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Savasana and Staph Infections: An examination of the effectiveness of 'natural' and commercial cleaners against *Staphylococcus carnosus* on yoga mats

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Abstract

Staphylococcus aureus is one of the most common bacterial strains found in community fitness centres in the United States. Recent studies have found a link between methicillin-resistant *S. aureus* (MRSA) infections and improperly sanitized fitness equipment, such as yoga mats. Therefore, effective disinfectant methods are crucial for the prevention of bacterial transmission. As opposed to commercial cleaners, 'all-natural' or 'organic' cleaners have been favoured by the yoga community. However, there is very little information on their effectiveness as disinfectants against various bacteria. The goal of this study was to examine the effectiveness of different 'natural' and commercial cleaning products in inhibiting the growth of *Staphylococcus carnosus* when disinfecting yoga mats infected with *S. carnosus*. All cleaners were compared to Lysol® All-Purpose Cleaner, a Health Canada certified disinfectant. We hypothesized that the 'natural' cleaners used on the yoga mats would not be as effective at inhibiting the bacterial growth of *S. carnosus* in comparison to the two commercial cleaners tested. It was found that the commercial cleaner containing sodium hypochlorite (bleach) was significantly more effective than all of the natural cleaning products.

Keywords — Fitness, Yoga Equipment, Cleaners, Disinfectants, Staphylococcus

1. Introduction

Antibiotic resistance in *Staphylococcus aureus* is a major health concern worldwide [1]. In the United States alone, methicillin-resistant *S. aureus* (MRSA) causes over 80,000 infections and 11,000 deaths per year [1]. MRSA can be transmitted via skin contact and through the touching of contaminated surfaces. Roughly one-third of the world's total population carries MRSA on their skin and in their nose [1, 2]. *S. aureus* becomes a threat when it enters the body, as it has the capacity to cause mild to life-threatening diseases and is resistant to most commercially available antibiotics [3]. *S. aureus* is one of the most common bacterial strains found in community fitness centres, and recent studies have found a link between MRSA infections and improperly sanitized yoga mats [1, 2]. Yoga mats are often shared within fitness facilities and between individuals, especially in highly trafficked recreational centers [1]. For most

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athletes, a yoga mat is essential, and usage involves intimate and prolonged mat contact, particularly with the hands and feet, during sweat-inducing activities that contaminate yoga mats and provide a viable mechanism for bacterial transmission [1].

In an attempt to bypass the use of toxic chemicals, 'all-natural' or 'organic' cleaners made with a number of different ingredients including essential oils have been favoured by the yoga community. However, there is insufficient information on their effectiveness as disinfectants [4]. Our study examined the efficacy of two 'natural' cleaners on a yoga mat infected with *Staphylococcus carnosus* in comparison to two commercial cleaners. To our knowledge, no prior studies have investigated bacterial adhesion and 'natural' cleaner efficacy on yoga mats made of polyvinyl chloride (PVC). The first chosen 'natural' cleaner was lemongrass essential oil. Previous studies have demonstrated it has general antibacterial properties [4]. The second was B CLEAN, a 'natural' antibacterial yoga mat spray. The 'natural' disinfectants were tested against Lysol® All-Purpose Cleaner, a Health Canada certified disinfectant [5], and Vim® Power and Shine™ Multi-Purpose Cleaner, a commercial disinfectant containing bleach that is used at Simon Fraser University's Fitness Centre.

Due to its prevalence on most gym surfaces, the genus *Staphylococcus* was selected [6]. We used the *S. carnosus* species, a Biosafety Level 1 classified bacteria. *S. carnosus* was selected because it belongs to the same genus as *S. aureus* but lacks the virulence genes, making it safer to work with for this study. Considering that *S. carnosus* is non-pathogenic, it makes for a suitable model organism to study pathogenic bacterial strains like *S. aureus* [7].

Based on the absence of common disinfectant chemicals present in lemongrass essential oil and B CLEAN [4], our hypothesis was that the effectiveness of the 'natural' cleaners on the yoga mat would not be as successful at inhibiting the bacterial growth of *S. carnosus* in comparison to both commercial cleaners.

2. Materials and Methods

Disinfectants*:

- Lysol® All-Purpose Cleaner (positive control)
- Water (negative control)
- Sage 100% lemongrass pure essential oil
- Vim® Power and Shine™ Multi-Purpose Cleaner
- B CLEAN Mat Spray

The disinfectants were tested at full concentrations because when tested on *S. aureus* they were less effective at diluted concentrations [4], and none of the directions found on any of the product's packaging instructs users to dilute the products. Lysol® was used as a positive control as it is a phenolic compound, which has repeatedly shown to inhibit the growth of many strains of bacteria, including *S. aureus* [8]. Water was used as a negative control.

A diagram of the methods is shown in Fig. 1. To begin, we pipetted 50 µl of *S. carnosus* onto the center of a 5 cm by 5 cm piece of sterile Athletic Works Yoga Mat (3 mm thick, made of PVC) which was placed in a sterile petri dish, and spread the bacterial broth using aseptic technique. We let the piece incubate for 30 minutes at 37°C, to imitate core body temperature and because this is the optimal inoculation temperature for the genus *Staphylococcus* [9]. Afterwards, we placed the mat piece in a petri dish and pipetted 1 mL of disinfectant from a sterile Erlenmeyer flask onto the center of the piece and spread it using aseptic technique. Based on our preliminary experiments, 1 mL was proven to be a sufficient amount to completely cover the surface of the mat. After 30 seconds, a 500 g weight was placed onto the center of a pre-autoclaved 15 cm by 15 cm Mainstays Bar Mop Dishcloth cotton towel and was used to wipe the yoga mat piece in a consistent manner 4 times. Next, we cut the mat piece, with sterilized scissors and forceps, into 9 squares to increase surface area exposure and to allow the remaining *S. carnosus* bacteria on both the surface and absorbed into the mat to be released when placed into a 50 mL falcon tube filled with 15 mL of Trypticase Soy Broth (TSB). We determined that 15-35 mL of TSB would be required for each replicate, as preliminary tests determined that the yoga mat piece displaced 6 mL of solution. We then manually shook the tube for 30 seconds to ensure full surface coverage of the mat pieces. The same person shook the tube each time to ensure consistency. We then performed several serial dilutions on the TSB and mat solution by transferring 0.1 mL of the 1/10 dilution from the falcon tube onto Trypticase Soy Agar (TSA) and spread it using aseptic technique. The whole procedure: Infect/Disinfect/Cut/Serial Dilute/Plate, was replicated 13 times for each disinfectant for a total of 65 replications. The plates were placed in the incubator at 37°C for 2 hours. The number of bacterial colonies were then counted, and the number of colony-forming units per mL (CFU/mL) were calculated. One limitation of this study was that we did not have the ability to confirm that all bacterial colonies found on the PVC yoga mats after incubation were *S. carnosus*. For the purpose of this study, all bacterial colonies found on the PVC yoga mats were assumed to be *S. carnosus*.

The statistical analysis was done using JMP® (Ver. 14. SAS Institute Inc., Cary, NC). We graphed the mean number of recovered bacteria *S. carnosus* with 95% confidence intervals for each disinfectant. We then ran a one-way analysis of variance (ANOVA) test to get the p-value. Lastly, we ran a post-hoc Tukey-Kramer HSD multiple-comparison test (MCT) to get the Connecting Letters Report.

3. Results

The mean number of bacteria present on the yoga mat pieces after disinfection varied between the disinfectants (Fig. 2; $F(4, 55) = 7.1648$, $p = 0.0001$). Vim® Power and Shine™ Cleaner was significantly more effective than lemongrass oil, B CLEAN, and water. It was not significantly more effective than Lysol® All-Purpose Cleaner. Thus, both commercial cleaners did not significantly differ in their ability to inhibit the growth of *S. carnosus*. No significant difference was observed between Lysol® All-Purpose Cleaner and lemongrass oil, nor between lemongrass oil, B CLEAN, and water. There was a significant difference observed between Lysol® All-Purpose Cleaner, B CLEAN,

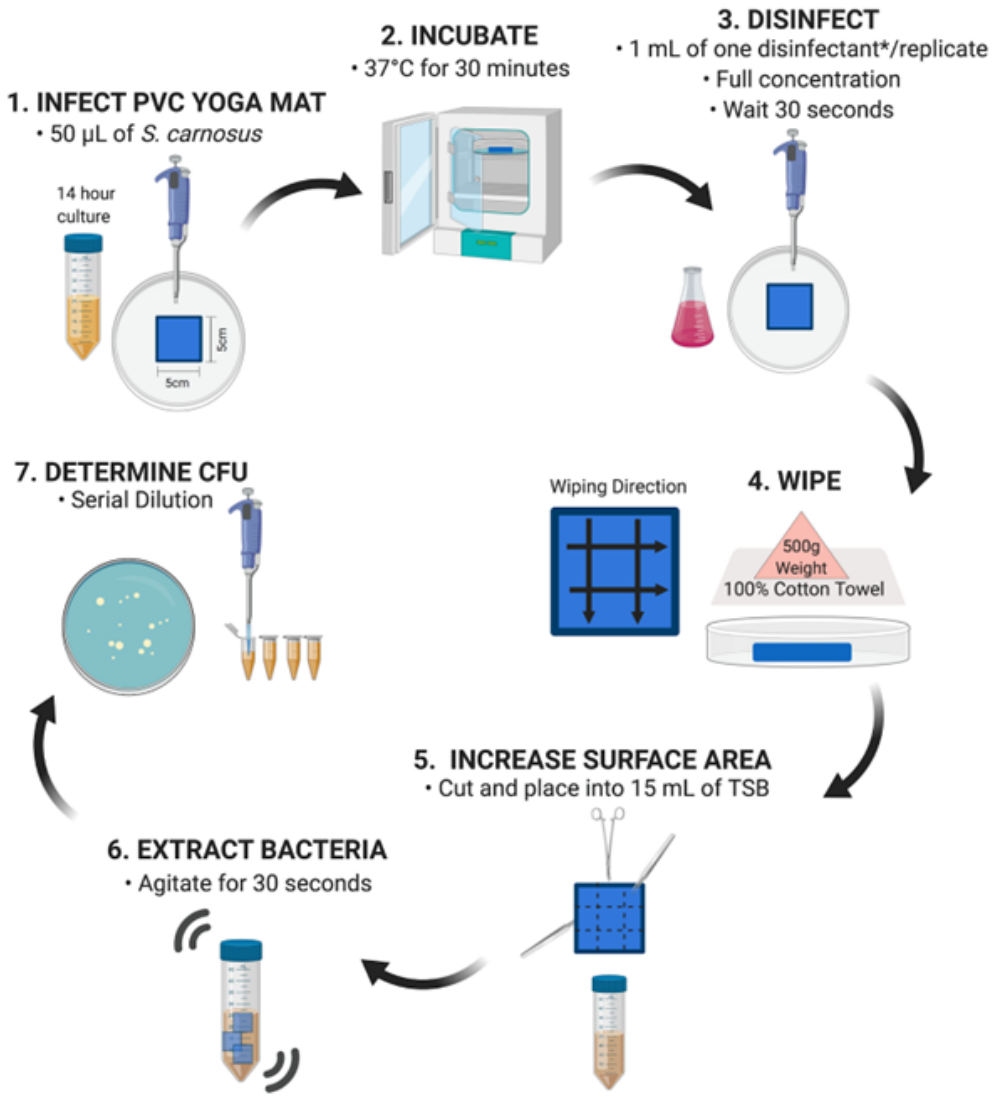

Figure 1: ***Diagram of Methods.*** *1. Infect 5 cm by 5 cm PVC yoga mat with 50 µl of S. carnosus broth. 2. Incubate the infected mat piece. 3. Disinfect with 1 mL of cleaner. 4. Wipe with a flat-bottom 500 g weight placed on a sterile cotton cloth. 5. Cut mat 4 times into 9 pieces. 6. Place pieces into Trypticase Soy Broth and shake. 7. Perform serial dilutions and plate on Trypticase Soy Agar plates. Image created with BioRender.com.*

and water. Water, the negative control, allowed for the most growth of *S. carnosus*. The cleaning agent that claimed to disinfect surfaces that allowed for the most growth of *S. carnosus* was B CLEAN. Vim® Power and Shine™ Cleaner was the most effective at inhibiting the growth of *S. carnosus* on PVC yoga mat (Fig. 2).

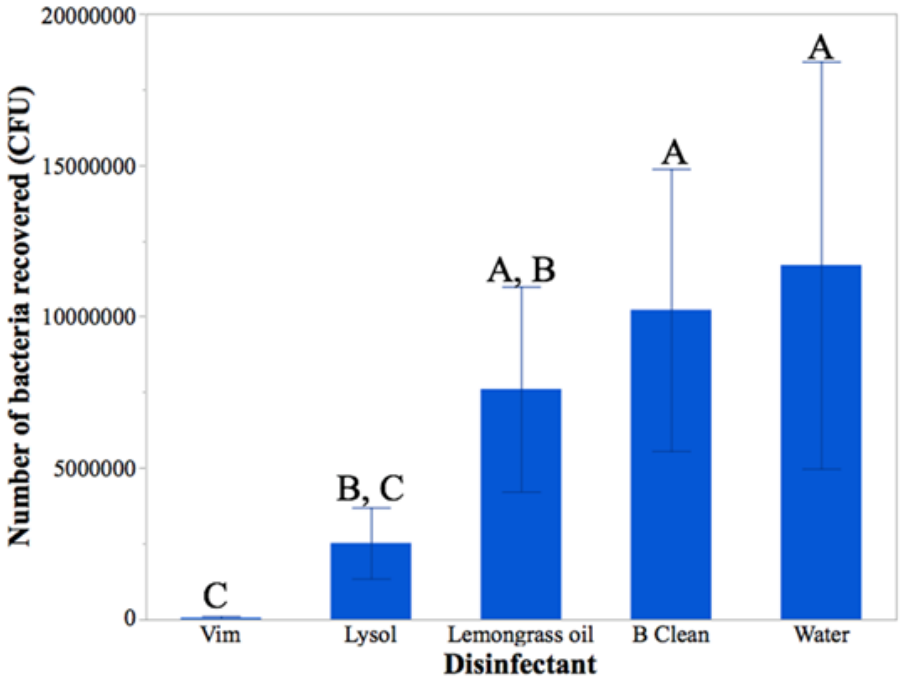

Figure 2: *Mean number of Staphylococcus carnosus present (CFU) on yoga mat after incubation for 30 minutes at 37°C and disinfection with Vim Power and Shine, Lysol All Purpose Cleaner, Saje Lemongrass Oil, B Clean Mat Spray, and Water. Mean disinfectant bars that do not share a letter significantly differ in disinfecting ability according to Tukey's HSD. Error bars represent 95% confidence intervals,* $\alpha = 0.05$.

4. Discussion

Our hypothesis was that the effectiveness of the 'natural' cleaners on the yoga mat would not be as successful at inhibiting the bacterial growth of *S. carnosus* in comparison to both commercial cleaners. The results differed from what we anticipated and contrast with the findings of relevant past studies. For instance, a previous study investigating the general effectiveness of 'natural' cleaners against *S. aureus* found no 'natural' cleaner to be more effective than Lysol® All-Purpose Cleaner [4]. Again, we hypothesized that the 'natural' cleaners would be less effective than the commercial cleaners due to their lack of common disinfectant chemicals [4]. Lysol® All-Purpose Cleaner contains phenolic compounds and ammonium chlorides, such as benzalkonium chloride, which are responsible for its strong disinfecting abilities [4, 5, 6, 7, 8, 9, 10]. However, our results suggest that Lysol was equally effective as lemongrass oil.

The disinfectant properties in Cymbopogon citratus (lemongrass) can be attributed to its chemical makeup [11]. A past study determined that the main components present in the leaves of lemongrass were hexadecenoic acid (Palmitoleic acid), hepta-9,10,11-trienioc acid, octadecenoic acid (Stearic acid), 2-ethenyltradecan-1-ol, eicosane aldehyde and 1-ethoxyoctadecane [11]. These phytocompounds are believed to exhibit

a wide range of functions including antibacterial, antiviral, and antifungal abilities [11]. Lemongrass oil should be further studied with regards to its ability to inhibit the growth of bacteria, including *S. carnosus* and *S. aureus*.

We were surprised to find that Lysol® All-Purpose Cleaner, our positive control, was equally as effective as Vim® Power and Shine™ Cleaner. This could be a result of Vim's primary active ingredient, sodium hypochlorite (bleach), which is commonly known for its antibacterial properties [12]. B CLEAN was found to be equally as effective as water, despite claiming to prevent bacterial growth, and it is significantly more expensive than water [13].

Overall, our data suggests that either of the commercial cleaners tested in this study could be effective at inhibiting the bacterial growth of *S. carnosus* on yoga mats. However, further research is needed to determine how variables such as contact times for the disinfectants, incubation times and temperatures, humidity levels, types of bacteria, and different mat materials can affect the cleaners' ability to eliminate pathogens. From this list, the further testing of disinfectant contact time could be particularly important. We speculated that most people spray and then immediately wipe their mats, so even our contact time of 30 seconds could be seen as generous if trying to mimic real life. Studies should also perform Minimum Inhibitory Concentration tests and determine the exact composition of each disinfectant – the precise concentrations of each ingredient contained within each bottle and each droplet – and explore the different combinations of essential oils and other natural ingredients in order to achieve maximum effectiveness against *S. carnosus* [4].

Although the results from this experiment did not support our hypothesis, in combination with results of previous external experiments, it revealed that these natural products do possess some potential to function as antibacterial disinfectants for yoga mats. As fitness becomes more popular, especially in the community setting, a need for effective sanitizing agents for yoga mats and fitness equipment will be in higher demand. Accordingly, the demand will also increase the production of alternative 'all natural' products because their nature suits the practice and lifestyle of yoga well. Future studies must be conducted to determine whether a 'natural' disinfectant, as successful as a chemical agent like Lysol® All-Purpose Cleaner or Vim® Power and Shine™ Cleaner, can be created for safe and effective use on yoga mats in gyms and in yoga studios.

5. Conclusion

This study yielded results consistent with other studies that have evaluated the antimicrobial activity of natural ingredients, but to our knowledge, this study was the first to test various disinfectants on PVC yoga mats [14]. Further research is needed to better comprehend how disinfectants work on PVC yoga mats. Different concentrations and combinations of essential oils and natural ingredients, as well as contact time before wiping, could all be investigated more extensively [4]. Further research on the antibacterial properties of essential oils, such as lemongrass oil, should also be carried out in order to appeal to the fitness community with regards to surface cleaners. Equipment specific studies, such as this one, are important to help determine the most

effective ways to reduce bacterial transmission in fitness facilities.

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