

Loofah Sponges as Reservoirs and Vehicles in the Transmission of Potentially Pathogenic Bacterial Species to Human Skin

EDWARD J. BOTTONE,* ANTHONY A. PEREZ II, and JAMEL L. OESER

Clinical Microbiology Laboratories, The Mount Sinai Hospital, New York, New York 10029

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Loofah sponges are natural products used as exfoliative beauty aids. As a consequence of tracing a case of *Pseudomonas aeruginosa* folliculitis to a contaminated loofah sponge, we assessed the role of loofah sponges in supporting the growth of a wide variety of bacterial species. Our data show growth enhancement of sterile loofah fragments for numerous gram-negative (*Pseudomonas*, *Xanthomonas*, and *Klebsiella*) and gram-positive (*Enterococcus* and group B *Streptococcus*) species of human and environmental origin. Furthermore, hydrated new, unused loofah sponges undergo a shift in bacterial flora from sparse colonies of *Bacillus* spp. and *Staphylococcus epidermidis* to a predominantly gram-negative flora. The growth-promoting potential of loofah sponges (and other exfoliatives) can be further augmented by desquamated epithelial cells entrapped in the loofah fibrous matrix. Therefore, as loofah sponges (and other exfoliatives) can serve as a reservoir and a vehicle for the transmission of potentially pathogenic species to the human skin, we recommend their decontamination with hypochlorite (10%) bleach at regular intervals.

Loofah sponges are derived from vegetable gourds of the cucumber family. Composed of a network of dried cellulose fibers, loofahs are used extensively as beauty aids designed to remove superficial epithelial cells during showering and bathing. While investigating a case of *Pseudomonas aeruginosa* folliculitis linked to a contaminated loofah sponge (2), we showed that sterile loofah sponges supported the growth of *P. aeruginosa* in the absence of any other food source. Because of these findings, we assessed the growth-promoting capability of loofah sponges for a variety of bacterial species. Based on the ability of loofah sponges to support and harbor many species that may be transmitted to the human skin, we offer a rationale for the periodic decontamination of these products.

MATERIALS AND METHODS

New loofah sponges made in El Salvador and marketed by Schroeder and Tremayne, Inc., St. Louis, Mo., were purchased and sterilized by autoclaving. Fragments of sterile loofah sponges (approximately 2 by 1.5 cm) were then individually placed into sterile tubes containing 2.5 ml of distilled water and inoculated with 10^3 to 10^4 CFU of a variety of gram-positive and gram-negative bacterial species per ml (Table 1). After incubation at 35°C for 24 h, colony counts were determined subsequent to 10-fold dilution. Controls consisted of inoculating each bacterial species into un-supplemented distilled water.

To assess the change in the microbial flora of new loofah sponges after hydration, the baseline microbial flora was determined for three sponges by touch inoculating the sponges to 5% sheep blood agar (BBL Microbiology Systems, Cockeysville, Md.) and MacConkey agar, and a sponge segment (1 by 2.5 in.) was then placed in a sterile glass container and immersed in sterile distilled water. After 24 h of incubation at room temperature, each segment was removed, touch inoculated onto sheep blood and MacConkey agars, and individually placed into a sterile vessel and left exposed to ambient air to dry. At daily intervals for

2 weeks, each sponge segment was touch inoculated as described above to determine the time interval to return to baseline flora. A similar study was carried out for 1 of 10 sponges currently in use which became grossly contaminated with *Klebsiella oxytoca* and *Alcaligenes xylosoxidans* (Fig. 1). This was deemed necessary, since many manufacturers recommend drying of the sponge between uses, probably as a means of preventing bacterial overgrowth.

Decontamination of the sponge was assessed by individually immersing in hypochlorite bleach (one part bleach to nine parts sterile distilled water) segments of a new sponge that was hydrated for 24 h, the in-use sponge overgrown with *K. oxytoca* and *A. xylosoxidans*, and new sponge segments exposed overnight to 10^8 CFU of a clinical (blood) isolate of *P. aeruginosa* in sterile distilled water. At 1-min intervals for 5 min, a sponge segment was removed, rinsed vigorously with sterile distilled water, touch inoculated as above, and placed into a tube containing 5 ml of brain heart infusion broth. Plates and broth were incubated at 35°C and examined for bacterial growth daily for 7 days.

RESULTS

In the absence of any other source of utilizable growth substrate, sterile loofah sponge fragments were growth promoting for *P. aeruginosa* and a variety of other gram-negative bacterial species (Fig. 2). Specifically, counts of *P. aeruginosa* increased from 10^3 to 10^4 CFU/ml to 10^8 to 10^9 CFU/ml in the presence of only the loofah sponge. Similar growth enhancement was noted for *Xanthomonas maltophilia*, *Serratia marcescens*, *Flavobacterium* species, and *Escherichia coli*. A modest growth enhancement (2 logs) was noted for *Enterobacter cloacae*, *Enterobacter aerogenes* and *Acinetobacter anitratus* (*A. baumannii*). No change in counts for any of the test species occurred in the un-supplemented distilled water controls.

Loofah sponge fragments were not growth promoting for *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pyogenes*, or the group G streptococcal strains tested. By way of contrast, *Streptococcus agalactiae* achieved a 2-log increase in growth over un-supplemented

* Corresponding author.

TABLE 1. Bacterial species tested for growth enhancement in sole presence of sterile loofah fragments

Type and species	No. of strains tested
Gram negative	
<i>P. aeruginosa</i>	10
<i>X. maltophilia</i>	8
<i>Flavobacterium</i> species.....	3
<i>S. marcescens</i>	3
<i>E. coli</i>	5
<i>E. cloacae</i>	5
<i>E. aerogenes</i>	5
<i>A. anitratus</i>	5
Gram positive	
<i>E. faecalis</i>	10
<i>E. faecium</i>	10
<i>S. aureus</i>	10
<i>S. epidermidis</i>	5
<i>S. pyogenes</i>	3
<i>S. agalactiae</i> (group B).....	3
<i>Streptococcus</i> group G.....	3
<i>Bacillus</i> species.....	5

controls, and *Enterococcus faecalis* and *Enterococcus faecium* achieved counts of 10^5 CFU/ml after overnight incubation. Interestingly, loofah fragments were as growth promoting for *Bacillus* species as for many of the gram-negative

species, with counts increasing from between 10^3 and 10^4 to 10^8 CFU/ml after 24 h of incubation (Fig. 2).

Hydrated new, unused loofah sponges underwent a shift in microbial flora from a baseline of scattered colonies of *Bacillus* spp. and *S. epidermidis* to an overwhelming growth of gram-negative species inclusive of *Pseudomonas fluorescens*, *E. agglomerans*, *E. cloacae*, *Citrobacter freundii*, *Flavobacterium* species, and *A. anitratus*. Common to all three sponges tested were *P. fluorescens*, *E. agglomerans*, and *Flavobacterium* species, while the remaining species were variably encountered (Table 2).

The time interval over which the new hydrated, the in-use, and the *P. aeruginosa*-challenged sponge segments showed significant decreases in CFU of gram-negative species exceeded 2 weeks. While there was a steady but gradual decline in the number of colonies of each gram-negative bacterial species during this time interval, counts never reached less than 1 CFU. Upon overnight rehydration of the sponge, however, CFU of the gram-negative flora soared to predrying levels.

Exposure of contaminated loofah segments to a 1:9 dilution of hypochlorite bleach for 1 min resulted in total decontamination of the sponge segments. Neither direct inoculation of the tested segments onto agar media nor inoculation to broth cultures grew any of the bacterial species (*P. aeruginosa* and *Klebsiella* and *Alcaligenes* species, etc.) prior to bleach exposure and vigorous washing with sterile distilled water. Furthermore, to ensure that

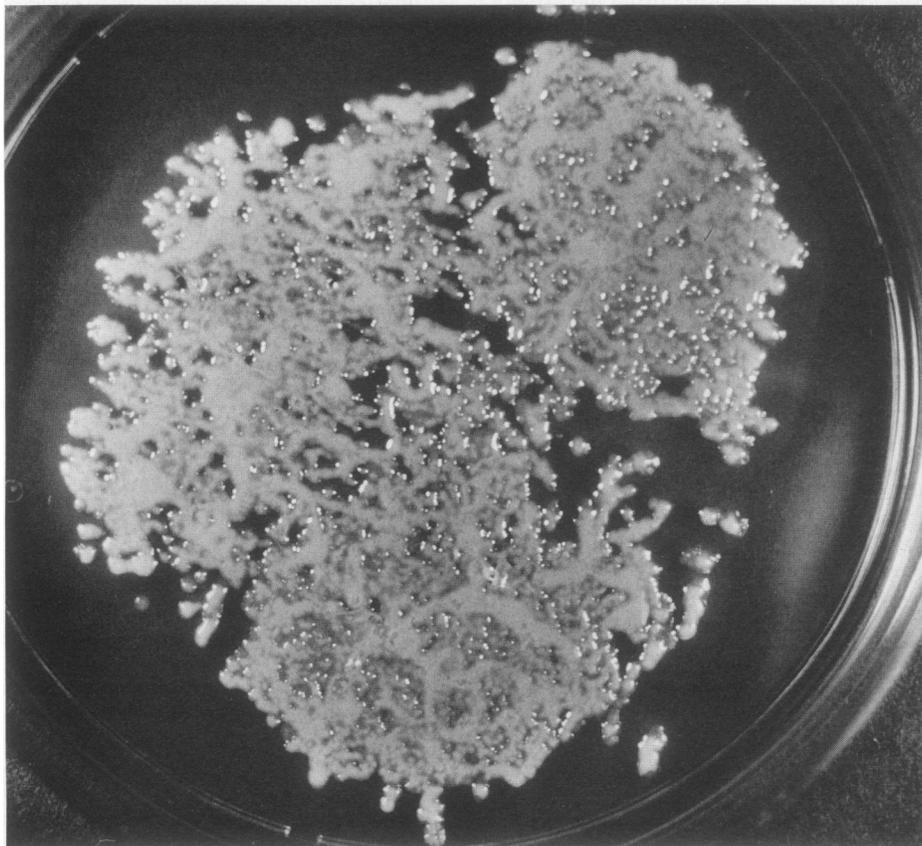


FIG. 1. Overwhelming growth of *K. oxytoca* and *A. xylosoxidans* 24 h after touch inoculation of sheep blood agar with an in-use loofah sponge.

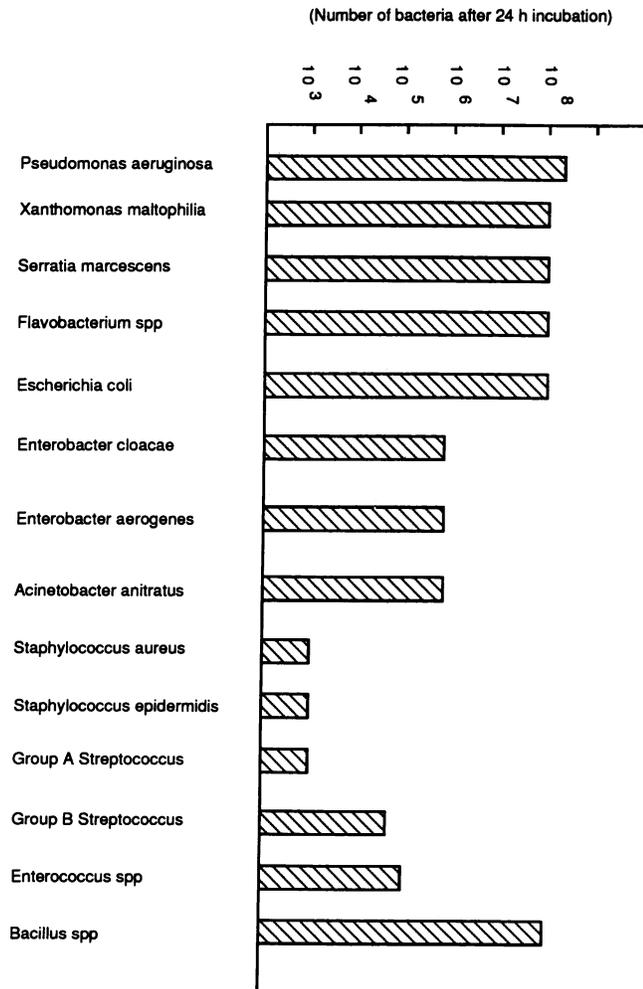


FIG. 2. Growth-promoting potential of sterilized loofah sponge segments inoculated with 10³ to 10⁴ CFU of each bacterial species.

negative broth cultures were a result of the killing of bacterial contaminants and not due to bleach carryover, each of the loofah-broth cultures was challenged with 10² CFU of *P. aeruginosa*. After a subsequent 24-h incubation, all tubes showed marked turbidity and *P. aeruginosa* could be recovered from all broths.

TABLE 2. Bacterial flora of new loofah sponge prior to and after wetting^a

Bacterial sp. recovered	Time recovered
<i>S. epidermidis</i>	Prior to wetting
<i>Bacillus</i> species.....	Prior to wetting
<i>P. fluorescens</i>	After wetting
<i>E. agglomerans</i>	After wetting
<i>E. cloacae</i>	After wetting
<i>C. freundii</i>	After wetting
<i>A. anitratus</i>	After wetting
<i>Flavobacterium</i> species.....	After wetting

^a Composite results of testing three new sponges.

DISCUSSION

Loofah sponges have gained immense popularity as exfoliative beauty aids designed to invigorate the human skin. As a natural product, however, loofah sponges play host to a variety of bacterial species, especially those of environmental origin.

Sterile loofah sponges are growth promoting for *P. aeruginosa* (2). In the present study, our observations have been extended to include a range of other bacterial species, e.g., *X. maltophilia* and *Flavobacterium* species, whose basic natural habitat is environmental. Of unique interest was the failure of sterile loofah fragments to support the growth of *S. pyogenes*, *S. aureus*, and *S. epidermidis*. In contrast to these species, which are predominantly human colonizers, loofah fragments supported *S. agalactiae* and *Enterococcus* species which, in addition to human colonization, have an animal reservoir as well (4, 5). Further testimony to a predilection in the bacterial growth-promoting potential of loofah sponges for environmental species can also be derived from the data obtained for *Bacillus* species. In the sole presence of loofah fragments, each of the five strains studied showed growth enhancement equivalent to that of the gram-negative species tested. While reasons for the growth enhancement of environmental species have not been elucidated, it is known that in nature loofah gourds falling from a vine are decomposed by environmental bacteria, suggesting a capability for the utilization by these bacteria of loofah components for growth.

Bacterial growth enhancement was also noted for in-use loofah sponges. In a survey of 10 in-use sponges (unpublished data), all showed an overwhelming growth of gram-negative species (inclusive of enterobacteria) when touch inoculated to blood and MacConkey agars. In three instances, *P. aeruginosa* was admixed with the diverse bacterial floras. It can be postulated that once in use, loofah sponges may be additionally contaminated with gram-negative bacterial species derived from the human body (e.g., *Klebsiella* species and *E. coli*), especially from the perineal area, and from the household setting itself. Furthermore, desquamated epithelial cells entrapped in the loofah mesh-work may additionally facilitate bacterial growth. In our experience (unpublished data) with inanimate exfoliatives such as pumice stones, growth of gram-negative bacterial species, including *P. aeruginosa*, is readily achieved after the stone has been used for skin debridement. The sequestering of desquamated skin in the crevices of the stone readily serves as a nidus for bacterial growth. On the basis of the preceding, it is apparent that bacterial overgrowth of loofah sponges and other exfoliatives literally takes place overnight. Chance contamination with *P. aeruginosa* may therefore represent a potential threat for the development of folliculitis.

New, dry loofah sponges contain a sparse bacterial flora consisting of *Bacillus* species and *S. epidermidis*. Upon hydration with sterile distilled water, however, there is a dramatic shift in the bacterial flora to predominantly gram-negative species, which are not detectable on initial touch inoculation of agar media. The source of these gram-negative species is therefore somewhat enigmatic. Initially, it may be argued that they are present in such small numbers as to be nondetectable by touch inoculation of agar media. Secondly, they may be embedded in the fibrous matrix of the loofah sponge and liberated upon hydration. In their manufacture, loofah sponges are soaked in water for several days to remove the gourd skin and seeds (1). They are then dried and

marketed. Hydration at this time could well introduce and even promote the growth of environmentally derived gram-negative and gram-positive (*Bacillus*) bacterial species, which embed in the cellulose fibers. Alternatively, prior to hydration during use, the bacterial component may be viable but not culturable (3). In this stage, bacteria can survive under adverse environmental conditions (such as drying) and grow under more favorable (hydration) conditions.

At this juncture, it is recognized that loofah sponges and similar beauty aids (e.g., pumice stones and synthetic sponges) require regular disinfection. Manufacturers recommend drying the sponge between uses. This practice, however, offers little in the way of reducing the numbers of bacterial species present. In most instances, sponges (and pumice stones) are used frequently, kept in the showering or bathing area, and are therefore constantly moist. Furthermore, prolonged drying (>2 weeks) is necessary to bring about a substantial diminution in bacterial counts which, upon subsequent rehydration, are elevated once again. We therefore strongly recommend that exfoliative sponges (natural or synthetic) and other exfoliative devices such as pumice stones undergo regular hypochlorite (bleach) decontamination. In this fashion, the epidemiologic link between exfoliatives and transmission of *P. aeruginosa* (2, 6, 7) and

other potentially pathogenic bacterial species to the human skin can be diminished.

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