continues to be one of the most serious complications that can occur in surgical patients (Altemeier, et al., 1976).

Many surgeons and bacteriologists has anticipated a greatly decreased incidence of postoperative wound infection after the introduction and general use of prophylactic therapy.

It should be remembered that the possibilities for in-

cisional wound sepsis in modern general hospital practice are numerous and ever threatening. In addition to the reservoir of the virulent and antibiotic resistant bacteria and the potential cross contamination and cross-infection between patients and the hospital's personnel. There are other factors contributing to this problem such as the condensation of the patients with a large variant of infections who have been admitted along with many other patients who are particularly prone to develop sepsis because of their unusual susceptibility (Altemeier , 1972).

Many Factors have been described as being important in reducing wound infection rates during major surgery. These include the use of prophylactic antibiotics, preoperative nutritional support, topical skin antiseptic agents used for surgical hand scrub and patient preparation (Moylan et al., 1987).

The present work was done with the aim at measuring the management of this problem.

AIM OF WORK

The present work is planned to give informations on the incidence of postoperative wound sepsis, the type of organisms encountered in the wound and their antibiotic sensivities in various surgical departments in Benha University hospital, and to trace the possible sources of infection.

The study also aims at evaluation of the control measures and to suggest the appropriate effective control program.

**REVIEW
OF LITERATURE**

REVIEW OF LITERATURE

Wounds are breaks in continuity of the soft parts of the body structures . Surgical incised wounds are clean wounds with minimal tissue damage sustained as the result of cut with knives (Rains and Ritchie , 1981) .

Infection is the invasion of the body by pathogenic organisms. For this to happen , a sufficient number of pathogens must enter the tissues , overcome the patient resistance and multiply . Sepsis is the term which describes the potential and real effect of infection , a process of decomposition (literally rotting) , putrefaction and poisoning which if unchecked can lead to fatal outcome (Gillespie , 1984) .

In primary surgical wound infection , the discharge of pus from a closed wound occurs when there is an imbalance between the number and virulence of bacteria contaminating the wound and efficiency of the patient defences .

The bacteria that contaminate surgical wounds during an operation originate either from exogenous sources (including the skin of the patient) or , more often , from endogenous sources (the opening of potentially contaminated hollow viscera) .

Whatever their origin , no infection can occur in dry closed surgical wound unless bacteria are deposited in that wound during the operation (Pollock , 1987) . There is no doubt that surgical infection is a serious complication of surgery .

Before 1850 , there was general ignorance and confusion about the causes of the terrifying picture of infection as a complication of wounds (Altemeier , 1982) .

Postoperative or post-traumatic putrifactive discharge accompanied by inflammation were regarded as inevitable after operation. Mortality due to deep or extensive wounds approached 70 to 80 percent . Pasteur developed the germ concept of infection between 1853 - 1867 . and the dreaded complications of infection that occurred to wounds thereafter . received new significance .

Lister repeated Pasteur's . experiment on fermentation to convince himself that wound putrefaction and other changes were caused by microbes. Lister searched successfully for an antimicrobial chemical that would inhibit or kill bacteria finding its way into the wounds .

The principle of asepsis was established and practiced by Lister between 1867 - 1875 .The introduction of antiseptics has been considered to be one of the great milestones of surgery .

During the years of 1880 - 1910 , pathogenic bacteria were being discovered , and it seemed to many that all surgical diseases were to be explained by this new science .Surgeons and bacteriologists reasoned that if bacteria caused infection in body tissue or wounds , all they had to do was killing the bacteria and wound infection would be a thing of the past .

The discovery and introduction of the modern antibiotic therapy during the second quarter of the twentieth century had a revolutionary effect on the treatment of many established infections , and had a tremendous impact on the practice of surgery (Altemeier ,1980) .

Antibiotic therapy has been used now for almost 40 years.Clinical and laboratory studies indicate that it has failed to reduce the overall incidence of infection in surgical practice (Altemeier , 1982) .

CLASSIFICATION OF SURGICAL WOUND INFECTION

Classification of infection is difficult but important. Since it aids in facilitating the search for the origin and causes of infection, permits earlier presumptive diagnosis before bacteriologic results are available, indicates earlier and more effective methods of treatment and provides a plan for collection of data related to the nature and control of infection (Cole, 1976).

Kaul and Jewett (1981) reported that operative wounds can be classified according to the clinical estimate of the degree of wound contamination at the time of the operation, and the subsequent contamination into four categories:

1 - Clean wounds :

In which : - no inflammation is encountered.

- no hollow viscus is entered

(i.e. respiratory ,gastrointestinal , genitourinary, tract are not opened) .

- no break in aseptic technique.

Examples for clean operations are herniorrhaphy, thyroidectomy ,mastectomy ,and removal of varicose veins. Clean wounds are elective ,primary closed , undrained wounds and usually heal with low incidence of

complications .

2- Clean - Contaminated wounds :

In which : - the hollow viscous were opened , but minimal manipulation of contents occurred or ,

- minor break in aseptic technique occurred or ,

- wounds are mechanically drained .

Examples of these operations are appendicectomy (non - perforated), operations on biliary tract in absence of infected bile , and operations on genitourinary tract in absence of infected urine .

Most problems with wounds occur in the clean contaminated group , because surgeon may fail to recognize the contamination and the potential risk.

3 - Contaminated wounds :

In which :- Hollow viscous was opened , with gross spillage .

- or , acute inflammation without pus formation was encountered .

- or , fresh traumatic wounds , less than 4 hours :

- or operation in which a major break in aseptic technique occur (e .g. open cardiac massage) .

Examples are operations in which entrance of biliary tract, genitourinary tract occurred in presence of infected bile or infected urine.

4- Dirty wounds :

In which : - perforated viscus was found

- or, pus is encountered.

- or, a traumatic wound untreated for not less than 4 hours.

Example is operation for purulent peritonitis. In dirty wounds, infection and other complications are expected to develop and steps are usually taken to provide adequate drainage and minimise risk of invasive infection.

The precise definition of this classification suggested that the organisms causing postoperative infection are present in the operative field before operation. Cruse and Foord, (1980), reported that surgical staff readily agreed to this classification and it is widely now accepted as a standard classification of operative wounds. Ronald, (1983), reported that the incidence of infection in clean operations was 1.5%, in clean-contaminated 7.7% and about 15.2% or more in contaminated and dirty operation.

FACTORS AFFECTING WOUND SEPSIS .

The first attempt to apply rigid statistical analysis to postoperative wound infections was made by Davidson et al., (1971) ,they found that , bacteria in the wound , dirty procedures , age of the patient , duration of surgery and the nature of the postoperative ward were independently significant in the development of wound sepsis .

Subsequent study by Cruse and Foord, (1973), confirmed that contamination status , age, duration of surgery , use of drains through the wound , and the length of preoperative hospital stay were important determinants of a wound infection . Additional risk factors associated with the development of wound infections include age , duration of surgery , duration of preoprative hospitalization , malnutrition , obesity , diabetes , immunologic dysfunction , and diminished local oxygen delivery to the wound either due to local damage or cardiopulmonary abnormalities (Hunt, 1981).

**** Factors affecting wound infection :-**

1 - Age :-

Wound healing is very fast in the young , but is normal in old age unless there is some associated debilitating diseases or ischaemia (Forrester , 1978).

Lewis et al., (1987),reported that advanced age is a true risk factor. The age of the patient was found to be inversely associated with the development of an operative site infection (Christou et al., 1987).

2 - Medical conditions :

Dineen ,(1969), noted an increase in wound infection with liver diseases . Other diseases such as malignant diseases , cardiovascular diseases ,also hypogammaglobulinemia , **deficient** cellular immunity are highly susceptible to infection,(Irving and Kathryn , 1972). Diabetes mellitus and uraemia are associated with impaired healing of unpredicated degree.

In diabetes ,the main problem is **attributed** upset carbohydrate metabolism in the poor tissue perfusion and increased susceptibility to infection .

Uraemia retards connective tissue formation and slows epithelial repair . (Forrester , 1978) .

3- OBESITY :

An infection rate of 1.8 percent was found in clean wounds . This number increased to 13.5 percent for obese patients (Cruse , and Foord , 1978) .

It is generally accepted that obesity is a risk factor for wound infection . One contributing physical factor may be an increase subcutaneous dead space (Kozol et al., 1986) .

4 - Remote infections :

Edward , (1976) , emphasized the importance of remote infection as a risk factor for the development of wound infections .

Among 1,865 patients with wound infections, he recorded a 61 percent incidence of associated remote infections with the most frequent sites of involvement , the urinary tract (30 %) , lower respiratory tract (25 %) , gastrointestinal tract (11%), and the blood stream (9 %) . Analysis of 383 patients from whom the same organisms were isolated from both the remote site of infection and the wound , revealed that 55 % of these pathogens were isolated

from the remote site of infection prior to the time they were cultured from the wound .

5 - SHAVING :

Although preoperative shaving was not associated with all the infections , it seemed to be a risk factor. Hair removal , especially by shaving , can injure the skin and such injury may increase the risk of infection by promoting skin colonization with bacteria (McCray et al . , 1986) .

Mead et al. , (1986) , in their studies were unable to show that timing of operative site shaving made a significant difference in clean wound infection rates . Interestingly , patients shaved in operating room had a trend toward higher clean wound infection rates (1.9%) than those shaved at the night before surgery (1.4 %) , they like others did find the lowest clean wound infection rates in patients who were not shaved at all (1.2 %) .

6 - EMERGENCY :

Dineen , (1961) , noted an increased wound infection rate in patients undergoing emergency and second operations . Farnell et al. , (1986) , reported that emergency operations were associated with a

significantly higher incidence of infection , it was 7.9 % in patient with emergency procedure and 4.3 % in patients who did not have emergency procedures .

Severely traumatised wound impairs antibacterial defence mechanisms , and infectious organisms can multiply in this wound (Ryan , 1976) .

7 - HOSPITALIZATION :

Cruse and Foord , (1973) , found a relationship between the duration of preoperative hospitalization and the development of wound infection. The increased wound infection rate associated with prolonged preoperative hospitalization may reflect colonization of the patient by more virulent , antibiotic-resistant hospital strains of bacteria . At the same time , hospital population consists of increasing numbers of chronically ill patients highly susceptible to postoperative infections developing as a result of deficiencies in their immunologic defence , pharmacologic or surgical manipulation and or protien caloric malnutrition .

Special considerations must be taken into account when managing these fragile patients , particularly with regard to the prevention and treatment of postoperative infections .

Pietsch and Meakins, (1977) ;Farnall et al., (1986) . noted that the duration of hospitalization in whom a wound infection did not develop was 9.5 days , where as that for patients in whom a wound infection develop was 19 days .

8 - The duration of operation procedure :

Long operation and surgical procedures involving hypotension and extensive blood loss are associated with higher delaying healing and infection rates, (Fekety and Murphy , 1972).

Cruse and Foord ,(1973) , reported that there is direct relationship between the length of operating time and infection rate . The clean wound infection rate roughly doubles with every hour of surgery .

The duration of operative procedure did significantly affect the frequency of infection in surgical wounds procedures that were less than 2 hours in length were followed by 1 % rate of infection this was different in operations that were longer than 2 hours. Operations longer than 4 hours produced an 8 % wound infection rate (Roth et al., 1986) .

Moylan et al., (1987), demonstrated that factors affecting wound infection rate include length of operations. Operating procedures longer than 90

minutes had an 8.4 % infection rate , while shorter procedures of less than 90 minutes had an 2.24 % infection rate .

9 - Use of drains :

Most studies show an increased risk of infection in clean wounds ,if abdominal drains are brought out through the wound itself ,(Cruse and Foord . 1973) .

The use of drains had a significant negative impact on the clean wound infection rate . with the lowest rates found when drains were avoided entirely (Mead et al ., 1986) .

10 - Length of surgical incision :

Walter and Israel (1984) reported that longer surgical incision , later healing and more liable to sepsis .

The direction of pull of the underlying a surgical incision , and the crease lines are related to the movement of underlying muscles and joints , therefore , crease lines constitute the best guide to the placing of the incision .

INCIDENCE OF POSTOPERATIVE WOUND SEPSIS

The definition of wound infection varies widely from one study to another, but most investigators define the wound to be infected if it drains purulent discharge, whether organisms could be cultured or not from the purulent material, (Simchen et al, 1984). Unfortunately, it is estimated that surgical wound infection account for 30 percent of all nosocomial infections (Weigelt, 1985).

A large prevalence survey of 43 hospitals in England and Wales showed a mean acquired infection rate of 9.2 percent in 1980 (Meers, et al, 1981). Many variables influence infection rate in individual institutions. These include age, sex, duration of hospitalization, presence of previous infection, underlying illness, operation, duration of surgery, continuous ventilatory support and immunosuppressive therapy (Haley et al, 1980). Mead et al. (1986) found that the overall wound infection rate is only 2.8 percent. It is apparent that the incidence of wound infection varies from hospital to hospital and varies with the type of operation (Davis, 1976).

Ronald , (1983) , reported that the incidence of infection varied with the type of operation and he found that it was 1.5 percent in clean operations , 7.7 percent in clean-contaminated operations , and 15.2 percent or more in contaminated and dirty operations .

The most common complication in patient undergoing appendicectomy is infection .Infection is noted in 10 % to 30 % of the patients with the wound being the most common site affected (Janik and Firor, 1979) .

Infection remains the major complication of renal transplantation , wound infection requires particular attention because of serious rates of morbidity and graft loss ,the reported incidence of surgical wound infection in recent reports ranges from 1.9 to 34 % (Susan et al., 1978).

Fitzgibbons et al., (1977) reported that the incidence of wound infection in abdominoperineal resection due to carcinoma of the rectum was up to 15.5 percent . Also that of wound infection in colostomy and primary anastomosis due to carcinoma noted in 6.3 percent (Welch , Donaldson 1974) .

Splenectomy after trauma was complicated by wound infection in 9.4 percent (Strauch , 1979) .

2.3 percent of patients who had acute cholecystitis , had wound infections after cholecystectomy (Moore et al., 1979) .

Matter and Bitmann ,(1979), reported that rates of infection in open fractures range from 2.3 percent to 25 percent and upward if only the most severe injuries are considered.

Edwards ,(1976), noted that the overall rate of wound infection on urology services has been reported to vary from 2.3 to 9.2 percent .

Finegold and Kibry ,(1984), reported that 95 percent of wound infections are due to bacteria , and about two thirds of bacterial wound infections are due to Gram-negative bacilli , the rest are mostly due to gram-positive cocci .

The continuing high incidence of Staphyl ococcus aureus was some what surprising , but the high incidence of Escherichia coli , Pseudomonas aeruginosa, and Proteus mirabilis infections was anticipated in view of the increasing incidence of gram-negative pathogens .

The incidence of anaerobic pathogens , such as Clostridium perfringens , Bacteroides , and Peptostreptococcus , probably is not representative of their actual frequency , because of the problems of

anaerobic cultivation and identification that prevail in many hospitals (Altemeier , 1979) .

****Financial Costs Of Postoperative Wound Infection :**

Haley ,(1985), reported that surgical wound infections account for approximately half of the extra-days and hospital costs attributable to all nosocomial infections .

Infected wounds delay healing , cause wound breakdown , and ultimately scarring and disarrangement. Scar may be weak and stretch and an incisional hernia may result, also constrictures and disfigurement may be result .

Serious and potentially fatal infections may occur after major surgery ,e.. g. septic complications of abdominal surgery involving the gastrointestinal tract may lead to death . Patients who develop septicaemia after surgery are at high risk , particularly if their operation has involved the implantation of foreign body as in cardiovascular surgery.

It is possible to quantitate the additional money spent when infection occurs, and to this should be added loss of wages and productivity and the number of working days lost , when the patient's stay in hospital and convalescence is prolonged (Watts , 1981) .

Weber et al., (1976) . stated that economic loss from wound infection in U.S.A. has been estimated to exceed 9.4 billion dollars annually .

Watts , (1981), in Australia found that the annual additional cost of infection could easily be as high as 20,000,000 dollars , when all branches of surgery considered .

Another cost is the costs of infection control which must be taken into account when determining regimens of prevention . Much of the expense of modern hospitals and operating theatre design is the result of the need to prevent infection in the surgical wounds . A part of these costs , other things that contribute to the cost of infection control must be added , e.g. drugs and disinfectants , antibiotics costs, complications and antibiotic resistant bacteria .

Every thing is done to reduce the infection rate, costs money and the effectiveness of any new procedure introduced must be evaluated (Watts , 1981).

SOURCES OF INFECTION

A- In operating room :

* Air-borne dissemination :

Air-borne dissemination is important as a source of postoperative wound infection . Today's concerns hold that air-borne organisms are important not only in causing infection when air-holding system is grossely contaminated , but also clean air itself has been unable to reduce infection rate below 1.5% (Putsep , 1979) .

Whyte et al., (1982), studied the relationship between the number of bacteria in operating room and the patient's skin at the wound site and reported that most important route of contamination was air-borne . The importance of air-borne organisms would be seen most clearly in the group with clean wounds , since influence of contamination with endogenous organisms is less in this group (Altemeier , 1979) .

However, Irving and Kathryn, (1972) , reported that spread of microorganisms via air relatively uncommon cause of bacterial wound infection , unless there is massive contamination of the ventilation system . Sufficient changes of air per hour are important since airborne organisms are diluted and purged in this

manner , (Schonholtz, 1976) .It has been recommended that for critical surgery , in which large implants are used , air should be changed between forty-five and ninety times per hour (Laufman, 1976) .

* * CONTACT

This infection arise from dissemination of particularly virulent bacteria ,which is generally introduced in hospital by carriers found among patients or hospital personnel (Leback , 1981). Bacteriological studies in operating room have shown that the number of bacteria in air and indirectly in the wound is related to activity and number of persons in the room (Ford et al., 1967) .

Putsep , (1979) , reported that the respiratory tract of the operating room staff, particularly the nasopharynx is frequently considered the main source of pathogenic organisms connected with postoperative wound infection.

Dineen (1973), stated that ,perhaps hair is a more common site than nasopharynx for colonisation of staphylococci .

Lowbury et al., (1968) , suggested that one of the main infective hazards at operations is the possible transfer of bacteria from the surgeon's hands to patient's wound.

Walter and Israel, (1984), showed that the most dangerous areas for transmitting staphylococcal infection are hands and wrists. The sweat that accumulates under gloves is found to be contain many organisms, the area of sweat just above glove is also dangerous. Imperfect sterilization, faulty masking and perforated gloves share in contamination of the wound (Brigden, 1980).

*** INSTRUMENTS :

Inadequately sterilized instruments are of greates danger in causing infection (Walter and Iserael, 1984).

Schonhoitz, (1976), reported that the surgical suction apparatus as a source of wound contamination has apparently been overlooked.

Greenough, (1986), showed that on study of sucker during hip replacement sucker contemination is related to how long the suction is in use. Consequently it is recommended that new suker should be used.

**** ENDOGENOUS INFECTION :

The principal source of organisms that produce incisional wound infections is probably the patient. Endogenous organisms are certainly a major factor underlying the rise of infection rates as wound classification progresses from clean to clean-contaminated to contaminated wounds (Altemeier . 1979).

Keighley and Burned , (1975) , stated that man is normally a host of a larger number of bacteria which comprise a complex flora of more than 500 different species but only a few of these are pathogenic .

These flora may be skin flora , flora of gastrointestinal tract , respiratory tract , or urogenital tract , the patient is infected by his own flora either through blood stream or lymphatics (internal transmission) or by self inoculation through his hands , clothes or with the help of attendants .

Kune et al. , (1983) , reported that out of 200 patients , 6.5 percent were *Staphylococcus aureus* skin carriers, and 3.5 percent out of them were skin carriers on admission , such patient have high subsequent rate of staphylococcal wound infection than those who are noncarriers or nasal and throat carriers.

Lewis et al., (1987) , stated that bile contamination is the major source of postoperative infection in biliary surgery .

Canzos et al. ,(1985), reported **in** patients **with +ve bile culture**, a rate of wound infections is 3 - 5 times greater than that of patients with negative intraoperative bile cultures .

B : IN THE SURGICAL WARD :

There are potential sources of infection in the ward than in the operation theatre. Infection is conveyed by contact through lapses of dressing technique .It is bad practice to keep a large supply of dressing in a metal drum using some and keeping the remainder (Walter and Israel , 1979). Surgical drains brought out through an incision offer a route for ingress of organisms from the patient's skin into the wound contaminated irrigating solutions represent another potential source for contact spread of wound infection (Altemier , 1979).

The organisms soon contaminate the bedding and are shed into the air. The same applies to the patients suffering from Staphylococcal pneumonia or Enterocolitis, these may harbour virulent strains which

can lead to ward epidemics (Walter and Israel, 1979).

Spread by contact between susceptible patient and infected patient either directly or indirectly is thought to be the commonest and the most important route of transmission of hospital organisms, hand transmission by doctors, nurses and other hospital personnel are accepted as the most common form of contact spread, therefore hand washing is stressed as a mean of preventing this transmission (Blowers et al., 1955).

PATHOGENS RESPONSIBLE FOR WOUND INFECTION .

Altmeier et al . . (1976) , reported that considerable confusion has persisted concerning the microbial etiology of surgical infections .

The pattern of invasive surgical infections has changed , and a marked increase in the incidence of gram-negative bacillary infections has occurred . The organisms commonly involved in the abdominal operations include Staphylococci , most commonly Staphylococcus aureus , Escherichia coli , Proteus , anaerobic bacteria including Bacteroides and occasionally group A β haemolytic streptococci (David & Goel . . 1980) .

Trumbore and Kaye , (1985) , demonstrated that in urologic patients , the pathogens encountered are Pseudomonas , Serratia , and Klebsiella species as well as group D streptococci . Roth et al . . (1986) , estimated that pathogens cultured from fracture site infections were Staphylococcus which was isolated most frequently . Pseudomonas . Enterococcus which were less frequent in closed fractures than in open fractures .

In National Nosocomial Infections Study (NNIS) , (1979) , reported postoperative wound pathogens , they isolated Staphylococcus aureus in 18.7 percent , Esherichia coli in 18.6 percent , Pseudomonas

aeruginosa in 8.8 percent , Proteus species in 8.8 percent , Bacteroides in 3.8 percent ,they also isolated A , B streptococci , species of Clostridium and other pathogens .

COMMON TYPES OF WOUND INFECTIONS :

* Staphylococcal wound infection :

Staphylococcal infections usually have an incubation period of four to six days . The responsible organism in majority of instances is coagulase-positive Staphylococcus aureus ,many of these strains are ~~B~~ haemolytic , liquify fibrin and gelatin and produce yellow pigment in cultures .

The majority of Staphylococcus aureus strains that cause endemic postoperative incisional wound infections are derived from strains colonizing the patient or attendants, also outbreaks occasionally derived from a member of the operating room staff who has active clinical staphylococcal disease or as an asymptomatic disseminating carrier .

In closed incisional wounds, the symptoms and signs of staphylococcal infection include redness about the margins , swelling , and increasing local pain , fever and leukocytosis are usually present (Cruse, 1986) .

In most hospitals, because antibiotics are used extensively, prevalent Staphylococci are resistant to commonly employed antimicrobial drugs (Jawetz et al., 1987) .

* Gram-negative bacillary wound infection :

Wound infections caused by gram-negative bacilli have a longer incubation period than staphylococcal or streptococcal infections and the period of 7 to 14 days is not unusual . Furthermore , the local signs of inflammation are less marked , and patients may present instead with signs of systemic sepsis . The genera and species most frequently identified in aerobic gram-negative bacterial infections are *Escherichia coli* , *Enterobacter* , *Proteus* and *Pseudomonas aeruginosa* . These organisms are commonly associated with the anaerobic bacterium *Bacteriodes fragilis* (Cruse , 1986) .

The source of most resistant strains in hospitals appears to be patients who are colonized or infected (Weinstein and Kabins , 1981) . Because the normal pharyngeal and intestinal flora of hospitalized patients may be displaced by multiply resistant enteric bacteria and *Pseudomonas aeruginosa* (urine , perineum and wounds may be similarly affected) , there

are often many colonized patients for each patient with recognized infection . This shift in flora often occurs within a very few days of admission and affects the older , generally ill or more debilitated patients (Gilmor et al. , 1982) .

Extensive outbreaks of infections may result when such contaminated equipment is shared by many patients (Weinstien and Kabins , 1981). Antibiotic resistant Enterobacteriaceae are common in hospitals , especially aminoglycosides resistant Klebsiella , Serratia , Entenbacter and E . coli (Weinstein et al . , 1980) .

Hart et al., (1981) , reported that some organisms such as Klebsiella may be more viable on human skin and thus may have a greater potential from person to person spread on the hands of hospital personnel . Schaberg et al. , (1976) , showed that Serratia commonly causes asymptomatic colonization of urinary tract and respiratory tract and chronically colonised patients are often a source for large numbers of cross-infection.

Shooter , (1971), reported that Pseudomonas is frequently colonizing patients (even before admission) and contaminating water and various foods , particularly salads and fresh vegetables .

* Aerobic streptococcal infection :

The majority of invasive streptococcal infections in wounds are caused by aerobic B-haemolytic and group - A streptococci, the infection is thus characterised by the development of thin ,watery pus. Streptococcal invasion of blood stream is frequent and relatively early .Aerobic streptococcal infections caused by organisms other than group A streptococci tend to be much less invasive group. D streptococci are encountered in clean- contaminated and contaminated wounds of the gastrointestinal tract (Cruse ,1986) .

* Anaerobic streptococcal infections :

Peptostreptococci may produce a variety of severe postoperative infections with or without bacteraemia , particularly after operative procedures on the genital, intestinal or respiratory tracts . The microorganism is difficult to grow bacteriologically and routine culture is inadequate for detecting its presence.

The pus produced by peptostreptococcal infection characteristically is thick and grey and has a fetid anaerobic odour (Cruse , 1986) .

*Gram - negative anaerobic bacilli :

Clostridial infections are particularly a serious complication of incisional wounds that are most likely to occur in the deficient blood supply , gross contamination by dirt and other foreign bodies , and significant delay in adequate surgical treatment .

The microorganisms that cause such infections are anaerobic and the most important type is *Clostridium perfringens* . Other *Clostridia*, such as *C. histoliticum* , *C. novyi* , *C. sporogenes* , *C. septicum* and *C. sordellii* may occur alone , but less frequently, or in combination with *C. perfringens* (Altemeier , 1979) .The gram-negative intestinal organisms of the genus *Bacteroides* are important groups, because they form the bulk of organisms in the intestine as well as being present as a part of the normal flora of the vagina .

They are non-sporing , strict anaerobes , and very sensitive to the toxic effect of oxygen .

They cause infection of wounds , the peritoneal cavity, and the uterus following labour and abortion .

Localised *Bacteroides* infections are characterised by a particularly foul odour . Not only must culture be performed under anaerobic conditions . but oxygen must also be excluded from the specimen during its transmit to the laboratory (Walter and Israel , 1979) .Most commonly isolates are the *B.fragilis* group particularly from the large intestine (Jawetz , et al., 1987) .

* Mixed infections :

A large group of wound infections that complicate surgical operations or trauma are caused by a mixed bacterial flora .This group has a polymicrobial causation ,the bacterial mixture may consist of aerobic, anaerobic ,gram-negative ,and gram-positive microorganisms whose origin most often is a lesion or perforation of the gastrointestinal ,respiratory or genitourinary tract .

The aerobic and anaerobic bacteria often relate to each other in symbiosis , and their synergistic action determines the characteristic nature of these septic process. Crepitation of the infected tissue may develop as a result of the bacterial action of the *Clostridia* , anaerobic *Streptococci* , or associated aerogenic or aerobic bacilli .Successful treatment depends on early diagnosis and adequate surgical drainage of the infected wound if it is closed (Cruse , 1986) .

THE ROLE OF THE LABORATORY IN INFECTION CONTROL

The microbiology laboratory obviously provides crucial support for infection control programs. Major contributions lie in providing early warning , providing retrievable records as data sources , and assessing specimen collection and handling. Laboratorians must act as consultants , as they can contribute valuably in areas such as environmental sampling , culture surveillance , consistent identification , and proper quality control (Jacobson, 1985).

* Collection and transport of specimens :

Weinsten and Mallison ,(1978), reported that specimen collection , transport and handling must be of sufficiently high quality to provide valid data . Specimens that are not collected or transported properly may give inaccurate results , even when handled as well as possible once they reach the laboratory . The laboratory must monitor specimen handling continually and work closely with the wards

and clinics to make sure that the possibility of contaminated specimens is minimized .

Certain laboratory findings suggest specific handling errors . For example , a frequent failure to isolate organisms from deep wounds or abcesses of patients who are not on antibiotics , or inability to recover pathogens seen on gram stain in cases of presumed anaerobic infections , suggests inadequate anaerobic transport media , delay in appropriate refrigeration of specimens in transit , or use of inadequate techniques for isolating anaerobes .

* Identification of isolates :-

Branchman , (1981), reported that etiologic diagnosis cannot be made with certainty in many cases because of defficulty in determination of the causative agents . The majority of cases today for which the cause is known involve gram-negative aerobic bacilli . The expansion of the list of possible microbial pathogens for hospitalized patients has made it more difficult for both microbiologist and clinician to deal effectively with infection .

Effective handling of such problems requires the laboratory staff to keep up with steadily unfolding panorama of organisms important in cross-infection and to implement and maintain culture and other techniques that will bring these to light. When organisms are to be identified completely , it is important that standard criteria and nomenclature be consistently applied .

Even more important , incomplete or incorrect identification of organisms may obscure real problems and make retrospective epidemiologic investigation impossible . For example , a report of "Klebsiella-Enterobacter group " fails to distinguish between two organisms (Klebsiella and Enterobacter) that have different epidemiologic patterns of infection within hospital (Schaberg et al. , 1976) .

Laboratories should maintain the capacity to identify gram-negative bacilli to the genus level with at least 95 percent accuracy , and such identification should be a routine part of laboratory procedure .

Investigations of an outbreak of infection may require isolation and identification of isolates in specimens not only from patients but also from personnel who might be colonized with outbreak strains and from environmental objects that might be similarly

contaminated (Goldman and Macone ,1980) .

Bartlett , (1974) , found that an effective clinical microbiology laboratory is essential to an effective infection control program , adequate quality control is essential to the practice of good clinical microbiology . Such a quality program begins with a comprehensive procedure manual that establishes standards for performance , including definition of acceptable and unacceptable quality of specimens and specimen containers , permissible delay between collection and receipt of the specimen in the laboratory , and times during which specimen in the laboratory are accepted for processing .

The action to be taken by workers when the specimens are ~~not in~~ accord with these standards also must be defined . These standards should be communicated to clinicians and nurses as well as to laboratory personnel . The periodic evaluation of skills of all workers , including evening , night and weekend workers, should be included in the program .

Such " pseudobreak" must be considered when laboratory culture or strain results do not correlate with clinical or epidemiologic findings (Maki,1980) .

However routine checks on the adequacy of sterilizer function, and periodic checks on the effectiveness of disinfection of certain equipment that directly contacts tissues other than skin may help to prevent infections from these sources (McGown ,1981).

**** ANTIBIOTICS SUSCEPTIBILITY TESTING :**

A standerized method of antimicrobial susceptibility testing subject to quality control evaluation is essential in any clinical microbiology laboratory and is equally critical to infection control studies . To investigate whether strains in the "cluster " are common or different , the usual practice is to examine results of biochemical tests and the pattern of susceptibility to antimicrobial agents (Schaberg et al., 1979).

Clinical failures were common when antibiotics were used for treatment of infections caused by resistant bacteria. Since bacteria could be isolated from site of infection , it becomes feasible to test these bacterial isolates for their susceptibility against the available antibiotics and thus attempt to select the most appropriate antibiotics (Loveless , 1986) .

The Kirby-Bauer single-disc-diffusion method or an equivalent test system is used in many laboratories for routine testing of antimicrobial susceptibility of bacteria (Jones , 1982).

The advantage of disc diffusion system are its relative reproducibility and the ease with which large numbers of antimicrobial agents may be tested . Difficulty arises with some bacteria that produce indistinct zones of inhibition .

In addition , aminoglycoside antibiotics do not produce reliable regression lines for the determination of susceptibility (Wartz , 1973) .

* * Selection of strains testing :-

For example , the request for testing of susceptibility should be carefully evaluated when the organisms isolated are endogenous flora present at sites in which they are not normally pathogens. Similarly , the testing of organisms from mixed culture should be avoided in most cases because of the unclear role of the various isolates (Bartlett , 1974) .

Laboratories must include testing methods that will rapidly and accurately detect the presence of such resistant strains , methods required vary from those routinely employed , and involved extensive quality

control evaluation to ensure precision (Ishida et al., 1982) .

**** Selection of drugs for routine and special testing :**

National committee for clinical laboratory standards, (1984), reported that ,in general only one drug of a particular class of antibiotics need be tested , since the result will pertain to all members of that class . The chosen agents should reflect both common usage practices of physician in the hospital and the spectrum of pathogens that are frequently encountered . Similarly, certain antimicrobial for which the hospital wishes to control usage may be tested but not reported routinely , or tested only after consultation (Kunin , 1981) .

Different groups of antimicrobials often used for gram-negative and gram-positive aerobic organisms. Drugs included in each panel should be periodically evaluated and updated .

The epidemiologic value of susceptibility patterns may be enhanced by inclusion of certain antibiotics that are not in routine clinical use . Such additional information can also provide valuable taxonomic and quality control information (Goldmann, 1980).

Anaerobic organism combines with aerobe , each providing factors that promote the growth of the other bacteria (Brook et al. , 1984) .

The more resistant organism may also produce substances that inhibit the activity of antibiotics on the sensitive organism (Thadepalli et al. , 1977) .

The use of drug combinations may alter the antimicrobial activity in vitro . Combination may be antagonistic , indifferent , or synergistic in their effect on bacteria (Loveless , 1986) .

Cosistant and accurate identification of organisms over time is necessary for susceptibility data to be useful for clinical and epidemiologic purposes .

In addition , errors in performance of susceptibility tests may result in information that is misleading about diagnosis and/or therapy . To minimize this possiblity , most antibiotics discs should be stored at 4 c to 8 c and discs containing synthetic penicillins and cephalosporins , required storage at - 20 c . Susceptibility tests depend on correct incubation temperatures , these should be checked routinely during the weak (McGown et al. , 1986) .

When attempting to select the approach antibiotic or combination of antibiotics , the physician must take into the account the source , location , and severity of infection .

Finally , knowledge of the pharmacokinetics and distribution of the specific antibiotic will affect the choice of antibiotic and appropriate dose.

Only this comprehensive , multifactor analysis will lead to the correct antibiotic choice and consistently produce a favourable clinical outcome for the patient with infection (Loveless , 1986) .

POSTOPERATIVE WOUND INFECTION CONTROL PROGRAMS .

In the theatre :

A . Operating room personnel :

1- Surgeons :

Hummel , (1976) , showed that surgeons who are carriers may continue to operate the practice safely , provided that they take the necessary extraprecautions such as :

- Scrubbing with suitable degerming agents .
- Frequent changing of clothing and laundering .
- Compulsive and thorough hand washing between contacts and treatments .
- Double masking .
- Inhibiting the nasal microbial flora preoperatively by sprays or nasal ointments before operating .
- Prompt changing of gloves during operations.
- The use of non touch technique.

Letts and Doermer , (1983) , stated that the purpose of surgical face-mask is , by filtration , to stop particles emitted from nose and mouth during respiration , talking , and sneezing from getting into the open surgical wound or the operating room environment .

Disposable masks are popular because of their convenience and the elimination of troublesome laundry problems ,and it is recommended that a fresh mask for each case (Hummel ,1976) .

One way to decrease bacterial escape from behind the masks is by decreasing the amount of conversations in the operating room and using of the hand signals. Another way is to wear the mask under a hood that completely over the sides and bottom of the nose (Letts and Doermer , 1983) .

Protective foot covering should be worn in the operating room area to prevent transmission of bacteria from shoes , and not worn outside the operating area (Hummel , 1976) .

Grimmond ,(1981) , noted that the aim of scrubbing is to remove transient bacteria and to reduce the resident flora . Soap is useful in removing the grease, dirt and transient bacteria . Liquid soap or detergent preparations are found to be more effective than antiseptic bar soaps . Effective agents to reduce the resident flora include 70 % alcohol , providone - iodine , hexachlorophene and chlorhexaidine . It should be remembered that germicides of skin are most effective only when fat have first been entirely removed .

Walter and Israel, (1984) stated that prolonged washing is not only unnecessary but is indeed harmful to the skin if frequently carried out . It has been recommended that 2 to 3 minutes scrubbing with careful attention to the nails , before the first operation , and 2 to 3 minutes without a brush before the subsequent operations of a list should be an adequate routine for a surgeon .

Beck , (1981) , stated that gowns and drapes are employed to separate sterile from non-sterile areas . Hummel (1976) noted that several varieties of disposable gowns which are satisfactory impermeable are now available .

Moylan et al., (1987) reported that the incidence of developing a wound infection was 2.5 times higher with a cotton system than with a disposable system . The results demonstrated not only significant reduction in wound infection rates but also major cost saving when a disposable gown and draps system was used in the operating room .

The majority of gloves currently in use are now disposable , but reusable gloves are still available . All gloves are packed with the cuff turned back so that they can be handled by the exposed part of the inside of the glove wet hands will not slip into the

glove , therefore the hands are usually dried either with a sterile towel or air dried before gloving .Any powder on the rubber gloves showed be removed e.g. by a sterile wet towel (Hummel , 1976) .

Putsep , (1979) , noted that wearing gloves on the appearance of visible tear or punctured ones during operation, the gloves must be removed and replaced with new gloves after washing hands with antiseptic detergent and a fresh gown must also be put on because sleeves become contaminated on changing gloves.

2 - Patients :

Mears , (1975) , stated that in addition to optimizing the general condition of an individual patient and minimizing the amount of preoperative and intraoperative time as much as clinical circumstances permit , the risk of postoperative wound infection can be reduced meaningfully by such measures as having the patient shower with hexachlorophene the night before surgery , using depilatory cream for removal of hair , and when the gastrointestinal tract is to be entered , use both mechanical bowel prep for reduction of intestinal fecal content and the oral non-absorbable antibiotics combination of erythromycin base and neomycin to reduce the quantity of intestinal bacteria.

Grimmond ,(1981) , showed that preoperative exposure to hospital environment should be minimized (less than 24 hours) and antimicrobials if required preoperatively should be given for limited periods to avoid selection of resistant strains .

Laufman and Winkelman ,(1976), reported that patient's bed clothes and blankets should not be allowed to enter into theatre and the patient should be transferred from the ward by cart and then transferred from the ward cart to the operating theatre cart , and the ward cart should not be allowed to enter inside the theatre because it may carry ward organisms along with its wheels .

Alexander et al.,(1985) , reported that satisfactory reduction of skin bacteria has been encountered with the use of one - minute preparation of 70 % alcohol or 2 % iodine in 90 % alcohol solution without a perior of soap or detergent rubbing (unless there is a gross dirt in the operative area) . They also reported that , the major advantage of using this one-minute skin preparation obvious plus the use of antimicrobial adhesive drapes makes a clean wound infection rate 1.3 percent and an over all infection rate of 2.5 percent .

3 - Some surgical techniques :

a- Draining :

Drain should be obviously left in place until they have served their purpose . The length of time will vary considerably , but generally , the drain should be removed when drainage has become small in amount (Postlethwait , 1981) .

b- Suturing :

Tobin , (1984) , reported that ,tapes are the skin closure of choice for clean contaminated wounds . Stillman et al., (1984) , reported that , the use of skin tapes should be the preferred method for wound closure , and it was found that while wound dressing is required for silk sutured wounds , it is unnecessary in taped wounds .

4 - Instruments:

The surgical instruments should be adequately sterilised . Autoclaving is the most preferred method for sterilization of instruments .

The great superiority of moist heat over hot air as a sterilizer is because it kills more rapidly and at a far lower temperature than dry heat , penetrates much better , and produces a negative pressure and brings more steam to the same site (Walter and Israel , 1984) .

Chemical sterilization of surgical instruments can not be relied on, and should be abolished .

5 - Air in the theatre :

Most operating rooms are ventilated by air drawn from outside doors. The incoming air must be filtered and distributed in the proper way, and a quantity of air proportional to that introduced must be exhausted to maintain a slight positive pressure within a room with closed doors (Laufman and Winkelmann , 1976). Ventilation with about 25 changes per hour probably creates a better mixing of air (Anderson et al. , 1983) .

Cleaning of operating room in the proper way is very important to decrease the incidence of wound contamination .

All surfaces must be dust free , dust particles on these surfaces may be disturbed by air current , or movement in the room . Therefore , all surfaces must be thoroughly cleaned and then disinfected by an appropriate disinfectant agent . It is necessary to remove all foreign and organic materials which may preserve bacteria and inhibit the antimicrobial action of antiseptic solutions .

In general , all surfaces are either wet mopped, wet vacumed , or wiped with a damp cloth to avoid creating microbic-laden aerosoles. Postoperative cleaning of operating rooms should be carried out after every case . Each room and scrub area should be cleaned daily .

In week end , all the operating room-floors should be scrubed with floor machine using detergent disinfectant solution . All walls should be cleaned thoroughly . Wet vacuum is used for picking up solutions (Laufman and Winkelman , 1976) .

A large number of agents have been tested as germicidal solutions, and the non-volatile iodophors, phenolics , and quaternary ammonium compounds have been reported to remain active for days when properly applied to a suitable surface . The solution should be left on the floor 5 minutes before wet - vacuum picked up to act on bacteria (Weber et al. , 1976) .

Walter and Israel .(1984) , stated that a contaminated room should be disinfected with hot formaline vapour.

In the ward :

* Isolation of infected patients :

Isolation is an important method of preventing and controlling infection in surgical patients (Polk , 1976). In order to prevent spread of infection to other patients e.g. patients with extensive draining wound infections , caused by common pathogens the infected patients should be isolated . In some hospitals a special and separate " septic ward " may be used for patients who require isolation (Walter and Israel , 1984) .

Plok ., (1976), reported that non-disposable articles (e.g dressing & instruments) should be cleaned with a disinfectant, wrapped ,and autoclaved following each use and before further processing .

* * Dressing :

To avoid transmission of pathogenic organisms and to decrease times of changing of dressing , an adhesive wound irrigation device is designed to facilitate the care of discharging wounds . Westaby and his colleagues, (1981), reported that the use of this adhesive irrigation device has the following advantages over the conventional dressing :

- Each device lasted for 4 to 5 days before change is required.
- It provides a closed system for wound irrigation .
- It reduces the exposure of purulent wounds to the atmosphere.
- This type of dressing is convenient and comfortable.

*** Measures of ward hygiene.

Walter and Israel ,(1984), stated that it is recommended to use cotton blankets and changed on the patients discharge, or when contaminated with infected material,sheets should be changed everyday .

Mallison,(1976), found that all furnatures of the ward should be cleaned so frequently that there is no visible dirt on them . Floor of room should start at the back of the room and working toward the door . All sinks, toilets ,and bathrooms should be washed daily with 1 % phenolic detergent disinfectant solution.

In contaminated rooms , in addition to other measures the cleaner should wear a gown and mask in all instances when cleaning a contaminating room and should be changed for each room cleaned . Also , solutions should be changed and clean clothes should be used for each room .

THE ROLE OF ANTIBIOTICS IN PROPHYLAXIS

The term "prophylactic antibiotic " implies that a microorganism is attacked by an antibiotic agent during the period of contamination before infection is already established (Sandusky , 1980).

Kington et al., (1986) , reported that the use of prophylactic antibiotics for selected surgical procedures is now accepted as a good surgical practice, and having adequate antibiotics concentration in the tissue prior to surgery significantly reduces the rate of wound infection . The aim of use of prophylactic antibiotics is to supplement host resistance by decreasing the number of viable bacteria , allowing efficient clearance by host defences .

Riess et al. ,(1984). reported that antibiotics are valuable in the prevention of postoperative wound infection if the appropriate antibiotic is given in the proper doses to carefully selected cases at the proper time by the proper route of administration .

Love,(1985) ,reported that the significance of their role in defending against anticipated infection . The critical factor is not only the antibiotic itself, but also the time of administration , which must be within a "decisive period" of three to four hours after

incision . Any inhibitor or enhancer of bacterial growth in a surgical incision must act during this critical period if it is to be effective.

Burke ,(1981) , reported that preoperative prophylactic antibiotics are only indicated where there is high probability that the patient resistance to bacterial invasion will not overcome the combined bacterial and physiologic challenge of the intended surgical procedure. He also reported that the antibiotic administration should be stopped as soon as the probability of bacterial contamination of the wound is over and the patient has returned to a normal state of physiology in the immediate postoperative period .

Dipiro et al.,(1983), and Nichol ,(1985), demonstrated that in recent years published recommendations for antimicrobial prophylaxis in surgery have suggested that preoperative antimicrobial administration with up to 24 hours of postoperative administration (two to four doses) was appropriate for surgical prophylaxis.

Tcherevenkev et al., (1986), showed that tissue defences~~are established~~, three hours following bacterial contamination, and the factors influencing the wound response to bacteria are virtually inoperative after that time . Therefore, prophylactic antibiotics are

effective in supplementing the host defences to overcome bacteria during this "decisive period" and are ineffective thereafter .

Nomikos et al., (1986) , recommended the use of preoperative systemic antibiotic , intraoperative, and its continuation for no longer than three days, while Oreskovich et al., (1982),and Caplan and Hoyt ,(1985), found that short courses of antibiotics were as effective as longer courses in prevention of wound infection, and they recommended the use of prophylactic antibiotics for one or two days only .

Burke,(1981), recommended to give the dose of prophylactic antibiotics intravenously with the beginning of anaesthesia ,and repeat it every two to three hours of operating time , with the final dose delivered as the patient leaves the recovery room .

* * Topical prophylactic antibiotics :

Application of prophylactic antibiotics topically has the advantage of producing high wound drug concentrations, with low systemic concentrations, and thus avoids dose related complications (Tobin , 1984) .

The topical antibiotics should be considered as an alternative to the use of systemic prophylactic antibiotics especially when it is desirable to avoid the latter, and as a complement to their use when wounds are liable to heavy contamination (Altemier and Alexander, 1981) .

Pitt et al., (1982) ,reported that a mixture of neomycin sulfate , polymixin B and bacitracin is used as a spray (Riko spray) and is effective against a wide variety of organisms. Topical antibiotics should be applied within 3 hours from the time of contamination.

Tobin, (1984), reported that both experimental and clinical studies support the use of topical antibiotics in contaminated wounds. Zelco and Moore ,(1980), reported that 10 % povidone-iodine applied once to the subcutaneous tissue before closure of the wound , was associated with reduction of the wound infection rate .

Lau et al. ,(1986), reported that with effective systemic antibiotics , 1 percent topical povidone-iodine causes more wound infection in late appendicitis and it is of no additional benefit in early appendicitis and it was shown to be cytotoxic..

* * Selection of antibiotics :

Ronald , (1983), reported that the cephalosporins have been used in many studies with considerable success. because they have an excellent record of safety, a wide spectrum of activity and proved efficacy for prophylaxis in many surgical procedures. He also reported that cefozal~~ine~~, administered as 1 gm. dose preoperatively and as one or two doses postoperatively at 6 hour intervals, is the agent of choice in many clean and clean contaminated surgical procedures .

These advanced spectrum cephalosporins are attractive choices for prophylaxis and therapy covering the maximum number of pathogens that cause surgical wound infection (Centers for Disease Control CDC surveillllance summarie~~ses~~, 1985) .

Jones et al.,(1987), confirmed that cefotaxime in single 1 gm dose was superior in preventing infectious morbidity and side effects and reduced hospital drug costs compared directly with multidose regimens of cefazolin or cefoxitin . For colorectal resection , multiple doses of cephalosporins appear equally effective , administered to a maximum of 24 hours postoperatively.

Roland et al. ,(1986), demonestrated that a single preoperative intravenous infusion of metronedazole in combination with ampicillin or doxycyclin is a simple and effective prophylactic regimen in colorectal surgery directed against anaerobic as well as aerobic bacteria . During last two years , our standard antibiotic regimen has been a single intravenous preoperative dose of latamoxef which we showed significantly better than our previous standard cephaloridine .

Ausobsky,(1983), and Sauven et al.,(1986), reported that a single dose of latmoxef at induction of anaesthesia provides better prophylaxis against abdominal wound infection than peritoneal and parietal tetracycline lavage.

Senior and Steigrad,(1986) , reported that cefoxitin is a semisynthetic cephamycin antibiotic .It is active against the clinically important gram-negative facultative bacteria except pseudomonas and many Enterobacter species as well as clinically important anaerobic organisms such as Bacteroides fragilis , it is inactive in vitro against most enterococci . Cephradine is a semisynthetic cephalosporin antibiotic .

Its range of activity is similar to that of cefoxitin except that cephradine has no activity against anaerobes, especially *Bacteroides fragilis*. Tenidazole is a metronidazole derivative that is claimed to be more bactericidal against *B. fragilis*.

Gould and Wise, (1985), reported that the antibiotics with activity against *Pseudomonas* include penicillins such as carbenicillin, ticarcillin, azlocillin, mezlocillins and piperacillin, aminoglycosides such as gentamicin, tobramycin, netilmicin and amikacin, and the newly developed third generation cephalosporins such as ceftazidime, cefsulodin and cefoperazone.

Bradley et al., (1985), reported that methicillin-resistant staphylococcus aureus is resistant also to penicillin, erythromycin, tetracyclines, sulphonamides, trimethoprim and gentamicin, but it is susceptible to vancomycin, rifampicin and fusidic acid.

* * Antibiotic use and antibiotic resistance :

Evidence from a number of studies suggest that the proportion of bacteria resistant to a given antibiotic may increase as use of the drug increases.

Many individuals have felt that a policy of restricting the use of the newest and most broadly active antibiotics would minimize the development of resistance to those drugs and hence prolong their useful life span (Davis, 1983) .

McGown,(1983),summarised seven types of evidence linking antimicrobial use in the hospital and antimicrobial resistance in hospital bacteria :

- 1- Antimicrobial resistance is more prevalent among bacteria causing infection in the nosocomial setting.
- 2 - Patients infected with resistant outbreak strains are more likely to have received previous antibiotic therapy.
- 3 - Changes in antimicrobial use may lead to parallel changes in the prevalence of resistance to that antibiotic .
- 4 - Area of most intense antibiotic use within the hospital generally had the highest prevalence of antibiotic resistance .
- 5 - Increased duration of exposure to antibiotics in the hospital generally increase the likelihood of colonization of infection with resistant organisms.

6 - The higher the dose of antibiotic given, the greater the likelihood of superinfection or colonization with resistant bacteria .

7 - The antibiotic therapy produces marked effects on the hosts endogenous flora and exerts selective pressure in favour of resistant organisms.

Sandusky ,(1980), stated that prophylactic antibiotics are not substitute for careful surgical technique. They can be used effectively only as an adjunct to adequate surgery, and they are not an option that permits lower standards of housekeeping antiseptics or asepsis .

MATERIALS

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METHODS

MATERIALS AND METHODS

The subject of this study were the patients in surgical departments of Benha University Hospital were included in the study during the period from November 1987 to the end of April 1988. In this period, patients admitted for surgical operations, in General Surgery, Urology and Orthopedic.

All cases were observed for postoperative wound

sepsis, wounds that developed sepsis were submitted for full bacteriological examination.

Also nasal swabs from doctors, nurses and workers who were in close contact with the infected cases were obtained and examined. Broth moistened swabs from anterior nares of the patients, and sites of incisions from skin of patients, were cultured and examined.

Specimens from bed linen, surgical dressing and disinfectants used in different wards were collected and bacteriologically examined. Nutrient agar plates

History of each investigated patient was recorded for :

Name	Age
Sex	Date of admission
Date of operation.	Type of operation
Time of development of postoperative infection.	
Using of prophylactic antibiotics.	

For every case, 2 samples from the exudate were taken by using a sterile cotton swab for aerobic cultivation and anaerobic swab for anaerobic cultivation. The anaerobic sample was moistened in Thioglycollate broth and transported to the laboratory. The aerobic swab was cultivated on ordinary aerobic culture media (nutrient agar, blood agar, MacConkey's agar), incubated at 37°C for 24 - 48 hours. anaerobic swab was inoculated on anaerobic culture media (Wilkins Chalgren Anaerobe Agar - M 619 (oxid) .

*** Wilkins Chalgren Anaerobe Agar (M 619) :-

This medium is recommended for general growth of anaerobes and for antimicrobial susceptibility testing. It allowed more consistent growth of anaerobics than Schaedler. (Eley et al., 1982).

Formula :-

	(Grams per liter)
Tryptone	10.0
Gelatine peptone	10.0
yeast extract	5.0
Dextrose	1.0
Sodium chloride	5.0
L . Arginine	1.0
Sodium pyruvate	1.0
Menadione	0.0005
Haemin	0.005
Agar	10.0
pH 7.1 \pm 0.02	

This is a preparation of non selective anaerobic culture media for all anaerobes .

21.5 grams of wilkins - chalgren, Anaerobe Agar was suspended in 500 ml . of distilled water, dissolved completely by boiling , then strilized by autoclaving at 121 c for 15 minutes , then cooled to 50 c. 25 ml defibrinated blood was then added aseptically , mixed gently and poured into sterile petridishes . We used for incubation anaerobically Oxoid anaerobic system which consists of gas generating kit , Anaerobic catalyst and anaerobic indicator .

**** Technique of anaerobic cultivation :-**

- The specimen was inoculated on freshly prepared Wilkins Chalgren Anaerobic Agar media.

- The plates was incubated in the anaerobic jar .

- The anaerobic catalyst were clipped to the underside of anaerobic jar, a strip of anaerobic indicator was placed in the anaerobic jar in order to check the correct anaerobic condition by changing its colour from pink to white indicate redox condition

which is suitable for growth of most anaerobic organ-

isms, 10 ml water was added to the sachet of gas generating kit, stood upright in the anaerobic jar and the lid was closed.

- The period of anaerobic incubation was 48 hours at 37 °C and the plates were examined after this period then if there was no growth, it was continued up to 5 days before plates were discarded, because up to 20 % of non-sporing anaerobes require prolonged incubation under unbroken anaerobic condition.

- Confirmatory tests on isolates were done and the

**** Bacteriological Examination of aerobic and anaerobic cultures:**

Any growth of discrete colonies were prepared on the appropriate aerobic or anaerobic media for further identification.

1- The bacterial growth was identified by colonial morphology as size ,shape , presence of pigments , haemolysis on blood agar , and lactose fermentation on MacConkey's media.

2- Films were prepared and stained with Gram-stain and examined microscopically .

3- Biochemical tests for complete identification of the isolated organisms were performed according to Cruickshank , (1975) .

*** Sugar fermentation test :**

Different types of sugars were used , glucose , lactose, maltose , mannite and sucrose . The sugar media were incubated at 37 c for 24 - 48 hours . After incubation ,the media were examined for presence of a colour change (indicating acid),and for gas formation in the Durham's tubes .

- Coagulase test :

It was done by 2 methods to determine the pathogenicity of the isolated staphylococci.

a- Slide method :

The slide was divided into two sections . A drop of normal saline was placed on each section , one or two colonies from agar plate of the tested strain were emulsified in the saline on both sides

to make smooth suspension . A drop of undiluted human citrated plasma was added and stirred gently with a wire and a drop of sterile saline was added to the other side . **Clumping factor** were identified by detection of clumping within 15 seconds , while no clumping in the other drop which serves as a control .

b - Tube method :

One ml. of 1/10 diluted citrated human plasma was placed in each of small Wasserman tubes . 0.1 ml . of an 18-24 hours of staphylococci broth culture was added to all tubes . The tubes were incubated at 37 °c and examined for coagulation 1 , 3 , 6 hours later . The conversion of plasma into gel was seen on tilting the tubes on horizontal position .

This reads positive result, the negative results were left at room temperature over night and examined . Control tests of known coagulase positive and coagulase negative cultures were set up with each batch of tests.

- Methyl red :

Glucose - phosphate - peptone medium was prepared , inoculated from a young peptone culture , incubated at 37 °c for 48 hours . 5 drops of methyl red reagent , was added . A bright red colour appeared immediately indicating positive result .

- Voges proskauer test :

Glucose - phosphate - peptone medium was prepared , inoculated , incubated , for 48 hours at 37°c . 1ml of 40 % potassium hydroxide , and 3ml of 5% α - naphthol in absolute ethanol , was added (Barritt's method) . Positive result was indicated by the development of pink colour in 2 - 5 minutes .

- Citrate utilization test :

A tube of koser's citrate medium was prepared , inoculated and incubated for 96 hours at 37°c. development of turbidity indicated positive result . We

used Simmon's citrate medium which was a modification of koser's medium with agar and an indicator added . positive result was indicated by changing colour and there was streak of growth .

- Gelatin liquefaction test :

Nutrient gelatin medium was prepared, inoculated by stab culture from an agar slope culture , after incubation at 37 °c , testing for liquefaction at intervals by removing the nutrient gelatin culture from the incubator ,holding them at 4 c for 30 minutes before reading the results .

- Indol test :

An indol test medium was prepared , inoculated and incubated at 37°c for 48 -96 hours . 2 drops of zylol 8-05ml of kovac's reagent was added,after shaking gently a red colour ring appeared immediately indicating positive results .

- Oxidase test :

It was performed by plate method ,culture was made on nutrient agar plate . A freshly prepared 1 % solution of tetramethyl -P- phenylene diamine dihydrochloride was poured on to the plate so as to cover its whole

surface and then decanted . Colonies developed dark purple colour within 5 - 10 seconds indicating a positive result .

- Urease production test :

A Christensen's medium slope was prepared , inoculated over the entire slope surface and incubated at 37 c. Examination was done after 4 hours and after over night incubation, purple pink colour indicated positive result.

- Catalase test :

Done by adding 1 ml. of 3 % H_2O_2 solution to the growth on slant , gas evolved indicated positive result.

4 - Motility test : by hanging drop preparation :

A ring of metal dipped in petroleum jelly was outlined on the cover , a drop of suspension containing organisms to be examined was put in the centre of the cover .

The slide was inverted over the coverslip , pressed and quickly turned round the slide so the coverslip was upper most , examined under microscope by low and high power .

It was essential to distinguish between true mobility, where the organisms changed its position in the field and Brownian movement which was an oscillatory movement of all small bodies (whether living or not) suspended in fluid .

** TYPES OF ISOLATED ORGANISMS.

A - Staphylococci :

Microscopic examination revealed Gram + ve cocci , non sporing , **clusters**, in pairs , in short chains or in irregular, **singly** (**clusters** arrangement is the most characteristic) , it was facultative anaerobe .

Organisms grew well on nutrient agar , blood agar , colonies were usually opaque ,circular , smooth and entire with golden yellow , white or pale colour .On MacConkey's were pinkish and very small to normal size. The pathogenic staphylococci were coagulase-positive .

B - Gram-negative organisms :

Microscopic examination revealed gram negative rods. On culture on MacConkey's media, lactose fermenting organisms (pink colonies) developed , were identified by their fermentative activity on sugars with production of acid and gas for all strains .E.coli strains were identified by indol test positive , methyl red positive , voges proskauer's test negative , citrate test negative , while Klebsiella strains were identified by indol test negative , methyl red test negative , v.p.test positive . citrate test positive . If non-lactose fermenting organisms (pale colonies) developed on culture on MacConkey's media , further identification tests were done Proteus strains cultured on nutrient agar , blood agar , revealed fishy smell , swarming growth urease test was the most important confirmatory test .

Pseudomonas pyocyanea strains revealed blue green pigment colouring the colonies and surrounding media intensive on culturing on nutrient agar or blood agar . Oxidase test was the most important confirmatory test , only most strains of Pseudomonas were strict aerobes , other organisms are facultative anaerobes .

C - Bacterioides :

They were identified by growing strictly anaerobic growth . By microscopic examination it was gram-negative bacilli non-sporing and highly pleomorphic. Some of them revealed β -hemolysis on blood agar and biochemical reactions were variable .

** Antibigram Pattern :

Antibiotic sensitivity test for each isolated aerobic and anaerobic pathogenic strains were tested by using disc diffusion method as following :

1 - A colony of isolated organism to be investigated was gently touched with a sterile loop , transferred to test tube containing 5 ml . broth then incubated for 2 - 5 hours untill cloudness was observed equal to standard prepared as follow :

0.5 ml of 0.048 molar barium chloride (11.7 gm of barium chloride / litre) added to 99.5 ml of 0.36 N H_2SO_4 . This density standard was in a tube of the same size as the culture tube ,this opacity was nearly corresponds to 10^5 - 10^6 cell/ ml .

2- A sterile cotton swab was dipped into the adjusted turbidity tube , then inoculated on surface of Muller Hinton agar by repeated streaks on the whole of a gar

surface , turning the disk at 60 each time to ensure the uniform inoculation .

3 - The antibiotic discs were carefully despended with a sterile forceps to the agar surface , it was arranged in 2-3cm apart to avoid overlapping of inhibition zones

Then the plate was inverted and incubated after 15 minute at 37 °c for 18 - 24 hours

4 - The previous procedure was done anaerobically for the anaerobic organisms by using Wilkins-Chalgren Anaerobic Agar culture media .

5- By using drawing templates , the diameter in mm.of the complete growth inhibition zone including the diameter of disc itself were carefully measured and recorded .

The different antibiotics used in the following concentrations were :

* For Aerobic isolated organisms :

AMIKIN	30 ug / disc
AMPICILLIN	10 ug / disc
AMOXICILLIN	10 ug / disc
ERYTHROMYCIN	10 ug / disc
GENTAMICIN	10 ug / disc
CHEPHALORIDINE	30 ug / disc
PENICILLIN G	10 units /disc
STREPTOMYCIN	10 ug / disc
TETRACYCLIN	30 ug / disc
TOBRAMYCIN	10 ug / disc
CHLORAMPHENICOL	30 ug / disc

* For anaerobic isolated organisms :

- PENICILLIN G	2 units / disc
- ERYTHROMYCIN	15 ug / disc
- NEOMYCIN	1000 ug / disc
- KANAMYCIN	1000 ug / disc
- COLISTIN	10 ug / disc
- RIFAMPICIN	15 ug / disc
- METRONIDAZOL	5 ug / disc
- CHLORAMPHENICOL	30 ug / disc