Role of semiquantitative roll over technique and flush technique in diagnosing central line associated bloodstream infection (CLABSI) and central line related local infections (CRLI) in MICU patients: A prospective study

Dhanashree P Inamdar¹*, Sujata Baveja²

¹Associate Professor, ²Professor and HOD, ³Dept. of Microbiology, ⁴Mamata Medical College and Hospital, Khammam, Telangana, ⁵Lokmanya Tilak Municipal Medical College and General Hospital, Sion, Mumbai, Maharashtra, India

*Corresponding Author: Dhanashree P Inamdar
Email: dhanashreeitmmc@gmail.com

Abstract

Introduction: Diagnosing Central line associated bloodstream infection (CLABSI) and Central line Related local infections (CRLI) involves doing central line tip culture. The routine method which is followed in every laboratory is the semiquantitative roll plate method. However, the use of a quantitative flush method will add a further advantage, as it may detect endoluminal microorganisms more easily which helps in diagnosing blood stream infections due to central line.

Materials and Methods: A prospective study was carried out in MICU of tertiary care hospital. Semiquantitative roll over technique and quantitative flush technique was followed to process central line catheter in patients suspected of CRLI and CLABSI.

Results: Of 210 samples processed from patients forty seven patients (47) were diagnosed with CRLI and seven (7) with CLABSI. Semiquantitative technique had superiority in diagnosing local infections whereas flush technique had superiority in diagnosing blood stream infections.

Conclusion: Both Semiquantitative and flush techniques are recommended for processing central line catheters for diagnosing CRLI and CLABSI. Early CLABSI can be missed sometimes in samples from ICU patients who have associated comorbid conditions. Thus those laboratories where only semiquantitative roll over technique is followed, quantitative flush technique is recommended which aids in diagnosing CLABSI which can be missed by roll over technique.

Keywords: Central line associated bloodstream infections (CLABSI), Central line related local infections (CRLI), Intensive care unit (ICU), Semiquantitative roll over, Quantitative flush technique.

Introduction

Central line catheter associated bloodstream infection (CLABSI) remains one of the leading causes of nosocomial acquired bacteremia in ICUs leading to increased mortality, whereas Central line related local infections (CRLI) leads to increased morbidity and hospital stay in turn increasing cost burden on patient.¹-⁵ Although ideally the diagnosis of CLABSI / CRLI is made before catheter removal, a definite diagnosis is done based on a culture from the catheter tip⁶,⁷ with clinical correlation. The reference standard established was a semiquantitative technique described by Maki et al. in 1977,⁸ with a cutoff of 15 CFU (colony forming units) to distinguish microbial contamination of catheters from significant colonization. This technique which is also called the roll plate method is done on distal 5 cm end of catheter tip which is rolled back and forth on an agar plate for culture. However, because catheter colonization and infection can be the consequence of the introduction of microorganisms during management of the catheter hubs or the infusion of fluids or drugs, this technique could be less appropriate for the detection of endoluminal infection following colonisation.

Other methods were developed subsequently, in an attempt to deal with this along with some limitations.⁹-¹⁵ One technique which contributed to proper diagnosis of CLABSI was quantitative endoluminal flush technique devised by Cleri¹¹ in 1980 with colony cutoff of 10³ CFU/ml. Nowadays, Infectious Diseases Society of America guidelines ⁶,¹⁶ recommends a breakpoint of 100 CFU/catheter segment. Bouza and colleagues also demonstrated a cutoff of 100 CFU to be superior to one of _1,000 CFU/catheter segment.¹⁷

Aims and Objectives

To diagnose CLABSI and CRLI in patients suspected of having infection in MICU by both Semiquantitative roll over technique and quantitative endoluminal flush technique and comparing them.

Materials and Methods

A prospective study was carried out in the department of microbiology in collaboration with Medical Intensive Care Unit (MICU) in a tertiary care hospital. Two hundred and ten consecutive adult patients on central venous catheter admitted in MICU constituted the study population

Inclusion Criteria: Adult patients on central venous catheters admitted in MICU and who developed systemic signs and symptoms of infections after 48 hours of admission.

Exclusion criteria:
1. Patients with sepsicaemia due to obvious causes other than central line.
2. Patients developing systemic signs and symptoms < 48 hrs of admission.

Study Procedure

All adult patients who met inclusion criteria were included in the study. Detailed clinical history of each
patient was noted as per the clinical proforma with every
day follow up to check vitals, local and systemic signs of
sepsis.

**Following specimens were collected:** Catheter tip, Blood
for blood culture, Urine, Endotracheal secretions (for
patients on ventilator), Sputum when indicated, Pus when
indicated.

**Catheter tip collection**

The skin was disinfected with 70% alcohol prior to
catheter removal. The catheter was held at the proximal end
and carefully removed from the patient with a sterile forceps,
taking care to avoid contact with the skin. The distal end was held over a sterile tube, and the tip was cut
with sterile scissors. The terminal two to three inches were
collected in the sterile test tube and transported to the
laboratory immediately.

Other relevant samples were collected based on standard
protocol. ¹⁹⁻²³

**Sample Processing**

**Catheter tip processing:** The central venous catheter so
collected was placed in sterile petri dish with the help of
sterile forceps and then with sterile scalpel, the distal 5cm
portion was cut. Semiquantitative extraluminal Maki’s roll
over plate method and quantitative endoluminal catheter
flush culture were used for processing the central line tip.

Semiquantitative extraluminal Maki’s roll over plate method

The catheter was rolled back and forth over 90mm area
of sheep blood agar plate for 4 times using sterile forceps.
Then further incubated at 37°C for 18-24 hours.

Quantitative endoluminal catheter flush culture

The same segment of the catheter was held with the
help of sterile forceps and 2ml of tryptic soy broth was
flushed intraluminally with the help of sterile needle and
syringe which was then diluted 10-fold, and 0.1ml of each
dilution was streaked onto Blood agar (BA) plate,
MacConkey agar (MA) plate and Sabouraud’s Dextrose
Agar (SDA) slant respectively. The BA plate was incubated
overnight in candle jar to provide optimum carbon dioxide
requirement at 37°C. MA plate and SDA slant were also
incubated overnight at 37°C.

**Interpretation:** Agar plates were examined after 18-24 hrs
of incubation. Significant growth was defined as ≥ 15
colony forming units (CFU) by Maki’s roll plate method or
≥ 1000 CFU/ml by the catheter flush method. Individual
bacterial colonies grown were further identified as per
standard protocol. ²⁴

**Results**

During the study period a total of 210 consecutive adult
patients with central venous catheter were analysed. Of
these fourty seven (47) patients developed Catheter related
local infection(CRLI) and seven (7) patients developed
Central line associated blood stream infections(CLBSI).

Diagnosis of central line related local and systemic
infections

No of catheters processed = 210

**Central line related local infections (CRLI) was
diagnosed as:** Recognized pathogen isolated from
blood culture and pathogen not related to infection from
another site (other than site of an intravascular device i.e. it
should not have been isolated from urinary tract / respiratory
tract / wound, etc)
or
One of the following
1. fever (>38 C)
2. chills
3. hypotension
And any of the following:
1. Common skin contaminant isolated from two blood
cultures drawn on separate occasions, and organism is
not related to infection at another site.
2. Common skin contaminant isolated from blood culture
from patient with intravascular access device and
physician institutes appropriate antimicrobial therapy.
3. Positive antigen test on blood or organism is not related
to infection at another site.

Of 210 patients observed for infections attributable to
catheter, only seven (7) patients met criteria to be diagnosed
as central line associated blood stream infections. Of total,
7(3.33%) were positive by both blood culture and
Quantitative endoluminal flush technique, while 5(2.38%)
were positive by Semiquantitative Maki’s roll over
technique. Two patients were missed by semiquantitative
method which was diagnosed by endoluminal flush
technique. None of them had growth in urine, ET
secretions, induced sputum or pus culture (Table 2).

Central line associated blood stream infections(CLBSI)
was diagnosed as:

- Any sign of local infection (induration, erythema, heat,
pain, purulent drainage) and
- Catheter tip colonization was defined as “Significant
growth of a microorganism by
  a. >15 colony-forming units from the catheter tip by
      semiquantitative method or
  b. >10⁷ by quantitative culture.”

Of total, 47(22.38%) were positive by Semiquantitative
Maki’s roll over technique (Fig. 1), while 43(20.4%) were
positive by Quantitative endoluminal flush technique (Fig.
2). Amongst signs and symptoms of local infection,
Erythema (59.5%) was the most common finding, followed
by oozing (38.2%), pain (21.2%) and induration (19.1%)
respectively with overlapping signs in some patients.
None of these patients were positive by blood culture, urine, ET
secretions, induced sputum or pus culture (Table 1).

Of 210 patients observed for infections attributable to
catheter, only seven (7) patients met criteria to be diagnosed
as central line associated blood stream infections. Of total,
7(3.33%) were positive by both blood culture and
Quantitative endoluminal flush technique, while 5(2.38%)
were positive by Semiquantitative Maki’s roll over
technique. Two patients were missed by semiquantitative
method which was diagnosed by endoluminal flush
technique. None of them had growth in urine, ET
secretions, induced sputum and pus respectively (Table 2).

**Catheter tip collection**

The skin was disinfected with 70% alcohol prior to
catheter removal. The catheter was held at the proximal end
and carefully removed from the patient with a sterile forceps,
taking care to avoid contact with the skin. The distal end was held over a sterile tube, and the tip was cut
with sterile scissors. The terminal two to three inches were
collected in the sterile test tube and transported to the
laboratory immediately.

Other relevant samples were collected based on standard
protocol. ¹⁹⁻²³

**Sample Processing**

**Catheter tip processing:** The central venous catheter so
collected was placed in sterile petri dish with the help of
sterile forceps and then with sterile scalpel, the distal 5cm
portion was cut. Semiquantitative extraluminal Maki’s roll
over plate method and quantitative endoluminal catheter
flush culture were used for processing the central line tip.

Semiquantitative extraluminal Maki’s roll over plate method

The catheter was rolled back and forth over 90mm area
of sheep blood agar plate for 4 times using sterile forceps.
Then further incubated at 37°C for 18-24 hours.

Quantitative endoluminal catheter flush culture

The same segment of the catheter was held with the
help of sterile forceps and 2ml of tryptic soy broth was
flushed intraluminally with the help of sterile needle and
syringe which was then diluted 10-fold, and 0.1ml of each
dilution was streaked onto Blood agar (BA) plate,
MacConkey agar (MA) plate and Sabouraud’s Dextrose
Agar (SDA) slant respectively. The BA plate was incubated
overnight in candle jar to provide optimum carbon dioxide
requirement at 37°C. MA plate and SDA slant were also
incubated overnight at 37°C.

**Interpretation:** Agar plates were examined after 18-24 hrs
of incubation. Significant growth was defined as ≥ 15
colony forming units (CFU) by Maki’s roll plate method or
≥ 1000 CFU/ml by the catheter flush method. Individual
bacterial colonies grown were further identified as per
standard protocol. ²⁴

**Results**

During the study period a total of 210 consecutive adult
patients with central venous catheter were analysed. Of
these fourty seven (47) patients developed Catheter related
local infection(CRLI) and seven (7) patients developed
Central line associated blood stream infections(CLBSI).
Table 1: Distribution of all patients who developed central line related local infections (n=210)

| Criteria                              | Positive | %Positivity |
|---------------------------------------|----------|-------------|
| Culture techniques (n=210)             |          |             |
| Semiquantitative Maki’s roll over     | 47       | 22.38%      |
| Quantitative endoluminal Flush        | 43       | 20.4%       |
| Signs/Symptoms of local infection (n=47) |         |             |
| Erythema                              | 28       | 59.5%       |
| Oozing                                | 18       | 38.2%       |
| Pain                                  | 10       | 21.2%       |
| Induration                            | 9        | 19.1%       |

Table 2: Distribution of all patients who developed central line associated blood stream infections (n=210)

| Criteria                              | Positive | %Positivity |
|---------------------------------------|----------|-------------|
| Culture techniques (n=210)             |          |             |
| Blood culture                         | 7        | 3.33%       |
| Semiquantitative Maki’s roll over     | 5        | 2.38%       |
| Quantitative endoluminal flush        | 7        | 3.33%       |
| Signs/Symptoms of systemic infection (n=7) |       |             |
| Fever                                 | 6        | 85.71%      |
| Chills                                | 2        | 28.5%       |
| Hypotension                           | 3        | 42.8%       |

Discussion

The relative risk for a catheter-related blood stream infection is 2 to 855 times higher with Central venous catheters (CVCs) than peripheral venous catheters. At the same time, appropriate infection control measures can help to reduce this problem. Although there are many studies about CVC related infection, very few Indian studies have analyzed methods for central line processing in detail. Hence the present study was undertaken with a purpose of diagnosing Central line associated bloodstream infection (CLABSI) and Central line Related local infections (CRLI) by semiquantitative roll over technique and flush technique in MICU patients on central line catheters.

This study compared the commonly used catheter culture methods: the roll plate method and other method used to culture vascular catheters i.e lumen flush technique. There were 47 patients who developed local infection due to central venous catheter. Semiquantitative Maki’s roll over technique was positive in all 47 patients but quantitative flush technique was positive in 43 patients. Growth was not seen in 4 samples processed by flush technique, but these were positive by roll technique. Lennert et al in 2009 on comparison of the roll plate method to the sonication method to diagnose catheter colonization and bacteremia in patients with long-term tunnelled catheters: randomized prospective study demonstrated that the use of the quantitative sonication technique to detect catheter tip colonization in patients with CVCs had no surplus value compared with the semiquantitative roll plate method. In another study by Charalambos et al in 1998, they also observed that semiquantitative technique distinguishes infection (greater than or equal to 15 colonies) from contamination and is more specific in diagnosis of catheter-related septicemia than culture of the catheter in broth.

In the present study, semiquantitative technique (22.38%) turned out to be a better indicator of infection for diagnosing CRLI than flush technique (20.4%) similar to the above study.
Also, in the study by Kristinsson et al.28 as well as in this one, flushing through the catheters was done by endoluminal flush technique. This increased the sensitivity of technique by removing organisms from the external surface of catheter and increasing chances of isolating organism which was actually in contact with blood leading to CLABSI which correlates with the present study as quantitative flush technique was done for diagnosing CLABSI.

The high frequency of catheter tip colonization in inserted location provides additional evidence that it can be due to hematogenous colonization which is more frequent than previously thought in ICU patient populations. Although many studies suggest that this route of infection is uncommon.11,27,29 However, a report by Maki and Will30 found that hematogenous seeding of central venous catheters was one of the most common mechanisms of catheter infection in ICU patients. The same study showed that for a patient with an infection at a distant site other than central line, which can cause bacteremia, removal of a central catheter may be required. But in the present study as other cultures were performed to rule out infection, CLABSI can be solely attributed to central line catheter. In a metaanalysis by Safdar et al.31 the sensitivity mean found for the qualitative culture was of 90%, while specificity was 72%. But, by semiquantitative culture lower sensitivity (85%) and higher specificity (82%) was noted. In a study done by Marconi et al32 in 2008 they concluded that the semiquantitative culture is a rapid and efficient technique for diagnosing catheter-related infection. Still, it requires a careful interpretation and its results need to be interpreted carefully for diagnosis and specific treatment. Many studies have compared quantitative sonication technique with semiquantitative roll over technique for processing central line catheters but only few studies in India have compared quantitative flush technique with semiquantitative one, which contributes to the uniqueness of the above study.

Conclusion
Semiquantitative and quantitative technique for culturing central line tips should be applied in every microbiology laboratory particularly in samples from ICU patients as they have associated comorbid conditions and diagnosis of CRLI/CLABSI could be hampered due to associated nosocomial infections. Both techniques not only aid in diagnosing infection but can differentiate local and systemic infection which helps in prompt treatment of patients who are actually suffering from CRLI/CLABSI.

Conflicts of Interest: None.

References
1. Edgeworth, J. D., D. F. Treacher, and S. J. Eykyn. 1999. A 25-year study of nosocomial bacteremia in an adult intensive care unit. Crit Care Med 27:1421–1428.
2. Jarvis, W. R., J. R. Edwards, D. H. Culver, J. M. Hughes, T. Horan, T. G. Emori, S. Banerjee, J. Tolson, T. Henderson, R. P. Gaynes, et al. 1991. Nosocomial infection rates in adult and pediatric intensive care units in the. Am J Med 91:1858–191S.
3. Kluger, D. M., and D. G. Maki. 1999. Abstr. 39th Intersci. Conf. Antimicrob. Agents Chemother., abstr. 514. Am Soc Microbiol Washington, DC.
4. Pittet, D., D. Tarara, and R. P. Wenzel. 1994. Nosocomial bloodstream infection in critically ill patients. Excess length of stay, extra costs, and attributable mortality. JAMA 271:1598–1601.
5. Wenzel, R. P., and M. B. Edmond. 2001. The impact of hospital-acquired bloodstream infections. Emerg Infect Dis 7:174–177.
6. Maki, D. G., C. E. Weise, and H. W. Sarafin. 1977. A semiquantitative culture method for identifying intravenous-catheter-related infections. N Engl J Med 296:1305–1309.
7. Ferreira J., Camargos P.A.M., Clemente W.T., Romanelli R.M.D.C. (2018) Clinical usefulness of catheter-drawn blood samples and catheter tip cultures for the diagnosis of catheter-related bloodstream infections in neonatology: A systematic review. American Journal of Infection Control, 46 (1), pp. 81–87.
8. Maki, D. G., C. E. Weise, and H. W. Sarafin. 1977. A semiquantitative culture method for identifying intravenous-catheter-related infections. N Engl J Med 296:1305–1309.
9. H. S., R. Colley, R. H. Bower, V. P. Dutry, J. T. Schwartz-Fulton, and J. E. Fischer. 1982. Association between microorganism growth at the catheter insertion site and colonization of the catheter in patients receiving total parenteral nutrition. Surg 92:720–727.
10. Brun-Buisson, C., F. Abrouk, P. Legrand, Y. Huet, S. Larabi, and M. Rapin. 1987. Diagnosis of central venous catheter-related sepsis. Critical level of quantitative tip cultures. Arch Int Med 147:873–877.
11. Cleri, D. J., M. L. Corrado, and S. J. Seligman. 1980. Quantitative culture of intravenous catheters and other intravascular inserts. J Infect Dis 141:781–786.
12. Kite, P., B. M. Dobbins, M. H. Wilcox, W. N. Fawley, A. J. Kindon, D. Thomas, M. J. Tighe, and M. J. McMahon. 1997. Evaluation of a novel endoluminal brush method for in situ diagnosis of catheter related sepsis. J Clin Pathol 50:278–282.
13. Kristinsson, K. G., I. A. Burnett, and R. C. Spencer. 1989. Evaluation of three methods for culturing long intravascular catheters. J Hosp Infect 14:183–191.
14. Linares, J., A. Sitges-Serra, J. Garau, J. L. Perez, and R. Martin. 1985. Pathogenesis of catheter sepsis: a prospective study with quantitative and semiquantitative cultures of catheter hub and segments. J Clin Microbiol 21:357–360.
15. Sherrerz, R. J., I. I. Raad, Belani, L. C. Koo, K. H. Rand, D. L. Pickett, S. A. Straub, and L. L. Fauerbach. 1990. Three-year experience with sonicated vascular catheter cultures in a clinical microbiology laboratory. J Clin Microbiol 28:76–82.
16. Sherrerz, R. J., S. O. Heard, and I. I. Raad. 1997. Diagnosis of triple-lumen catheter infection: comparison of roll plate, sonication, and flushing methodologies. J Clin Microbiol 35:641–646.
17. Bouza, E., N. Alvarado, L. Alcala, M. Sanchez-Conde, M. J. Perez, P. Munoz, P. Martin-Rabadan, and M. Rodriguez-Creixems, 2005. A prospective, randomized, and comparative study of 3 different methods for the diagnosis of intravascular catheter colonization. Clin Infect Dis 40:1096–1100.
18. Issam R, Hend AH, Badie A, Chatzinikolaou I, Marcella MJ, and Tarrand J. Differential Time to Positivity: A Useful Method for Diagnosing Catheter-Related Bloodstream Infections. *Ann Intern Med* 2004;140:19-22

19. Washington W Jr., Stephen A, Janda W, Koneman E, Procop G, Schreckenberger P, Woods G. Koneman’s color atlas and textbook of diagnostic microbiology, ed 6, Washington, 2006, Lippincott, ch43:20-21

20. Washington W Jr., Stephen A, Janda W, Koneman E, Procop G, Schreckenberger P, Woods G. Koneman’s color atlas and textbook of diagnostic microbiology, ed 6, Washington, 2006, Lippincott, ch43:22-23.

21. Endotracheal Suctioning of Mechanically Ventilated Adults and Children with Artificial Airways. AARC Clinical Practice Guideline. *Respir Care* 1993;38(5):500–504.

22. Washington W Jr., Stephen A, Janda W, Koneman E, Procop G, Schreckenberger P, Woods G. Koneman’s color atlas and textbook of diagnostic microbiology, ed 6, Washington, 2006, Lippincott, ch43; 30-31

23. Washington W Jr., Stephen A, Janda W, Koneman E, Procop G, Schreckenberger P, Woods G. Koneman’s color atlas and textbook of diagnostic microbiology, ed 6, Washington, 2006, Lippincott, ch43:19-22

24. Collee JG, Fraser AG, Marmin BP, Simmons A. Mackie and McCartney practical medical microbiology. 14th edition. Churchill Livingstone Inc. 1996.

25. Pawar M, Mehta Y, Kapoor K, Sharma J, Gupta A, and Trehan N. Central Venous Catheter-Related Blood Stream Infections: Incidence, Risk Factors, Outcome, and Associated Pathogens. *J Cardiothorac Vasc Anesth* 2004;18(3): 304-308.

26. Lennert S, Abdelilah B, Eric B, Bart J et al. Comparison of the Roll Plate Method to the Sonication Method To Diagnose Catheter Colonization and Bacteremia in Patients with Long-Term Tunnelled Catheters: a Randomized Prospective Study. *J Clin Microbiol* 2009;47(4):885–888.

27. Charalambous C, Swoboda SM, Dick J, Perl T, Lipsett PA. Risk Factors and Clinical Impact of Central Line Infections in the Surgical Intensive Care Unit. *Arch Surg.* 1998;133(11):1241–1246. doi:10.1001/archsurg.133.11.1241

28. Kristinsson, K. G., I. A. Burnett, and R. C. Spencer. 1989. Evaluation of three methods for culturing long intravascular catheters. *J Hosp Infect* 14:183–191.

29. Gahlot R, Nigam C, Kumar V, Yadav G, Anupurba S. Catheter-related bloodstream infections. *Int J Crit Illn Inj Sci*. 2014;4(2):162-167.

30. Maki, D. G., and L. Will. 1990. Risk factors for central venous catheter related infection within the ICU. A prospective study of 345 catheters, abstr. 715, p. 205. In Program and abstracts of the 30th Interscience Conference on Antimicrobial Agents and Chemotherapy. *Am Soc Microbiol* Washington, D.C.

31. Safdar, N.; Fine, J.P.; Maki, D.G. (2005). Meta-analysis: methods for diagnosing intravascular device-related bloodstream infection. *Ann Intern Med*, 142 (6), 451-466.

32. Camila Marconi, Maria de Lourdes RS Cunha, João C Lyra, Maria R Bentlin, Jackson EN Batalha, Maria Fátima Sugizaki, Lígia MSS Rugolo. Comparison between qualitative and semiquantitative catheter-tip cultures: laboratory diagnosis of catheter-related infection in newborns. *Br J Microbiol* 2008;39:262-267.

**How to cite this article:** Inamdar DP, Baveja S. Role of semiquantitative roll over technique and flush technique in diagnosing central line associated bloodstream infection (CLABSI) and central line related local infections (CRLI) in MICU patients: A prospective study, *Int J Med Microbiol Trop Dis* 2019;5(1):47-51