Time-related Concordance Between Swab and Biopsy Samples in the Microbiological Assessment of Burn Wounds

Ebrabim Salebifar, PharmD; Ghasemali Khorasani, MD; Shabnam Ala, PharmD

Abstract: The aim of this study was to investigate the concordance between swab and tissue biopsy samples in terms of microbiological isolates and their time-related changes. A total of 156 samples (78 swab and 78 biopsy) were collected from 39 cases of partial- or full-thickness burns and compared at days 7 and 14 after admission regarding the type of microorganisms and their time-related changes. Pseudomonas aeruginosa and Citrobacter freundii were the two most common microorganisms found by both sampling methods. While the majority of swab and biopsy samples were concordant in day 7, the rate of concordance in day 14 was less than day 7—87.1% versus 66.6%, respectively. After comparing the ratio of P. aeruginosa and C. freundii in positive swab and biopsy cultures on days 7 and 14, unlike the swab samples, the biopsy samples yielded similar results both times (75% P. aeruginosa and 25% C. freundii, respectively). The results of this study show that the swab is a sufficient tool for burn wound monitoring during the first week and could defer the need for invasive biopsy sampling. For patients who remain in the burn unit for a longer period, biopsy samples are justified for monitoring the bacterial activity in burn wounds.

A severe burn is a serious injury that bacterial colonization often complicates. Infection impairs the healing process, and microorganism invasion of neighboring healthy tissue can lead to sepsis, which is the most frequent cause of death in burn injuries. Early detection of invasive burn wound infection could be helpful in reducing the mortality for these patients. Although tissue biopsy is considered the most appropriate sampling method for identifying wound infection and its causative pathogens, the procedure is potentially traumatic and is not routinely available, particularly for slow- or non-healing wounds that require frequent, long-term care. For these reasons, the use of a more conventional and readily available sampling method such as swab sampling could be considered as an alternative method for monitoring burn wounds. Since there are arguments to support the use of the swab sample as a useful method for routinely assessing the microbiology of appropriate wounds, we conducted this study to investi...
gate the value of swab samples versus biopsy samples in terms of microbiological isolates and their time-related patterns of change.

**Material and Methods**

**Setting and patients.** Our burn center is a part of a 75-bed tertiary referral university-dependent hospital located in northern Iran. This prospective cross-sectional study was conducted on patients with partial- or full-thickness burns who were enrolled from September 2006 through March 2007. The Mazandaran University of Medical Sciences review board approved this study. Surface swabs and wound biopsies for bacteriological assessment of the burn wounds were performed at the end of weeks 1 and 2. Seven of the 39 patients in this study were septic based on the following clinical criteria: body temperature < 36°C or > 39°C, blood pressure < 90 mmHg or a reduction of 40 mmHg or more, and pulse rate > 90 beats per minute. Blood samples were taken 3 times and cultured in patients who had signs and symptoms of sepsis.

**Biopsy samples.** Under anesthesia, about 1 cm² of tissue, along with the underlying live tissue, was removed from each wound and suspended in 2-mL physiological saline and homogenized. Samples were inoculated on blood and MacConkey agar. A standard biochemical test was used to identify bacterial isolates; no growth up to 48 hours was considered negative. The biopsy samples in the second week were taken from close proximity to the sampling area from the first week.

**Surface swab samples.** Silver sulfadiazine (if any was present) was first removed with sterile, saline-soaked gauze. An area of 4 cm² was swabbed using two sterile swab sticks. Swab samples were taken from the wound area where the degree of burn was highest. If there were wounds with distinct color change, this was preferred to other wounds due to higher chance of infection. For a dry wound, the swab was moistened with sterile saline before swabbing. Once collected, it was homogenized in 4-mL sterile saline. Further processing was done for biopsy culture. The swab samples in the second week were taken from the same wounds that were swabbed in the first week.

**Microbiology.** The swabs were dipped in Stuart’s transport medium and then plated on blood agar, eosin methylthionine blue (EMB), and chocolate agar media. The isolates were identified using conventional identification media after incubation for 18-48 hours at 37°C. An oxidase test was used to differentiate the

### Table 1. Patient demographics (n = 39).

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>33.9 ± 14.74</td>
</tr>
<tr>
<td>Male</td>
<td>22 (66.4)</td>
</tr>
<tr>
<td>TBSA &lt; 10%</td>
<td>5 (12.8)</td>
</tr>
<tr>
<td>TBSA 10%–20%</td>
<td>8 (20.5)</td>
</tr>
<tr>
<td>TBSA &gt; 20%</td>
<td>26 (66.7)</td>
</tr>
</tbody>
</table>

**Cause of burn**

<table>
<thead>
<tr>
<th></th>
<th>Value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hot fluids</td>
<td>2 (5.1)</td>
</tr>
<tr>
<td>Flame</td>
<td>21 (53.8)</td>
</tr>
<tr>
<td>Electrical</td>
<td>4 (10.3)</td>
</tr>
<tr>
<td>Gas explosion</td>
<td>11 (28.2)</td>
</tr>
<tr>
<td>Acid</td>
<td>1 (2.6)</td>
</tr>
</tbody>
</table>

### Table 2. Comparison of swab and biopsy sample cultures on days 7 and 14.

<table>
<thead>
<tr>
<th></th>
<th>Day 7 (n = 30)</th>
<th>Day 14 (n = 30)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frequency</td>
<td>Frequency</td>
<td>Frequency</td>
</tr>
<tr>
<td>Biopsy &amp; swab</td>
<td>1</td>
<td>2.6</td>
</tr>
<tr>
<td>Biopsy &amp; swab</td>
<td>33</td>
<td>84.6</td>
</tr>
<tr>
<td>Biopsy + swab</td>
<td>2</td>
<td>5.1</td>
</tr>
<tr>
<td>Biopsy + swab</td>
<td>2</td>
<td>5.1</td>
</tr>
<tr>
<td>Biopsy + swab</td>
<td>1</td>
<td>2.6</td>
</tr>
<tr>
<td>Total</td>
<td>39</td>
<td>100</td>
</tr>
<tr>
<td>Concordance between swab and biopsy</td>
<td>87.1%</td>
<td>66.6%</td>
</tr>
</tbody>
</table>

1. The isolated pathogen from biopsy sample was the same as the swab sample.
2. Although both swab and biopsy samples yielded a positive result, the isolated pathogen was not the same between sampling methods.

### Table 3. Comparison of microorganisms isolated by swab and biopsy sampling on days 7 and 14.

<table>
<thead>
<tr>
<th></th>
<th>Day 7 (n = 39)</th>
<th>Day 14 (n = 39)</th>
<th>Comparison</th>
</tr>
</thead>
<tbody>
<tr>
<td>Swab</td>
<td>Biopsy</td>
<td>Swab</td>
<td>Biopsy</td>
</tr>
<tr>
<td>Negative</td>
<td>35</td>
<td>35</td>
<td>28</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>1</td>
<td>3</td>
<td>7</td>
</tr>
<tr>
<td>C. freundii</td>
<td>3</td>
<td>1</td>
<td>4</td>
</tr>
</tbody>
</table>

*Swab on days 7 and 14.
* *Bioppy on days 7 and 14.

**Pseudomonas aeruginosa** from other Enterobacteriaceae. **Pseudomonas aeruginosa** is a Gram-negative bacillus with a positive oxidative test that often has a fruity odor and may produce fluorescent pigments on Mullan Hinton agar, indol production, citrate agar, triple sugar iron agar (TSA), methyl red Voges-Proskauer (MRVP), urea agar, and lysine agar (lysine

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Results
A total of 156 samples including 78 swab and 78 biopsy samples were collected from 39 patients (Table 1). The total body surface area (%TBSA) damaged was more than 20% in two-thirds of the patients. Flame injury was the most common cause of burn.

The swab and biopsy comparisons are presented in Table 2. The rate of concordance between swab and biopsy (eg both samples negative or both samples positive with the same microorganism) was 87.1% and 66.6% at days 7 and 14, respectively.

*P. aeruginosa* and *C. freundii* were the two microorganisms found by both sampling methods. The rate of positive cultures for both swab and biopsy increased from day 7 to day 14: 10.3% to 28.2% for swab and 10.3% to 41% for biopsy; *P = 0.011* (swab), *P = 0.04* (biopsy).

The number of *P. aeruginosa* isolates was compared to all isolated microorganisms between days 7 and 14 for both sampling methods. For swab, it was 1 of 4 (25%) on day 7, and 7 of 11 (63.6%) on day 14; whereas, for the biopsy samples, it was 3 of 4 (75%) on day 7, and 12 of 16 (75%) on day 14. Biopsy yielded an equal ratio on days 7 and 14 regarding the number of isolated *P. aeruginosa* relative to all isolated microorganisms (Table 3).

Discussion
Evaluation and treatment of microflora in the burn wound to prevent life-threatening complication such as sepsis are important issues in the care of burn patients. The reliability of different sampling methods for the microbial assessment of wound infection is still debated. In our study, *P. aeruginosa* and *C. freundii* were the two most commonly isolated microorganisms in both sampling methods. While the swab and biopsy samples were closely concordant in day 7, the rate of concordance on day 14 was lower than it was on day 7 (87.1% versus 66.6%).

Theoretically, there should be advantages to using the biopsy culture technique compared to surface swab cultures. The bacteria identified within the burned tissue are more relevant to invasive burn wound infection, and possible sepsis, than the bacterial colonization on the wound surface. Additionally, it may be more accurate to administer antibiotics based on tissue culture findings rather than surface cultures in patients with a burn who are also septic and do not have a positive blood culture.

Although treating the surface of the burn with topical antimicrobials before swab sampling may change the microflora on the surface even if it is washed off, the effect is not problematic on the biopsy cultures.

Uppal et al. reported that in 95% of the cases both sampling methods yielded the same organism. In their study, biopsy was found to be more valuable, as it gives the critical load of an organism beyond which metastatic invasion of the organism takes place. Thus obviating the repeated need for blood culture in burn patients. Interestingly, no microorganism was isolated from the blood cultures. The low level of sensitivity of blood cultures, despite its high level of specificity, has been reported elsewhere.

The most frequently isolated microorganisms (both swab and biopsy) in the present study were *P. aeruginosa* and *C. freundii*.

Although *P. aeruginosa* was found to be the leading cause of infection in burn wounds, the emergence of *Citrobacter* as a common microorganism is a new finding that has not been reported by other studies in both developed and developing countries. *Citrobacter* species are members of Enterobacteriaceae family and have been associated with nosocomial infections in the urinary and respiratory tracts of debilitated hospital patients. *Citrobacter* has also been associated with intra-abdominal infections, hospital-acquired bacteremia, and meningitis.

To determine the source of *Citrobacter*, 350 swab samples taken from hospital equipment and instruments, operating room and intensive care unit floors, and from the hands, nasal passages, and clothing/shoes of hospital staff. Five isolates of *C. freundii* were found on a staff member’s boot and a water faucet, implying the source of the wound infection came from an environmental source.

The high prevalence of *P. aeruginosa* has been reported in other studies and could be explained by the fact that this opportunistic microorganism grows mainly in moist areas, such as a burn wound, and also could be the result of prolonged hospital stays and administration of broad-spectrum antibiotics. *P. aeruginosa* infection is particularly problematic since it is inherently resistant to many drug classes and is able to acquire resistance to all effective antimicrobial drugs.

Although it has been reported that *S. aureus*, especially methicillin-resistant *S. aureus* (MRSA), is a significant microorganism in burn wound infections, it was not isolated in our study.
We found that the incidence of Gram-negative bacilli wound infections, other than \textit{P. aeruginosa} and \textit{C. freundii}, is not problematic compared to other studies that reported a high incidence of Gram-negative bacilli infections, especially \textit{Acinetobacter}.\textsuperscript{13,14,15,16}

To our knowledge, the time-related concordance between swab and biopsy samples in burn wounds has not been studied thoroughly. Aiptoparik et al\textsuperscript{17} reported time-related changes in the bacterial profile of burn wounds and body flora including nasal, axillary, inguinal, and umbilical regions. Coagulase-negative staphylococci (CNS) and \textit{S. aureus} were the most prevalent isolates found in all body regions, according to cultures performed at admission. Over time, the CNS isolates decreased, while there was a marked increase in \textit{S. aureus} and \textit{P. aeruginosa} infections. They did not differentiate their wound isolates from other isolates, and unlike the present study, they did not take biopsy samples.

To evaluate the positive percentage and the role of different microorganisms in the wound infection of patients during their stay in the burn unit, we compared the isolated microorganisms of both biopsy and swab samples at days 7 and 14. For both swab and biopsy samples, the rate of positive results was higher at day 14 compared to day 7. Although all positive cultures yielded only \textit{P. aeruginosa} and \textit{C. freundii} in 2 weeks, the role of each microorganism was different between the two sampling methods. Positive swab samples for \textit{P. aeruginosa} were fewer than swabs positive for \textit{C. freundii} at day 7; but by day 14, the number of positive samples for both \textit{P. aeruginosa} and \textit{C. freundii} were similar.

**Conclusion**

An 87.1\% concordance was found between swab and biopsy samples at day 7 and 66.6\% at day 14. Based on the results of this study, swab sampling can be considered a good tool for monitoring burn wounds within the first week of treatment, and could defer the need for invasive biopsy sampling. For patients who remain in the burn unit for a longer period, biopsy samples are justified for monitoring the bacterial activity in burn wounds.

**Acknowledgement**

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**References**


