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

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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.1−3 Although ideally the diagnosis of CLABSI / CRLI is made before catheter removal, a definite diagnosis is done based on a culture from the catheter tip4−7 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 10^3 CFU/ml. Nowadays, Infectious Diseases Society of America guidelines6,16 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 forty 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:**

1. Any sign of local infection (induration, erythema, heat, pain, purulent drainage) and
2. 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. >10³ 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).

**Central line associated blood stream infections(CLBSI) 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 and pus respectively (Table 2).
Table 1: Distribution of all patients who developed central line related local infections (n=210)

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

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

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Positive</th>
<th>%Positivity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Culture techniques (n=210)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Blood culture</td>
<td>7</td>
<td>3.33%</td>
</tr>
<tr>
<td>Semiquantitative Maki’s roll over</td>
<td>5</td>
<td>2.38%</td>
</tr>
<tr>
<td>Quantitative endoluminal flush</td>
<td>7</td>
<td>3.33%</td>
</tr>
<tr>
<td>Signs/Symptoms of systemic infection (n=7)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fever</td>
<td>6</td>
<td>85.71%</td>
</tr>
<tr>
<td>Chills</td>
<td>2</td>
<td>28.5%</td>
</tr>
<tr>
<td>Hypotension</td>
<td>3</td>
<td>42.8%</td>
</tr>
</tbody>
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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\(^\text{25}\) 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\(^\text{27}\) 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 Will\(^{30}\) 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 Saad 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 al.\(^{32}\) 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**


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