AEROBIC BACTERIOLOGICAL STUDY IN SURGICAL SITE INFECTIONS WITH SPECIAL REFERENCE TO MRSA, VRSA & VRE

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Abstract

Keywords: MRSA, Surgical Site Infections, VRSA.

Context: Surgical Site Infections (SSI) has been responsible for the increasing cost, morbidity and mortality related to surgical operations. They continue to be a major problem even in hospitals with most modern facilities and standard protocols of preoperative preparation and antibiotic prophylaxis.

Aim: The aim of this study to identify the aerobic bacteriological profile of Surgical Site Infections and its antibiogram, with special reference to Methicillin and Vancomycin resistance patterns of Staphylococcus aureus and Enterococci in this area.

Settings and Design: This is a prospective study carried out in the Department of Microbiology for one and half year at Siddhartha Medical College/Government General Hospital, Vijayawada, Andhra Pradesh, India.

Methods: Specimens were collected from those post operative wounds which were showing signs/symptoms of an infection during the stay of the patient's in the hospital and also during their follow up visits to the outpatient department after their discharge from the hospital according to CDC guidelines. All the specimens collected were transported immediately to the laboratory for further processing according to standard protocols.

Results: The prevalence of SSI is 8.94% (136 out of 1520 surgeries). Out of 136 clinically diagnosed SSIs, 113 organisms isolated. Out of 113 organisms, Staphylococcus aureus (28.31%) was the most predominant organism followed by Pseudomonas aeruginosa (17.69%). Among 32 strains of Staphylococcus aureus, 11(34.3%) MRSA and 1(3.12%) strain of VISA were detected.

Conclusion: An effective national and state level antibiotic policy and draft guidelines should be introduced to preserve the effectiveness of antibiotics and for better patient management.

Introduction

A major 30% - 50% of antimicrobials prescribed in hospital practice are for surgical prophylaxis to prevent post-operative wound infection. A reduction in the infection rate to a minimum level could have significant benefits in terms of both patient comfort and medical resources used.^[1]

The common pathogenic bacteria in SSIs include *Staphylococci*, *Pseudomonas species*, *Streptoccci*, *Enterococci*, *Esch.coli*, *Klebsiella*, *Enterobacter*, *Citrobacter*, *Acinetobacter*, *Proteus*, etc., [2][3]

Staphylococcus aureus is the commonest cause of SSI. An increased number of Methicillin Resistant Staphylococcus aureus (MRSA) strains have been seen worldwide.^[3]

Enterococci emergence in the past two decades is in many respects attributable to their resistance to many commonly used antimicrobial agents and ease with which they appear to attain and transfer resistant genes,^[4] thus giving rise to Enterococci with High-Level Aminoglycoside Resistance (HLAR) and glycopeptide resistance.

Data regarding Surgical Site Infections and its etiology, diagnostic criteria and treatment options are lacking in this region, hence the present study was undertaken to identify the aerobic bacteriological profile of Surgical Site Infections and its antibiogram.

Materials and Methods

This is a prospective study carried out in the Department of Microbiology, Siddhartha Medical College/Government General Hospital, Vijayawada, Andhra Pradesh, India for one and half year. Study includes 136 clinical specimens from patients who have developed Surgical Site Infections. The present study was taken up after the review and approval by the institutional ethical committee.

An informed oral consent was taken from the patients or their attendants.

Inclusion criteria

Patients underwent clean and clean-contaminated surgeries electively; contaminated and dirty surgeries on emergency basis.

Exclusion criteria

- 1. Wound site pre operatively infected will be excluded
- 2. Burn injuries and donor sites of split skin grafts.
- 3. Patients undergoing re-operations

Specimens were collected from those post operative wounds which were showing signs/symptoms of an infection during the patients stay in the hospital and also during their follow up visits to the outpatient department after their discharge from the hospital according to CDC guidelines [5].

All the specimens collected were transported immediately to the laboratory for further processing.

The samples collected were processed as follows [6]:

- a) Direct microscopic examination of gram stained smear.
- b) Inoculation of the samples onto different culture media for specific aerobic organisms and incubated for 24 hours at 37°C.
- c) Recognition of colonies followed by Identification of bacteria
- e) Antibiotic sensitivity testing which includes as per CLSI guidelines:
- a. Kirby-Bauer Disk Diffusion test using HiMedia Laboratories products, Mumbai, India.
- b. Detection of MRSA, VRSA, VRE
- c. E-test Vancomycin/Cefoxitin Dual strip by HiMedia Laboratories, Mumbai, India.
- d. Agar Screening and Agar Dilution methods Vancomycin powder by Himedia Laboratories, Mumbai, India.
- e. Detection of Beta lactamases using HiMedia antibiotic discs

Sample Size Calculation:

 $N=p(1-p)(Z_{\alpha/2}/\epsilon)^2$

N=Sample size

P=Prevalence rate in %

Z=Confidence interval of 95% which is equivalent to confidence of 1.96

E= Error.

Statistical Analysis:

All the data were entered into spread excel sheet and analyzed. All Quantitative descriptive variables were expressed as number, percentages. Statistical analysis was assessed by Graph pad software. The p value <0.05 is considered as significant.

Results

The prevalence of SSI is 8.94% (136/1520). Of the 136 clinically diagnosed cases of SSI, 59 (43.3%) cases belonged toclass I wound, 39 (28.6%) to class II, 32 (23.5%) to class III and 6 (4.41%) cases belonged to class IV wounds. In present study, 119 (87.5%) of 136 cases of SSI were diagnosed between 3rd and 8th postoperative day. 79 (58.08%) cases were diagnosed between 5th and 8th day with the peak on 5th day. Out of 136 cases of SSI, 28 (20.58%) patients had diabetes, 16 (11.76%) patients were smokers, 7 (5.14%) patients had diabetes and were smokers and postoperative drain was used on 15 (11.02%) patients.

Out of 136 clinically diagnosed cases of SSI, 95 (69.8%) were males and 41 (30.14%) were females with a male to female ratio of 2.37: 1. Maximum number of SSI patients was in the age group of 31-40 years followed by age group of 21-30 years (Table 1).

Table No. 1 Age and Sex distribution of patients with SSI

		Male Male	9		Female			Total	
Sl.No. S.No.	Age Group in Years	Clinicall y Diagnose d	Cultur e positiv e	% of culture positiv e	Clinicall y Diagnose d	Cultur e positiv e	% of culture positive	No	%
1.	0-10	5	3	2.20	2	1	0.73	7	5.14
2.	11-20	14	8	5.88	3	1	0.73	17	12.5
3.	21-30	16	16	11.7	9	7	5.14	25	18.3
4.	31-40	23	20	14.7	7	8	5.88	30	22.0
5.	41-50	15	11	8.08	7	5	3.67	22	16.1
6.	51-60	13	9	6.61	8	5	3.67	21	15.4
7.	61 and above	9	7	5.14	5	4	2.94	14	10.2
Total		95	75	55.1	41	28	20.5	103	100

Of the 136 clinically diagnosed cases of SSI, 103 (75.73%) samples were culture positive and 33 (24.26%) samples yielded no growth.

In the present study of 136 samples, 95 (69.85%) of positive Gram stained smears developed growth on culture and 6(4.41%) samples yielded no growth. Among the negative Gram stained smears, 8 (5.88%) were culture positive and 27(19.85%) were culture negative. At P=0.05 and df =1, the Chi Square value obtained as 76.5, it was significant (Table 2).

Table No.2 - Direct microscopy and culture positivity

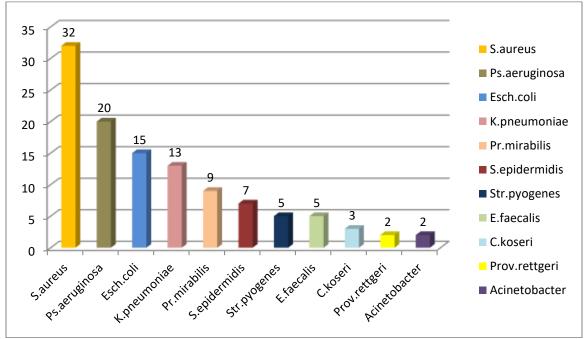
Sl.No.	Gram Stain		Positive culture	Negative culture	Total
		PC & GPC	31	2	
1	Positive	PC & GNB	54	4	101
		PC, GPC &GNB	10	0	

2	Nagativa	Few PC, No Org	8	10	35
2	Negative	No PC, No Org	0	17	33
Total	Total		103	33	136

SSI were more common in Contaminated and dirty wounds with percentages 93.7% and 100% respectively than in clean (55.9%) and clean contaminated (87.1%) wounds.

In this study, a total of 113 organisms isolated among these, 103 (91.1%) samples yielded a single organism on culture and 10 (8.8%) samples yielded 2 organisms (mixed). Out of 113 Culture positive samples, *Staphylococcus aureus* (28.31%) was the most predominant organism followed by *Pseudomonas aeruginosa* (17.69%) which, in turn followed by *Escherichia coli* (13.27%) and *Klebsiella pneumoniae* (11.50%) (Chart 1).

Chart 1. Types of aerobic bacteria isolated



Staphylococcus aureus (36.3%), was the most common organism isolated from class I wound infection, followed by *Pseudomonas aeruginosa* (15.1%). *Pseudomonas aeruginosa* (20.5%) was the predominant organism isolated from class II wound infection. The most common organism isolated from class III and IV wound infection was *Staphylococcus aureus* 26.6% and 50% respectively.

50% of gram negative isolates have shown susceptibility to Imipenem, Gentamicin, Amikacin, Tetracycline, Cotrimoxazole, Ceftriaxone, Ceftazidime, Ceftazidime/Clavulanic acid (Table 3).

Table 3. Sensitivity pattern of Gram Negative isolates

			1 11010 01 2	ensuring p	wite in of	3. tim 1.08	mirre isom				
Organis	AMX	PIT	CTX	CAZ	CAC	TE	COT	CIP	AK	GEN	IPM
ms											
	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)
Ps.aerugi	0(0)	16(80	-	9(45)	11(55)	13(65	11(55	8(40)	13(65	15(75	16(80)
nosa)))))	
(n=20)											

Esch.coli (n=15)	2(13.3)	-	8(53.3)	9(60)	8(53.3	10(66	8(53. 3)	6(40)	8(53. 3)	8(53. 3)	15(10 0)
K.pneum oniae (n=13)	3(23.07)	-	7(53.8)	8(61.5)	7(53.8	9(69. 2)	7(53. 8)	5(38. 4)	9(69. 2)	10(76	13(10 0)
Pr.mirab ilis (n=9)	1(11.1)	-	3(33.3)	6(66.6)	5(55.5	6(66. 6)	3(33. 3)	2(22. 2)	5(55. 5)	6(66. 6)	9(9.99
C.koseri (n=3)	0	-	2(66.6)	2(66.6)	2(66.6	2(66. 6)	2(66. 6)	1(33. 3)	3(100	3(100	3(100)
Prov.rett geri (n=2)	0	-	1(50)	1(50)	1(50)	1(50)	2(100	2(10 0)	1(50)	2(100	2(100)
Acinetob acter (n=2)	0	1(50)	-	0	1(50)	2(100	2(100	1(50)	2(100	2(100	2(100)
Total – 64	6(9.3)	58 (90.6)	21 (32.8)	35 (54.6)	35 (54.6)	43 (67.1)	35 (54.6)	25 (39.0 6)	41 (64.0 6)	46 (71.8)	55 (85.9)

(AMX-Amoxycillin, PIT-Piperacillin/Tazobactum, CTX-Cefotaxime, CAC-Ceftazidime/ Clavulanic acid, TE-Tetracycline, COT-Cotrimoxazole, CIP-Ciprofloxacin, AK-Amikacin, GEN-Gentamicin, IPM-Imipenem).

50% of gram positive isolates have shown susceptibility to vancomycin, Linezolid, Gentamicin, Levofloxacin, Azithromycin, Clindamycin, Erythromycin, Amoxyclav (Table 4).

Table 4. Sensitivity pattern of Gram Positive isolates

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Organis	P	AMC	CN	CL	E	AZM	TE	COT	LE	GEN	LZ	VA
ms												
	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)
S.aureus	3(9.3)	21(65.6	21(65.6	23(71.	18(56.	21(65	12(37	11(3	18(56	18(56	31(96	32(100
(n=32)))	8)	2)	.6)	.5)	4.3)	.2)	.2)	.8))
S.epider	0	1(14.2)	7(100)	5(71.4)	6(85.7	6(85.	5(71.	5(71.	4(57.	6(85.	7(100	7(100)
midis)	7)	4)	4)	1)	7))	
(n=7)												
E.faecali	1(20)	3(60)	-	2(40)	2(40)	3(60)	2(40)	2(40)	2(40)	4(80)	5(100	5(100)
s (n=5))	
Str.pyog	3(60)	4(80)	3(60)	3(60)	4(80)	4(80)	3(60)	3(60)	4(80)	5(100	5(100	5(100)
enes))	
(n=5)												
Total =	7	29	28	33	30	34	22	21	28	33	48	49
49	(14.2)	(59.1)	(57.1)	(67.3)	(61.2)	(69.3)	(44.8)	(42.8	(57.1)	(67.3)	(97.9)	(100)
	, ,	, ,	, ,		, ,		, ,)				

(P-Penicillin, AMC-Amoxyclav, CN-Cefoxitin, CL-Clindamycin, E-Erythromycin, AZM Azithromycin, TE-Tetracycline, COT-Cotrimoxazole, LE-Levofloxacin, GEN-Gentamicin, LZ-Linezolid, VA- Vancomycin).

Out of 32 Staphylococcal strains, 4(12.5%) were constitutive MLSB (Macrolide-Lincosamide-Streptogramin B) resistant, 5(15.6%) were inducible MLSB resistant (inducible clindamycin resistant) and 5(15.6%) belonged to MS phenotype. 18(56.25%) strains were susceptible to both erythromycin and clindamycin.

In our study, among 32 strains of *Staphylococcus aureus*, 10 (31.2%) strains were methicillin resistant by cefoxitin disc diffusion test and 11 (34.3%) by cefoxitin E-test (Fig 1). 50% of MRSA isolates shown susceptibility to Vancomycin, linezolid, gentamicin, levofloxacin, azitromycin, erythromycin, clindamycin (Table 5).



Fig 1. Combined vancomycin & cefoxitin E test strips testing of S.aureus isolates

Table 5. Sensitivity pattern of MRSA and MSSA isolates

				3. Schstiff	Ny paritro	9						
Organis	P	AMC	CN	CL	E	AZM	TE	COT	LE	GEN	LZ	VA
ms												
	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)	S(%)
MRSA	0(0)	0(0)	0(0)	6(54.5)	7(63.6	7(63.	4(36.	4(36.	8(72.	8(72.	10(90	11(100
(n=11))	6)	3)	3)	7)	7)	.9))
MSSA	3(14.	15(71.4	21(100)	11(52.	11(52.	14(66	8(38.	7(33.	10(47	10(47	21(10	21(100
(n=21)	2))		3)	3)	.6)	1)	3)	.6)	.6)	0))
Total =	3(9.3)	15(46.8	21(65.6	17(53.	18(56.	21(37	12(37	11(3	18(56	18(56	31(96	32(100
32))	1)	2)	.5)	.5)	4.3)	.2)	.2)	.8))

Out of 32 Staphylococcal strains, Only 1(3.12%) strain was shown growth in 4ug/ml vancomycin Muller Hinton Agar plate, considered Vancomycin Intermediate Staphylococcus aureus, it was also MRSA and remaining strains are all sensitive to vancomycin by all methods (Fig 2, 3 & 4).

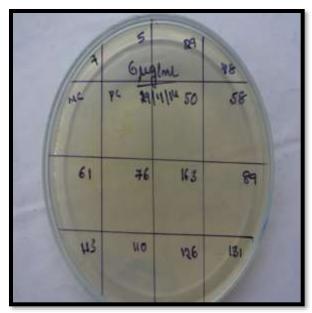


Fig 2. Agar Screening Method

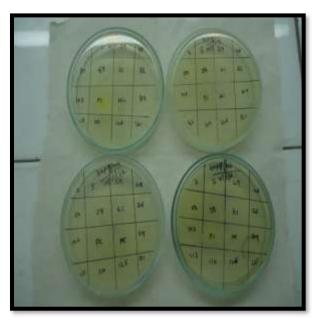


Fig 3. Agar Dilution Method

All 5 strains of Enterococcus faecalis were sensitive to vancomycin detected by vancomycin disk diffusion method, Agar screening, Agar dilution method and E- test strips.

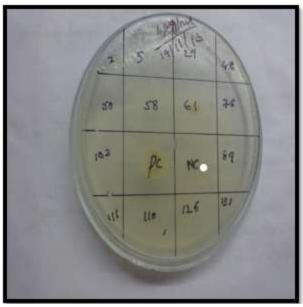


Fig 4. Showing VISA by Agar Dilution Method

Discussion

Surgical Site Infection (SSI) continues to be a major problem even in hospitals with most modern facilities and standard protocols of preoperative preparation and antibiotic prophylaxis ^[1]. SSIs are usually caused by the exogenous and endogenous microorganism that enters the operative wound during the course of the surgery ^[2].

The present generations of surgeon has seen increasing numbers of serious infections related to a complex combination of factors, including the performance of more complicated and longer operations; an increase in number of geriatric patients with accompanying chronic or debilitating diseases; many new surgical procedures with implants of foreign materials; a rapidly expanding number of organ transplants requiring the use of immunosuppressive agents; and increased use of diagnostic and treatment modalities that cause bacterial exposures or the suppression of normal host resistance.

Surgical Site infections were confirmed by bacteriological study, the overall infection rate was 6.77% as per this study (**Table 6**).

Table 6. Incidence of SSI by various studies

Author	Year	Country	Infection Rate
Lilani SP [9]	2005	India	8.95%
Rafael Lima Rodrigues de	2011	Brazil	3.4%
Carvalho et al [11]			
Narasinga rao Bandaru	2012	India	9.81%
[12]			
Farhan Sattar et al [13]	2016	Pakistan	33.68%
Marie Josee	2018	Rwanda	10.9%
Mukagendaneza et al [14]			
Present Study	2014	India	6.77%

Lilani SP et $al^{[7]}$ described that Surgical site infections are the third most commonly reported nosocomial infection and they account for approximately a quarter of all nosocomial infections.

This correlates with the study by Chia JYH *et al*^[12] in which 64% of the cases were diagnosed between the 5th and 8th postoperative day. Lilani SP *et al*^[7] observed that mean postoperative stay in patients who developed infections were almost four times (24.82 days) as compared to patients who did not develop SSI where the mean postoperative stay was 6.19 days.

In another study in Pune, there was a marginal preponderance of male patients developing SSI (7.4%) over female patients with SSI $(5.1\%)^{[13]}$. In contrast in Aligarh, females (27%) showed preponderance of SSI than males $(18\%)^{[14]}$. Similar findings were demonstrated by Mead *et al*^[15], who observed an increased Clean wound infection in patients less than 1 year old (2.7%) or greater than 50 years old (2.8%) versus those 1 to 50 years old (0.7%). Narasinga rao Bandaru *et al*^[9] observed that age of more than 50 years was found to be at risk factor for the postoperative wound infection.

This correlates with a study by Lilani SP $et\ al^{[7]}$, in which 14 (82.36%) out of 17 cases of SSI were culture positive and 3 (17.64%) were culture negative. In a study conducted by Similarly, out of 52 samples studied by Kownhar H $et\ al^{[16]}$, 46 (88.46%) were culture positive and 6 (11.54%) samples yielded no growth. Culture negativity may be due to antibiotic therapy prior to culture of material from an apparently infected site or due to the presence of fastidious or atypical organisms that do not grow on standard culture media or grow so slowly that plates are discarded before growth is apparent.

Staphylococcus is predominant pathogen in Surgical site Infection from many years. It may be due to presence of more Staphylococcal carriers among hospital staff who could be silent carriers or due to strong Virulence factors of Staphylococci and also emergence of Multidrug resistant staphylococci like MRSA.

Studies of previous years (before 1990) shown that *Escherichia coli* was second most predominant pathogen but Studies from 1990 onwards shown that *Pseudomonas aeruginosa* was the second most predominant. This may be due to fact that *Pseudomonas* is an emerging pathogen in hospital acquired infections & also in procedures where most sophisticated device are using **(Table 7)**.

Table 7. Prevalence of Bacterial isolates of various studies from surgical site infections

Organism	Edwards et al ²² (1976)	Udgaonk ar ²³ (1986)	Kownhar et al ²¹ (2008)	Gayathree naik et al ²⁴ (2011)	Setty NH et al ²⁵ (2014)	Maire Josee M et al ¹⁴ (2018)	Present study
S.aureus	30.3%	28.13%	37%	32.2%	55.5%	6%	28.31%
S.epidermidi s	21.5%	22.16%	-	1.48%	-	3%	6.19%
Enterococcu s	22.6%	0.6%	03%	1.6%	-	1	4.4%
Group A Beta hemolytic Streptococc us	-	1.01%	-	1.9%	-	-	4.4%
Pseudomon as aeruginosa	13.8%	13.36%	37%	12.8%	36.11%	-	17.69%
Escherichia coli	27.8%	21.25%	4.8%	8.9%	16.67%	15%	13.27%
Klebsiellapn eumoniae	11.75%	14.71%	8.0%	4.7%	27.78%	55%	11.50%
Proteus vulgaris	-	16.59%	4.8%	2.7%	-	-	-
Proteus mirabilis	9.2%	-	-	1.6%	13.89%	12%	7.9%

Citrobacterf reundii	-	1.16%	1.6%	2.3%	16.67%	-	2.65%
Enterobacte raerogenes	4.9%	-	-	1.2%	-	-	1
Acinetobact er	-	-	3.2%	5.8%	-	9%	1.76%

According to Goswami NN et al^[21] Pseudomonas aeruginosa were sensitive to ciprofloxacin (83.78%), gatifloxacin (51.35%), and meropenem (51.35%). Escherichia coli was sensitive to levofloxacin (72.41%) and ciprofloxacin (62.07%). Klebsiellapneumoniae was sensitive to ciprofloxacin (63.16%), levofloxacin (63.16%), gatifloxacin (63.16%), and linezolid (56.52%). Proteus mirabilis was sensitive to ciprofloxacin (75%) and linezolid (62.50). Proteus vulgaris was sensitive to ampicillin+sulbactam (57.14%) followed by levofloxacin (50%). As per their work Linezolid showing sensitivity against Gram negative bacteria. Escherichia coli and Klebsiella pneumoniae showed sensitivity to ciprofloxacin in similar to the present study. For Pseudomonas aeruginosa they used meropenem, observed more resistant where as in the present study Imipenem was used, the reisistance rate is slightly lower. Due to some difficulties Meropenem, Etrapenem and Linezolid were not used in the present study. From different studies there is an emergence of meropenem, and third generation cephalosporin resistant Pseudomonas aeruginosa. In contrast to present study Lilani SP et al^[7], reported that Pseudomonas aeruginosa exhibited 100% resistance to gentamycin. Other Gram negativebacilli were found to be 100% resistant to tetracycline followed by ampicillin (83.33%).

In line with this study Gayathree naik $et\ al^{[19]}$ reported that out of the 83 $Staphylococcus\ aureus$ isolates, Penicillin Sensitivity was seen in 7 (12.3%), Erythromycin sensitivity was seen in 36 (43.4%), Ciprofloxacin sensitivity was seen in 38 (45.8%), Cotrimoxazole sensitivity was seen in 58 (69.9%), Cefaperazone sensitivity was seen in 53 (66.3%), Netillin sensitivity was seen in 63 (75.9%) and Oxacillin sensitivity seen in 75 (90.4%) respectively. All the 83 (100%) isolates were sensitive to Vancomycin. Goswami NN $et\ al\ ^{[21]}$ reported that S. aureus was sensitive to Rifampicin (89.58%), levofloxacin (60.42%), and Vancomycin (54.17%).

Joyce SB *et al*^[22] also observed a high percentage of Penicillin resistance among strains of *Staphylococcus aureus* (91%) and *Enterococcus faecalis*(100%) causing SSI. 73% of the *Staphylococcus aureus* strains were resistant to Cotrimoxazole, 51% were resistant to ciprofloxacin and 11% of the strains were resistant to Gentamicin and Amikacin. *Pseudomonas aeruginosa* was resistant to Cotrimoxazole (94%), followed by103 Ciprofloxacin (58%), Gentamicin (15%) and Amikacin (9%).

The Prevalence of MRSA has varied from hospital to hospital in various countries. The incidence of MRSA in India ranges from 30-70% [23]. This wide range of MRSA prevalence rate is extremely difficult to explain these conflicting data with regards to both time and place of study, the variation is probably due to Differential clonal expansion and drug pressure in the community (**Table 8**).

Table 8. Rate of isolation of MRSA& MSSA among SSI in different study groups

S.No	Author	Year	MSSA	MRSA
2	Kownhar H et al ²¹	2008	79.3%	21.7%
4	Bhattacharya S ³¹	2012	74.55%	25.45%
5	Ranjan KP ³²	2013	72.04%	27.96%
6	Present study	2014	65.62%	34.37%

Gayathree naik et $al^{[19]}$ reported that the MRSA strains (100%) were sensitive to Vancomycin, Rifampicin, Teicoplanin, Linezolid, this finding is line with present study.

Bhattacharya S *et al*^[24] stated that the MRSA strains have been found to be 100% sensitive to linezolid and tigecycline followed by fucidin (92.51%), mupirocin (88.39%), levofloxacin (75.66%) and doxycycline (72.28%). No vancomycin resistant strains were detected, but 3 strains (1.12%) were found to be intermediately susceptible to it (VISA).

<u>Flannery EL</u> *et al*^[26] from university of Michigan, USA described that because cocolonization with MRSA and VRE precedes VRSA development, MRSA/VRE cocolonization in the device group occurred most frequently in wounds (4.1 per 100 resident-months).

Antony SJ et al^[27] from university school of medicine, Texas given that case series describing an outbreak of VRSA/VISA associated infections in orthopedic related procedures that occurred on a medical mission trip in Antigua, Guatemala.

Furthermore, Extensive use of vancomycin creates a selective pressure that favors the outgrowth of rare, vancomycin-resistant clones leading to heterogenous vancomycin intermediate *S. aureus* (hVISA) clones, and eventually, with continued exposure, to a uniform population of vancomycin-intermediate *S. aureus* (VISA) clones. These heterogeneous VISA (hVISA) are more common; reports from around the world indicate that 0.5%–20% of MRSA are heteroresistant^[28]. Currently, no standardized method for identifying hVISA exists. Population analysis profiling (PAP) has been proposed as the most precise method of determining heteroresistance. The present study didn't mention about hVISA because of few difficulties like could not get Control strain -MU and also PAP method is a gold standard for detection of hVISA. There are no much reports about VRSA/VISA in south India related to Surgical Site infections. Regarding Vancomycin Resistant Enterococci only 5 *Enterococci* strains were isolated out of 136 samples. For which Vancomycin resistant was studied by using all the four, methods and found them to be sensitive to Vancomycin. Still the sample size is too low and VRE has to be studied further.

VRSA should be cautiously interpreted as this might be because of false positive result by bacteria like acinetobacter^[29] and the resistance to vancomycin was not counterchecked by other laboratory.

Conclusion

From this study we conclude that MRSA are the commonest etiological agents of Surgical Site infections. Emergence of MRSA, VRSA and Beta lactamase producers like *Pseudomonas spp*, *Escherichia spp*, *Klebsiella spp*, added more severity to this clinical condition. Regular antimicrobial susceptibility surveillance is essential for area-wise monitoring of the resistance patterns. An effective national and state level antibiotic policy and draft guidelines should be introduced to preserve the effectiveness of antibiotics and for better patient management.

There should be a policy of mandatory infection control practices in every hospital, surveillance of HCWs and patients and also there is a need to emphasize the rational use of antimicrobials and strictly adhere to the concept of "reserve drugs" to minimize the misuse of available antimicrobials.

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