BACTERIAL CONTAMINATION OF LIQUID HAND SOAPS USED IN PUBLIC RESTROOMS

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Abstract
The objective of this study was to determine the occurrence of heterotrophic and coliform bacteria in liquid hand soaps collected from public restrooms in the United States. Samples included public restrooms in restaurants, health clubs, office buildings and retail stores. The liquid soap samples collected were from refillable dispensers (also referred to as “open systems” or “bulk soap systems”). Of 541 samples, 133 (25%) had bacterial numbers greater than 500/mL, and 87 samples (16%) contained coliform bacteria. Approximately 65% of the bacteria isolated from the soap belonged to the coliform group. The average number of bacteria detected in the soap was 3.0x10⁵ CFU/mL, with a range of 500 to 5.3x10⁷ CFU/mL. The average number of coliform bacteria was 3.9x10⁴ CFU/mL, with a range of <10 to 6.5x10⁷ CFU/mL. Opportunistic pathogens identified in the liquid soap samples included Klebsiella ornithinolytica, Klebsiella pneumoniae, Enterobacter aerogenes, Proteus mirabilis and Escherichia coli. No bacteria were detected in dispensers that required sealed soap replacement. All of the dispensers in the soap samples were Gram-negative bacteria. This is most likely because of the presence of sodium laurel sulfonate in the soap, which inhibits the growth of Gram-positive bacteria. The results suggest that some liquid soap dispensers become colonized by Gram-negative bacteria over time, possibly because of the degradation of preservatives in the soap.

Introduction
Washing hands with soap and water is a universally accepted method to reduce the microbial load on the hands and is usually the first step of personal hygiene. However, many people who use public restrooms use soap dispensers that are refillable using a stock soap solution. The CDC recognized in 1975 that the use of these types of dispensers can result in a salient environment for the growth of potentially disease-causing microorganisms. Current hand hygiene guidelines still do not recommend the use of refillable dispensers. The liquid soap used in these dispensers can become contaminated regardless of the preservative used when the microbial population exceeds the preservatives defenses. When product contamination has been reported, contamination was more likely to have occurred extrinsically (after manufacturing) than intrinsically (during manufacturing). The likelihood of extrinsic contamination is greatest when the product is open to repeated exposure to bacteria from the user or the environment, hence, the packaging and the dispensing method plays a significant role in product safety.

Materials and Methods
Liquid soap samples were collected from public restrooms in five cities (Boston, MA (107), Atlanta, GA (120), Columbus, OH (109), Los Angeles, CA (94), and Dallas, TX (111)). Samples were organized into 5 categories: office, health clubs, food service, retail locations and other (education, libraries, etc.). All samples were confirmed to be from open-refillable systems.

The samples were collected in sterile 50 mL conical tubes and shipped to the laboratory on ice. 1 mL of DE neutralizing broth (Remel, Lenexa, KS) was added to each sample tube and shaken vigorously for 40 seconds. Heterotrophic plate counts (HPC) were obtained by the spread plate method on R2A media (Difco, Sparks, MD). Plates were incubated at 30°C for 5 days. Any sample showing bacterial growth was confirmed for Coliform bacteria.

Coliform analysis and enumeration was performed using the spread plate method on mEndo agar (Difco, Sparks, MD). Plates were incubated at 35°C for 5 days. Any sample showing bacterial growth was confirmed for Coliform bacteria.

For each sample, a spread plate was performed using the spread plate method on TSA plates (Difco, Sparks, MD) for isolation and identification. TSA plates were incubated at 35°C for 24 hours. Identification of bacteria was obtained using API20E strips (BioMerieux, Marcy-l’Etoile, France).

Frequency analysis was performed using the spread plate method on TSA plates with 5% sheep blood (BD) and sheep blood agar (BD). Plates were incubated at 35°C for 24 hours. Beta hemolytic isolates were enumerated and identified as a TSA plate and incubated for 24 hours at 35°C. Isolated colonies underwent further confirmation testing utilizing catalase production, microscopic morphology, capsulate production (tube and slant tests) and antibiotic (penicillin G) sensitivity.

Results
The total number of liquid soap samples analyzed in this report were 541, consisting of 428 soap samples from the sink area and 113 soap samples from showers. Samples with <500 colony forming units (CFU/mL) were not considered since industry standards allow for this amount of bacteria in liquid soap.

<table>
<thead>
<tr>
<th>Total Number of Open Refillable Soap Samples</th>
<th>Number of Samples with Bacteria</th>
<th>Number of Samples with Coliforms</th>
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<tbody>
<tr>
<td>541</td>
<td>133 (25%)</td>
<td>87 (16%)</td>
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Heterotrophic bacterial numbers detected in the liquid soap samples ranged from 500 to 5.3x10⁷ CFU/mL. The average number of bacteria found on 500 mL of soap was 3.02x10⁵ CFU/mL. Coliform bacteria ranged from <10 to 6.5x10⁷ CFU/mL in liquid soap samples, with an average of 3.9x10⁴/CFU/mL of soap. No Staphylococcus aureus were detected in any of the liquid soap samples analyzed.

Summary
A total of 133 liquid soap samples were from the shower area of health clubs, 65 from men’s showers and 48 samples from women’s showers.

Conclusions
High levels of bacterial contamination (average 3.02x10⁵ CFU/mL) were found in 25% of the liquid soap samples in this study. Since these samples represent a diverse cross-section of geographic locations and individual sites, it is concluded that refillable, open, “bulk” liquid soap systems commonly found in the U.S. are routinely contaminated with bacteria. Many of the bacteria isolated are opportunistic pathogens which can cause a variety of health issues including respiratory infections, bloodstream infections, urinary tract infections and skin infections. The type and level of bacteria found in these systems present a potential health risk to users, especially to any immunocompromised individuals.

Acknowledgments
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