Analysis of Organisms Found at Incision Site Intra-Operatively and its Implications with Post-Operative infections

Dr. Pravin Suryawanshi *, Dr. A. Q. Khan **, Dr. Saiyyad Altaf G ***, Dr. Amey Patil ****

* Porf. & Head Of Department, Chief Endoscopist, M.G.M Medical College, Aurangabad.
** AssoProf. Head Of Unit, Surgery Dept, Laparoscopic Surgeon, M.G.M Medical College, Aurangabad.
*** Lecturer, Surgery, M.G.M. Medical College, Aurangabad.
**** Chief Resident, Surgery, M.G.M. Medical College, Aurangabad.

Abstract- Post-operative surgical site infection (SSI) is a major source of morbidity and mortality increasing the suffering of the ill. It adds enormous burden to the already strained medical services system more so in a developing country like India. Various protocols are in place to minimize the detrimental effects of SSI. Fomites are recognized source of SSI. We in our study have found out culture and sensitivity profile of the organism found at the incision site intraoperatively. Staph. Aureus, P. Aeurignosa, E.Coli, Klebsella, were found to be most common. The culture and sensitivity profile of these organisms were found to be same as the national, international, studies. This was an attempt to recognize relation between these organisms and organisms cultured in post-operative infections. We found post operative infection rates were 5.97% in Clean cases and 8.51% in Clean Contaminated cases. India as a developing country lacks in the research and development activities. We tried to do a pilot study pioneering to explore the milieu of incision site as indicator of SSI. Our study is also dictating the anti-biotic protocol which we feel is must of every institute.

I. INTRODUCTION

Postoperative surgical site infections remain a major source of illness and a less frequent cause of death in the surgical patient.1 These infections number approximately 500,000 per year, among an estimated 27 million surgical procedures,2 and account for approximately one quarter of the estimated 2 million nosocomial infections in the United States each year.3 The incidence of infection varies from surgeon to surgeon, from hospital to hospital, from one surgical procedure to another, and-most importantly from one patient to another.4 During the mid1970s, the average hospital stay doubled, and the cost of hospitalization was correspondingly increased when postoperative infection developed after six common operations.5 These costs and the length of hospital stay are undoubtly lower today for most surgical procedures that are done on an outpatient basis, such as laparoscopic (minimally invasive) operations or those that require only a short postoperative stay. In these cases, most infections are diagnosed and treated in the outpatient clinic or the patient's home. However, major complications such as deep organ, tissue space infections continue to have a grave impact, increasing the duration of hospitalization as much as 20-fold and the cost of hospitalization fivefold.6 Previous authors have reported the extra charge for SSI ranging from $ 858 to $17,708 per infection and the excess length of stay attributable to SSI ranging from 1 to 44 days.7-16 The most critical factors in the prevention of postoperative infections, although difficult to quantify, are the sound judgment and proper technique of the surgeon and surgical team, as well as the general health and disease state of the patient.17 Other factors influence the development of postoperative wound infection, especially in clean surgical procedures, for which the infection rate (<3%) is generally low. Infections in these patients may be due solely to airborne exogenous microorganisms.18 Other older Indian studies by Agarwal (1972) and other authors done at different periods of time reported an overall higher infection rate ranging from 10.2% to 25%.19

Postoperative infections caused by drug-resistant pathogens are more difficult to treat.20 Mshana SE found that extended spectrum beta-lactamase (ESBL) are on the rise.21 Many Factors are responsible for this rise of resistance as discussed by Levy.22-23 Indiscriminate use of drugs, flawed hospitals protocols, infection control.24-25 This is the reason every health center should have their own policy.

II. AIMS AND OBJECTIVES

1. To find out which organism prevail at the suture site intra-operatively.
2. To evaluate the correlation of organisms present at the suture site intra-operatively and it future implication with infection if any.
3. To evaluate the differential in the spectrum of organisms when different procedures are considered.

III. RESEARCH ELLABORATIONS

History of Wound infections:

An ‘antiseptic’ operation in Dundee – note the carbolic spray

Hippocrates (Greek physician and surgeon, 460-377 BC), known as the father of medicine, used vinegar to irrigate open wounds and wrapped dressings around wounds to prevent further injury. His teachings remained unchallenged for centuries. Galen (Roman gladiator surgeon, 130-200 AD) was first to recognize that pus from wounds inflicted by the gladiators heralded heal

www.ijsrp.org
Unfortunately, this observation was misinterpreted, and the concept of pus pre-empting wound healing persevered well into the 18th century. The link between pus formation and healing was emphasized so strongly that foreign material was introduced into wounds to promote pus formation-suppuration. The first written evidence of a connection between inflammation, wound healing and suppuration is accredited to the Egyptians by Breasted as cited by Henry et al. The use of honey for chemical debridement of wounds was also the practice of Egyptian physicians, and is a treatment continued into modern times. The concept of wound healing remained a mystery, as highlighted by the famous saying by Ambroise Paré (French military surgeon, 1510-1590), “I dressed the wound. God healed it."

Koch (Professor of Hygiene and Microbiology, Berlin, 1843-1910) first recognized the cause of infective foci as secondary to microbial growth in his 19th century postulates. Semmelweis (Austrian obstetrician, 1818-1865) demonstrated a 5-fold reduction in puerperal sepsis by hand washing between performing post-mortem examinations and entering the delivery room. Joseph Lister (Professor of Surgery, London, 1827-1912) and Louis Pasteur (French bacteriologist, 1822-1895) revolutionized the entire concept of wound infection. Until the 1860’s surgical site infection was so severe that surgeons rarely operated. L’operation, comme operation a reussiemais le maladeestmort. The operation succeeded, but the patient is dead. As surgeons became more skilful, and introduction of anaesthesia in the 1850’s allowed more complex operations – so this became a more heart-rending comment. The word ‘Hospitalism’ was introduced by Sir James Simpson in Edinburgh to describe what we now call hospital-acquired surgical-site infection. Shock, erysipelas (streptococcal infection) or pyaemia (staphylococcal infection) and gangrene were the big post-operative killers. Erichsen, from University College Hospital in London provided 13 recommendations for its prevention - many of which remain valid to-day. However it was his pupil Joseph Lister who first made surgery possible using his ‘antisepic method’ based on a phenol-impregnated wound dressing. This was later developed into the ‘aseptic surgery’ of to-day. Lister recognized that antisepsis could prevent infection. In 1867, Lister placed carbolic acid into open fractures to sterilize the wound and to prevent sepsis and hence the need for amputation. In 1871, Lister began to use carbolic spray in the operating room to reduce contamination. However, the concept of wound suppuration persevered even among eminent surgeons, such as John Hunter, 1728-1793.

**Present Scenario:**

Surgical site infections can be classified into three overarching categories: Superficial incisional; deep incisional; and organ/space, or intracavitary. It is believed that the difficulty in obtaining positive culture is related to sampling the wrong locations (e.g., the associated fluids rather than surfaces) and the notion that bacteria within the bio film enter a “dormant” state due to local nutrient depletion within the biofilm. Starvation-induced “dormancy” has been shown to make bacteria in staphylococcal biofilm clusters significantly less susceptible to oxacillin. Kathju and et al. concluded that chronic SSI can arise from underlying bacterial bio films, which can invest implanted foreign bodies and associated soft tissue surfaces. Surgical site infection (SSI) is a common surgical complication; culture-negative SSI presents a particular problem in management. Examination of explanted foreign bodies (sutures) using confocal laser scanning microscopy (CLSM) and fluorescent in situ hybridization (FISH) after surgical exploration of a chronic culture-negative SSI. Confocal microscopy (CM) demonstrated bacilli and cocci attached to the surface of the explanted sutures in a mixed bio film. Florescent in situ hybridization confirmed that Staphylococci were components of the mixed bio film. Removal of the foreign bodies (sutures) resolved the chronic infection.

In prosthetic surgery, the presence of the foreign body (for example, a vascular graft after arterial bypass surgery or a prosthetic joint in orthopaedic surgery) reduces the number of pathogenic organisms required to cause an SSI. In this environment, normally non-pathogenic organisms such as Staphylococcus epidermidis (coagulase-negative staphylococcus) may also cause an SSI. Operations on sites that are normally sterile (‘clean’) thus have relatively low rates of SSI (generally less than 2%), whereas after operations in ‘contaminated’ or ‘dirty’ sites, rates may exceed 10%.

Studies of the epidemiology of SSIs are complicated by the heterogeneous nature of these infections: the incidence varies widely between procedures, between hospitals, between surgeons and between patients. Data from the United States Centers for Disease Control National Nosocomial Infections Surveillance (CDC NNIS) system show that SSIs are the third most frequently reported nosocomial infections, accounting for 14–16% of such infections among hospitalized patients and 38% among surgical patients. Similarly, European data suggest that the incidence of SSIs may be as high as 20% depending on the procedure, the surveillance criteria used and the quality of data collection. The increasing use of minimally invasive (laparoscopic) surgery has resulted in a decrease in the incidence of SSIs. For example, in patients undergoing cholecystectomy, the SSI rate following laparoscopic procedures has been reported to be 1.1%, compared with 4% following open procedures. Similarly, in patients with acute appendicitis, the SSI rate has been reported to be 2% with minimally invasive procedures and 8% with open procedures.

Possible reasons for the lower incidence of SSIs with minimally invasive procedures include the smaller incision, earlier mobilization, reductions in postoperative pain, better preservation of immune system function, and decreased use of central venous catheters.

**Definitions to Aid:**

Since skin is normally colonized by a range of microorganisms that could cause infection, defining an SSI requires evidence of clinical signs and symptoms of infection rather than microbiological evidence alone. SSIs frequently only affect the superficial tissues, but some more serious infections affect the deeper tissues or other parts of the body manipulated during the procedure. The majority of SSIs become apparent within 30 days of an operative procedure and most often between the 5th and 10th postoperative days. However, where a prosthetic implant is used, SSIs affecting the deeper tissues may occur several months after the operation. Although the outcome measure for SSI used by many studies is based on standard definitions such as those described by the Centers for Disease Control and Prevention (CDC) or the Surgical Site Infection...
Criteria for defining a Surgical Site Infection (SSI):

Superficial Incisional SSI: Infection occurs within 30 days after the operation and infection involves only skin or subcutaneous tissue of the incision and at least one of the following:
1. Purulent drainage, with or without laboratory confirmation, from the superficial incision.
2. Organisms isolated from an aseptically obtained culture of fluid or tissue from the superficial incision.
3. At least one of the following signs or symptoms of infection: pain or tenderness, localized swelling, redness, or heat and superficial incision is deliberately opened by surgeon, unless incision is culture-negative.
4. Diagnosis of superficial incisional SSI by the surgeon or attending physician. Do not report the following conditions as SSI:
   1. Stitch abscess (minimal inflammation and discharge confined to the points of suture penetration).
   2. Infection of an episiotomy or newborn circumcision site.
   3. Infected burn wound.
   4. Incisional SSI that extends into the fascial and muscle layers (see deep incisional SSI).

Note: Specific criteria are used for identifying infected episiotomy and circumcision sites and burn wounds.

Deep Incisional SSI: Infection occurs within 30 days after the operation if no implant is left in place or within 1 year if implant is in place and the infection appears to be related to the operation and infection involves deep soft tissues (e.g. fascial and muscle layers) of the incision and at least one of the following:
1. Purulent drainage from the deep incision but not from the organ/space component of the surgical site.
2. A deep incision spontaneously dehiscence or is deliberately opened by a surgeon when the patient has at least one of the following signs or symptoms: fever (>38ºC), localized pain, or tenderness, unless site is culture-negative.
3. An abscess or other evidence of infection involving the deep incision is found on direct examination, during reoperation, or by histopathologic or radiologic examination.
4. Diagnosis of an organ/space SSI by a surgeon or attending physician.

Notes:
1. Report infection that involves both superficial and deep incision sites as deep incisional SSI.
2. Report an organ/space SSI that drains through the incision as a deep incisional SSI.

Organ/Space SSI: Infection occurs within 30 days after the operation if no implant† is left in place or within 1 year if implant is in place and the infection appears to be related to the operation and infection involves any part of the anatomy (e.g., organs or spaces), other than the incision, which was opened or manipulated during an operation and at least one of the following:
1. Purulent drainage from a drain that is placed through a stab wound into the organ/space.
2. Organisms isolated from an aseptically obtained culture of fluid or tissue in the organ/space.
3. An abscess or other evidence of infection involving the organ/space that is found on direct examination, during reoperation, or by histopathologic or radiologic examination.
4. Diagnosis of an organ/space SSI by a surgeon or attending physician.

Bacteriology of Surgical Wound

In most SSIs, the responsible pathogens originate from the patient’s endogenous flora. The average human adult has 2m2 of skin surface. The chemical composition and moisture of skin varies and dictates what type of bacteria will grow on it and how much. The surface of skin (epidermis) is not a favorable place for microbial growth because it is often dry, salty, and has low pH. Most microorganisms are associated with sweat glands and hair follicles because of the moist and nutritious environment. Urea, amino acids, salts, lactic acids, and lipids are secreted through the skin and provide microorganisms with what they need to grow. Published by Clinical Reference Systems, a division of HBO & Company. Some of the most commonly found microorganisms on the skin are Corynebacterium diphtheriae, Staphylococcus aureus, Micrococcus luteus, Staphylococcus epidermis, and Pityrosporum ovale. Most bacteria are beneficial to the skin because they prevent colonization of the skin by pathogens and they control the other organisms on the skin. But if a cut is present, these bacteria can enter the body and cause damage. The most commonly isolated organisms are S. aureus, coagulase-negative staphylococci, Enterococcus spp. and Escherichia coli; however, the pathogens isolated depend on the procedure. An increasing number of SSIs are attributable to antibiotic-resistant pathogens such as meticillin-resistant S. aureus (MRSA) or Candida albicans. This development may reflect the increasing number of severely ill or immunocompromised surgical patients, and the widespread use of broad spectrum antibiotics. Pathogens may also originate from preoperative infections at sites remote from the operative site, particularly in patients undergoing insertion of a prosthesis or other implant.

Staphylococcus aureus is gram positive, forms clusters, non-motile, non-spore-forming, and is a facultative anaerobe. This bacteria tests positive for coagulase, catalase, and forms yellow colonies on agar. It is found mainly in the nose and on skin. Ailments that are caused by the bacteria include food poisoning, toxic shock syndrome, skin lesions (boils, styes, and furuncles), pneumonia, and other diseases.

Micrococcus luteus is a strict anaerobe that produces yellow to cream-white water insoluble pigment on agar (as shown above). This bacteria is nitrogen reductase negative and oxidase positive. It can cause septic shock, pneumonia, and urinary tract infections in an immune-deficient person.

Staphylococcus epidermis is non-motile, gram-positive cocci, arranged in irregular clusters, and unlike Staphylococcus aureus, it is coagulase-negative. This bacteria is an opportunistic...
Pathogens that does not cause problems unless it enters the bloodstream via cuts, catheters, or needles, for example.

*Pityrosporum ovale* is a yeast that gets into hair follicles and sebaceous glands and causes acne (shown above) and dandruff. This fungus causes the disease *Pityriasis versicolor*. It appears as dandruff or multi-colored scaley patches on the skin. UV light is used in detection of this disease, the color given off by the skin is analyzed. An increasing proportion of SSIs are caused by antimicrobial-resistant pathogens, such as methicillin-resistant *S. aureus* (MRSA), or by *Candida albicans*. The increased proportion of SSIs caused by resistant pathogens and *Candida* spp. may reflect increasing numbers of severely ill and immunocompromised surgical patients and the impact of widespread use of broad spectrum antimicrobial agents. Outbreaks of SSIs have also been caused by unusual pathogens, such as *Rhitoporus zyge*, *Clostridium perfringens*, *Rhodococcus bronchialis*, *No cardiafaricina*, *Legionella pneumophilia* and *Legionella dumoffii*, and *Pseudomonas multivorans*. These rare outbreaks have been traced to contaminated adhesive dressings, elastic bandages, colonized surgical personnel, tap water, or contaminated disinfectant solutions. When an outbreak of SSIs involves an unusual organism, a formal epidemiologic investigation should be conducted.

### Pathogens commonly associated with different surgical procedures (adapted from Mangram et al.)

#### Type of surgery

<table>
<thead>
<tr>
<th>Common pathogens:</th>
<th>Staphylococcus aureus; CoNS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Placment of graft, prosthesis</td>
<td><em>S. aureus</em>; CoNS</td>
</tr>
<tr>
<td>Cardiac</td>
<td><em>S. aureus</em>; CoNS</td>
</tr>
<tr>
<td>Neurosurgery</td>
<td><em>S. aureus</em>; CoNS</td>
</tr>
<tr>
<td>Breast</td>
<td><em>S. aureus</em>; CoNS</td>
</tr>
<tr>
<td>Ophthalmic</td>
<td><em>S. aureus</em>; CoNS; streptococci; Gram-negative bacilli</td>
</tr>
<tr>
<td>Orthopaedic</td>
<td><em>S. aureus</em>; CoNS; Gram-negative bacilli</td>
</tr>
<tr>
<td>Non-cardiothoracic</td>
<td><em>S. aureus</em>; CoNS; <em>Streptococcus pneumoniae</em>; Gram negative bacilli</td>
</tr>
<tr>
<td>Vascular</td>
<td><em>S. aureus</em>; CoNS</td>
</tr>
<tr>
<td>Appendectomy</td>
<td>Gram-negative bacilli; anaerobes</td>
</tr>
<tr>
<td>Biliary tract</td>
<td>Gram-negative bacilli; anaerobes</td>
</tr>
<tr>
<td>Colorectal</td>
<td>Gram-negative bacilli; anaerobes</td>
</tr>
<tr>
<td>Gastroduodenal oropharyngeal</td>
<td>Gram-negative bacilli; streptococci; anaerobes (e.g., <em>peptostreptococci</em>)</td>
</tr>
<tr>
<td>Head and neck</td>
<td><em>S. aureus</em>; streptococci; <em>Oropharyngeal anaerobes</em> (e.g., <em>peptostreptococci</em>)</td>
</tr>
<tr>
<td>Obstetric and Gynaecological</td>
<td>Gram-negative bacilli; enterococci; Group B streptococci; anaerobes</td>
</tr>
<tr>
<td>Urological</td>
<td>Gram-negative bacilli.</td>
</tr>
</tbody>
</table>

**MRSA (Methicillin – resistant Staphylococcus Aureus)** is a resistance strain of *Staphylococcus Aureus* microbes (example penicillin and tetracycline). It’s usually harmless, but may occasionally get into the body and cause infection. 20-30% people carry MRSA on their skin, nose and throat without knowing it, these patients are said to be colonized. The type of surgery, prolonged hospital stay and also a long time in ICU can increase the risk of being infected by MRSA. Occasionally, MRSA gets into the body through breaks in the skin such as cuts, wounds, surgical incisions or in dwelling catheters. At the same time MRSA has the ability to survive in dry, dusty environment, it can also be spread through airborne, and therefore the most common infection route between patents is health care workers. In addition to the patient’s endogenous flora, SSI pathogens may originate from exogenous sources such as members of the surgical team, the operating theatre environment, and instruments and materials brought within the sterile field during the procedure. Such pathogens are predominantly aerobes, particularly Gram-positive organisms such as *staphylococci* and *streptococci*. The risk of an SSI developing after microbial contamination of the surgical site will depend on the dose and virulence of the pathogen and the patient’s level of resistance, according to the relationship. Microbial contamination of the surgical site is a necessary precursor of SSI. 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, many 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 for most SSIs, the source of pathogens is the endogenous flora of the patient’s skin, mucous membranes, or hollow visera. 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.

### Exogenic Sources:

The source of 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 are primarily aerobes, especially gram-positive organisms (e.g., staphylococci and streptococci).

Risk of SSI=(Dose of bacterial contamination % Resistance of patient)\times \text{virulence}

The risk of SSI is considered elevated when the level of contamination exceeds 100000 organisms per gram of tissue, although lower doses may be required if foreign material such as suture is present. The virulence of the organism relates to its ability to produce toxins or other factors that increase its ability to invade or damage tissue. Mortality rates in patients infected with highly virulent pathogens such as MRSA may be as high as 74\%.

\text{It has been found that various pathogens are found with varying frequencies in case of surgical site infections as mentioned by Hidron et al. most commonly organism found being Staphylococcus Aureus. This also depends on the organ system involved. As can be derived from Mangram et al having said that very few studies discuss this variable. 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, many gram-negative bacteria produce endotoxin, which stimulates cytokine production. In turn, cytokines response can trigger the systemic inflammatory response syndrome that sometimes leads to multiple system organ failure.}

Factors Affecting SSI:

- Exogenous sources of SSI infection are usually aerobic (e.g., staphylococci) or anaerobic bacteria (e.g., E. coli), but may include strict anaerobes 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 typically the SSI isolates. Table 1 lists operations and the likely SSI pathogens associated with them. Seeding of the operative site from a distant focus of infection can be another source of SSI pathogens, particularly in patients who have a prosthesis or other implant placed during the operation.

- Such devices provide a nidus for attachment of the organism. Exogenous sources of SSI 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 (refer to “Intra-operative Issues”).

- Fungi from endogenous and exogenous sources rarely cause SSIs, and their pathogenesis is not well understood.

Pathogenesis Organisms Causing SSI - January 2006

- Staphylococcus aureus 30.0%
- Coagulate-negative staphylococci 17.8%
- Enterococcus spp. 11.2%
- Escherichia coli 9.6%
- Pseudomonas aeruginosa 5.6%
- Enterobacter spp 4.2%
- Klebsiella pneumoniae 3.0%
- Candida spp. 2.0%
- Klebsiella oxytoca 0.7%
- Acinetobacter baumannii 0.6%

Factors Affecting SSI:

- The most important aspect of surgical site infection is recognizing the risk factors in to play. These factors can be broadly divided into modifiable risk factors like shaving, bathing before surgery, aseptic precautions, antibiotic prophylaxis etc. and unmodifiable risk factors like extreme of age, immune-compromised state, and presence of contamination. These factors can again identified as Pre-operative, Intra-operative, Post-operative.

Important Modifiable Risk Factors:

- Antimicrobial prophylaxis.
- Inappropriate choice (procedure specific).
- Improper timing (pre-incision dose).
- Inadequate dose based on body mass index, procedures >3h, or increased blood loss.
- Skin or site preparation ineffective.
- Removal of hair with razors.
- Colorectal procedures.
- Inadequate bowel prep/antibiotics.
- Improper intraoperative temperature regulation.
- Excessive OR traffic.
- Inadequate wound dressing protocol.
- Improper glucose control.
- Colonization with preexisting microorganisms.
- Inadequate intraoperative oxygen levels.

IV. RISK FACTORS FOR SURGICAL SITE INFECTIONS (SSI’s)

There are four main factors which influence the infection rates in surgical wounds, they include, Patient variables, Preoperative preparation, Operative procedure and Postoperative care, as summarized in Appendix-5 (Flanagan 1997). From these factors, the following were identified as the main risk factors for SSI’s.

Patient Variables Diseases:

- The contribution of diabetes to SSI risk is controversial, because the independent contribution of diabetes to SSI risk has not typically been assessed after controlling for potential confounding factors. Also, increased glucose levels in the immediate postoperative period were associated with increased
SSI risk. More studies are needed to assess the efficiency of perioperative blood glucose control as a preventive measure. Other diseases are like 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.

**Hyperglycaemia and hypoglycaemia:**

Elevated blood sugar concentration impaired the function of phagocytic cells in experimental studies. Constant checking of blood sugar levels for patients with diabetes is important to maintain the blood sugar at a constant level. Intra-operative and postoperative blood sugar control remains a logistical problem. Furthermore, the ideal blood sugar level remains undefined. It generally is agreed that maintaining euglycemia (i.e., normal blood glucose concentration) for the patient is desirable. Future developments in real-time, on-line measurement of blood glucose will allow this conundrum to be resolved (Donald, 2007).

**Weak Immune System:**

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 postoperative nosocomial infection, impaired wound healing. 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).84

**Preoperative care**

**Prolonged Hospital Stay**

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

**Perioperative Transfusion**

Mangram and co-workers (1999) states that, it has been reported that perioperative transfusion of leukocyte-containing allogeneic blood components is an apparent risk factor for the development of postoperative bacterial infection including SSI. However, there is currently no scientific basic for withholding necessary blood transfusion from surgical patients as a means of either incisional or organ/ space SSI risk reduction.

**Operative procedure:**

**Type of Surgery:**

When a surgery has to be done on an infected wound, the chances of SSI are increased. 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 (Surgical Site Infection, 2008).

**Foreign Objects:**

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’s 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:**

According to Mangram and co-workers (1999), the type of postoperative incision care is determined by whether the incision is closed primarily, left open to be closed later, or left open to heal by second intention. When the wound is closed primarily, it is covered by sterile dressing for at least 24-48 hours therefore reducing the chances of infection. 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 patient condition may prevent primary closure, (e.g. edema at the site). When a surgical incision is left to heal by second intention, it is packed with sterile moist gauzes and covered with sterile dressing. It 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 also increasing the risk of SSI, at the same time patients receiving steroids or other immunosuppressive drugs perioperatively may be predisposed to developing SSI. Even though nicotine is found to be a reason among other patients to be the cause of surgical site infection, it’s not 100% proven to have relations with causes of SSI (Mangram et al 1999).

**Asepsis:**

There are several aseptic agents available for preoperative preparation of the skin at the incision site. 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 gross 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:**

Mobile phones have been the source of communication within the hospital. According to a recent research by Ulger, Esan, Dilek, Yanik, Gunaydin and Leblebicious (2009,1-2) 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 there are no cleaning guidelines available that meet hospital standards. However, mobile phones are used routinely all day long but not cleaned properly as health care workers may/do not wash their hands as often as they should. They found out that health care 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.
V. MATERIAL AND METHODS

STUDY CRITERIA

Inclusion Criteria:
1. Patients admitted in MGM College & Hospital.
2. Patients undergoing two types of surgeries where, CDC Classification:
   - Clean Wounds.
   - Clean Contaminated Wounds.

Exclusion Criteria:
1. Patients suffering from skin infections.
2. Patients not willing to take part.
3. Patients having Dirty Wounds

Type Of Study:
Single observer, cross sectional study, prospective study.

Methodology

1. Study will be carried out amongst the patients admitted in surgery ward in MGM Medical College And Hospital, Aurangabad.
2. Informed, Valid, Written consent will be taken after the patient is explained the study in her/his language.
3. Two groups will be considered.
   - Where minimal gastro-intestinal handling was done e.g. Appendicectomy and Cholecystectomy.
   - Abdominal wall is only opened e.g. Hernioplasty, Ventral wall Lipoma Hernia.
4. Pre-Operative, Intra-Operative, Post-Operative variables like antibiotic, time of shave and preparing parts will be standardized.
5. Sample will be taken from two sites, operatives field and sub cutaneous planes just wash is given and closure done.
6. Fever will be noted if present.
7. If drain is put, drain fluid culture and sensitivity will be done if signs of infections manifests.
8. Sterile swabs will be used to collect samples from suture site at three different places, the ends and middle portion.
9. Culture will be done on according to standard procedure protocol as followed in MGM College Micro-Biology Department.
10. Routine Sensitivity for standard drugs will be done.
11. Different data collected in correlated tabular forms and analytically processed.
12. Post-Operative swabs will be collected from
   - Serous or Non-Purulent Discharge from the wound.
   - Pus Discharge from wound.
   - Serous or Non-Purulent Discharge from wound with signs of inflammation.
   - Wound deliberately opened by surgeon to drain collection (serous/purulent).

Implications:
1. If the rate of infection and organisms cultured are in accordance with national and international studies.
2. The organisms cultured remain the same irrespective of which organ system was operated on.
3. If the organisms cultured from the drain fluid and suture site overlap for a given patient.
4. If the Sensitivity and Resistance profile match accepted standards for the given set up.
5. What was the condition of the suture site on the day of discharge, either healthy, infected, slough formation.

VI. OBSERVATIONS AND RESULTS

The prospective study involves culture and sensitivity of 114 patients undergoing Clean (Group A), Clean and Contaminated (Group B) surgeries in the Department Of Surgery at our institute. Patients where from the three general surgical units, and the first procedure of the day in the three allotted operating rooms. Swabs were collected by the investigator. Age Distribution of the subjects is given below.

### Table – 1
AGE DISTRIBUTION OF PATIENTS

<table>
<thead>
<tr>
<th>Age Distribution</th>
<th>Group A</th>
<th>Group B</th>
<th>Total</th>
<th>Percentage % (n=114)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 – 10</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>0.89</td>
</tr>
<tr>
<td>11 – 20</td>
<td>3</td>
<td>7</td>
<td>10</td>
<td>8.8</td>
</tr>
<tr>
<td>21 – 30</td>
<td>21</td>
<td>11</td>
<td>32</td>
<td>28.07</td>
</tr>
<tr>
<td>31 – 40</td>
<td>15</td>
<td>8</td>
<td>23</td>
<td>20.17</td>
</tr>
<tr>
<td>41 – 50</td>
<td>13</td>
<td>10</td>
<td>23</td>
<td>20.17</td>
</tr>
<tr>
<td>51 – 60</td>
<td>8</td>
<td>4</td>
<td>12</td>
<td>10.53</td>
</tr>
<tr>
<td>61 – 70</td>
<td>4</td>
<td>6</td>
<td>10</td>
<td>8.8</td>
</tr>
<tr>
<td>71 - 80</td>
<td>2</td>
<td>1</td>
<td>3</td>
<td>2.63</td>
</tr>
</tbody>
</table>

The maximum number of cases are between 21 to 60 yrs (78%). The mean age in Group A was 39.2 years while in Group B was 39.8 years. Chart-1 shows the mean time required to finish the procedure from incision to skin closure. The mean stay for Group A was 4.7 days and Group B was 5.9 days.

**Chart-1 : Average Age, Time required for procedure, Hospital stay.**

![Average Age, Time required for procedure, Hospital stay](image-url)
Incidentally as observed in Chart 2 the sex ratio is quite evenly matched. 63 male patients and 51 female patients in all. Amongst the 114 subjects Group A comprised of 58.77% (67) while Group B consisted of 41.22% (47). 13 out of 67 in Group A came back as sterile, in Group B sterile samples were 10. Illustrated in Chart-3.

**TABLE – 2**

<table>
<thead>
<tr>
<th>Column1</th>
<th>Culture Positive</th>
<th>Bi-Microbial</th>
<th>Organisms Cultured</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>54</td>
<td>10</td>
<td>64</td>
</tr>
<tr>
<td>Group B</td>
<td>37</td>
<td>11</td>
<td>48</td>
</tr>
<tr>
<td>Total</td>
<td>91</td>
<td>21</td>
<td>112</td>
</tr>
</tbody>
</table>

Amongst all the culture positive examples, 10 in Group A and 11 in Group B grew two types of micro-organisms on a single culture. In gist total organisms cultured in Group A are 64 and Group B are 48. So for all statistical analysis the total will be 64 and 48 for Group A and Group B respectively. As one of the two organism is bound to give rise to the infection.
Chart 4 shows the detailed analysis of the different types of organism cultured in the two groups. Most common being Staph. Aureus, P.Aeurignosa, Klebseilla and E.coli in descending order in Group A. Group B has Staph. Aurues, E.coli being the most common followed by P.Aeurignosa, Klebseilla. While Gram Negative Bacilli, Protues, Pseudomonas species, Citobacter were cultured in different frequencies.

Chart 5 : Proportion of M.R.S.A among Stap. Aureus

<table>
<thead>
<tr>
<th>Column 1</th>
<th>Group A</th>
<th>Group B</th>
</tr>
</thead>
<tbody>
<tr>
<td>S.A</td>
<td>25</td>
<td>12</td>
</tr>
<tr>
<td>M.R.S.A</td>
<td>6</td>
<td>5</td>
</tr>
<tr>
<td>Total</td>
<td>31</td>
<td>17</td>
</tr>
</tbody>
</table>

Chart 5 states the Methicillin Resistant Stap. Aureus and the total number of Stap. Aureus in the two groups.

Chart – 6 Percentage of most common organisms cultured.

TABLE – 3
SENSITIVITY & RESISTANCE PROFILE OF MOST FREQUENT ORGANISMS

<table>
<thead>
<tr>
<th>Organism</th>
<th>Group A</th>
<th>Group B</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. Aureus</td>
<td>Linezolid- 26</td>
<td>Co-Trimoxazole- 5</td>
</tr>
<tr>
<td></td>
<td>Amikacin- 25</td>
<td>Ciprofloxacin-17</td>
</tr>
<tr>
<td>P. Aeruginosa</td>
<td>Doxycycline-25</td>
<td>Clindamycin-9</td>
</tr>
<tr>
<td></td>
<td>Ciprofloxacin-17</td>
<td>Amikacin-12</td>
</tr>
<tr>
<td></td>
<td>Penicillin-15</td>
<td>Linezolid-12</td>
</tr>
<tr>
<td></td>
<td>Clindamycin-17</td>
<td>Clindamycin-9</td>
</tr>
<tr>
<td>P. Aeruginosa</td>
<td>Levofloxacin-14</td>
<td>Erythromycin-7</td>
</tr>
<tr>
<td></td>
<td>Linezolid-6</td>
<td>Amikacin-11</td>
</tr>
<tr>
<td></td>
<td>Cefuroxime-5</td>
<td>Cefuroxime-3</td>
</tr>
<tr>
<td>E. Coli</td>
<td>Imipenum- 5</td>
<td>Amox-Clav-5</td>
</tr>
<tr>
<td></td>
<td>Cefuroxime-4</td>
<td>Piper- Tazo -5</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>Imipenum-9</td>
<td>Cefoperazone-6</td>
</tr>
<tr>
<td></td>
<td>Cefuroxime-9</td>
<td>Co-Trimoxazole-4</td>
</tr>
<tr>
<td></td>
<td>Levo Flaximycin- 5</td>
<td>Piper- Tazo -5</td>
</tr>
</tbody>
</table>

Table-3 discuss the drug sensitivity and resistance profile of the most commonly cultured organism amongst the two groups.
Chart 7 denotes the frequency of different organisms cultured for a particular procedure. Four most common procedures have been included in the analysis.

**TABLE – 4**
**SURGICAL SITE INFECTION RATE**

<table>
<thead>
<tr>
<th></th>
<th>Infected</th>
<th>Infection Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A (n=67)</td>
<td>4</td>
<td>5.97%</td>
</tr>
<tr>
<td>Group B (n=47)</td>
<td>4</td>
<td>8.51%</td>
</tr>
</tbody>
</table>

Table 4 denotes the number of cases which prospectively showed signs of infection and second swab were sent for the same.

**TABLE – 5**
**POST PROCEDURE INFECTION ANALYSIS**

<table>
<thead>
<tr>
<th>Culture</th>
<th>Sensitive</th>
<th>Resistantat</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>S.A.</td>
<td>Lin, Doxy, Eryt, Levo, Clin, Cefoxitin</td>
</tr>
<tr>
<td></td>
<td>S.A</td>
<td>Lin, Doxy, Clin, Ami, Levo</td>
</tr>
<tr>
<td></td>
<td>P.A</td>
<td>Lin, Ami, Doxy</td>
</tr>
<tr>
<td></td>
<td>Sterile</td>
<td></td>
</tr>
<tr>
<td>Group B</td>
<td>P.A</td>
<td>P iT, Czidin, Ami</td>
</tr>
<tr>
<td></td>
<td>S.A.</td>
<td>Eryt, Levo, Clin, Cipro, Lin Ami, Doxy</td>
</tr>
<tr>
<td></td>
<td>Gram -ve Bacilli</td>
<td>Imi, Ami, P iT,</td>
</tr>
<tr>
<td></td>
<td>S.A</td>
<td>Lin, Clin, Eryt, Cifoxitin</td>
</tr>
</tbody>
</table>

Table 5 gives the break of the results of all the swab cultures sent for the infected cases.

**VII. DISCUSSION**

The impact of surgical site infection on increasing the morbidity on already strained health care system and its burden on the society is well known. Culture and sensitivity studies have played a pivotal role in better understanding of the SSI for the medical care giver and researchers. Routine periodic Culture and Sensitivity of the fomites present in the operating room to monitor the bacteriological flora has been employed for many years and is the current standard practice all over the globe. In this study our quest has been to make a pilot analysis of the
organisms grown from the swabs taken from the operating plane and subcutaneous tissue.

All the 114 cases were randomly selected from the three general surgery units, patients were free of any confounding risk factors like diabetes mellitus, known immune compromised state, skin infections, etc. None of the procedures were emergency. Procedures were carried out in the three allotted theatres for general surgery department, and all the 114 cases where the first procedure of the day so as to eliminate the bias of more contaminated OR environment as the day progresses other aspects are control of operating room by restricting the number of people allowed in the operating room, closing the doors to the operating room to prevent in and out traffic, and limiting unnecessary movement and talking once in the operating room. This was all done in an attempt to get a simple standardization.86,87,88

The most common organisms cultured were S. Aureus, P. Aeruginosa, Klebsiella and E. Coli. percentage of organisms shown in Table-6.

<table>
<thead>
<tr>
<th>Group A (n=64)</th>
<th>Group B (n=48)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staph. Aureus</td>
<td>48.44%</td>
</tr>
<tr>
<td>P. Aeruginosa</td>
<td>17.19%</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>17.19%</td>
</tr>
<tr>
<td>E. Coli</td>
<td>7.81%</td>
</tr>
<tr>
<td>Sterile Cultures</td>
<td>20.31%</td>
</tr>
</tbody>
</table>

These are the organisms cultured from the subcutaneous plane and operating plane. While the organisms cultured from the post-operative surgical site were similar as seen in Table-5. This difference has to be kept in mind throughout the discussion. The results are more or less in accordance to what others found in their studies of postoperative infections. In a study conducted in Pondicherry by Dr. A.Ramesh et al, they found that the Staph.Aureus and Klebsiella were the most predominant cultures. S.Aureus 30% in Clean, 50% in Clean Contaminated cases. While Klebsiella 16.67% in Clean, 30% in Clean Contaminated cases.89 In a tertiary care hospital in India the most common organisms cultured were S.Aureus, E.Coli, Pseudomonas.90 Similarly Dr. Lilani from Grant Medical College & Sir J.J. Group of Hospitals found S.Aureus & P.Aeruginosa to be the commonest Cultured Organisms in SSI.19 In our study the Post-operative infection rates were 5.97% in clean cases and 8.51% in clean contaminated cases as shown in Table-4. Similarly study by Lilani found SSI rate as 3.03% & 22.41%.

Table-7 shows the postoperative wound site infection rate of few of the studies under taken in India and Other developing countries.

<table>
<thead>
<tr>
<th>Infection Rates in Various Studies</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>In India</strong></td>
</tr>
<tr>
<td>Anvikar, GMC, Aurangabad clean surgeries 4.04% clean contaminated 10.06%</td>
</tr>
<tr>
<td>Mumbai 8.95%</td>
</tr>
<tr>
<td>S.Sahu(2009) E.Coli Most Common Elective abdominal surgery</td>
</tr>
<tr>
<td>Agarwal (1972) 10.20%</td>
</tr>
<tr>
<td>Rao Venkataraman MS. (1975) 25%</td>
</tr>
<tr>
<td><strong>Across the borders</strong></td>
</tr>
<tr>
<td>Wanger MB et al.(1997) Brazil 8.70%</td>
</tr>
<tr>
<td>Soleto et al. (2003) 12%</td>
</tr>
<tr>
<td>Bolivia 25%</td>
</tr>
<tr>
<td>Peru (2005) 25%</td>
</tr>
</tbody>
</table>
In our study S. Aureus was found to be sensitive to Linezolid, Clindamycin, Amikacin, Doxycycline both in Group A and Group B in different frequencies. While same S. Aureus was found resistant to Co-Trimoxazole, Ciprofloxacain and Penicillin. Jyoti et al in a tertiary hospital in Mumbai found Linezolid and Vancomycin to be most effective. Similarily in Italy the antibiogram showed Vancomycin and Teicoplanin to be most effective.

In regards to the Gram Negative Organisms i.e Pseudomonas and Klebsiella, Amikacin, Piperacillin Tazobactum, Levofloxacin, Imipenum where most effective. While Cefuroxime, Amoxycillin Clavulanic acid where most ineffective. Even these results are in accordance to the above two studies we have compared. Highest resistance was found to be to penicillin which was once the mainstay of treating bacteriological infection.

**TABLE – 8 GROUP-A : ANALYSIS FOR ORGANISMS IN CASE OF INFECTION**

<table>
<thead>
<tr>
<th>Master Chart Sr. No</th>
<th>Diagnosis</th>
<th>Intra Operative site culture</th>
<th>Post procedure infection culture</th>
</tr>
</thead>
<tbody>
<tr>
<td>28</td>
<td>Thigh Lipoma</td>
<td>P. Aeruginosa</td>
<td>S. Aureus</td>
</tr>
<tr>
<td>16</td>
<td>Incisional Hernia</td>
<td>P. Aeruginosa, Klebsiella</td>
<td>S. Aureus</td>
</tr>
<tr>
<td>21</td>
<td>Rt. Fibroadenoma</td>
<td>S. Aureus</td>
<td>P. Aeruginosa</td>
</tr>
<tr>
<td>15</td>
<td>Rt. M.R.M</td>
<td>S. Aureus</td>
<td>Sterile</td>
</tr>
</tbody>
</table>

**TABLE – 9 GROUP-B : ANALYSIS OF ORGANISMS CULTURED IN CASE OF INFECTION**

<table>
<thead>
<tr>
<th>Master Chart Sr. No.</th>
<th>Diagnosis</th>
<th>Intra Operative site culture</th>
<th>Post procedure infection culture</th>
</tr>
</thead>
<tbody>
<tr>
<td>55</td>
<td>Acute Appendicitis</td>
<td>S. Aureus, E. Coli</td>
<td>P. Aeruginosa</td>
</tr>
<tr>
<td>17</td>
<td>Cholecystitis</td>
<td>S. Aureus</td>
<td>S. Aureus</td>
</tr>
<tr>
<td>105</td>
<td>Cysto-Gastrostomy</td>
<td>Klebsiella</td>
<td>Gram –ve Bacilli</td>
</tr>
<tr>
<td>62</td>
<td>Resection Anastomosis</td>
<td>E.Coli</td>
<td>S. Aureus</td>
</tr>
</tbody>
</table>

From Table-8 and 9 we have deduced that out of the 8 number of cases of surgical site infection only once the Intraoperative site organism and post procedure infection site organism was found to be same in case of Patient Serial number 17 that is 12.5% of the 8 cases. Various Factors are responsible for this variation determination of which is out of scope of this study. While we got one postoperative sterile culture even when SSI was present. Rasnake MS et al have concluded that the potential causes of culture-negative SSIs include prior antimicrobial therapy; the presence of fastidious or slow-growing microorganisms such as mycobacteria, Mycoplasma spp., and Legionella spp.; infection caused by mundane bacteria that may
be dismissed as "contaminants"; factitious infection; and others.100

Throughout the exercise we have realized the need of a more elaborate protocol required for all the operations theatres. To include other surgical departments. Find out if their results match ours. Include more number of cases and undertake a longer study duration. We have excluded patients with confounding factors, it will be imperative to find out the course of SSI in these patients. Undertaking the recommendations of various studies, evidence based strategies, and a in house protocol for preventing and managing surgical site infections remains the ultimate goal.

VIII. SUMMARY AND CONCLUSIONS

The present study was done to analyze the organisms cultured at suture site intraoperative. Clean (Group A) and Clean Contaminated (Group B) where included in the study. To find the sensitivity & resistance profile of the same. We have noted the percentage of post-operative infections in both the groups.

1. Amongst the 114 patients in my study Mean Age in Group A and B was 39.2 yrs. and 39.8 yrs. respectively. There were 34 males in Group A and 29 males in Group B while 33 females in Group A and 28 females in Group B.

2. The mean stay in Group A was 4.7 days and 5.9 in Group B, the mean procedure time was 59.18 minutes and 72.55 minutes in Group A and Group B respectively.

3. Amongst all the organisms cultured Staphylococcus Aureus, P. Aeruginosa, Klebsiella and E. Coli were the most common cultured.

4. The sensitivity and resistance profile was more or less similar to national and international studies.

5. The most common procedure in the study were Acute Appendicitis, Inguinal Hernia, and Fibroadenoma.

6. The post-operative procedure rates were 5.97% in Group A and 8.51% in Group B.

7. Amongst the 8 cases of SSI only once the organism

REFERENCES


www.ijsrp.org


AUTHORS

First Author – Dr. Pravin Suryawanshi- Porf. & Head OF Department, Chief Endoscopist , M.G.M Medical College, Aurangabad.

Second Author – Dr. A. Q. Khan- AssoProf. Head Of Unit,Surgery Dept. Laparoscopic Surgeon M.G.M Medical College, Aurangabad.

Third Author – Dr. Saiyyad Altaf G - Lecturer, Surgery, M.G.M. Medical College, Aurangabad.

Fourth Author – Dr. Amey Patil- hief Resident, Surgery, M.G.M. Medical College, Aurangabad. E-mail – ameypatil@gmail.com, Ph. +91 7620048002