Povidone Iodine and Skin Disinfection before Initiation of Epidural Anesthesia

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Background: Povidone iodine (PI) solution is used commonly for skin disinfection before epidural and spinal anesthesia. Although there have been reports indicating the presence of microbial contaminants in PI solution, none have evaluated the prevalence of PI contamination. The aims of this study were to assess the frequency of bacterial contamination of previously opened bottles of PI solution and to compare the effectiveness of new and previously opened bottles of PI solution for skin disinfection.

Methods: Twenty previously opened and ten previously unopened multiple-use bottles of PI solution were evaluated for microbial contamination. In addition, final swabs and PI solution used for skin disinfection in 80 patients undergoing elective epidural analgesia were evaluated.

Results: The inside of the bottle cap or the PI solution from 40% of the multiple-use PI bottles in use were contaminated. There was no growth from any previously unused PI bottles. Povidone iodine from newly opened bottles provided more effective skin decontamination than did solution from previously opened bottles.

Conclusions: Multiple-use PI bottles in normal use may become contaminated by bacteria. In addition, PI solution from previously opened bottles was less effective than PI from previously unopened bottles. Based on these findings, if PI solution is chosen for skin antisepsis before initiation of epidural and spinal anesthesia, only single-use containers should be used. (Key words: Bacteria; contamination; infection.)

ADEQUATE skin antisepsis before initiation of epidural and spinal anesthesia is essential because of complications that could result from bacterial contamination in an immunologically compromised area.1,2 Povidone iodine (PI) solution is often used to provide such antisepsis. It is common practice among anesthesiologists to use PI solution from multiple-use bottles, with the implicit assumption that antisepctic solutions do not support bacterial growth and that the active ingredients maintain their effect after the bottle has been opened. Several reports have indicated the presence of microbial contaminants in PI solution,3,4 but none of these have evaluated the prevalence of PI solution contamination. Further, no report of the potential for PI solution contamination has appeared in the anesthesiology literature. The aims of our study were twofold: (1) to assess the frequency of bacterial contamination of previously opened bottles of PI that were in use in our hospital, and (2) to compare the efficacy of skin disinfection produced by PI solution taken from new and also previously opened bottles.

Methods

Microbiological Assessment of Povidone Iodine Bottles

Twenty previously opened, multiple-use, 473-ml bottles of 10% PI (Betadine; Purdue Frederick Co., Norwalk, CT) were collected during a single sweep from the labor and delivery suites at St. Luke’s-Roosevelt Hospital Center. The length of time any bottle had been open was unknown, but all bottles were in current use. The bottles were numbered and submitted to the microbiology laboratory for evaluation, along with ten unused (control) bottles of PI solution. Various amounts of solution were removed from the control bottles to blind the microbiology laboratory to the previously opened or unused status of the bottles. Each bottle was evaluated as follows.

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Anesthesiology, V 88, No 3, Mar 1998
SKIN DISINFECTION BEFORE EPIDURAL ANESTHESIA

Povidone Iodine Solution. After the bottle cap was removed, 10 ml PI solution was removed from each PI bottle using a sterile technique. Aliquots (100 μl) of these solutions were inoculated into chocolate agar, trypticase soy agar with 5% sheep blood, MacConkey agar, cooked-meat glucose broth, and thioglycolate broth.

Bottle Cap. The insides of the PI bottle caps were cultured to identify organisms that might contaminate the PI solution during pouring, particularly those that might preferentially grow on the cap surface rather than in solution (e.g., slime-producing coagulase-negative staphylococci).

The cap from each bottle was removed, and a sterile culture swab moistened with sterile saline was run along the inside of the cap. The swab was then used to inoculate chocolate agar and cooked-meat glucose broth.

All plates were incubated at 37°C in 5% carbon dioxide for 72 h and reviewed daily for growth; cooked-meat glucose and thioglycolate broths were incubated and reviewed daily for 14 days. All media were obtained from BBL (Becton-Dickinson, Cockeysville, MD). All isolates were identified using standard methods, including conventional tests and API20E (bioMerieux Vitek, Hazelwood, MO).

Effectiveness of Povidone Iodine Solution as an Antiseptic

The study was reviewed and approved by the hospital’s institutional review board before initiation. Final prep swabs (“third sponge sticks”) and bottles of PI solution used as antiseptic for 80 patients undergoing elective epidural anesthesia during a 20-week period were evaluated to identify bacterial contamination and to compare bacteria remaining on the skin after routine skin preparation. Previously opened or new bottles were used as the source for the PI solution based on the choice of the labor and delivery nurse assisting in the setup; the nurses were not aware of the study at this point. The skin of each patient’s back was prepped in our usual manner with three sequential PI-soaked sponge sticks. The anesthesiologist waited at least 20 s between each sponge stick application to allow the PI solution on the skin to dry. The third sponge stick was not allowed to touch any area that had not been prepared with the first two PI-immersed sponges. After each patient’s back had been prepared, the third sponge stick was immediately placed within a sterile 60-ml syringe and transported to the microbiologist along with the bottle of PI used. The PI solution and bottle cap were evaluated as outlined here before, with the exception that thioglycolate broth was not included. In addition, the sponge applicator was inoculated to trypticase soy agar with 5% sheep blood, chocolate agar, and MacConkey agar by directly applying the sponge to the surface of each plate sequentially. Plates were incubated at 37°C in 5% carbon dioxide for 72 h and reviewed daily for growth. In addition, we demonstrated the integrity of our transport system by evaluating ten new bottles of PI and ten sponges that were immersed in those PI solutions but not used for skin preparation. Varying amounts of PI solution were removed from all new bottles that were sent to the microbiology laboratory to maintain blinding.

Statistical Analysis

For the microbiological assessment of previously used and unopened PI bottles, inferences about the population proportion of contaminated bottle cap specimens was based on the binomial probability distribution. For the antiseptic effectiveness of PI, the difference between the proportions of contaminated solutions from previously opened and new bottles was tested using the chi-square test and by Fisher’s exact test for the difference between the proportions of contaminated caps from previously opened and new bottles. The risk of contamination from any PI source (cap, solution, or swab) was estimated using the odds ratio. Differences <0.05 were considered significant.

Results

Microbiological Assessment of Previously Used and Unopened Povidone Iodine Bottles

Of the 30 bottles of PI that were sent to the microbiology laboratory for culture, 10 had not been opened before (controls). None of these controls yielded growth from either the cap or the PI solution. The volumes of PI solution remaining in the 20 bottles that were in current use varied from 100-450 ml. There was no correlation between the remaining volume of PI solution and the presence or absence of bacterial growth. Four plastic bottle caps and PI solutions from four different bottles yielded organisms including Staphylococcus epidermidis, Staphylococcus haemolyticus, Bacillus, and Stenotrophomonas (Xanthomonas/Pseudomonas) maltophilia. Thus 8 of the 20 PI bottles (40%) were contaminated. The semiquantitative

Anesthesiology, V 88, No 3, Mar 1998
cultures from the four PI solutions represented growth at levels of 10–10⁴ (10–100) CFU/ml. A significant difference was found when comparing the incidence of positive bacterial cultures among the 10 new bottles (no growth) and the 8 positive cultures found in the 20 previously opened bottles (two-sided Fisher’s exact test, P = 0.04). Because it is commonly assumed that organisms do not grow in the presence of an antiseptic solution such as PI, the proportion of contaminated cap and bottle specimens was tested against a negligible expectation (P₀ = 0.001) and found to be significantly higher (z = 52.9, P < 0.001).

**Effectiveness of Povidone Iodine Solution as an Antiseptic**

A marked difference was noted in the recovery of organisms from the third sponge sticks when the PI solution used for skin preparation came from new rather than previously used bottles. Of the 40 third sponges obtained from procedures in which the PI solution used for skin preparation came from previously used bottles, 16 (40%) yielded bacteria including *Bacillus* species, *Enterococci, S. epidermidis, S. haemolyticus*, and *S. (Xanthomonas/Pseudomonas) maltophilia*. In comparison, bacteria were isolated only from 2 of the 40 third sponges (5%) obtained from procedures in which the PI solution used for skin preparation came from new bottles (one *Bacillus sp.* and one *S. epidermidis*). *Bacillus* were the most frequently recovered species.

The number of colonies were counted for those cultures that were positive. As shown in table 1, the two positive cultures from women whose skin was cleaned with PI solution from new bottles grew only one colony each. In contrast, nearly three quarters of positive cultures from women whose skin had been cleaned with PI solution from previously opened (used) bottles grew more than one colony. Thus, the proportion of swab cultures with more than one colony was significantly higher for PI solutions from multiple use bottles than for PI solution from new bottles (Fisher’s exact test, P = 0.0001). One culture from the previously used PI bottle group had a much higher number (n = 12) of colonies. Even when this culture was eliminated from analysis, the difference between type of bottle and proportion of cultures with more than one colony was still significant (Fisher’s exact test, P = 0.0002).

The proportion of swabs yielding any bacterial growth was significantly higher when previously opened PI bottles (P < 0.001) were used. There was no bacterial growth from PI solution from any of the ten previously unused control bottles or the concurrent swabs. The risk of bacterial growth from any source (bottle cap or solution or back swab) was increased 13 times when PI solution from a previously opened bottle (odds ratio = 13; 95% confidence interval, 3.8–44.1) was used.

With one exception, in each case the bacteria isolated from the third swabs were not isolated from the concurrent PI solution used. Thus these bacteria were considered to represent skin flora and not bacteria from the PI solution. In the single exception, the same organism was isolated from both the swab and the PI solution, suggesting that contaminated PI was the source of the organism that had potentially contaminated the patient’s skin.

**Table 1. Organisms Isolated from Back Swabs but Not Found in Concurrent PI Solution**

<table>
<thead>
<tr>
<th>New vs. previously Used PI Bottle</th>
<th>Volume in Bottle</th>
<th>Organisms Isolated</th>
<th>Colonies per Swab Impression</th>
</tr>
</thead>
<tbody>
<tr>
<td>Used</td>
<td>200</td>
<td><em>S. haemolyticus</em></td>
<td>1</td>
</tr>
<tr>
<td>Used</td>
<td>200</td>
<td><em>Bacillus</em></td>
<td>1</td>
</tr>
<tr>
<td>Used</td>
<td>200</td>
<td><em>Bacillus</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
<td>200</td>
<td><em>Bacillus</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
<td>400</td>
<td><em>Bacillus</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
<td>100</td>
<td><em>Bacillus</em></td>
<td>5</td>
</tr>
<tr>
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<td>200</td>
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<td>3</td>
</tr>
<tr>
<td>Used</td>
<td>200</td>
<td><em>Bacillus</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
<td>200</td>
<td><em>Pseudomonas</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
<td>200</td>
<td><em>Bacillus</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
<td>300</td>
<td><em>S. epidermidis</em></td>
<td>2</td>
</tr>
<tr>
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<td>350</td>
<td><em>Bacillus</em></td>
<td>2</td>
</tr>
<tr>
<td>Used</td>
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<td><em>Bacillus</em></td>
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</tr>
<tr>
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<td><em>Bacillus</em></td>
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</tr>
<tr>
<td>New</td>
<td>na</td>
<td><em>Bacillus</em></td>
<td>1</td>
</tr>
<tr>
<td>New</td>
<td>na</td>
<td><em>S. epidermidis</em></td>
<td>1</td>
</tr>
</tbody>
</table>

**Discussion**

Bacterial growth occurring in disinfectant solution has been reported previously, but the frequency of contamination and implications for skin disinfection before epidural anesthesia has not been addressed. The adequacy of skin disinfection before epidural anesthesia is of even greater concern as use of long-term epidural catheters increases.

The first phase of our study, in which 40% of the bottles of PI solution currently in use in the labor and
SKIN DISINFECTION BEFORE EPIDURAL ANESTHESIA

delivery suite yielded growth of bacteria from either the PI solution or the inside of the bottle cap, indicates that contamination of previously opened containers of PI solution is not infrequent. The appearance of the bottle and cap was not a good indicator of bacterial contamination. For example, the bottle that grew *S. (Xanthomonas/Pseudomonas) maltophilia* appeared no different than bottles and caps that were found to be bacteria-free. Isolation of organisms from the cap may be particularly relevant for disinfection before placement of an epidural catheter because these bacteria are adapted for growth on a synthetic surface.

Also of note, the antibacterial activity of PI solution appeared to be decreased for previously opened bottles. There was a dramatic difference in our ability to isolate organisms from the third sponge stick when a previously opened rather than a previously unopened bottle of PI solution was used. None of the organisms that were isolated from the sponge sticks was present in the PI solution, except for one case in which *S. haemolyticus* was isolated from the swab and PI bottle cap. Therefore, the organisms from the swabs represented bacteria from the patient's skin that had not been eliminated by the PI solution. The predominance of *Bacillus* species recovered from the swabs is not surprising because this organism is commonly found on skin and in the environment.

Because PI bottles were not dated when they were first opened, we could not correlate directly time in use with likelihood of contamination. The proportion of bottles of PI solution that were contaminated was much greater during the first phase than the second phase of the study. When the labor and delivery staff became aware of the results of the first phase of the study, all bottles of PI solution with evidence of repetitive previous use (spillage on the outside of bottle or cap or contents less than a quarter full) were removed from use. Eventually, all multiple-use PI bottles were removed and replaced with single-use containers.

Bacteria can survive in povidone iodine solution, as shown in this study and several previous reports.14 Previously opened iodophor-containing solutions may lose their antimicrobial effectiveness because of a partitioning of the iodine between the micelle structure of the surface active agent and the water phase.15 This may explain why the PI solutions from previously opened bottles were not as effective as new bottles in this study.

A potential disadvantage of the design of our study to determine whether PI solution from previously opened bottles became less effective for skin disinfection was that a culture was not taken directly from the patient's skin after preparation with PI, but rather the third sponge stick was used to estimate residual skin contamination. The sponge stick that was ultimately cultured had been previously bathed in PI solution. Although transport time to the laboratory and delay before plating was minimal, the PI solution saturating the swab could continue to have antimicrobial activity during transport. Thus the actual bacterial counts may have been higher than those observed, leading to underestimation of the level of bacterial contamination of the skin. Conversely, an advantage of the design is that third sponges from all unselected patients undergoing skin disinfection before epidural anesthesia were evaluated, and thus no confounding variables associated with socioeconomic or educational factors that might be related to willingness to give informed consent were introduced.

None of the patients in our study, regardless of PI solution used, developed a postepidural infection. However, one PI solution and the accompanying third sponge both yielded *S. haemolyticus*, suggesting that if contaminated PI solution is used, there is potential to introduce a pathogen onto the patient's back during skin preparation.

Although the results of a recent study showed that isolation of viable organisms from excised skin specimens after disinfection with iodine solution occurred in many patients,9 there are few reports of iatrogenic abscess or meningitis in the anesthesia literature.8-10 In a retrospective review of 505,000 women receiving obstetric epidural anesthesia, not a single case of meningitis was reported.11 However, two cases of postepidural meningitis have been reported in a review by Ready and Helfer.9 The causative organisms in these cases were listed as *Streptococcus fecalis*, now called *Enterococcus faecalis*, and an alpha-hemolytic streptococcus, previously called *S. uberis*. A recent case report by Davis et al.9 described a patient in whom bacterial meningitis developed that was caused by *Streptococcus agalactiae* (B-hemolytic *Streptococcus* group B) after an uncomplicated epidural anesthetic for labor. In a similar report, *Streptococcus sanguis* meningitis developed in a patient after an epidural with inadvertent dural puncture.12 Possible portals of entry for bacteria that cause postepidural abscesses or other infection include skin contamination as a result of improper skin preparation, as well as contamination by the operator, contamination of the local anesthetic, or hematogenous dissemination of bacteria.13

Although rare, the consequences of undiagnosed or
misdiaagnosed epidural infection are grave and include paraplegia and death.\textsuperscript{14,15} To reduce the risk of infectious complications of epidural anesthesia, attention should focus on any means of prevention, including optimal skin disinfection. We suggest that multiple-use containers of PI solution should no longer be used for skin disinfection, especially when the anesthetic technique involves the spinal cord or other normally compromised sites.

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References