Original Article

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Surgery Section

Prevalence of Aerobic Organisms in Surgical Site and Ulcerations at a Tertiary Rural Hospital in Mandya, Karnataka, India

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ABSTRACT

Introduction: In an era in which the cost of treatment is an increasing source of concern in wound management in surgery, wound infection increases costs and hospital stay.

Aim: To study the microbial spectra, antibiotic sensitivity, different modalities of wound management and the outcome of treatment in surgical side infections and ulcerations.

Materials and Methods: This cross-sectional study was conducted at a tertiary rural centre with a total of 60 cases of wounds with various aetiologies for duration of two years at AIMS, Mandya, Karnataka, India, from January 2020 to December 2021 to study demographics, prevalence and management outcome. Categorical data was represented in the form of frequencies and proportions.

Results: Total 60 wound cases were further divided into three groups with 20 patients each having non specific ulcer, Surgical Site Infection (SSI) and Diabetic Ulcers (DU). The SSI was most common in patients belonging to age group 20-31 years. A total of 65% of patients who had SSI were observed among clean contaminated surgeries. Methicillin Resistant Coagulase Negative Staphylococci (MRCNS) was the most common organism causing infection in postoperative wounds, showing maximum sensitivity to vancomycin and resistance to amoxicillin. A 70% of the diabetic wounds underwent skin grafting and 70% of SSI patients underwent secondary suturing and 75% patients having an Arterial or Venous ulcers (A/V) were allowed to heal by secondary intention.

Conclusion: A thorough understanding of microbial spectra and their antibiotic sensitivity is required in addition to effective wound management to improve the outcome of ulcer management.

Keywords: Antibacterial agents, Extended spectrum beta-lactamases producers, Methicillin-resistant coagulase-negative staphylococci, Meropenem, *Pseudomonas*, Vancomycin, Wound infection

INTRODUCTION

Wound infection is one of the most common nosocomial infections. Despite all the advancements and perioperative care, wound infection continues to be a serious problem all over the world. High rates of wound infection are associated with a higher morbidity and mortality and also increased cost of medical care and hospital stay. Hence, a constant awareness of the threat of infection is a worry for the entire surgical fraternity [1].

Another dilemma faced by the surgeons is the proper choice of antibiotics. Due to the indiscriminate use of antibiotics, antimicrobial resistance has developed. Clear understanding of the pathogens and their pathogenicity with judicious selection of antibiotics will help the surgeon not only to tackle this problem, but will also turn out to be effective in long run [2]. In any surgical intervention, wound complications occur from time to time and impede the postoperative course. Majority of these complications are of minor importance, responding immediately to therapy, however a smaller group of patients develop a postoperative wound complication of major importance [3].

Chronic wounds, especially non healing types are one of the most common surgical conditions encountered by the surgeon, which fail to heal in the expected time. Delay in healing most often occurs in the inflammatory phase. The peculiarity of a chronic wound is that despite of all the care, the wound does not heal especially in diabetic, venous and arterial ulcers [4]. Proper perioperative aseptic precautions, optimum care of wounds and judicious use of appropriate antibiotics are required to reduce SSI and promote early wound healing [5]. With this in mind, this study was undertaken at Adichunchanagiri Hospital and Research Centre, BG Nagara, Bellur, Mandya district to study the prevalent organisms infecting the wounds. The objective of this research was to study the prevalence of aerobic organisms in SSI and skin ulceration patients with emphasise on isolation of the organism, culture and antibiotic sensitivity, initiation of treatment with appropriate antibiotic and outcome, procedure undertaken for the wound care and its treatment and number of days of stay in hospital.

MATERIALS AND METHODS

This cross-sectional study was conducted for duration of 24 months, from January 2020 to December 2021 at AIMS, Mandya, Karnataka, India. Institutional Ethical Committee clearance was obtained. (IEC Number-AIMS/IEC/2206/2019). Written informed consent was obtained.

Sample size calculation: During this period, 60 patients between 21-85 years who fulfilled the below mentioned inclusion and exclusion criteria were studied. With assumed prevalence of 20%, confidence limit to 10% and confidence level of 95% the estimated size was 62.

Inclusion criteria: All non specific ulcers, open wounds with or without diabetes mellitus and those patients having SSI were all included in the study.

Exclusion criteria: Actively healing wounds, malignant ulcers, wounds with suspected anaerobic and fungal infection and ulcers associated with gangrene were excluded from the study.

The wounds were grouped according to the associated co-morbidity and clinical features of the wound as:

- **Group A:** All non specific ulcer (venous and arterial ulcers)
- **Group B:** Patients with SSI
- **Group C:** Diabetic foot ulcers

Study Procedure

After admission of the patients with skin ulceration, required laboratory and radiological investigations were done. On the day of

admission patient was started whenever required oral or IV antibiotic ceftriaxone depending on the condition of the wound. The next day, two wound swabs were aseptically collected from the wound after irrigation of the wound and its surrounding area with normal saline; this is done before the application of any topical antibacterial solution and sent to microbiology laboratory within 10 minutes to prevent any superadded infection.

The samples collected were processed. Culture done on culture media (e.g., blood agar, MacConkey agar and chocolate agar) incubated overnight at 37°C. Direct microscopic examination of gram stained smear to identify the organism type was done. Organisms were identified by following the standard protocol. The antibiotic susceptibility was carried out by disc diffusion method on Muller Hilton Agar (MHA) [1]. In SSI (postoperative wound infection) swab was collected on the day signs of infection or serosanguinous discharge was noted. After collection of the swab, wound was dressed daily with sterile dressing using povidone iodine -10% solution, hydrogen peroxide, sterile gauze and pad. The patient was treated with appropriate antibiotic according to the culture sensitivity report along with daily dressing. The outcome of the wound like graft acceptance, healing of infected surgical site by secondary intention or by secondary suturing was noted.

Parameters evaluated were: diabetic status, isolated organism and their antibiogram, abdominal Ultrasound Sonography (USG) and Computed Tomography (CT) scan were done, whenever indicated to exclude organ space infection and a proforma was used to collect information from the patients with consent and results were analysed and compared with other studies.

STATISTICAL ANALYSIS

Data was entered into Microsoft excel data sheet and was analysed using Statistical Package for Social Sciences (SPSS) 22.0 version software. Categorical data was represented in the form of frequencies and proportions. Chi-square test was used as test of significance for qualitative data. Continuous data was represented as mean and standard deviation. The ANOVA was used as test of significance to identify the mean difference between three groups. MS excel and MS word was used to obtain various types of graphs p-value (Probability that the result is true) of <0.05 was considered as statistically significant after assuming all the rules of statistical tests.

RESULTS

Age distribution: In this study majority, of patients belonged to the age group of 41-50 years (15 patients). This was followed by 51-60 year age group (13), 61-70 year age group (12), 21-30 year age group (10), 71-80 year age group (5), 31-40 year age group (3) and >80 year age group (2). Out of these, SSI and DU were maximum in age group of 21-30 years and 41-60 years, respectively [Table/Fig-1].

Age group (in years)	Diabetic ulcer (n)		Surgical site infection (SSI) (n)		Arterial/Venous ulcer (n)		Total
	Male	Female	Male	Female	Male	Female	
21-30	1	0	5	1	3	0	10
31-40	0	0	1	1	1	0	3
41-50	2	4	3	1	4	1	15
51-60	4	2	2	0	3	2	13
61-70	3	2	2	1	2	2	12
71-80	1	0	2	0	2	0	5
>80	0	1	1	0	0	0	2
Total	11	9	16	4	15	5	60

[Table/Fig-1]: Age and gender distribution.

Mean age in different ulcers were 53 (A/V), 48.8 (SSI) and 57.8 (DU) and standard deviation (SD) was 15 (A/V), 20 (SSI) and 14 (DU); ANOVA, F-value-1.58, mean age with SD=52.9±18 and range being 21-95. There was no statistical difference between the groups with respect to age [Table/Fig-2].

Variable		DU (n)	SSI (n)	A/V (n)	Total (n%)
Mean age±SD (In years)		57.8±14	48.8±20	53±15	52.9±18
Gender distribution	Male	11	16	15	42 (70%)
	Female	9	4	5	18 (30%)
Smoking history		7	8	13	28 (46.7%)
Diabetes mellitus		20	14	7	41 (68.3%)

[Table/Fig-2]: Patient demographics.
DU: Diabetic ulcer; SSI: Surgical site infection; A/V: Arterial and venous ulcer

Sex distribution: Out of 60 cases, males were 42 (70%) and females were 18 (30%) in the study. There was no statistically difference between the sexes of the individuals in the three groups. The p-value (0.188) and Chi-square value (3.33) [Table/Fig-1].

Smoking history: The history of smoking was present in 28 cases. Cases were smokers in having DU 7 (35%), SSI 8 (40%), and A/V ulcer 13 (65%) groups respectively. Maximum smokers were seen in the A/V group (Chi-square value- 4.15) [Table/Fig-2].

Patients with diabetes in the present study: The number of cases with DM in the present study were 41 accounting to 68.3%. Other than the obvious diabetic group (20 patients), seven patients in A/V group and 14 patients in SSI group also had diabetes mellitus [Table/Fig-2].

Incidence of SSI in different types of surgery: Among the 60 cases studied, 7 (35%) clean wounds patients and 13 (65%) patients clean contaminated wounds developed infection respectively. The more the contamination of the wound the more is the chances of developing infection. However, there were no contaminated and dirty wounds in the study.

Manifestations of SSI postoperatively: In the present study, 3 (15%) cases presented with infection on 3rd day, 6 (30%) cases on 4th day and 9 (45%) cases on 5th day and 2 (10%) cases on 6th day. Thus, the maximum number of SSI cases occurring within first five days of surgery.

Microbial profile in ulcers: *Staphylococcus* was the common organism isolated including MRSA and MRCNS in SSIs. None of the strains were resistant to vancomycin. Other organisms isolated were ESBL producers *Klebsiella* and *Escherichia coli (E. coli),* followed by *Pseudomonas*. ESBL producers were the common organism isolated in diabetic foot ulcers. DU usually has polymicrobial infection.

The common organism isolated from Non Specific Arterial/Venous ulcers (A/V) were *Pseudomonas* followed by MRSA and MRCNS. Non specific ulcers also show polymicrobial infection [Table/Fig-3].

Antibiotic sensitivity: In non specific ulcers, 80% of *Pseudomonas* (6 out of 7) isolated were sensitive to vancomycin. All of the isolates of MRSA (7 out of 7) were sensitive to meropenem, imipenem, linezolid and vancomycin and MRCNS (3 out of 3) were sensitive to linezolid, vancomycin and piperacillin-tazobactam.

In SSI, MRCNS (11 out of 11) isolated were sensitive to meropenem and vancomycin and MRSA (4 out of 4) were sensitive to meropenem, gentamycin, imipenem, vancomycin and piperacillin tazobactam. All isolates of *P. aeruginosa* (4 out of 4) were sensitive to linezolid and vancomycin.

In DU, all *klebsiella* isolated (9 out of 9) were sensitive to meropenem, gentamycin and piperacillin- tazobactam and *E. coli* (6 out of 6) were sensitive to meropenem and linezolid. All isolated MRSA (2 out of 2) were sensitive to linezolid [Table/Fig-4].

Modalities of treatment: The different procedures followed during the stay in the hospital were wound debridement, VAC dressing, wound irrigation or vinegar dressing. Most of the diabetic wounds were treated by daily or alternate wound debridement, SSI were treated by wound irrigation and daily dressing and most of the arterial and venous ulcers were treated by vinegar dressing [Table/Fig-5].

Organism isolated	SSI	DU	A/V
Citrobacter	-	1	-
Citrobacter+MRCNSA	1	-	-
Citrobacter+*Pseudomonas*	1	-	-
E. coli (ESBL Producer)	1	3	1
E. coli+*Enterococcus*	-	1	-
E. coli+*Klebsiella*	-	1	-
E. coli+Proteus Vulgaris	-	1	-
Klebsiella (ESBL Producer)	-	5	2
Klebisella+*S. aureus*	1	-	-
Klebsiella+Citrobacter	-	1	-
Klebsiella+Enterobacter	-	1	-
Klebsiella+Proteus Vulgaris	-	1	-
MRCNSA	8	-	3
MRCNSA+*Escherichia coli* (ESBL Producer)	1	-	-
MRCNSA+*Pseudomonas*	1	-	-
MRSA	4	2	7
Pseudomonas	2	3	7
Grand total	20	20	20

[Table/Fig-3]: Microbial spectre.
MRCNSA: Methicillin-resistant coagulase-negative staphylococci; ESBL: Extended spectrum beta-lactamases; MRSA: Methicillin-resistant *staphylococcus aureus*

and 75% patients of arterial/venous ulcer were allowed the wound to heal by secondary intention [Table/Fig-5].

Period of hospitalisation: The patients having diabetic wounds had a longer stay in the hospital followed by the arterial/venous ulcer. The mean duration of hospital stay in three groups was statistically significant [Table/Fig-5].

DISCUSSION

Skin ulceration has always been a major problem. In the present study, an attempt has been made to know the microbial spectra responsible for the postoperative wound infections and infection of chronic leg ulcers and their antibiotic sensitivity pattern.

Age distribution: A study conducted by Carvalho RLR et al., showed that the mean age of patients having SSI was 54.2 years±16.4 (18-99), with a median of 55 years [6]. In the present study, patients with DU were within the age group of 41-60 years affecting males more than females which is correlates with the above authors.

Venous Leg Ulcers (VLU) that develop as a consequence of chronic venous hypertension occur with a prevalence of 0.12-0.32% most commonly in those aged over 65 years. As per other studies the prevalence of leg ulceration remains relatively constant and continues to affect 0.14-2% of the population, depending on the methodology used [7]. In the present study, patients having venous ulcer were within the age group of 41-60 years which is similar to the above

Ulcer	Organism isolated	Antibiotic sensitivity																											
		AZ		AM		AK		CH		CA		CE		CF		CT		M		E		G		I		L		LZ		V		PC	
		s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r	s	r
Non specific ulcer	*P. aureginosa*	2	5	1	6	6	1	2	5	2	5	1	6	5	2	0	7	5	2	5	2	5	2	5	2	6	1	4	3	6	1	5	2
	Klebsiella	2	0	0	2	2	0	0	2	0	2	0	2	2	0	0	2	2	0	2	0	2	0	2	0	2	0	2	0	1	1	2	0
	MRSA	0	7	6	1	6	1	5	2	2	5	2	5	1	6	1	6	7	0	7	0	2	5	7	0	4	3	7	0	7	0	7	0
	MRCNS	0	3	2	1	3	0	1	2	0	3	0	3	2	1	1	2	1	2	0	3	1	2	0	3	0	3	3	0	3	0	3	0
SSI	MRCNS	3	8	2	9	7	4	6	5	2	9	3	8	5	6	7	4	11	0	3	8	3	8	6	5	9	2	10	1	10	1	6	5
	E. coli	0	2	0	2	2	0	0	2	0	2	0	2	1	1	1	1	2	0	0	2	2	0	1	1	1	1	2	0	2	0	2	0
	MRSA	3	1	1	3	4	0	3	1	3	1	1	3	3	1	3	1	4	0	3	1	4	0	4	0	3	1	3	1	4	0	4	0
	P. aureginosa	0	4	0	4	2	2	1	3	1	3	1	3	2	2	1	3	3	1	1	3	2	2	2	2	2	2	4	0	4	0	2	2
Diabetic ulcer	*Klebsiella*	5	4	2	7	8	1	6	3	4	5	4	5	7	2	5	4	9	0	8	1	9	0	7	2	7	2	7	2	7	2	9	0
	E. coli	3	3	3	3	5	1	0	6	2	4	2	4	3	3	2	4	6	0	4	2	2	4	5	1	3	3	6	0	5	1	5	1
	P. aureginosa	0	3	1	2	1	2	0	3	0	3	0	3	1	2	0	3	2	1	1	2	1	2	1	2	1	2	0	3	1	2	1	2
	MRSA	0	2	0	2	2	0	2	0	2	0	0	2	0	2	0	2	2	0	1	1	2	0	2	0	2	0	2	0	2	0	2	0

[Table/Fig-4]: Antibiotic sensitivity of the isolates.
AZ: Azithromycin; AM: Amoxycillin; AK: Amikacin; CH: Chloramphenicol; CA: Ceftazidime; CE: Cefotaxime; CF: Ciprofloxacin; CT: Ceftriaxone; M: Meropenem; E: Erythromycin; G: Gentamycin; I: Imipenem; L: Levofloxacin; LZ: Linezolid; PC: Piperacillin+Tazobactam; V: Vancomycin

Parameters		DU	SSI	A/V	Total
Modalities of treatment	VAC dressing	8	-	-	8
	Wound debridement	12	6	8	26
	Wound irrigation and dressing	-	-	10	10
	Daily dressing	-	5	2	7
	Vinegar dressing	-	9	-	9
Treatment outcome	Healed by secondary intention	5	4	15	24
	Secondary suturing	1	14	0	15
	Grafting	14	2	5	21
Mean period of hospitalisation (In days)		26.9±5	13±3	16.7±5	p-value# <0.5 #ANOVA-F-value- 42.9

[Table/Fig-5]: Treatment modalities and outcome.
#: ANOVA

Outcome of treatment: A 70% of the diabetic wounds were treated by grafting, 70% of SSI patients were treated by secondary suturing

studies. The higher incidence in our Indian study may be because of productive age group population in our country working in the field or in other occupation involving prolonged hours of standing.

Sex distribution and ulcer: Morikane K et al., in their study showed the rate of SSI was higher in male patients [7]. As per the literature the incidence of varicose veins in females is three times more when compared to males. Callow AD and Ernst CB in Switzerland recorded a ratio of 1:1 [8]. Callam MJ, in England and Margolis DJ et al., recorded a ratio of 1:2 [9,10]. In the present study, 42 male and 18 female patients presented with wound infection accounting to 70% male and 30% female. The SSI and chronic leg ulcers are seen more in males than in females. Males are more affected than females because, working outdoor make them more vulnerable for trauma and sequelae.

Associated risk factors: Studies have shown that risk factors such as Body Mass Index (BMI), smoking, PVD, neuropathy and pre-existing chronic diseases are associated with SSI [11]. SSI rates are higher in patients who are more debilitated or who have systemic diseases, such as diabetes mellitus as shown by Franco LMC et al., in his study [12]. In a series of cases conducted by de Castro Franco

LMC et al., out of 57.1% of patients presenting with SSI 13% were diabetic, indicating a 10 fold increase in the risk of SSI [5].

Risk factors for development of VLUs include older age, female sex, obesity, trauma, immobility, congenital absence of veins, Deep Vein Thrombosis (DVT), phlebitis, and factor V Leiden mutation [13].

In the present study, patients having diabetes mellitus and smokers have more incidence of wound infection. Among 60 cases of the present study, 35% of diabetic, 40% of SSI and 65% of arterial and venous ulcers were smokers. A 68.3% of present patients had diabetes mellitus. Poor glycaemic control and reduced immunity are additional factors for the development of infection in diabetics.

Type of surgery and associated SSI: One of the most important factors influencing the incidence of postoperative wound infections is the type of surgery and degree of contamination [Table/Fig-6] [14-17].

Author and year	Type of wound	No. of cases	%
Cruse PJ and Foord R [14] (1973)	Clean	732	1.5
	Clean contaminated	720	7.7
Agarwal PK [15] (1984)	Clean	-	36.17%
	Clean contaminated	-	57.14%
	Contaminated	-	100%
Anvikar KR et al., [16] (1999)	Clean	91	2.6
	Clean contaminated	104	4.8
Yadav UK et al., [17] (2016)	Clean contaminated	-	18.2%
	Contaminated	-	33.3%
	Dirty	-	45.5%
Present study (2019)	Clean	6	35%
	Clean contaminated	14	65%

[Table/Fig-6]: Comparison of incidence of wound infection in relation to risk class by various authors is as follows [14-17].

The present study is comparable with various authors' studies and it correlates with the study of Rhoads DD et al., [18]. Findings in the various studies have shown that there is significant rise in infection rate with increased degree of operative contamination [19].

Postoperative day infection in SSI: In the present study, 18 out of 20 cases of SSI was diagnosed between 3rd and 5th postoperative day. The peak was noted on 5th postoperative day.

Microbial spectra in ulcers and antibiotic sensitivity:

Rhoads DD et al., study showed the various organisms isolated from different skin ulceration as follows [18]:

DU: *Enterococcus*, *Pseudomonas*, *Streptococcus* and *Staphylococcus*

Surgical site: *Staphylococcus*, *Enterococcus* and *Pseudomonas*

VLU: *Staphylococcus*, *Streptococcus* and *Pseudomonas*

Kowli SS, showed, *Staphylococcus aureus* was the predominant organism isolated with an average of 45.6% and Agarwal PK published *E. coli* is the most common organism causing postoperative wound infection [15,20]. Carvalho RLR et al., isolated *Staphylococcus aureus* (24.3%; 43/177) and *Escherichia coli* (15.3%; 27/177) as the main microorganisms causing SSI [6].

Golia S et al., showed *Staphylococcus aureus* as the most common isolate followed by *Escherichia coli* and Coagulase Negative Staphylococci (CoNS) [21]. Other organisms isolated were *Pseudomonas aeruginosa, Enterococcus, Klebsiella pneumoniae* and *Proteus mirabilis*. Among them, 88.8% of *S. aureus* and 50% of CoNS isolates were methicillin resistant strains. 80% of *E. coli* and 100% of *Klebsiella* species were ESBL producers [Table/Fig-7] [22-25].

The present study is comparable with other author's studies. *Staphylococcus aureus* is most commonly found in SSI probably as it transiently colonises the nares of 30-40% of the population, has a predominant role in hospital cross infections and also emerges with virulent antibiotic resistant strains.

Author (Year)	Country	Microorganism Isolated
Kanth KT et al., [23] (2013)	India	*S. aureus* (18.1%), *P. aeruginosa* (16.2)
Dinda V et al., [24] (2013)	Kenya	*S. aureus, klebsiella*
Guta M et al., [25] (2014)	Ethiopia	*S. aureus, klebsiella*
Present study (2019)	India	MRCNS (40%) MRSA (20%)

[Table/Fig-7]: Microbial profile obtained by various authors in SSI [23-25].

In the present study, most of the organisms were sensitive to vancomycin followed by linezolid, piperacillin-tazobactam, meropenem, imipenem, levofloxacin, gentamycin, amikacin, ciprofloxacin and ceftriaxone. This pattern of antibiotic sensitivity correlated with the study of Anvikar AR et al., [16].

Kowli SS showed gentamycin, cloxacillin, cotrimoxazole, chloramphenicol as the most sensitive antibiotics postoperatively for gram positive aerobes and cephaloridine, gentamycin, kanamycin as sensitive against gram negative aerobes [20]. Mengesha RE et al., proved 83.1% of gram negative and 100% of gram positive isolates were sensitive to gentamycin and vancomycin respectively [26]. Krishna Kanth T et al., in his study showed CoNS was susceptible to linezolid, minocycline, sparfloxacin and teicoplanin and *S. aureus* showed susceptibility to teicoplanin, amoxiclav and sparfloxacin [Table/Fig-8] [25,27].

Author	Study (year)	Organism	Sensitive	Resistant
Raza MS et al., [27]	Nepal (2013)	*S. aureus*	Vancomycin	
Guta M et al., [25]	Ethiopia (2014)	*S. aureus*	Gentamycin	
Present study	India (2019)	CoNS	Vancomycin	Amoxicillin, cefotaxime

[Table/Fig-8]: The antibiotic sensitivity pattern obtained by various studies in SSI [25,27].

Diabetic Ulcer (DU): DU present with polymicrobial infection. In most of the study's high prevalence of multi-drug resistant pathogens was observed in DU [28]. Ranjini J showed that gram negative bacilli were the predominant isolates in diabetic wounds (78.98%). The resistance patterns prevalent among gram negative organisms include resistance to extended spectrum cephalosporins and penicillin due to production of ESBLs and Amp C β-lactamases [Table/Fig-9] [21,29-33].

Author	Study	Organism	Percentage
Gadepalli R et al., [29]	India (2006)	*E. coli*	54.5%
Varaiya AY et al., [30]	2008	*E. coli*	46.5%
		Klebsiella	44.4%
Shobha KL et al., [31]	2009	*K. pneumonia*	27.3%
		E. coli	25.2%
		Pseudomonas spp	21.42%
		Acinetobacter spp	25%
Umashankari J et al., [32]	2012	*Klebsiella*	59.5%
		E. coli	40%
Golia S et al., [21]	2013	*E. coli*	40.8%
		Klebsiella	38.8%
Umadevi S et al., [33]	2016	*K. pneumoniae*	20.5%
		P. aeruginosa	17.0%
		S. aureus	17.0%
		Escherichia coli	14.6%
		CoNS	7.0%
Present study	India (2019)	*K. pneumoniae*	35%
		E. coli	15%

[Table/Fig-9]: The microbial spectra by various authors in Diabetic Ulcers (DU) [21,29-33].

Earlier other studies have documented gram positive bacteria as the predominant organisms associated with diabetic foot infections [34]. Hence, a shifting trend of gram positive bacteria being replaced by gram negative bacteria as most common agents causing diabetic foot infections has been noted. A number of studies have found that *S. aureus* is the main causative pathogen in diabetic foot ulcers, but two recent investigations reported a predominance of gram-negative aerobes [29,34]. In India, gram negative bacteria are commonly isolated from diabetic foot infections when compared to western countries where gram positive cocci are predominant [35]. The ESBL production is one of the common mechanisms of drug resistance among the Enterobacteriaceae.

Diane M et al., in their study showed that the predominant aerobic organisms in diabetic foot ulcers were oxacillin-susceptible *S. aureus* (14.3%), oxacillin-resistant *S. aureus* (4.4%), coagulase negative *Staphylococcus* species (15.3%), *Streptococcus* species (15.5%), *Enterococcus* species (13.5%), *Corynebacterium* species (10.1%), members of the family Enterobacteriaceae (12.8%), and *Pseudomonas aeruginosa* (3.5%) [36]. The most sensitive antibiotic for diabetic foot ulcers in the present study was meropenem. Other antibiotics were linezolid, piperacillin-tazobactam, vancomycin, imipenem, erythromycin, gentamycin, amikacin, ciprofloxacin and levofloxacin.

Umadevi S et al., in their study showed that the members of *Enterobacteriaceae* as well as *Pseudomonas* and Acinetobacter were found to be susceptible mainly to amikacin, piperacillin-tazobactam and imipenem [33]. *S. aureus* and *Enterococcus* spp. were susceptible mostly to vancomycin, with varying susceptibility to tetracycline. A 56% of the isolates belonging to Enterobacteriaceae were producing ESBL and 65.5% of *S. aureus* were methicillin-resistant. Thus amikacin, piperacillin-tazobactam, imipenem was active against gram negative bacilli, while vancomycin was found to be active against gram positive bacteria [36].

Varaiya AY et al., in his study has shown antibiotic sensitivity patterns in which *E. coli* and *Klebsiella* are 100% resistant to ampicillin and sensitivity to amikacin and gentamycin are 66.2% and 31.3% for *E.Coli* and 63.5% and 35.6%, for *Klebsiella*, respectively [30].

As reported by Varaiya AY et al., imipenem is 100% sensitive for all ESBL strains of Enterobacteriaceae. Anitha S and Natarajan V, isolated 60 Enterobacteriaceae from diabetic foot ulcer patients among which 27 (45%) isolates were ESBL producers [30,37]. The ESBL production is predominantly seen in *E. coli* (48.1%) followed by *Klebsiella* (44.4%), *P. mirabilis* (3.3%) and *P. vulgaris* (3.3%). The ESBL producers were sensitive to imipenem (88.9%), amikacin (77.8%) and gentamycin (55.6%) whereas highly resistant to ampicillin, amoxiclav, cefuroxime and ceftriaxone.

Arterial ulcer/venous ulcers: Bacterial contamination of VLU is well characterised and clinical infection is associated with wound breakdown and impaired healing. Cooper R et al., in his study showed a wide range of wound colonising organisms [38]. They were mainly *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Streptococcus*, *Staphylococcus* being one of the most common pathogens colonising venous ulcers.

Moore K et al., in his study showed that *S. aureus* (64.3%), *Corynebacterium striatum* (60.6%), *Pseudomonas aeruginosa* (32.6%), *Proteus mirabilis* (16.1%) were the most common organisms colonising [39].

A study conducted by Gethin G and Cowman S, showed staphyloccocus was the most common wound isolate at baseline, being found in 41 (38%) of all wounds. At baseline, methicillin-resistant *Staphylococcus aureus* (MRSA) was 24% of them [40]. Brook I et al., in his study showed CVLU have a polymicrobial aerobic-anaerobic flora [41]. The predominant aerobic organisms were *Staphylococcus aureus* (26 isolates), group D Streptococci and *Escherichia coli*. The predominant anaerobes were *Peptostreptococcus*, Bacteroides fragilis group and *Propionibacterium acnes*. Hansson C in his study showed *Staphylococcus aureus* was the most common organism followed by *Enterococcus faecalis*, *Enterobacter cloacae* and some fungi in chronic VLU [42]. In a bacterial profiling study, Gjodsbol K et al., found that chronic VLUs showed *Staphylococcus aureus* (93.5%), *Enterococcus faecalis* (71.7%), *Pseudomonas aeruginosa* (52.2%), CoNS (45.7%), Proteus species (41.3%), and anaerobic bacteria (39.1%) [43].

Fazli M et al., conducted a study on spatial organisation of *Pseudomonas aeruginosa* and *Staphylococcus aureus* in chronic VLUs of nine patients [44]. Out of the nine patients four patients had *S. aureus*, another four patients had *P. aeruginosa* and one patient had a wound that contained both *S. aureus* and *P. aeruginosa*. In the present study, the most common organism isolated from non specific arterial and venous ulcers were *Pseudomonas* followed by MRSA and MRCNS. The most sensitive antibiotic for arterial and venous ulcers was Vancomycin. Other sensitive antibiotics were linezolid, piperacillin-tazobactam, meropenem, imipenem and levofloxacin.

Different modalities of treatment: The different procedures undergone by the wound during the stay in hospital was wound debridement, VAC dressing, wound irrigation or vinegar dressing. More than two-thirds (71.2%) of diabetic wound patients received surgical treatment, mainly in the form of debridement seen in a study conducted by Gadepalli R et al., [29].

- In the present study most of the diabetic wounds were treated by daily or alternate day wound debridement
- SSI were treated by wound irrigation and daily dressing
- Most of the arterial and venous ulcers were treated by vinegar dressing.

Results from various procedures: In the present study,

- 70% of the diabetic wounds were treated by grafting,
- 70% of SSI patients were treated by secondary suturing and
- 75% patients having an arterial or venous ulcer were allowed to heal by secondary intention.

Days of stay in hospital: In a study conducted by Yadav UK et al., the median length of hospital stay with SSI was 16 days while the median length of hospital stay without SSI was 10 days [17]. In a study conducted by de Castro Franco LM et al., the median length of hospital stay for SSI was three days, the longest hospital stay being (31 days) for SSIs [5].

The long duration of hospitalisation can be explained by the refractory to the treatment of the lesions owing to the diminished resistance of the body, hyperglycemia, impaired hormonal defense mechanisms and resistance of the organism to antibiotic therapy.

- In the present study, patients having SSI were admitted for a mean of 13±3 days, DU patients admitted for 26.9±5 and DU patients stayed the longest in the hospital.
- A/V ulcer patients admitted for 16.7±5 days

This research strength, being a cross sectional study, was cost effective, reliable and versatile containing multiple variables.

Limitation(s)

However, limitation of this study is that it consisted of 60 study participants (small study group) at a single center with a short study period. The authors recommend a multi-centric study with a larger study population to generalize the study findings.

CONCLUSION(S)

In spite of all the efforts wound infection is the most common problem in the surgical wards, it not only prolongs the hospital stay of the patient but increases the daily expenditures. This affects the patients socially, psychologically, economically and restraining them from performing routine work. This requires a thorough understanding of various patient related, microbial and surgical factors which have an aggregate effect on causation of wound

infection. In our study, the most common organism found in SSI, DUs and non specific ulcers were MRCNS, ESBL producer namely *Klebsiella* and *Pseudomonas* respectively and showed maximum sensitivity to vancomycin, meropenem respectively. The sensitivity pattern changes from hospital to hospital, population to population and time to time. The antibiotic sensitivity test results must be followed to avoid the emergence of the resistant strains. Since the risk factors and determinants of wound infection are largely modifiable or preventable, it is the collective effort of the surgeons to prevent this infection.

REFERENCES

[1] Leaper DJ. Surgical Infection. Bailey And Love's Short Practice of Surgery, 2004, 25th Edition; P 32-48.
[2] Briggs M, Closs, SJ. The prevalence of leg ulceration: A review of the literature. EWMA J. 2003;3(2):14-20.
[3] Large M, Stubbs E, Benn R, Beard-Pegler MA, Harbour C, Vickery AM. A study of coagulase negative staphylococci isolated from clinically significant infections at an Australian hospital. Pathology. 1989;21:19-22.
[4] Schwartz Si, Comshires G, Spencer Fc, Dally Gn, Fisher J, Galloway Ac. Principles of Surgery, 9th Edition. Chapter 6 "Surgical Infections" Ny: McGraw Hill Companies; 2010.
[5] de Castro Franco LM, Cota GF, de Almeida AGI, Duarte GMH, Lamounier L, Pereira PFS, et al. Hip arthroplasty: Incidence and risk factors for surgical site infection. Canadian Journal of Infection Control. Spring. 2018;33(1):14-19.
[6] Carvalho RLR, Campos CC, Franco LMC, Rocha AM, Ercole FF. Incidence and risk factors for surgical site infection in general surgeries. Rev Latino Am. Enfermagem. 2017;25:e2848.
[7] Morikane K. Epidemiology and risk factors associated with surgical site infection after different types of hepatobiliary and pancreatic surgery. Surg Today. 2017;47:1208-14.
[8] Callow AD, Ernst CB. Vascular surgery: Theory and practice New York: Appleton and Lange publications; 1996;1427-573.
[9] Callam MJ. Epidemiology of varicose veins. British Journal of Surgery. 1994;81:167-73.
[10] Margolis DJ, Berlin JA, Strom Bl. Risk factors associated with the failure of a venous leg ulcer to heal. Arch Dermatol. 1999;135:920-26.
[11] Centers For Disease Control and Prevention (CDC). Procedure Associated Module: surgical site infection event. Atlanta, 2016;29.
[12] Franco LMC, Ercole FF, Mattia A. Infecção cirúrgica em pacientes submetidos a cirurgia ortopédica com implante. Rev SOBECC. 2015;20(3):163-70.
[13] Etufugh CN, Phillips TJ. Venous ulcers. Clin Dermatol. 2007;25:121-30.
[14] Cruse PJ, Foord R. A five-year prospective study of 23,649 surgical wounds. Arch Surg. 1973;107:206-10.
[15] Agarwal PK. Incidence of postoperative wound infection Aligarh. Indian journal of surgery. 1984;46(6):326-33.
[16] Anvikar AR, Deshmukh AB, Karyakarte RP, Damie AS, Patwardhan NS, Malik AK, et al. A one year prospective study of 3280 surgical wounds. Indian J Med Microbial. 1999;17(3):129-32.
[17] Yadav UK, Mishra G, Jha R, Bhatta PN. Surgical site infection in laparotomy, Shrestha Journal of Universal College of Medical Sciences. 2016;4(1):22-25.
[18] Rhoads DD, Wolcott RD, Sun Y, Dowd SE. Comparison of Culture and Molecular Identification of Bacteria in Chronic Wounds. Int J Mol Sci. 2012;13:2535-50.
[19] William G. Cheadle. Surgical Infections. 2006;7(1):s07-11.
[20] Kowli SS. Hospital infection. Indian Journal of Surgery. 1985;47(11):475-86.
[21] Golia S, Hittinahalli V, Reddy KM, Karjigi KS. Detection of ESBL producers from pus samples of diabetic skin and soft tissue infections. Int J Biol Med Res. 2013;2732-35.
[22] Golia S, Asha S, Kamath B, Nirmala AR. A study of superficial surgical site infections in a tertiary care hospital at Bangalore. Int J Res Med Sci. 2014;2(2):647-52.
[23] Kanth KT, Vasndhara T, Chandra DS, Kumar BA, Adiveni T, Padmini P, et al. A study of epidemology and microbiology of surgical wound infections in RMMCH. International Journal of Medical and Applied Sciences. 2013;2(2):01-07.
[24] Dinda V, Gunturu R, Kariuki S, Hakeem A, Raja A, Kimang A. Pattern of pathogens and their sensitivity isolated from surgical site infections at the Aga khan university hospital, Kenya. Ethiop J Health Sci. 2013;23(2):141-49.
[25] Guta M, Aragaw K, Merid Y. Bacteria from infected surgical wounds and their antimicrobial resistance in Hawassa University Referral Teaching Hospital, Southern Ethiopia. AJMR. 2014,8(11):1118-24.
[26] Mengesha RE, Kasa BG, Saravanan M, Berhe DF, Wasihun AG. Aerobic bacteria in post-surgical wound infections and pattern of their antimicrobial susceptibility in Ayder Teaching and Referral Hospital, Mekelle, Ethiopia. BMC Research Notes. 2014;7:575.
[27] Raza MS, Chander A, Ranabhat A. Antimicrobial susceptibility patterns of the bacterial isolates in postoperative wound infections in a tertiary care hospital, Kathmandu, Nepal. OJMM. 2013;3(3):159-63.
[28] Alavi SM, Khosravi AD, Sarami A, Dashtebozorg A, Montazeri EA. Bacteriologic study of diabetic foot ulcer. Pak J Med Sci. 2007;23:681-84.
[29] Gadepalli R, Dhawan B, Sreenivas V, Kapil A, Ammini AC, Chaudhry R. A Clinico-microbiological study of diabetic foot ulcers in an indian tertiary care hospital. Diabetes Care. 2006;29(8):1727-32.
[30] Varaiya AY, Dogra JD, Kulkarni MH, Bhalekar PN. *Escherichia coli* and *Klebsiella Pneumonia* in Diabetic Foot Infections. Indian J Pathol Microbiol. 2008;51:370-72.
[31] Shobha Kl, Ramachandra YL, Rao G, Majumder S. Extended Spectrum beta-Lactamases (ESBL) in gram negative bacill at a tertiary care hospital. Journal of Clinical and Diagnostic Research. 2009;3:1307-12.
[32] Umashankari J, Jeya M, Sekar C. Microbiological Study of Diabetic Foot Ulcers and Analysis of ESBL Activity of the Bacterial Isolates. J Phar Res. 2012;5:2329-32.
[33] Umadevi S, Kumar S, Joseph NM, Easow JM, Kandhakumari G, Sreenivasan S, et al. Microbiological study of diabetic foot infections. Indian Journal of Medical Specialities. 2011;2(1):12-17.
[34] Lipsky BA, AR Berendt, HG Deery, JM Embil, WS Joseph, AW Karchmer, et al. Diagnosis and treatment of diabetic foot infections. Clin Infect Dis, 2004;39:885-910.
[35] Saseedharan S, Sahu M, Chaddha R, Pathrose E, Bal A, Bhalekar P, et al. Epidemiology of diabetic foot infections in a reference tertiary hospital in India. Brazilian Journal of Microbiology. 2018;49(2):401-06.
[36] Citron DM, Goldstein EJC, Merriam CV, Lipsky BA, Abramson MA. Bacteriology of Moderate-to-Severe Diabetic Foot Infections and In Vitro Activity of Antimicrobial Agents. J Clin Microbiol. 2007;45(9):2819-28.
[37] Anitha S, Natarajan V. Prevalence of ESBL Producing Enterobacteriaceae in Diabetic Foot Ulcers. IAIM. 2017;4(11):122-127.
[38] Cooper, R, Molan, P, Harding, KG. Antibacterial activity of honey against strains of *Staphylococcus aureus* from infected wounds. J Royal Soc Med. 1999;92:283-85.
[39] Moore K, Hall V, Paull A, Morris T, Brown S, McCulloch D, et al. Surface bacteriology of venous leg ulcers and healing outcome. J Clin Pathol. 2010;63(9):830-34.
[40] Gethin G, Cowman S. Bacteriological changes in slough venous leg ulcers treated with manuka honey or hydrogel: An RCT. J Wound Care. 2008;17(6):241-44.
[41] Brook I, Frazier EH. Aerobic and anaerobic microbiology of chronic venous ulcers. Int J Dermatol. 1998;37(6):426-28.
[42] Hansson C, Hoborn J, Möller A, Swanbeck G. The microbial flora in venous leg ulcers without clinical signs of infection. Repeated culture using a validated standardised microbiological technique. Acta Derm Venereol. 1995;75(1):24-30.
[43] Gjødsbol, K, Christiensen JJ, Karlsmark T, Jørgensen B, Klein BM, Krogfelt KA. Multiple Bacterial Species Reside in Chronic Wounds: A Longitudinal Study in the International Working Group On the Diabetic Foot The International Consensus On the Diabetic Foot. The International Working Group on the Diabetic Foot, Amsterdam, Netherlands (1999).
[44] Fazli M, Bjarnsholt T, Møller KK, Jørgensen B, Andersen AS, Krogfelt KA, et al. Non random Distribution of *Pseudomonas* aeruginosa and *Staphylococcus aureus* in Chronic Wounds. Journal of Clinical Microbiology. 2009;47(12):4084-89.

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