Original Article

TO STUDY THE PREVALENCE , RISK FACTORS OF SURGICAL SITE INFECTIONS AND THE MOLECULAR CHARACTERIZATION OF *EXO T* GENE IN DRUG RESISTANT *PSEUDOMONAS AERUGINOSA* PATIENTS ATTENDING A TERTIARY CARE CENTRE, INDIA.

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Abstract

Introduction: Surgical site infection (SSI) is still a common and widespread disease that leads to severe morbidity and death, longer hospital stays, and ultimately higher healthcare costs. *P.aeruginosa* clinical isolates have been shown to express the virulent gene Exo T, which is significant in the aetiology of infection.

The current study aimed to identify the resistance gene *Exo T* gene in *Pseudomonas aeruginosa* and to ascertain the prevalence of SSI and associated risk factors among those who had undergone any type of surgical operation.

Aim and Objective: To Study the Prevalence , Risk Factors of Surgical Site Infections and the Molecular Characterization of *Exo T* gene in Drug Resistant *Pseudomonas aeruginosa* patients at a Tertiary Care Centre, India.

Material and Methods: This was a cross-sectional study conducted in a hospital setting over the period of 1 year i.e,, August 2022 to August 2023 at a Tertiary Care Centre. All surgically treated adult patients of both sexes who were older than 16 years old were included. Patients who received a second surgery at the same location for any reason, patients receiving immunosuppressant medication, people with immunodeficiency diseases, people currently taking antibiotics, and people with infections elsewhere were all disqualified from participating. If there was signs of a wound infection 48 hours after surgery, the patient was diagnosed with SSI. The DNA was extracted using the Qiagen DNA Extraction kit and the resistant gene *Exo T* was detected using the conventional PCR.

Results: In the present study a total of 170 patients underwent different types of surgeries. The prevalence of SSIs during the study period was 8.2%. SSIs were more common in abdominal surgeries with the Males (64.2%) have a higher risk of getting SSI than females (35.7%) . Patients who underwent emergency surgery have a higher risk of getting SSI than those who underwent elective surgery . Those with diabetes had a higher risk of getting SSI than those who were non-diabetics. In the present study it was also observed that *Klebsiella pneumoniae* (28.5%) was the most common isolate followed by *E.coli* , *Pseudomonas aeruginosa* with 21.4%, *S.aureus* with 14.25% and least for *Staphylococcus epidermitidis* and *Klebsiella oxytoca* with 7.14%. It was observed that the site of the infection most common affected was the superficial site with 57.1%. The Molecular characterization confirms that out of 3 Pseudomonas 2 showed the presence of *Exo T* gene (66.6%).

Conclusion: SSIs were more likely to follow abdominal operations. Patients who were male, in the 30-year-old age range, had emergency surgery, had diabetes, and/or had a protracted hospital stay prior to any kind of surgery were more likely to experience surgical site infections (SSIs). It was also found that the proven role of *Exo T* virulence genes in the pathogenicity of *P. aeruginosa* would help in treatment and prognosis of Pseudomonas infections

Keywords: Surgical site infection, Prevalence, risk factors, Molecular Characterization, DNA, PCR, Exo T,

INTRODUCTION

Wound infection is the term used to describe the invasion of organisms into tissues as a result of a breakdown in the local and systemic host defences, which can lead to cellulitis, lymphangitis,

abscess, and bacteremia. Infections that arise in surgical sites are known as surgical site infections (SSIs) [1]. Surgical site infections (SSIs) are defined as infections that occur within 30 days following surgery and cause damage to the incision or deep tissue at the operation site. If an implant is left in place after the procedure, the infection may occur within a year [2]. The National Nosocomial Infection Surveillance Programme (NNIS) [3] has identified three kinds of infections: deep infections, superficial infections, and organ/space infections.

The patient's own natural flora, germs from the hospital environment that the patient contracts during treatment, particular underlying disorders, trauma, or burns that could disrupt the mucosa or skin surface are all potential sources of SSIs. SSIs are severe postoperative complications that account for 20% of infections connected with healthcare and happen in about 2% of surgical procedures. According to numerous research, SSIs are the third most frequent nosocomial infection after respiratory tract and urinary tract infections [2-5]. According to recent studies, the SSI rate varies from 19.4% to 36.5% globally, but only from 3% to 12% in India [6-7].

SSI is still a widespread problem that drives up healthcare costs by extending hospital stays, increasing morbidity and mortality rates, and so on. Risk factors for SSIs include smoking, obesity, diabetes mellitus, and length of hospital stay. The potential development of a surgical wound infection is determined by the intricate interplay of multiple factors. A common cause of infection in most surgical wounds is endogenous. The surgical team's nostrils or skin flora are usually the source of exogenous infections, which are then transferred by the surgeon's hands or improper sterilisation of the operating room, which includes preoperative, intraoperative, and postoperative care [8].

The rising prevalence of multidrug-resistant (MDR) strains of *P. aeruginosa* has complicated medical management, which is a global issue. *P. aeruginosa* produces biofilms in the airways if it is not eliminated during the initial infection phase. One of the main factors contributing to chronic infections is the formation of biofilm, which are organised bacterial communities embedded in an extracellular polymeric matrix adhered to surfaces .

The virulence factors can be chemical or proteinaceous, and either cell-associated or secreted. Proteinaceous virulence factors are often secreted through one of the five protein secretion systems so far described as *P. aeruginosa*: type I, II, III, V [9] and the recently discovered type VI [10].

MDR, XDR and PDR variants manifest a high level of intrinsic resistance to antimicrobial drugs by the help of efflux pump, biofilm formation, aminoglycoside modifying enzymes and sometimes by mutation in chromosomal gene (ESBL and AmpC hyper expression). Pseudomonas spp. is also able to obtain the resistance by means of horizontal gene transfer mechanism which is responsible for class B carbapenamase (MBL).

Genes responsible for drug resistance are located on integrons which is frequently located in plasmids or transposons and these genes can shift very often and contributes to the dissemination of resistance mechanism around the world [10].

P. aeruginosa also has a large number of other virulence factors such as exotoxin A (*exoA*), alkaline protease (*aprA*), exoenzyme S, U, and T (*exoS*, *exoU*, *exoT*), elastase and sialidase, which are *exoA* gene and virulence factor *exoS* secretions by a type III section system [8].

ExoT is a bi-functional protein possessing an N-terminal GTPase-activating protein (GAP) domain, which inhibits RhoA, Rac1, and Cdc42, small GTPases, and a C-terminal ADP-ribosyltransferase (ADPRT) domain, which targets Crk adaptor proteins and PGK1 glycolytic enzyme Due to its multiple targets, ExoT performs a number of distinct virulence functions for *P. aeruginosa* [10].

Establishing worldwide incidence of SSIs in general surgical patients is imperative to understand the extent of the condition, its burden on society, and the demographic and clinical risk factors that predispose general surgical patients to develop SSIs. Drug-resistant phenotypes have evolved as a result of Pseudomonas spp.'s capacity to create a wide range of drug resistance mechanisms. This presents a hurdle for our clinician when treating an infection this serious. This kind of scenario leads to the identification of phenotypes that generate various medication resistance mechanisms in order to prevent treatment failure and hospital acquired infections [9].
Therefore the present study was undertaken to study the Prevalence , Risk Factors of Surgical Site Infections and the Molecular Characterization of Exo T gene in Drug Resistant *Pseudomonas aeruginosa* patients at a Tertiary Care Centre, India.

MATERIAL AND METHODS

Study settings and duration

This was a hospital-based, cross-sectional study carried out in the Department of Microbiology and the Surgery Department. The study was carried out over a period of 1 year. Ethical clearance was duly obtained from the Institutional Ethical Committee.

Study population and sampling technique

As per the convenience sampling technique, all the cases admitted to the surgical wards (including both elective and emergency surgery) during the study period and those who met the eligibility criteria were included in the study.

Sample size calculation

The prevalence of SSI observed in the study by Tabiri et al. was 11.5% [11]. Based on this study, consideration.

SAMPLE SIZE :- SS (n)= $4PQ/L^2$ Where,**P=**Prevalence,**Q=** 100-p,**L=** Allowable error, If the allowable error is 5 % **SS (n) = 4×11.5x88.5**

Sample Size (n) = 4071/25= 162

So, in order to coverup the lost- to-follow-up, drop-out rate and non-response rate the sample size taken in our research study was 170 .

Inclusion criteria

All patients of both genders above 16 years who underwent surgery and were admitted to the surgical wards during the study period were included in the present study.

Exclusion criteria

All pediatric cases were excluded from the study. Patients who underwent second surgery at the same site for any reason, patients on immunosuppressant therapy or any known immunodeficiency disease, patients on antibiotics already for any other infections, and patients with infection elsewhere in the body were also excluded from the study.

Ethical clearance

The study was carried out after getting ethical approval from the Institutional Ethical Committee and written consent was obtained from every study subject.

Data collection procedure

Data about the age of the patients, gender, demographic details, clinical details including the name of the procedure, date and duration of surgery, the experience of surgeons, preoperative hospital stay, nature of the surgery, postoperative hospital stay, and the onset of illness (SSI) were collected by reviewing the patient's case sheet.

The surgical wound dressings were removed 48 hours after the procedure. Indications of a wound infection was taken into account if the patient displayed local inflammatory changes at the wound site, such as edoema, redness, warmth, or discharge. If before applying the bandage, samples were taken to determine if there had been any discharge. Inflammatory changes alone were present but did not have any discharge, the wounds were watched for the emergence of until the patient was sent home, the wound. If inflammatory symptoms emerged within 48 hours, patients were followed with the assistance of the corresponding surgeons. These patients also received education and followed up for the creation of SSIs through mobile phone for the development of SSIs over a period of 30 days.

After cleaning the suspected wound infections with sterile normal saline and 70% alcohol, a sterile swab was used to collect the material. Within two hours, the laboratory received two swabs taken from the depth of the wound, or the aspirates were collected in a sterile disposable syringe. We noted and observed the materials' colour, consistency, and odour.

A direct thin smear was made from each wound swab and/or aspirates on a clean grease-free glass slide and was air dried. It was then heat-fixed, and Gram staining was done with positive and negative control.The presence of pus cells and microorganisms was observed under the oil immersion (100X) objective. The samples were cultured onto nutrient agar, 5% sheep blood agar, and MacConkey agar plates by adopting standard microbiological techniques. After 24 hours of incubation aerobically at 37°C, plates were read, and the isolates were identified based on colony morphology, Gram stain, motility, and biochemical tests.

Antibiotic susceptibility testing was performed by Kirby bauer Disk diffusion method as per the CLSI guidelines 2022. The antibiotic discs was placed on the Muller–Hinton agar which were previously inoculated with test strains and incubated at 37 °C for 16–18 h. After incubation, inhibition zones was recorded as the diameter of the clear zones around the disc and interpreted

according to performance standard for antimicrobial disk susceptibility test as per the CLSI guidelines [12].

GENOTYPIC METHOD:

The Molecular Detection of DNA extraction was done to detect the presence of *exo T* gene in *Pseudomonas aeruginosa* from the surgical site isolates.

Molecular Identification of *Exo T gene*

The DNA was isolated using the Qiagen DNA Extraction Kit as per the manufactures guidelines. The DNA was eluted in 60 µl elution buffer and preserve at -20 °C till PCR analysis. For amplification of the target gene, PCR was carried out in a 50 µL reaction mixture. The primers were purchased from "**Saha gene"** and was reconstituted with sterile double distilled water based on the manufacturer's instruction.

Material and instruments required for the DNA Extraction:

Micro centrifuge upto 10000g.

- Vertex Mixer
- Dry bath for incubation at 55°C
- Biosafety cabinet
- Pipettes (1ml,200µl,10µl)
- Pipettes tips with filters
- 1.5 ml microcentrifuge tubes (DNase/RNase free)

Figure No. 1: The *Exo T* primers from the Saha gene

Polymerase Chain Reaction (PCR): After the DNA Extraction, the PCR was done for the gene detection. The sequences of the primers used in PCR for detection of exoT gene and its molecular weight are mentioned in the Table no. 1.

Gene	Primer sequence	Amplicon size	Length (bp)
Exo T	Forward 5'-AATCGCCGTCCAACTGCATGCG-3'	22	152
	Reverse 5'-TGTTCGCCGAGGTACTGCTC-3'	20	

Table 1: The Primer sequence used for the detection of *Exo T* genes

Polymerase Chain Reaction (PCR) cycling conditions : After the DNA Extraction, the PCR was done. The sequences of the primers used in PCR for detection of *exoT* gene and its molecular weight are mentioned in the Table no.1 and 2 [13,14].

ATCC 27853 was used as Positive Control (PC), while nuclease free water will be used as Negative control (NC).

Gene	Initial Denaturation	No.of Cycles	Denaturation in Each Cycle	Annealing	Primer Extention	Final Extention
exoT	95ºC, 2 min	36	95ºC, 30 sec	58ºC,30 sec	72ºC, 30 sec	72ºC,5 min

Table 2 : The Polymerase Chain Reaction (PCR) conditions..

The above was the cycling conditions used for the PCR.

Gel electrophoresis : The Agarose Gel Electrophoresis was performed in order to identify the Purified PCR Product which was previously identified by its amplified DNA fragments. The resulting PCR product was subjected to 1 % agarose gel electrophoresis and visualized by Gel Doc™ EZ Gel Documentation System (Bio-Rad Laboratories Inc., Hercules, CA, USA). A 1 kb DNA Ladder (Thermo Fisher Scientific ™, Waltham, MA, USA) was used as the marker to evaluate the PCR product of the sample [15].

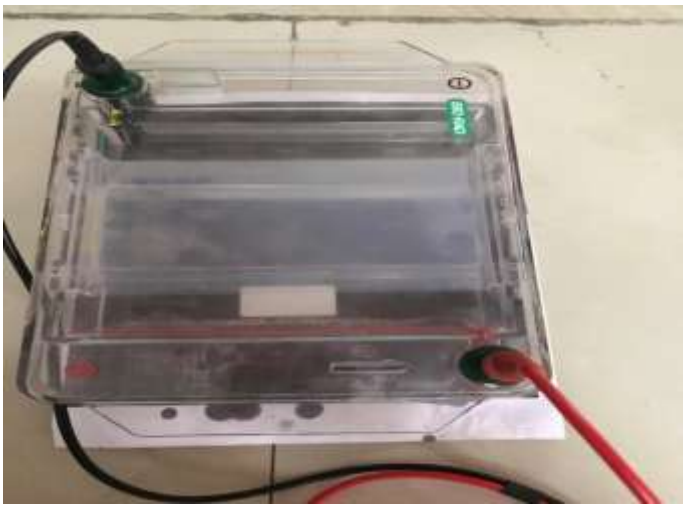


Fig No.2: Gel Electrophoresis for the DNA Extraction

Data analysis

The data obtained were entered in Microsoft Excel (Microsoft Corp., Redmond, WA), and the results were analyzed using SPSS (Statistical Package for the Social Sciences) version 21 (IBM Corp., Armonk, NY). All the data collected in the current study was categorical, so they were expressed in a table as frequency and percentage. Also, the figures were expressed as a pie chart. The association between risk factors and the presence of SSI was assessed using the Chi-square test. With a 95% confidence interval, a p-value of less than 0.05 was considered statistically significant.

RESULTS

A total of 170 patients underwent different types of surgeries, including elective as well as emergency procedures, during the study period. About 14 SSIs were documented, and hence, the overall prevalence of SSI rate during the study period was 8.23% The number of cases that developed SSIs in relation to the type of surgery is shown in Table 3.

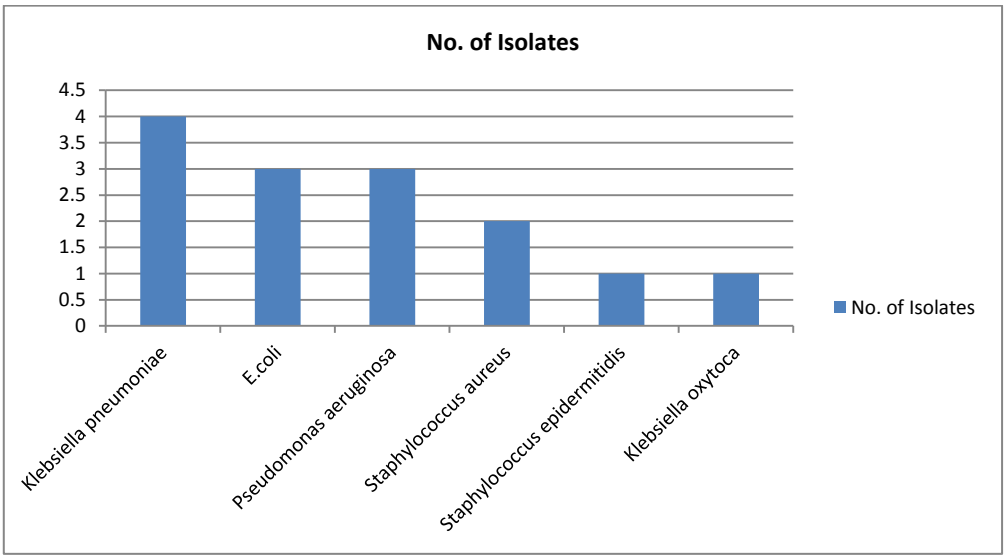
Site of Surgery	Types of Surgeries	No. of Surgeries, n (%)	SSI, n (%)
Abdomen	Appendectomy	10 (5.88%)	1(6.25%)
	Hernia repair	9 (5.29%)	2 (12.5%)
	Exploratory laparotomy	7 (4.117%)	3 (18.75%)
	Cholecystectomy	6 (3.52%)	1 (6.25%)
	Lower segment cesarian section	25 (14.7%)	1 (6.25%)
	Hysterectomy	14 (8.23%)	1 (6.25%)
Pelvis	Sphincterotomy	3 (1.76%)	1 (6.25%)
	Hemorrhoidectomy	3 (1.76%)	1 (6.25%)
	Fistulectomy	4 (2.35%)	2 (12.5%%)
	Hip replacement	3 (1.76%)	Nill
Urogenital	Transurethral resection of prostate	4 (2.35%)	Nill
	Urethroscopy lithotripsy	4(2.35%)	Nil
Breast and axilla	Modified radical mastectomy	3 (1.76%)	1(6.25%)
	Fibroadenoma excision	5 (2.94%)	Nil
Skin, bone, and joints	Knee replacement	4 (2.35%)	Nil
	Varicose vein	4 (2.35%)	Nil
	Open reduction and internal fixation	3(1.76%)	Nil
Eye	Intraocular lens implantation	40 (23.52%)	Nil
Ear, nose, throat	Tonsillectomy	12 (7.05%)	Nil
	Mastoidectomy	4 (2.35%)	Nil

Neurosurgery		3 (1.76%)	Nil
Total		170	14

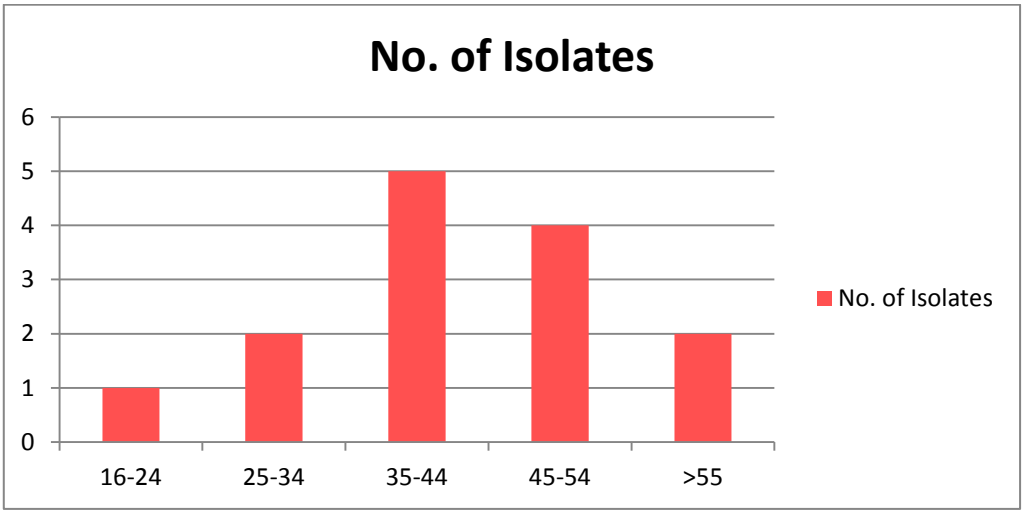
Table 3: Prevalence of SSI according to the Types of Surgery (n)

Type of organisms Isolated	Number of Isolates	Percentage
Klebsiella pneumoniae	4	28.5%
E.coli	3	21.4%
Pseudomonas aeruginosa	3	21.4%
Staphylococcus aureus	2	14.25
Staphylococcus epidermitidis	1	7.14%
Klebsiella oxytoca	1	7.14%
Total	14	100%

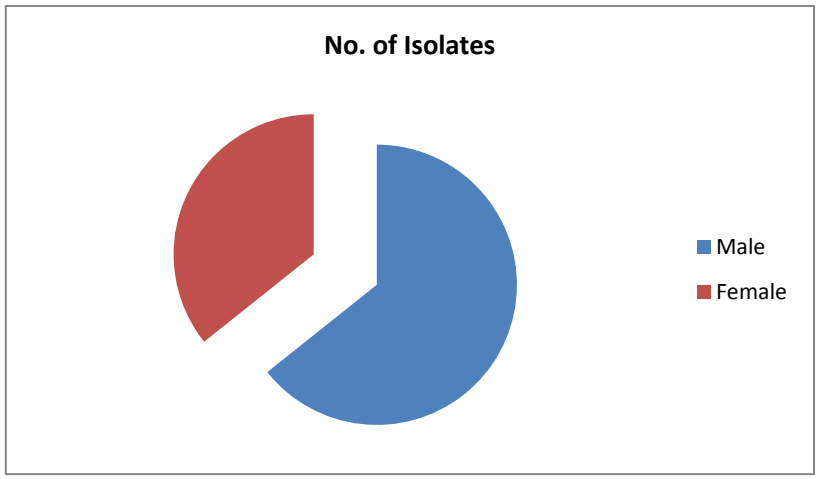
Table 4: The Type of Isolates causing SSIs



Graph No. 1: Graphical Representation of the Number and the Type of Isolates



Graph No. 2: Graphical Representation of the Agewise Distribution of the Isolates



Graph No. 3: Graphical Representation of the Genderwise Distribution of the Isolates

S. No	Risk Factors		Frequency of SSI	Percentage
1	Age group (years)	16-24	1	7.14
		25-34	2	14.2
		35-44	5	35.7
		45-54	4	28.5
		≥ 55	2	14.2
2	Gender	Male	9	64.2
		Female	5	35.7
3	Type of surgery	Emergency	12	85.7
		Elective	2	14.2
4	Extend of wound	Superficial	8	57.1
		Deep	6	42.8
		Organ	0	0
5	Diabetes mellitus	Yes	10	71.4
		No	4	28.5
6	Smoking	Yes	7	50
		No	6	42.8
7	Alcoholism	Yes	8	57.1
		No	6	42.8
8	Anemia	Yes	8	57.1

9	Hospital stay	No	6	42.8
		1-7 days	8	57.1
		>7 days	6	42.8
10	Drain	Yes	10	71.4
		No	4	28.5

Table 5: Distribution of Risk factors of the Study Population according to SSI (n = 14)

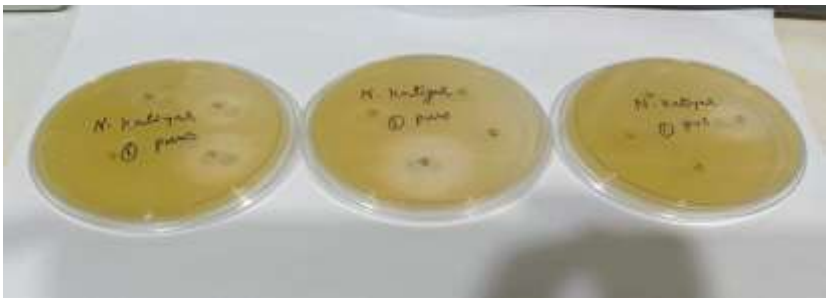


Fig No. : 3 : The Antimicrobial Disk Susceptibility Test

It was observed that the Sensitivities of Colistin was (95.8%), Piperacillin‑tazobactam (72.9%), Amikacin (77 %), and cefepime (72.9%) were found to be the most effective Antibiotics. The resistance to ciprofloxacin was (43.7%), Levofloxacin (50%), Gentamicin(64.5%), Imipenem (64.5%), Tobramycin(68.7%), Ceftazidime (68.7%) [Table No. 9] .

The AST was performed where it was observed that Colistin and Polymixin-B was found to be the most sensitive drugs. Among the aminoglycosides, amikacin has the highest sensitivity against *P. aeruginosa* . Amikacin was designed as a poor substrate for the enzymes that bring about inactivation by phosphorylation, adenylation or acetylation, but some organisms have developed enzymes that inactivate this agent as well. Amikacin seems to be a promising therapy for Pseudomonal infection. Hence, its use should be restricted to severe nosocomial infections, in order to avoid rapid emergence of resistant strains.

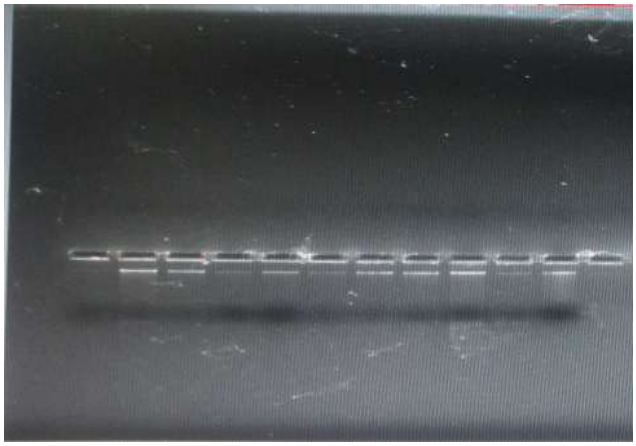


Fig:No. 4: The DNA Extraction in Agarose gel

In the present study it was observed that out of 3 Pseudomonas isolates there were 2 (66.6%) which expressed ExoT gene.

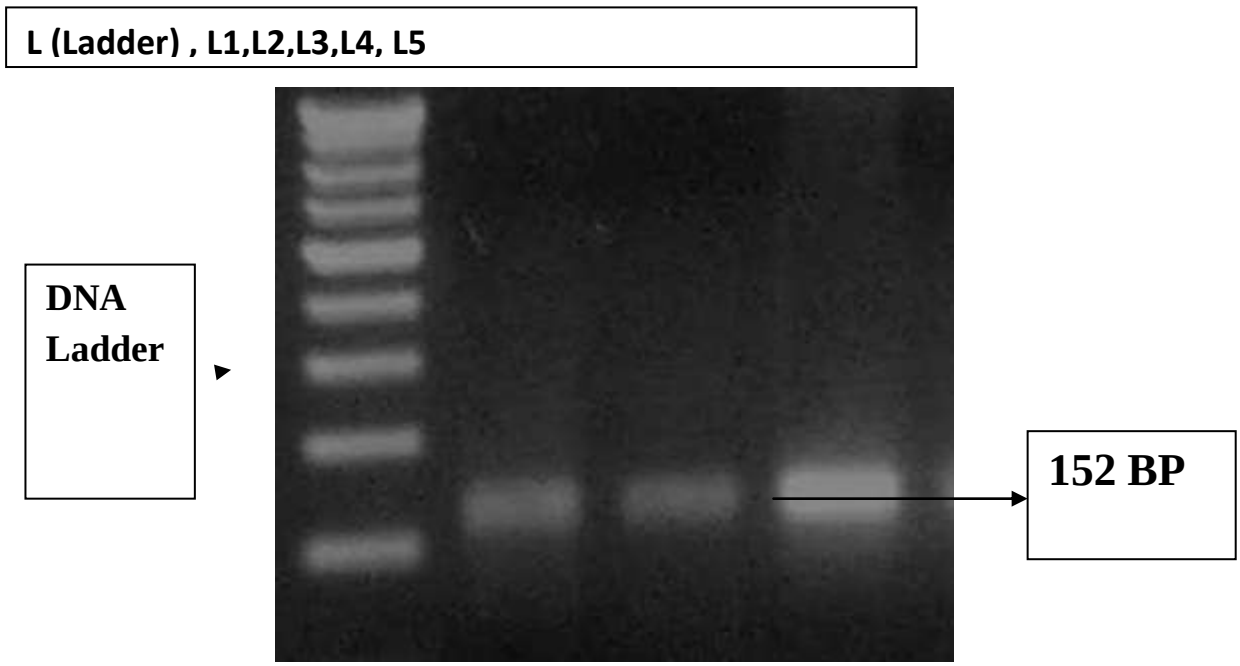


Fig No. 5: The Gene Exctraction ExoT gene:
L corresponds to the DNA Ladder; L1- L2 are the sample positive for ExoT gene; L3 corresponds to the Positive Control for ExoT gene; L4 is the Negative Control gene for ExoT gene

DISCUSSION

Surgical site infections (SSI), which have been reported to have an incidence rate of 2-20%, are a common surgical site consequence and one of the most common nosocomial infections [16]. They bear the blame for elevated treatment expenses, prolonged hospital stays, and notable rates of morbidity and mortality. Even in hospitals with the most up-to-date amenities, surgical site infections (SSI) remain a serious issue despite technological advancements in infection control and surgical techniques [17]. Exogenous and/or endogenous microorganisms that infiltrate the surgical site during the procedure (primary infection) or afterwards (secondary infection) are typically the source of these illnesses. Usually developing five to seven days after surgery, primary infections are more dangerous [18]. *Pseudomonas aeruginosa* is an important causative agent of human infection, especially in a host with compromised defense mechanisms. *It* is a common nosocomial pathogen, notorious for its multidrug resistance (MDR) and life threatening infections in critically ill patients. Lately, carbapenems are being used as the last resort antimicrobial treatment for serious infections due to MDR *P. aeruginosa* [10].

Bacteriological studies have shown that SSIs are universal and the etiological agents involved may vary with geographical location, between various procedures, between surgeons, from hospital to hospital or even in different wards of the same hospital [16].

Out of the total of 170 patients showed local signs and symptoms and were suspected to have postoperative wound infections. These cases were evaluated and followed up. Among them, the

culture positive was observed in 14 cases and hence was considered as cases of SSI in our hospital; thus, the overall prevalence rate of SSIs was 8.2% in the present study .. The current status of SSIs identified in their hospital concurs with the studies of Golia et al. [19] and Iqbal et al. [20]who reported the prevalence rate as 4.3%, 5.4%, and 7.3%, respectively, which were in accordance to the current study. There were other studies performed by the other research investigators which were in contrast to the present study where, Al-Mulhim et al. [21] reported in their study that the overall prevalence rate of SSIs was 2.5%, which was lesser than one third of our present study rate.
There was another study which was also in contrast to the present study by Setty et al. [22] which reported the prevalence rate to be quite high with 21.66% and 22.2% respectively.

In the present study it was observed that the ratio of Males 9 (64.2%) was more as compared to that of females 5 (35.7%). This study was similar to the study performed by the other research investigator Vikrant Negi et al.,[23] where Males (74.6%) were more commonly affected than females (25.5%) and the sex ratio male: female was 2.9:1. A study by Hernandez et al., in 2005 conducted in a Peruvian Hospital reported more occurrences among males (65.6%) [24].
In contrast, a study done by Shanmugam et al. reported almost equal occurrences among females (52%) and males (48%) [25]. The increasing occurrence among males was attributable to the nature of the infected wounds with which they come to surgical departments.

In the current study it was observed that the maximum number of isolates found were in the age group of 35-44 years of age followed by 45-54 years of age and least in the age group of 16-24 years of age. This study was similar to the study performed by the other author [23] where the 31 - 50 years was affected the most. The patients with age >50 years had a higher incidence of SSI (51.8%) in comparison to an incidence of 12.4% among the patients who were ≤30 years of age. Advancing age is an important factor for the development of SSIs, as in old age patients there is low healing rate, low immunity, increased catabolic processes and presence of co-morbid illness like diabetes, hypertension, etc [26].
Other research investigators, Owens et al. [27] and Bharatnur et al. reported that a greater number of SSIs occurred among 36-50 years (1.3 times higher risk of acquiring SSIs than the ones who were in the age group of 10-35 years). Similarly, a high rate of infection was noted in the later age groups by Mundhada et al. [28]. It was also observed that there were increase cases in the emergency ward (85.7%). The increased rate of SSI in emergency surgeries may be due to a very narrow time span without proper patient preparation and surgical preparedness as well as contaminated wounds as in cases of road traffic accidents. The same have been cited in most of the studies done earlier on SSIs. Tabiri et al. also reported that emergency cases had a higher number of SSIs (23.8%) as compared to elective cases (7.4%)[11]. In another study done by Dessie et al., SSIs were reported in 61.7% of emergency cases and 38.3% of elective cases [29].
In the present study, it was observed that superficial and deep SSIs were observed with the ratio of 57.1% and 42.8% respectively. There was no SSIs observed in the organ site. There was another study which was similar to our study reported that superficial incision SSI was more prevalent (215 cases, 55.9%) followed by deep incisional SSI (169 cases, 44%), and van Walraven et al. [30,31] reported the same that a majority of these (n= 8188, 57.5% of all SSIs) had a superficial component. This is discordant with the study by Dessie et al., who reported superficial SSI as 42.1% and deep

SSI as 57.9% (112 cases) [29].

In the present study it was observed that *Klebsiella pneumoniae* (28.5%) was the most common isolate followed by *E.coli* , *Pseudomonas aeruginosa* with 21.4%, *S.aureus* with 14.25% and least for *Staphylococcus epidermitidis* and *Klebsiella oxytoca* with 7.14%. There was the another study which was in support to the present study where *E. coli* (46.4%) was the commonest gram negative bacteria isolated followed by *P. aeruginosa* (15.9%) and *Citrobacter spp* (15.9%) [23]. Similar observations have been reported by various other authors also [32-34]. Few studies have reported *P.aeruginosa* as the most frequent isolate in SSI [35,36] which remains a third most isolated strain in this study.

In the present study the expression of Exo T gene was studied in *Pseudomonas aeruginosa* where out of 3 (66.6%) expressed the gene. This study was in support with the study by Dadmanesh et al. [37] where *exoT* rat as 69.21%. Another study stated that exoT gene occurred in 20 (66.67%) isolates of *P. aeruginosa*, while 10 (33.33%) showed negative amplification results [38].

There was another study which was in contrast to the present study where the rate of Exo T was 5% [39].

Exo T virulence genes have been shown to play a significant role in *P. aeruginosa* pathogenicity, which is important for understanding the prognosis of pseudomonas infections and developing a vaccine that will effectively prevent them. This could be useful in epidemiological research to determine the best course of treatment for *P. aeruginosa*-caused illnesses. These results could aid in the identification of pathogenic gene targets for immune intervention, which could control how severe the host response.

In the recent years there has been a growing prevalence of gram negative organisms as a cause of serious infections in many hospitals. In addition irrational use of broad spectrum antibiotics and resulting anti microbial resistance (AMR) has further deteriorated the condition in this regard.

CONCLUSION

Finding virulent pathogen markers to identify acute and chronic infections early on is still a crucial field that requires much investigation. These kinds of investigations and study results help avoid bacterial infections and can be very helpful in managing Pseudomonas infections. Hence, Surveillance of SSI along with feedback from surgeons will help to reduce the SSI rate and this surveillance system should be developed in all hospitals and also guidelines for antibiotic use among surgical patients should also be developed and strictly followed which may provide the estimate of incidence of SSI.

LIMITATION

The fact that fungal cultures and anaerobic bacterial profiles were not performed on the wound swabs from SSIs was one of the study's weaknesses. Additional prospective study in this field is feasible.

DECLARATIONS:

Conflicts of interest: There is no any conflict of interest associated with this study
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