Postoperative Nosocomial Infections and Antimicrobial Resistant Pattern of Bacteria Isolated Among Patients Admitted at Ad-din Women's Medical College Hospital, Dhaka.

Mursheda Akter¹, Afzallunessa Binte Lutfor², Munir Hassan³, Ferdausi Chowdhury⁴, Arpita Nug⁵, Nayeema Akter⁶, Shimu Shaha⁷, Musrat Rahman⁸

¹Associate Professor, Department of Microbiology, Ad-din Women's Medical College, ²Professor & Head, Department of Microbiology, Addin Women's Medical College, ³Professor & Head, Department of Microbiology, Dhaka National Medical College & Hospital, ⁴Assistant Professor, Department of Microbiology, Ad-din Women's Medical College, ⁵Professor, Department of Microbiology, Ad-din Women's Medical College, ⁶Assistant Professor, Department of Gynae & Obstetric, Ad-din Women's Medical College, ⁷Assistant Professor. Department of Microbiology, Ad-din Women's Medical College, ⁸Assistant Professor & Head, Department of Pediatric surgery, Ad-din Women's Medical College

Abstract:

Objective: To find out the causative agents and antibiotic resistant pattern of organisms isolated from surgical site wound infection. Place of study: This study was carried out in microbiology department, Ad-din Women's Medical Collage, Moghbazar Dhaka. Method of study: The was carried out from January to December 2011 in microbiology department Ad-din women's medical college hospital. One hundred and thirty three clinically suspected surgical site wound infection cases were included in the study. Wound swab sample from surgical site was collected by sterile cotton swab. All the samples were immediately sent to the microbiology laboratory. Culture and sensitivity were done by standard bacteriological method. Result: Out of 133 SSIs cases, 82(51.57%) were culture positive. Out of 82 culture positive cases, 38(45.23%) were Gram positive and 46 (54.76%) were Gram negative. Predominant organisms were Esch.coli 20(23.80%) and Staphylococcus aureus 20 (23.80%). Most of the organisms were resistant to Cephalosporin group while Carbapenem, Colistin, Piperacillin Tazobactum and Amikacia showed low level of resistance. Quinolones were more effective against Gram positive than Gram negative bacteria. Methicillin resistant Staphylococcus aureus were 11 (55%). Conclusion : Esch.coli and Staphylococcus aureus were the major pathogen isolated from SSIs in our study. Cephalosporins were least effective drug. Carbapenem is the most effective drugs. For prevention of SSIs reinforcement of infection control policy for the health care worker is very important.

Introduction:

Surgical site infections (SSI) are the third most frequently reported nosocomial infections accounting for 14 to 16% of all infections in hospitalized patients. Surgical site infection is defined as an infection that occurs at the incision site within thirty days after surgery.¹ Infection of

the surgical site remains a complication of surgical procedures resulting in increased cost, morbidity and mortality.² In fact wound infection adds approximately more than 6000 US dollars to the hospital cost and more than 7 days hospital stay with consequent delay in work return. ^{3,4}. The risk of developing a surgical site infection

depends upon the balance between the factors determining the number of bacteria contaminating the site and the factors determining the resistance of the site against the infection. 5 Despite improvements in operating room practices, instrument sterilization methods, better surgical technique and best efforts by the practitioners, surgical site infections (SSIs) remain a major cause of nosocomial (hospital acquired) infections and rates are increasing globally6.7. The most common organism involved is Staphylococcus as it is most common normal flora of skin8. Major problem faced by the surgeon, is the wound infection caused by multi drug resistant bacteria. Organisms include both Gram positive cocci and Gram negative bacilli9.3.. Infection of the surgical site can be tackled properly by isolation and identification of bacteria causing infection and their susceptibility pattern to different antimicrobial agents so that these can be treated with appropriate antibiotics10. The emerging antibiotic resistant bacteria such as MRSA (Methicillin resistant Staphylococcus aureus), ESBL (Extended Spectrum of Beta lactamase) and their abilities for rapid evolution against antibiotics have increased the need for continuous monitoring and reporting of resistance and susceptibility pattern. Microbiologists have a role in providing guidance to the surgeon regarding the use of proper prophylactic antibiotic.

Materials and Method :

This retrospective study was carried out in the Department of Microbiology, Ad-din Women's Medical College, Dhaka, from January to December 2011. A total 159 wound swab/ pus were included in this study. Sample was collected by sterile tipped cotton swab, and the samples were sent to the microbiology laboratory immediately. The samples were inoculated on to blood agar and MacConkey's agar media, incubated aerobically at 37°C for 24 hours. Those plates showing no growth were incubated for another 24 hours. Isolated organisms were identified by their colony morphology, staining characteristics and other relevant biochemical tests as per standard methods. ^{11,12} All bacterial isolates were tested for antimicrobial susceptibility by the disc diffusion method against different antimicrobial agents .¹³ Mueller Hinton agar media was used for antibiotic susceptibility testing for all bacteria and blood agar media was used only for Streptococcus spp. For screening of Methicillin resistant *Staphylococcus aureus* (MRSA) oxacillin disc vas used. Within 30 minutes of applying the discs all plates were incubated aerobically at 35 °C for 16 to 18 hrs.¹⁴ *Staphylococcus aureus* resistant to oxacillin is considered as MRSA.

Results:

Department	No. of sample	No. of positive	No. of isolates	Gram positive	Gram negative
		culture		bacteria	bacteria
OBG	133	62(47.61%)	62	31(50%)	31(50%)
General Surgery	26	20(76.92%)	22	07 (31.81%)	15(68.18%)
Fotal	159	82 (51.57%)	84	38(45.23%)	46(54.76%)

 Table I: Rate of culture positive wound swab from different department

Table I shows that out of 159 samples, 133 were from SSI of patients from Gynae and Obstetric department and 26 were SSI of patients from General surgery department. Among 133 samples had undergone gynaecological and obstetric operation, 62 (46.61%) were culture positive and out of 26 samples who under went general surgical operation, 20 (76.92%) were culture positive. Among total 159 cases 82 were culture positive, from 82 culture positive cases total number of isolates were 84. Out of 84 isolates, Gram positive organisms were 38 (45.23%) and Gram negative organisms were 46 (54.76%).

 Table II: Type of organisms isolated from surgical site

 wound infection in Gynae and Obstetric department.

Organisms isolated	Number of organisms(percentage)		
Staph.aureus	16 (25.80%)		
Coagulase negative Staphylococcus	14 (23.58%)		
Esch.coli	11 (17,74%)		
Pseudomonas spp.	09 (14.51%)		
Acinetobacter spp.	07 (11.29%)		
Klebsiella spp.	03 (4.83%)		
Proteus spp.	01 (1.61%)		
Enterococcus spp.	01 (1.51%)		
Total	62		

Table II shows that, out of 62 isolates from Gynae and Obs SSI patients, *Staphylococcus aureus* 16 (25.80%), *CoNS* 14 (23.58%), *Esch.coli* 11 (17.74%), *Pseudomonas* spp. 09(14.51%). *Acinetobacter spp.* 07(11.29%), *Klebsiella spp.* 03(4.83%) *Proteus* and *Enterococcus* spp. 01 (1.61%) was recovered.

Table III: Organisms isolated from SSI of General surgery department

Organisms	Number
Esch.coli	09 (40.90%)
Staph.aureus	04 (18.18%)
Pseudomonas	03 (13.63%)
Morganella spp	01 (4.54%)
Coagulase negative staphylococcus	01 (4.54%)
Acinetobacter spp.	01 (4.54%)
Enterococci	01 (4.54%)
Beta Haemolytic streptococci	01 (4.54%)
Unidentified Gram negative bacilli	01 (4.54%)
Total	22

Table III shows among 22 isolates from SSI of surgery department, *Esch.coli* 9 (40.90), *Staphylococcus aureus* 04 (18.18%), *Pseudomonas spp.* 3 (13.63%) were recovered. *Morganella spp., CoNS, Acinetobacter, Enterococcus spp, Beta haemolytic streptococci* and unidentified Gram negative bacteria was 01(4.54%) from each respectively.

Table IV : Distribution of organisms isolated from surgical site wound infection (n=84).

Organisms	No. of organisms (%)
Staphylococcus aureus	20 (23.80)
Esch.coli	20 (23.80)
CoNS	15 (17.85)
Pseudomonas spp.	12 (14.28)
Acinetobacter spp.	08 (9.52)
Klebsiella spp.	03 (3.5)
Enterococcus spp.	02 (2.38)
Morganella spp.	01 (1.19)
Proteus spp.	01 (1.19)
Beta haemolytic	01 (1.19)
streptococcus	01 (1.19)
Unidentified Gram negative bacilli	
Total number of isolates	84

Table IV shows out of 84 isolates equal (20 or 23.83%) number of *Staphylococcus aureus* and *Esch.coli* were isolated, followed by *CoNS* 15(17.85%), *Pseudomonas spp.* 12(14.28%), *Acinetobacter spp.* 08 (9.52%), *Klebsiella spp.* 03 (3.5%), *Enterococci* 02 (2.38%) and *Morganella spp.*, *Proteus spp. Beta haemolytic streptococci* and unidentified Gram negative bacteria was 01(1.19%) respectively.

Antimicrobials	S.aureus	E.coli	CoNS	Pseudomonas spp.	Acinetobacter	Klebsiella spp
	n=20	n=20	n=15	n=12	n=08	n=03
Ampicillin	20 (100%)	18 (90%)	15 (100%)	ND	08 (100%)	03(100%)
Amoxyclavulonic acid	12 (60%)	18 (90%)	1(66.66%)	ND	08 (100%)	03(100%)
Cephradine	17 (85%)	19 (95%)	12 (80%)	ND	08 (100%)	03(100%)
Cefuroxime	16 (85%)	19 (95%)	11(86.66%)	ND	08 (100%)	03(100%)
Ceftriaxone	12 (60%)	14 (70%)	08(53.33%)	08 (67%)	07(87.5%)	02(66.66%)
Cefixime	14 (70%)	17 (85%)	09 (60%)	10 (83%)	08 (100%)	02(66.66%)
Ceftazidime	ND	17 (85%)	ND	10 (83%)	08 (100%)	03(100%)
Ciprofloxacin	7 (35%)	18 (90%)	03 (20%)	02 (17%)	05(62.5%)	02(66.66%)
Levofloxacin	6 (30%)	18 (90%)	03(20%)	02 (17%)	05(62.5%)	01(33.33%)
Cotrimoxazole	14 (70%)	17 (85%)	08(53.33%)	ND	07(87.5%)	01(33.33%)
Gentamycin	06 (30%)	11 (55%)	05(33.33%)	03 (25%)	07(87.5%)	01(33.33%)
Amikacin	ND	03 (15%)	ND	02 (17%)	03(37.5%)	00 (0%)
Pipericillin tazobactum	ND	06(30%)	ND	02(17%)	02(25%)	02(66.66%)
Imipenem	01 (5%)	00	01(6.66%)	01 (8%)	01 (12.5%)	00 (0%)
Oxacillin			7 (46.66%)			
Colistin					00	00
Doxycycline	16 (80%)	16(80%)	10(66.66%)	ND	00	
Cloxacillin	12 (60%)	ND	8 (53.33%)	ND	ND	ND
Erythromycin	17 (85%)		12 (80%)			
Azithromycin	16 (80%)		11(73.33%)			
Vancomycin	00(0%)		00 (0%)	*		

Table V: Resistance	pattern o	of	common isolates	to different	antimicrobial agents
---------------------	-----------	----	-----------------	--------------	----------------------

Figures within parenthesis indicates percentages. CoNS= Coagulase Negative Staphylococcus

Table V shows the antibiotic resistance pattern of common organisms isolated from SSI. All common organisms shows 100% resistant to Ampicillin except *Esch.coli* which shows 90% resistance. *Acinetobacter spp* and *Klebsiella spp* 100% resistant to Amoxyclavulonic acid respectively followed by *Esch.coli* 90%, *Staph. aureus* 60% and CoNS 66.66% resistance. All isolates of *Acinetobacter* and *Klebsiella spp*. were (100%) resistant to Cefuroxime followed by *Esch.coli* 95%, *CoNS* 86.66% and

Staph.aureus 85%. Acinetobacter spp were (87.5%) resistant to Ceftriaxone followed by Esch.coli 70%. Pseudomonas spp. 67% , Klebsiella spp. 66.66%, Staph.aureus 60% and CoNS 53.33% . Acinetobacter spp also showed 100% resistance to Cefixime followed by Esch.coli 85%, Pseudomonas spp 83%, Staph.aureus 70%, Klebsiella spp 66.66% and CoNS 60%. Ceftazidime was used only for Gram negative bacteria and all (100%) Acinetobacter spp. and Klebsiella spp. were resistant to it.

followed by Esch.coli 85% and Pseudomonas spp. 83%. In case of ciprofloxacin 90% of Esch.coli showed resistance, followed by Klebsiella spp. 66.66%, Acinetobacter Staph. aureus 35%, CoNS 20% and spp.62.5%, Pseudomonas spp 17%. Levofloxacin had almost same resistant pattern as ciprofloxacin, only it is much less resistant in case of Staph.aureus (35%). Acinetobacter showed highest resistance to Cotrimoxazole 87.5% followed by Esch.coli 85%, Staph.aureus 70%, CoNS 53.33% and Klebsiella spp 33.33%. Acinetobacter spp. also highly to Gentamycin 87.5%. Resistance pattern of resistant Gentamycin in other organisms are Esch.coli 55%, CoNS and Klebsiella spp. 33.33% respectively, Staph. aureus 30% and Pseudomonas spp. 25%. Amikacin was used only for Gram negative bacteria . Less than forty percent Acinetobacter spp.(37.5%) are resistant to Amikacin followed by 17% Pseudomonas spp. and 15% Esch.coli. Organisms showed less and varied resistance pattern to Imipenem, Acinetobacter spp. 12.5% Pseudomonas spp. 8%, CoNS 6.66%, Staph.aureus 5% and no Esch.coli were resistant. However, resistant strain of Staph.aureus and CoNS to Vancomycin was not observed. Colistin was used in case of Acinetobacter spp. and Pseudomonas spp. and were found to be sensitive. Sixty percent of (60%) of Staph.aureus and 53.33% of CoNS resistant to Cloxacillin. About 30% Esch.coli, 17% Pseudomonas, 25% Acinetobacter and 66.66% Klebsiella are resistant to Pipericillin tazobactum.

Table VI: Methicillin resistant *Staphylococcus aureus* isolated from SSI

Staphylococcus aureus	Methicillin resistant	• Methicillin sensitive	
uureus	Staphylococcus	Staphylococcus	
	aureus(MRSA)	aureus (MSSA)	
20	11 (55%)	9 (45%)	

Table VI shows out of 20 *Staphylococcus aureus* 11(55%) were Methicillin resistant (MRSA) and 09 (45%) were Methicillin sensitive (MSSA).

Discussion:

Infection is an important cause of morbidity and mortality in surgical patients. The incisional SSI classified in to 2 types: superficial and deep. The superficial infection involves only subcutaneous adipose layer, deep infection is less frequent but serious consequences.¹⁵ These are more prevalent after emergency surgical procedures. It could be attributable to the fact that most of these patients are low socioeconomic group and maximum number of patients are malnourished.^{3, 4}The pathogens isolated from SSI are usually bacteria.

In present study out of 159 cases, 82 (51.57%) were culture positive. It is much lower than other study (76.36%). ¹⁶ (Santos et al) Most of the surgery in their study were elective surgery. Out of 82 culture positive sample, 62 (47.61%) from gynae and obstretic dept. and 20 (76.92%) from General surgery dept. Culture positive rate was more in wound infection samples from General surgical department than Gynae and Obstetric department. Similar findings were experienced in another study.¹⁷ . In present study it may be due to delay in transportation of wound swab samples, antiseptic wound wash prior to giving samples, improper collection and use of multiple broad spectrum antibiotics after operation.

In current study S. aureus was a major pathogen from patients under went gynecological and obstetrics operations (25.80%), followed by CoNS (23.58%). These are the most common isolated bacteria from SSIs of patient who under went emergency type of surgery and which might have been surface contamination by these bacterium from the skin and from the environment.17 E. coli was the most common pathogen, isolated from patients who underwent general surgery such as appendectomy and other surgery where intestinal manipulation occurred . Exposing endogenous flora prevalent organisms in our study were both Esch.coli and Staph.aureus which yielded equal number (23.80%) followed CoNS (17.85%), Pseudomonas spp (14.28%). by Klebsiella spp(3.4%),Acinetobacter spp (9.52%). Enterococcus spp.(2.38%) Proteus spp. Morganella spp. Beta haemolytic streptococci and unidentified Gram negative bacilli 1.19% respectively . S. aureus, CoNS and E. coli were prevalent organisms associated with surgical wound infections. 18 Another study done in Bangladesh isolated Esch.coli 53.33%, S.aureus 25.56%, Pseudomonas 13.33%.¹⁹ Other researchers in different study isolated Esch.coli Pseudomonas spp 27.01%. Proteus spp 56.75% . 13.51%., Staphylococcus aureus 8.10%, Klebsiella spp. 5.40%, Acinetobacter spp. 4.05% from surgical site wound infection. 20 Almost similar findings observed in our study (Tab IV)

Cephalosporins were ineffective against most of the organisms isolated in our study (table V). Similar findings were observed in other studies.²¹ Safer and more widely used drugs for both Gram positive and Gram negative bacteria are Ceftriaxone and Cefixime, 60% of Staphylococcus aureus showed resistant to Ceftriaxone and 70% to Cefixime. Esch.coli found 70% resistant to Ceftriaxone, and 85% to Cefixime . Ceftazidime was tested only against Gram negative bacteria, more than 80% of all Gram negative isolates found resistance to it. Only 5% of Esch.coli are sensitive to 1st and 2nd generation Cephalosporins. Similar finding was observed in Bangladesh (Mohiuddin et al., 2010).²⁰ Over use of Cephalosporins in last two decades as documented in other studies may be the cause (Morgan, 2006).22

The current study showed that the Gram positive organisms were much more sensitive to Quinolones than Gram negative (table V). However Pseudomonas spp. were 17% resistant to Quinolones. It was also 25% resistant to Gentamycin, 17% to Amikacin and 8% to Imipenem. Other researchers also experienced almost similar findings.23 . In present study none of the Esch.coli and Klebsiella were resistant to Imipenem however 5% of Staphylococcus aureus, 6.66% of CoNS and 12.5% of Acinetobacter were resistant to Imipenem. Next to Imipenem, Piperacillintazobactum is most effective drug for Acinetobacter spp and Amikacin for E.coli. Colistin was used only for Acinetobacter spp., non of the isolates of Acinetobacter spp. were found resistant to Colistin. In Egypt 25% of Acinetobacter were resistant to Colistin in MIC method but non of the Acinetobacter spp. found resistance to Colistin in disc diffusion method. In their study they observed 87.5% of Acinetobacter were multidrug resistant.²²

The current study showed that 55% of Staphylococcus aureus were MRSA (Methicillin resistant Staphylococcus aureus) none of the isolate showed resistance to Vancomycin. Similar findings were observed in different studies done in different places. ^{17, 23} This is a matter of great concern because treatment of such infections warrants costly antibiotics. Antimicrobial susceptibility pattern in our study showed that most of the isolated pathogens were multidrug resistant.

Conclusion :

This study give us an evidences to the current state of organisms isolated from superficial surgical site infection and their resistant pattern in our hospital. Due to high incidence of MRSA and multidrug resistant bacteria reported in our study, there is a need for continuous monitoring to determine the susceptibility pattern of common isolates which are found in our hospital. Data showed that the Cephalosporins are ineffective against SSIs and it is the time for surgeons to choose new antibiotics effective against today's pathogens for both prophylaxis and empirical therapy. Reinforcement of infection control measures is also strongly recommended in order to prevent healthcare-associated infections.

Bibliography :

- Man gram AJ. Horan TC, Pearson ML, et air. "Guideline for Prevention of Surgical Site Infection 1999" issued by ('enters for Disease Control and Prevention through its 1 2—member Hospital Infection Control Practices Advisory Committee. Am. J. Infect. Control, 1999;27:27-30.
- Bay-Nielsen M. Risk factors for surgical wound infection Ugeskr. Laeger.. 1996;158:5749-53
- Olson M, Lee JT. Continuous 10 year wound infection surveillance. Arch. Surg., 1990;125:794-803.
- Olson M. O'Connor Schwartz ML. Surgical wound infection A 5 years prospective study of 20,193 wounds at Minneapolis A Medical Centre, Am Surg 1984;199: 253-9.
- Noer HH Interest and possibilities of post-operative registration of wound infections in Danish orthopaedic department' A Study Survey. Int. J. Clin.. Monit. Computer., 1990;1:21-26.
- Alvarado CJ. 2000. The Science of Hand Hygiene: A Self- Study Monograph, University nnof Wisconsin Medical School and Sci-Health C
- Awan SM, Dhari JF, Laghari A A, Bilal F, Khaskheli M Noor. Schalkwyk J V, Eyk N V. Antibiotic Prophylaxis in Obtetric Procedure. SOGC Clinical Practice Guideline. No. 247, September 2010.mmunication. March.
- Weber DJ, Raasch R, Rutala WA, Nosocomial infection in the ICU, the growing importance of antibiotics resistance pathogens. Chest 1999; 115(3 suppl): 34S-41S.
- Bergogne BE, Deere D. Joly ML. Opportunistic nosocomial multiply resistant bacterial nfecti ons-their treatment and prevention. J - Antimicrob. Chemother., 1993;32:Suppl A:39-47.

30

- Gorback SL, Cartless JG, Nichols L. Epidemiology and prevention of surgical infections. 1st ed, Boston, Little Brown and Co., 1984, pp.
- Collee JG. Laboratory Strategy in the Diagnosis of infective syndrome. In Mackie and Mc Cartney Practical Medical Microbiology. 14th ed. Duguid JP. Fraser AG, Marimon BP (editors). Churchill Pepys. 1981- Livingstone. New York. 1996.
- Chessbrough M. Medical Laboratory Manual for Tropical Countries. Vol.II. ELBS Cmbridgeshire. England. 1984..
- Bauer A.W, Kirby W.M.M, Sherris J.C and Truck M. Antibiotic Susceptibility Testing by a Standardized Single Disk Method. The American Journal of Clinical Pathology. 1996;36(3): 493-496.
- Chessbrough M. District Laboratory Practice in Tropical Countries. Part I. II. Cambridge University Press. UK. 2000.
- Lee J. Wound infection surveillance. Surg Clin North Am. 1992;6:643-56.
- K. R. N Santos, L. S. Fonseca, G. P Bravo Neto and P. P Gontijo Fitho Surgical site infection : Rates, etiology and resistant patterns to antimicrobials among strains isolated at rio de janiro university hospital. Volume 25, Number 4(1997), 217-220, DOI: 10, 1007/BF01713147.
- Amare B. Abdurrahman Z, Moges B, Ali J, Muluken L, et al., (2011) Postoperative Suegical site bacterial infections and susceptibility pattern of Gondor University teaching hospital , Northwest Ethiopia. Journal of Bacteriology and Parasitology, 2:126. Dol: 10.4172/2155-9597, 10000126.

- CDC NNIS System(1996) National Nosocomial Infectios Survaillance (NNIS) Semiannual Report from October 1986-April 1996. Am J Infect Control 24: 380-388.
- 19. Mohammad M, Sabeena S, Mohammad A M. Detection of resistance gene marker *int11* and antimicrobial resistance pattern of *E.coli* isolated from surgical site wound intection in Holy Family Red Crescent Medical Collage Hospital. Bangladesh J Med Micobiol 2010: 04 (02) : 19-23. Bangladesh Society of Medical Microbiologists.
- Md. Mohiuddin, Ashraful Haq, Md. Mozammel Hoq, Farida Hoq. Microbiology of Nosocomial Infection in Tertiary Hospitals of Dhaka City and its Impact. Bangladesh J Med Micobiol 2010: 04 (02) :32-38. Bangladesh Society of Medical Microbiologists.
- Abeid Mahmood(Department of Pathology, PNS, Safia, Karachi). Bacteriology of Surgical Site Infection and Antimicrobial Susceptibility Pattern of the Isolates at a Tertiary Care Hospital in Karachi. Journal of Pakistan Medical Association. August 2000.
- 22. Morgan M. Surgery and cephalosporins: A marriage made in haven or time for Divorce? The Internet J Surgery 2006;8 (1), URL: http://www.ispub.com/journal/the_internet journal _of surgery/volume_8_number_1/article /surgery _ and _ cephalosprins_a marriage _made in _heaven_or _ time _ for _ divrce.html.

 Wassef M.A, Hussein A, Abdul Rahman E.M, EI-Sherif R.H. A prospective surveillance of surgical site infectio: Study for efficacy of preoperative antibiotic prophylaxix. African Journal of Micrbiology Research Vol. 6(12), pp. 3072-3078, 30 March, 2012