Management of Infections with Mycobacterium Other Than Tuberculosis (MOTT) as a Complication of Surgical Procedures.

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Research Article

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Abstract

BACKGROUND:

Atypical mycobacteria (MOTT) have emerged as significant human pathogens, causing post-surgical wound infections. The aim of this study is to assess the causative organisms of such infection and their treatment response.

METHODS:

After matching the criteria, 28 cases were taken for this prospective, single center, observational study. The diagnosis was confirmed by bacterial culture.

RESULTS:

Among 28 patients, 16 were females (median age of 45.5 years). Patients had undergone laparoscopic cholecystectomy (n=13), laparoscopic appendicectomy (n=3), laparoscopic hernioplasty (n=2), open appendicectomy (n=2), open mesh hernioplasty (n=7), exploratory laparotomy (n=1). No major comorbidities or immunosuppression was identified. All patients were initially treated with repeated incision and drainage and started conventional antibiotics until culture and sensitivity report was available. All except one patient had culture confirmed MOTT infection. Combination antibiotics (clarithromycin, linezolid and ofloxacin) given for initial 3 months. 12 well responded within 3 months. 9 required additional few months to get complete cure. 4 patients cured after 6 months of treatment and 3 patients did not come for follow up.

CONCLUSIONS:

Delayed onset chronic wound infection by Atypical mycobacteria is preventable. These organisms are not responsive to conventional antitubercular drugs but to specific drug regimens.

Introduction

Postoperative infections caused by saprophytic Mycobacterium are not uncommon, and their prevalence is increasing. The clinical presentation includes cellulitis, abscess formation, draining sinuses, and postoperative wound infection. Patients often do not have fever, chills, or other manifestations that are characteristic of a systemic infection, which makes clinical diagnosis difficult. Mycobacteria are ubiquitous and have been found in both tap water and soil as well as skin surfaces on man and animals. There are controversies regarding the pathways of infection. It is believed that contaminated surgical instruments, reusable surgical instruments in particular, such as the Laparoscopic instruments might be the possible sources of infection. Mycobacteria usually cannot be thoroughly killed with the usual disinfectants, and surgical equipment must be autoclaved. If surgical instruments are contaminated with Mycobacteria and cannot be autoclaved, special sterilization methods should be used. Although autoclaving surgical instruments is the best method for preventing Mycobacterial contamination of wounds, reusable instruments such as cannulas, tubings, and fibreoptic cables that can be damaged by autoclaving should be immersed completely in 2% activated gluteraldehyde for minimum 10 minutes to destroy vegetative organisms or minimum 10 hours it will destroy all forms of microbes and or ortho-phthalaldehyde solution for minimum 5 minutes (for high level disinfection) or sterilized with ethelene oxide gas (36 to 48 hours). In addition, mucus or residue from cells on surgical instruments may lead to a significant decrease in disinfecting activity, which means that decontaminating baths must be renewed according to their frequency of use. Rinsing
instruments in sterilized distilled water or with 70% alcohol after disinfection and before use is an excellent method of preventing instrument colonization. Because most MOTT (Mycobacterium Other Than Tuberculosis) infections are iatrogenic, adequate sterilization of surgical instruments are needed. It is known that effective treatment of MOTT infection includes surgical treatment concurrent with combination antibiotic therapy. An antibiotic combination based on susceptibility of individual isolates must be administered for a sufficiently long period of time to ensure complete wound healing and no recurrence. Conventional anti-tubercular drugs have been reported to be ineffective.

It has been shown that atypical Mycobacteria, particularly \textit{M. chelonae} and \textit{M. fortuitum}, both of which belong to the group of rapidly growing \textit{Mycobacterium}, widely colonize soil and water. This \textit{Mycobacterial} complex primarily presents itself as localized cutaneous infection 3–4 weeks after surgery. There is, however, very little evidence of disseminated disease following infection with this complex except in immunocompromised hosts. These bacteria have an affinity for the dermis and the subcutaneous area. Protective factors within the peritoneum destroy the \textit{Mycobacterium} and prevent infection within the peritoneal cavity. In theory, these microorganisms can be isolated through culture of affected tissue but takes a longer time to grow and is difficult to culture.

Infections with atypical \textit{Mycobacterium} have been primarily reported after laparoscopic procedures. A recent study has shown that atypical \textit{Mycobacterium} such as \textit{M. chelonae} and \textit{M. smegmatis} are showing increased resistance to these chemicals due to defects in porin expression in the bacterial cell walls. Porins are cell wall proteins found in \textit{Mycobacteria} and have been known to create channels that allow the passage of small hydrophilic molecules, such as antibiotic drugs, through the highly hydrophobic \textit{Mycobacterial} cell wall. Mutations leading to defects in porin expression prevent the delivery of antibiotics such as β-lactams, fluoroquinolones and chloramphenicol into the \textit{Mycobacterial} cell thus conferring drug resistance.

Surgical wound infection by atypical Mycobacteria may be of following types:

1. Portsite infection: Wound discharge, nodule and erythema around the port site are the most common presentation of non-mycobacterial infection. There are five clinical stages of atypical mycobacterial port site infection (PSI).

   \textit{First stage}: A tender nodule appears in the vicinity of the port site, and its usual timing of appearance is around four weeks following the surgery.

   \textit{Second stage}: Increase in the size of the nodule, and increased tenderness of the site along with other signs of inflammation with eventual formation of a discharging sinus.

   \textit{Third stage}: Reduced pain sensation following discharge of the purulent material and necrosis of the skin surrounding the port site.

   \textit{Fourth stage}: Chronic sinus discharging white or serous fluid.

   \textit{Fifth stage}: Hyper-pigmentation of the skin surrounding the sinus and appearance of multiple nodules at different places.

2. Wound infection following mesh hernioplasty: Usual presentation is chronic discharging sinus. Discharge is usually serous in nature.
The objective of this study is to assess the causative organisms of such infection and to study their treatment response.

**Methods**

This is an institution based (single center) prospective, observational study. Ethical approval for the study was obtained from the Institutional Ethics Committee, NRSMCH, Bankura, West Bengal, India. The study population comprised of 28 cases of surgical wound infection (who undergone surgery from January 2017 to June 2018 in general surgery department of N. R.S medical college) presenting with signs of persistent inflammation at the incision site and no pyogenic organism was recovered. The cases did not respond to usual antibiotics and dressing for more than 4 weeks. They had granulomas in tissue obtained from the surgical wound or surrounding structures or a positive acid-fast staining but negative culture for *Mycobacteria*. The subjects satisfied the inclusion and exclusion criteria mentioned below. The primary data for this study were the investigation reports of the subjects.

**INCLUSION CRITERIA:**

Patients with postoperative wound infections with signs of inflammation of the skin and abscesses or drainage at the wound site in addition to not responding to abscess drainage and antibiotics used for pyogenic infections. And organisms isolated from an aseptically obtained culture of fluid or tissue from the incision.

**EXCLUSION CRITERIA:**

Exclusion criterion was all acute postoperative wound infections of less than 3 weeks from the time of surgery.

**STUDY TECHNIQUE**

Based on detailed history, thorough clinical examination provisional diagnosis was made. Patients were then subjected to investigations such as routine investigations: Haemoglobin, Total Leukocyte Count, Differential Count, Blood urea, Creatinine, fasting blood glucose, LFT, viral markers and special investigations: Gram staining, AFB staining, AFB culture of the secretions from the wound site. Antibiotic treatment and surgical treatment were then offered according to study protocol. Each case was observed regarding outcome of management. They were followed up in the OPD (General surgery) to assess completeness of wound healing.

**MANAGEMENT DETAILS:**

Combination antibiotic therapy plus surgical treatment were applied to all the patients. But neither the standard regimen nor the exact dosage of antibiotics has been firmly established.

1. **ANTIBIOTIC THERAPY:**

An antibiotic combination based on susceptibility of individual isolates was administered for a period of time to ensure complete wound healing and no recurrence. Conventional antitubercular drugs have been reported to be ineffective. The choice of antibiotics depends on in-vitrosusceptibility patterns. Some antimicrobial agents, including quinolones, amikacin have been found to be effective against *M. fortuitum*

2. **SURGICAL MANAGEMENT:**

a) Port site infection: Surgical excision of skin at port site under local anesthesia.
b) Mesh infection: removal of mesh – debridement of wound – wound to be kept open – waiting for healthy granulation tissue to appear > secondary suturing of the wound.

The statistical analysis was carried out using available standard statistical software. Odds ratio with 95% confidence interval (CI) and multivariate analysis has used to establish the interrelationships between pre-operative and intra-operative findings. All statistical tests have two tailed and P value <0.05 has taken as significant.

**Results**

Total twenty eight patient with post-operative delayed wound infection with MOTT were identified. All of the 28 patients met inclusion criteria were selected in the study after obtaining written consent. The following observations were made during the course of the study according to age. *[Table 1]*. Median value of age distribution was 45.5 years.

| AGE (YEAR) | NO. OF PATIENTS |
|------------|-----------------|
| 18 - 30    | 4               |
| 31 - 40    | 6               |
| 41 - 50    | 9               |
| 51 - 60    | 7               |
| 61 - 70    | 2               |
| TOTAL      | 28              |

The majority of patients were female (16 females and 12 males) with median age of 45.5 year (range 18 –70). *[TABLE 2]*

| SEX      | NO  |
|----------|-----|
| MALE     | 12  |
| FEMALE   | 16  |
| TOTAL    | 28  |

Those patients had undergone different surgeries like laparoscopic cholecystectomy (n = 13), laparoscopic appendicectomy (n = 3), laparoscopic hernioplasty (n = 2), open appendicectomy (n = 2), open mesh hernioplasty
(n = 7), exploratory laparotomy (n = 1). No major disease comorbidities or causes of immunosuppression (e.g., HIV infection) were identified except three patients were diabetic and two were hypertensive. TABLE 3

| SURGERY                                | NO |
|----------------------------------------|----|
| LAPAROSCOPIC CHOLECYSTECTOMY           | 13 |
| LAPAROSCOPIC APPENDICECTOMY            | 3  |
| LAPAROSCOPIC MESH HERNIOPLASTY         | 2  |
| OPEN MESH INGUINAL HERNIOPLASTY        | 7  |
| OPEN APPENDICECTOMY                    | 2  |
| EXPLORATORY LAPAROTOMY                 | 1  |

All the patients were initially treated with repeated incision and drainage and were started with conventional antibiotics like amoxicillin/ clavulanic acid or second or third generation cephalosporin until culture and sensitivity report available. [FIGURE 1] shows wound of astudy subject, who underwent exploratory laparotomy followed by splenectomy in a case of blunt trauma abdomen. Wound infection occurred after 2 months following operation.

And [Figure 2] shows the wound of same patient after 3 months of continued combination antibiotic treatment and repeated debridement.

Skin findings varied widely, including sinus tracts, non-healing ulcers, subcutaneous abscesses or firm nodules of varying size and erythema or chronic discharge from prior surgical wound.

All patients had cultured- confirmed MOTT except one who was only AFB positive by ZN stain. Species identified were *Mycobacterium fortuitum* and *M chelonae*.

After getting the culture and sensitivity reports, combination antibiotics (clarithromycin, linezolid and ofloxacin) given for initial 3 months. 12 out of 28 patients well responded with 3 months of treatment. 9 patients required additional few months to get complete cure. Four patients cured after 6 months of treatment and three patients did not come for follow up. Antibiotics were continued for additional three months after patient got cure to prevent relapse of infection. [TABLE 4]. A comparative analysis of present study with other studies is given in [Table 5]
Table 4: Distribution of study population according to clinical outcome

| OUTCOME                                         | NO. |
|------------------------------------------------|-----|
| CURED                                           | 21  |
| CURED AFTER 6 MONTHS OF TREATMENT               | 4   |
| LOST FOLLOW UP                                  | 3   |

**Discussion**

An antibiotic combination is better than single-drug therapy in case of postoperative infection by atypical Mycobacteria. [9]

Recent studies have demonstrated that clarithromycin, cefoxitin, and imipenem were also effective against these microorganisms [10,11,12]. Based on reported NTM [MOTT] sensitivities, amikacin with ciprofloxacin or amikacin with cefoxitin [10] should be administered to the patient while waiting for microbial sensitivity results, if there is a strong suspicion of atypical mycobacterial infection [10,13]. According to results of a study from Brown-Elliott and Wallace, the *M. fortuitum* group is less drug resistant than *M. abscessus* and *M. chelonae*. [13] Thus, treatment of infections caused by the *M. fortuitum* group has been much easier and generally more effective than treatment of *M. abscessus* and *M. chelonae* infections. Postoperative wound infections caused by MOTT generally appear few weeks to some months following the procedure. [14] In some case series the incubation period ranged from 20 to 66 days. On the contrary infections due to other pyogenic bacteria have a shorter incubation period as compared to MOTT which have a longer incubation period ranging from several days to several months. [8] The absence of clinical response after the administration of antimicrobial agents against commonly invading bacteria (e.g., Staphylococci, Streptococci) and the sterility of routine cultures of samples taken from the infected sites were clues for MOTT infection.

Time between the onset of symptoms and the microbiological diagnosis also took long time. Therefore, a high index of suspicion is imperative to make the diagnosis. A study by Joon Young Song *et al.* stated that since the symptoms are relatively mild and indolent, the clinical diagnosis of atypical mycobacterium is often delayed and took more than two months from initial manifestation. [15] Also, in the revised literature, most publications conclude that clinical diagnosis of mycobacterial skin and soft tissue infections is not easy to perform and that the diagnosis is often delayed. Delays of more than one year have been reported. A high degree of clinical suspicion and appropriate microbiological techniques are necessary to avoid delays in diagnosis. [10]

Clinically the infections caused by MOTT in post-operative wound infections are almost similar to pyogenic abscesses with induration, micro-abscesses, and discharge from sinuses and erythema. Systemic manifestations
| Author                  | Size of study population | Clinical Features                                                                                                                                                                                                 | Bacteriological Features                                                                                                                                                  | Management                                                                                                                                 |
|-------------------------|--------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Jury B. Kaltia et al [18]| 25                      | Delayed onset post-operative wound infection (3 weeks after surgery), appearance of erythema and oedema followed by breakdown of wound and suppurating / discharging sinuses, absence of symptoms / illness and lack of response to antibiotics used for pyogenic infections. | Swab from 20 out of 25 patients revealed growth of rapidly growing *Mycobacterium* spp. (*M. fortuitum-chelonae* complex). Remaining 5 culture negative cases received prior treatment with one or the other fluoroquinolones which are known drugs for treatment of mycobacterial infection. | Only 7 patients who turned up in the OPD could be followed up and all of them were cured when treated with fluoroquinolones along with clarithromycin and amikacin for 2 to 3 months along with vigorous surgical debridement. |
| Julio et al [19]        | 1, who underwent laparoscopic inguinal herniorrhaphy | The local findings were erythema, local warmth, painful nodules and minimal purulent discharge from deep fistulous tract. No improvement was observed even after 10 days of treatment and local dressings with 10% povidone iodine. | Microscopic examination of biopsy material showed acid fast bacilli in Ziehl Neelsen stained smear. Culture results were positive for mycobacteria. Polymerase chain reaction (PCR) restriction enzyme analysis of the hsp65 gene, rpoB partial gene sequencing identified the strain as *Mycobacterium massiliense*. | This patient underwent mesh removal with debridement of extensive granulomatous inflammatory tissue in pre-peritoneal pelvic area. The antibiotic scheme consisted of clarithromycin, amikacin, and minocycline for 3 months and thereafter clarithromycin, minocycline and moxifloxacin. Antibiotics were maintained for additional 3 months. Patient had a full recovery following this course of treatment. |
| Mahvash Haider et al [20]| A 40 years old immunocompetent female underwent laparoscopic cholecystectomy | Three weeks later she developed port site granuloma with persistent seropurulent discharge. Empirical oral antibiotics were started but provided no relief. | After 3 months the wound was explored and tissues were sent for histopathological examination and smear microscopy along with culture-sensitivity. *Mycobacterium abscessus* was identified. | Amikacin and Clarithromycin was started for a period of one month. Then clarithromycin alone was continued. Discharging sinuses improved and healed completely by the end of six months of clarithromycin therapy. |
| Kavitha Kannaiyan et al [21] | 19 patients (13 female and 6 male patients, aged 18 to 60 years) who underwent laparoscopic cholecystectomy, hernioplasty, diagnostic | Abscess and chronic non healing wound infection. | *Mycobacterium fortuitum* and *M. chelonea* were predominantly found. | All patients underwent surgical procedure to drain existing abscesses, removal of immature nodule, removal of mesh and wound debridement. Linezolid and |
laparoscopy, reduction mammoplasty | clindamycin were started for all patients. Of 19 patients 11 patients received clarithromycin for 3 months, 5 patients for 5 months, 2 patients on and off for 24 months and 1 patient was lost follow up. 16 patients were completely cured and 2 patients were not cured in this study

| JS Rajkumar et al[22] | 38 years old lady who underwent laparoscopic ovarian cystectomy | Few weeks later, the patient developed multiple discharging sinuses and abscesses. | Non mycobacterium tuberculosis rapid grower | Patient underwent eight successive surgical explorations for multiple skin and soft tissue sinuses along with medicinal treatment of NTM (amikacin, clarithromycin and ofloxacin). Treatment was continued for a period of six months. Abdominoplasty and meshplasty was done 2 months after complete control of infection

| Present study | 28 patients. patients had undergone different surgeries like laparoscopic cholecystectomy ( n=13 ), laparoscopic appendicectomy ( n=3), laparoscopic hernioplasty ( n=2 ), open appendicectomy ( n=2 ), open mesh hernioplasty ( n= 7 ), exploratory laparotomy | Skin findings varied widely, including sinus tracts, non-healing ulcers, subcutaneous abscesses or firm nodules of varying size and erythema or chronic discharge from prior surgical wound. All the patients were initially treated with repeated incision and drainage and were started with conventional antibiotics like amoxicillin/clavulanic acid or second or third generation cephalosporin until culture and sensitivity report available. | All patients had cultured confirmed MOTT except 1 who was only AFB positive by ZN stain. Species identified were Mycobacterium fortuitum and M chelonae. | After getting the culture and sensitivity reports, combination antibiotics (clarithromycin, linezolid and ofloxacin ) given for initial 3 months. 12 out of 28 patients well responded with 3 months of treatment. 9 patients required additional few months to get complete cure. 4 patients cured after 6 months of treatment and 3 patients did not come for follow up. Antibiotics were continued for additional 3 months after patient got cure to prevent relapse of infection.

chills are rare. The clinical features in our study were also similar with erythematous nodules, indurations, micro-abscesses and discharging sinuses. All our patients presented with only local manifestations that started with painful nodules which gradually increased in size, which then would fistulize and open on the skin draining pus while none of them had any systemic manifestations.
The source of infection in our case series is not clear. As per latest article by Maurer et al., a number of sources could have been the possible source of infections which included contaminated gentian violet, rinsing solutions, antiseptic solutions, injectable medications, unsterile surgical instruments or poor wound care. However in case of hernioplasty patients in our study as the organisms were also isolated from mesh, the source could be either the mesh or the transient presence of the mycobacteria in the surgical environment.

In other patients who had undergone caesarean section it is theoretically possible as per other study that nontuberculous mycobacteria might have gained access to the surgical wound from the public water system at the time of showering or it is equally possible that these organisms are present on the skin and are not eliminated by skin preparation preoperatively, thus gaining access through the skin incision.

Mechanical cleaning of blood and charred tissue that accumulates in the joints of the instruments may be not done properly after surgery. Thus, these contaminated instruments used during the surgical procedures might have left microorganisms implanted on the subcutaneous tissue which germinates and after an incubation period of 3–4 weeks giving rise to clinical symptoms.

Successful treatment of MOTT requires both surgical treatment and combination of antibiotics. This Combination of antibiotics as determined by susceptibility should be prescribed for an adequately long time so that the wound heals and also to ensure that no recurrence occurs. It has been reported that conventional anti-tubercular drugs are ineffective in treating these cases. Antibiotics should be given based on their susceptibility report and also combination of antibiotics is preferable over single regimen. M.fortuitum responds to antibiotics like amikacin, quinolones, doxycycline and sulphamethaxole. Latest studies reveal that clarithromycin, cefoxitin and imipenem were useful for treating MOTT. Almost all the patients in our study were cured with a combined approach of drainage and clarithromycin based combination therapy. In hernioplasty patients mesh was removed. In one female patient who developed incisional hernia as a complication of this infection, underwent hernia repair and excision of abscess cavity.

It has been recommended that in order to prevent recurrence, antibiotic treatment should be given for a minimum of at least three months, or to be continued for at least 3 to 6 weeks after the wound get healed. Recent research work has also recommended that antibiotic treatment should be given for 6 to 12 months though the optimal length of treatment has not been yet established.

So, it is important to re-emphasize upon the surgeons about the importance of following strict sterilization protocols including cleaning laparoscope instruments as per the manufacturer's instructions. Proper sterilization of medical equipments, proper skin cleansing preoperatively are essential prerequisite to prevent these infections. Clinicians should be aware to include MOTT in the differential diagnosis of surgical site infections in order to make early diagnosis and prompt treatment.

The limitations of this study were as follows: the exact source of infection could not be made out for further prevention of infection involving future surgical procedures and also molecular characterization was not done. Since PCR is costly for most patients, identification is still done commonly by conventional methods in most of the laboratories in countries like India.

To conclude, delayed onset chronic wound infection by Atypical mycobacteria is preventable. These organisms are not responsive to conventional antitubercular drugs but to specific drug regimens.
Declarations

FUNDING: No funding source/grant was available. No external fund was available. All investigations and treatment were done free of cost in the government teaching hospital named NRSMCH, WB, India.

Conflict of interest: The authors declare that they have no competing interests.

Ethical approval: Obtained from the Institutional Ethics Committee, NRSMCH, Bankura, West Bengal, India. Approval letter (Memo No. NMC/958 dt 23.02.17) is available for review by the editor of the journal. Written consents from individual patients were also obtained.

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Figures
Figure 1

Wound of a study subject, who underwent exploratory laparotomy followed by splenectomy in a case of blunt trauma abdomen.
Figure 2

The wound of same patient after 3 months of continued combination antibiotic treatment and repeated debridement.