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Prevalence and Disinfection of Bacteria Associated with Various Types of Wristbands

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Abstract

The potential role of personal items in the transmission of pathogens is poorly understood. In this study, we cultured bacteria of public health importance found on wristbands, determined whether there is a correlation between wristband material and prevalence, and tested three household disinfectants for efficacy in reducing bacteria on wristbands made of plastic, rubber, metal, and cloth, using standard microbiological assays. Total cultivable bacteria, Staphylococci, *Enterobacteria* (*Escherichia coli*), and *Pseudomonas* on 20 smartwatch wristbands were cultured from randomly recruited subjects. Nearly all wristbands (95%) were contaminated, with the highest average numbers of 3.46E+4 cfu/cm^2 and 1.52E+4 cfu/cm^2 on rubber and plastic bands respectively. Metallic gold and silver wristbands had zero to 18 cfu/cm^2. While the high prevalence of *Staphylococcus spp* (85% of wristbands)—skin microbiota; was not unexpected, the occurrence of *Pseudomonas spp* (30%), and enteric bacteria (60%), even at relatively low numbers is of public health significance. Bacterial load on individual subjects varied remarkably with males and females harboring average total bacteria of 4.045 and 3.42 $\log_{10}$ cfu/cm^2 of wristband, respectively. The most important predictor of wristband bacteria load was the texture of wristband material and activity (hygiene) of the subject at sampling time. Potential pathogens—*Staphylococcus aureus* (8143 cfu/cm^2) and *Pseudomonas spp*. (1126 cfu/cm^2) were most abundant on cloth and rubber wristbands, respectively, while the presence of the *E. coli* group was associated with animal handling activity by a veterinarian. Lysol Disinfectant Spray and 70% Ethanol were highly effective regardless of wristband material with >99.99% kill rate and a log cfu/cm^2 reduction of 3 - 4.0 and 3 - 4.5 respectively within 30 seconds. Apple Cider Vinegar (ACV) was not as potent. Only 2 - 3.5 log cfu/cm^2 drop was obtained after 120 seconds of exposure. Further susceptibility assays with standard reference

bacteria showed that Lysol and 70% alcohol effectively killed > 99.99% (>8 log CFU drop) of *Escherichia coli* strain 7001, *Staphylococcus aureus* strain 6538, and *Pseudomonas aeruginosa* strain 10662 within 30 seconds of contact. Vinegar had a similar efficacy on the gram negatives but little or no effect on *Staph aureus* (only a 2-log CFU/ml reduction in 5 minutes!) The high prevalence of potential pathogens, some of which could be reservoirs of antibiotic resistance reveals a weak link in infection control and underscores the need for regular cleaning of personal and hand-held accessories with adequate considerations of their texture.

Keywords

Apple Cider Vinegar, Disinfectants, *Escherichia coli*, Lysol *Pseudomonas aeruginosa*, Public Health, *Staphylococcus aureus*, Wristbands

1. Introduction

The human skin serves not only as a barrier between the body's interior and the environment, but also as a habitat for a variety of beneficial organisms, especially bacteria. Bacterial symbiosis with humans may fall anywhere on a continuum between mutualism and parasitism, depending on several variables, including biochemical and virulent attributes of the symbiont and immune system status of the host. When these interactions are parasitic, however, bacteria can pose a serious direct threat to human health. Even the normal microbiota has been shown to serve as reservoirs of antibiotic resistance.

Because of these dangers, routine sanitation remains an important preventative tool for bacterial infections. Wristbands associated with wrist devices, such as smartwatches and fitness trackers, are often worn during activities—while sweating, swimming, holding animal pets or even sleeping. However, they are often not sanitized regularly, and can thus be an important fomite for bacterial transmission and infection. Previous studies have shown that wristwatch use increases quantities of bacteria retained on the hands of hospital workers [1]. As a result, they can become an easy route for nosocomial spread of infection in hospitalized people who are often more susceptible to infectious diseases. Wristbands, then, could potentially put both patients and healthcare workers at risk without regular sanitation. In the general community, touching contaminated wristbands could also lead to infection.

Skin-associated bacteria include Corynebacterium, Propionibacterium *Staphylococcus epidermidis* and *S. aureus*. The latter is known to act as a human pathogen, causing boils, sepsis, pyomyositis, botryomycosis, and endocarditis, among others. Additionally, some strains are resistant to antibacterial agents (e.g., methicillin-resistant *Staphylococcus aureus*, or MRSA), which complicates treatment [2]. Other common human skin residents include those of the genus *Pseudomonas*, especially *P. aeruginosa*. This opportunistic pathogen commonly

causes wound and burn infections as well as pneumonia and urinary tract infections (UTIs) associated with a urinary catheter [3]. Enteric bacteria, including those of the genera *Enterobacter*, *Klebsiella*, and *Escherichia* (like *E. coli*), which normally inhabit human intestines, can also act as opportunistic pathogens when exposed to extraintestinal environments. *Klebsiella*, especially *K. pneumoniae*, infects immunocompromised subjects, causing pneumonia, sepsis, and UTIs when inadvertently transferred from the gastrointestinal tract [4]. *E. coli* has similarly been found to cause sepsis, peritonitis, meningitis, and UTIs [5].

These potential health risks would suggest that routine cleaning of wristband surfaces can prove useful as an infection preventive measure. However, the extent of contamination of wristbands is not known. In addition, the material of the wristband could affect both bacteria present and the efficacy of cleaning and disinfection. Different disinfectants, depending on their active ingredients, kill bacteria in different ways, such as by disrupting cell membrane integrity, denaturing proteins, or interfering with metabolic activities [6].

Routine cleaning of wristbands is commonly ignored due to perceived lack of need and ignorance of the sanitation process. In this project we examine the hygienic state of various types of wristbands worn by active individuals and determine the best protocols for their proper disinfection. Bacterial counts, identity, and their distribution on various types of wristband surfaces are determined, followed by a bacteria susceptibility assay study focused on screening the effectiveness of three different disinfectant solutions—(Lysol Disinfectant Spray, which received the Emergency Use Authorization by the United States Environmental Protection Agency (EPA) to combat the SARS-CoV-2 [7]; 70% ethanol, which is commonly used in hospitals and research facilities and in alcohol wipes; and apple cider vinegar, which is often used in household cleaning products)in killing bacteria associated with wristbands of various texture.

2. Materials and Methods

2.1. Prevalence of Bacteria of Public Health Importance on Wristbands

2.1.1. Sample Collection

Twenty wristband swab samples were randomly collected (by co-author—MC of WPTV with an IRB waiver from Florida Atlantic University) from the wristbands of anonymous volunteer participants. No specific inclusion or exclusion criteria were adopted for the study. Volunteers actively engaged in their daily routines were recruited and sampled on the spot. **Table 1** presents the demographics of participants and types of wristbands worn as well as their activity at the time of sampling. In all, the wristband materials were various metallic (5), rubber (5), plastic (5), cloth (3) and leather (1). A sterile cotton swab lightly soaked with 1X phosphate-buffered saline (PBS) was used to swab an area of 1 cm^2 from each of the twenty wristbands. Each cotton swab was then placed into a sterile 15 mL falcon tube containing 1 mL sterile PBS and sent to the lab on ice for processing within 2 hours.

Table 1. Volunteer subjects, type of wristband worn and their activity at the time of sample collection.

Subject #	Age	Gender	Ethnicity	Activity	Wristband Material
1	Adult	Male	White	Working at desk	Metal
2	38	Female	Black	Arriving at work	Metal (Silver)
3	40	Male	White	Sitting at desk	Metal (Gold)
4	65	Female	White	Collecting samples	Rubber
5	29	Male	White	Working with video equipment	Cloth
6	28	Male	White	Driving	Metal
7	36	Male	White	Working in office	Rubber
8	30	Male	White	Firefighter, Assistant Chief	Metal
9	62	Female	White	Firefighter	Plastic
10	46	Male	White	Firefighter	Plastic
11	26	Male	White	Firefighter	Cloth
12	47	Male	White	Firefighter	Plastic
13	24	Male	Hispanic	Firefighter	Plastic/Rubber
14	35	Female	White	CrossFit	Cloth
15	39	Male	White	CrossFit	Plastic
16	33	Female	White	CrossFit	Rubber
17	25	Female	White	CrossFit	Rubber
18	19	Male	Hispanic	Vet doing surgery	Leather
19	50	Female	White	Vet assistant with dog	Rubber
20	31	Female	White	Vet working	Rubber

2.1.2. Enumeration of Total Bacteria Counts, Pseudomonads, Staphylococci and Enterobacteriaceae

After a brief vortex to release the bacteria on the swabs, a 10-fold serial dilution was carried out using sterile PBS in microfuge tubes. Aliquots were top spread onto Trypticase Soy Agar (TSA) for total viable counts, Mannitol Salt Agar (MSA) for Staphylococci, Pseudomonas Isolation Agar (PsA) for Pseudomonads, and MacConkey Agar (Mac) for Enterobacteriaceae/*E. coli* in duplicates. The plates were incubated at 37˚C for 24 - 48 hours and counted. Colonies characteristic of bacteria groups of interest were counted and recorded for each plate.

Mannitol Salt Agar (MSA) Assay: Mannitol Salt Agar is a selective and differential medium that inhibits the growth of most gram-negative and some gram-positive bacteria through its high salt content, selecting staphylococci. This medium also differentiates between mannitol-fermenting staphylococci, like *S.*

aureus, which changes the color of the medium from red to yellow, and other staphylococci, which produce red or pink colonies with no color change in the medium.

Pseudomonas Isolation Agar (PsA) Assay: Pseudomonas Isolation Agar is a selective medium for the isolation of *P. aeruginosa*. The medium induces production of the secondary metabolite pyocyanin in *P. aeruginosa*, which inhibits the growth of other organisms such as *S. aureus* [8].

MacConkey Agar (Mac) Assay: MacConkey Agar is a selective and differential medium that inhibits growth of gram-positive bacteria, selecting for enteric bacteria. Additionally, due to the pH indicator neutral red, lactose-fermenting bacteria, like *Enterobacter*, *Klebsiella*, and *Escherichia* produce mucoid, pink colonies, in contrast to other enteric bacteria, such as Shigella and Pseudomonas which produce colorless colonies [9].

Further identification of representative characteristic colonies on the selective differential agar was accomplished by microscopy and classic microbiological procedures.

2.2. Screening of Household Hygiene Products for Antibacterial Efficacy

2.2.1. In-Use Assay for Disinfecting Wristbands

Freshly prepared solutions of common commercial disinfectant—Lysol Disinfectant Spray (79% ethanol, 0.1% alkyl (50% C_{14}, 40% C_{12}, 10% C_{16}) dimethyl benzyl ammonium saccharinate, undiluted), 10% Butane and 1% - 5% Propane; lab made 70% Ethanol solution, and Heinz Apple Cider Vinegar (acetic acid diluted with water to 5% acidity) were employed in ways that simulated actual use (wiping/cleaning) scenarios. Phosphate buffered saline (1X PBS) contained 137 mM NaCl, 2.7 mM KCl, 8 mM Na_2HPO_4, and 2 mM KH_2PO_4. Actively worn wristbands—rubber, cloth, metal, and plastic were employed in duplicates for the study (**Figure 1**). To determine the bacteria load on the wristbands before disinfection (zero time), PBS-moistened cotton swabs were used to swab 1 square cm of each of the eight wristbands. They were then vortexed in PBS (to dislodge bacteria) followed by a ten-fold serial dilution in microfuge tubes. Aliquots of 100 ul were plated onto and an all-purpose medium—Tryptic Soy Agar in duplicates and incubated at 37˚C for 24 - 48 hours. For efficacy at 30 seconds of exposure, 1 sq cm custom-made disinfectant discs made from sterile absorbent thick paper were prepared for each disinfectant/apple cider acetic acid and placed on the wristbands for 30 seconds and removed. Phosphate-buffered saline swab samples were then taken from the cleaned surfaces and processed as in zero-time samples above. The exercise was repeated on successive sq cm areas on a given wristband for exposure times of 30 and 120 seconds. At each exposure time point, the original PBS swab was plated as control for the corresponding wristband. The difference between the average colony forming units (CFU/cm^2) computed for the control and treatment (disinfectant and exposure time) is reported as the log cfu reduction values. The data for the metal wristbands were

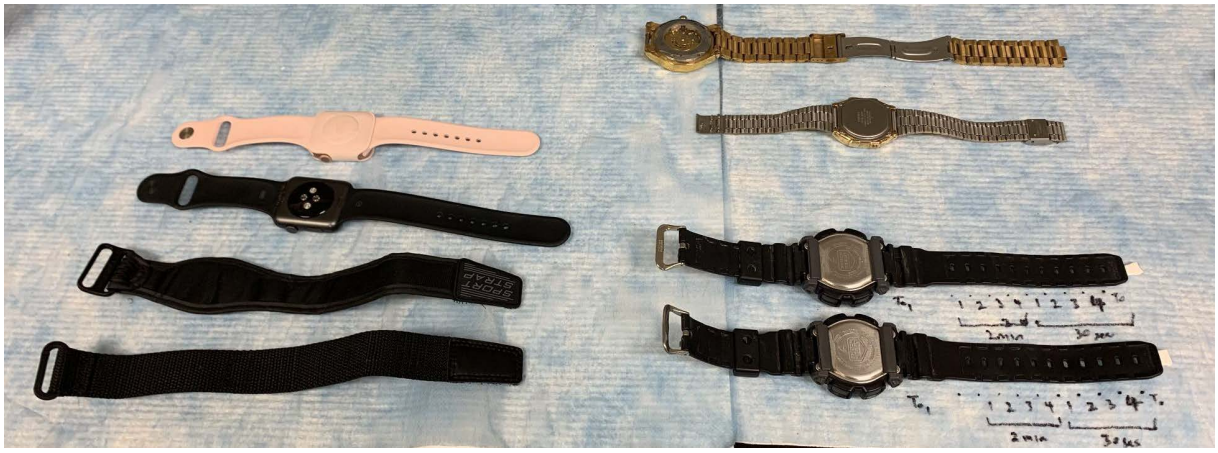


Figure 1. Wristbands of various textures were used in the experiment (rubber (top left), cloth (bottom left), metal (top right), and plastic (bottom right)).

not included in data analysis.

2.2.2. Antibacterial Efficacy of Household Disinfectants on Reference Bacteria Using Quantitative Dilution Assay

Escherichia coli strain 7001, *Staphylococcus aureus* strain 6538, and *Pseudomonas aeruginosa* strain 10662 were purchased from Microbiologics (St Cloud Minnesota, USA). Lyophilized pellets were aseptically inoculated unto tryptic soy broth (TSB) and incubated for 24 hours; followed by a subsequent subculture of confirmed axenic isolates into TSB for 4 - 5 hours at 37˚C. Once in log growth-phase, aliquotes were used for the susceptibility assay while the original population of each bacterium was determined simultaneously by viable plate count techniques after serial ten-fold dilutions and plating of TSA.

For the germicidal treatment, a 100 µL aliquot of the log-phase bacteria was added to 900 µL of each disinfectant (Lysol Disinfectant Spray solution, 70% Ethanol, Vinegar), and PBS as control. They were thoroughly mixed, and after each time interval (30 seconds, 2 minutes, and 5 minutes), 100 µL was removed and rapidly serially diluted in 1X PBS, then spread-plated onto TSA. The plates were incubated at 37˚C for 24 - 48 hours, after which bacterial colonies were counted. Colony forming units per mL in the control (PBS tubes) minus the colony forming units in each disinfectant for a given exposure time equals the killed population. A $\log_{10}$ cfu value of this difference over the exposure time is the log reduction rate.

3. Results

3.1. Prevalence of Bacteria on Wristbands

Total Bacterial Counts: **Figure 2** displays the relative densities of total heterotrophic plate counts recovered from various wristbands. Viable bacteria varied widely between subjects and even wristbands made of the same materials. On the average however, the trend of bacteria load was cloth ≥ plastic ≥ rubber ≥ leather > metal. The lowest total bacterial counts were from a metal (gold) wristband,

with 0 cfu/cm^2.

Staphylococcal Bacterial Counts: **Figure 3(a)** displays the occurrence of viable staphylococcal plate counts for the wristbands. No significant differences (P = 0.05) were noted on the average because of high variability of staph counts between subjects and within wristband materials; except for the metallic bands

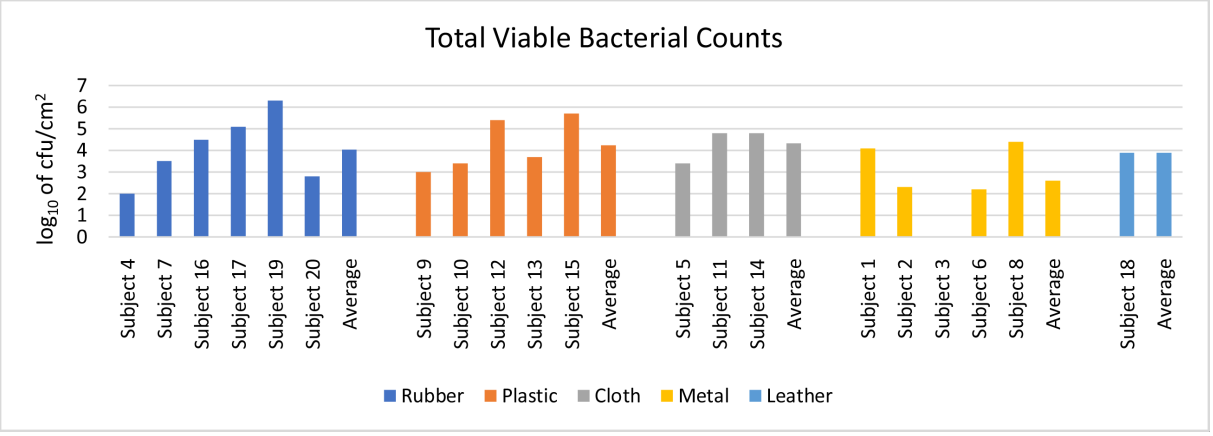
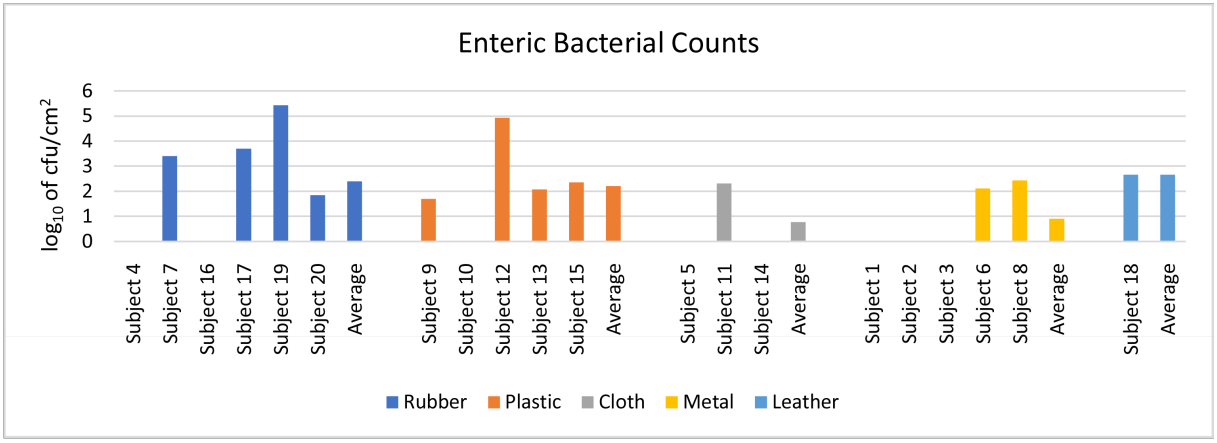
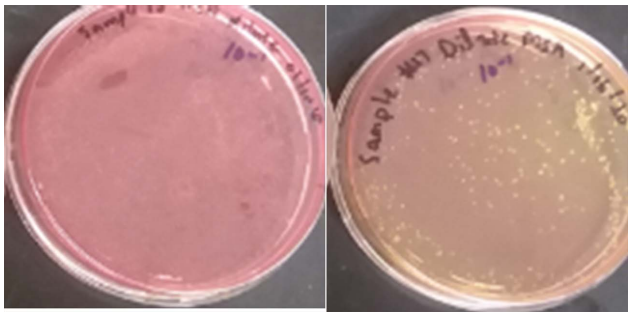


Figure 2. Total counts of bacteria ($\log_{10}$ cfu/cm^2) recovered from the surface of various types of wristbands. Subjects are grouped according to wristband material.



(a)



(b)

Figure 3. (a) Staphylococcal bacterial counts on MSA ($\log_{10}$ cfu/cm^2) from wristbands of 20 randomly recruited subjects grouped according to type of wristband worn. (b) Mannitol Salt Agar after 30 hrs of incubation, showing Subject #15—no *Staph aureus* (left) and *Staph aureus* positive Subject 17 (right).

which recorded significantly lower levels of bacteria. The value of the leather wristband relies on a single subject, and so may not be representative. Mannitol-fermenting staphylococci, such as *S. aureus*, were identified on plates as distinctly yellow colonies (Figure 3(b)) and confirmed microscopically after catalase +ve test.

Bacterial Counts: No viable *Pseudomonas* bacteria were identified from any of the cloth, leather, and metal wristbands except for Subject 8 metallic band. The average log cfu for rubber bands and plastic were similar (Figure 4). Most wristbands did not appear to have cultivable *Pseudomonas*, with only six samples (three rubber, two plastic, and one metal) out of 20 showing growth on the selective-differential agar.

Enteric Bacterial Counts: Gram negative, oxidase negative lactose fermenting rods on the MacConkey agar assays showed highly elevated levels of enteric bacteria obtained for Subject 19 (26,760 cfu/cm^2), a veterinary assistant wearing a rough rubber wristband and for subject #12, a firefighter's plastic wristband (Figure 5). Several samples (eight of the twenty) did not contain any enteric

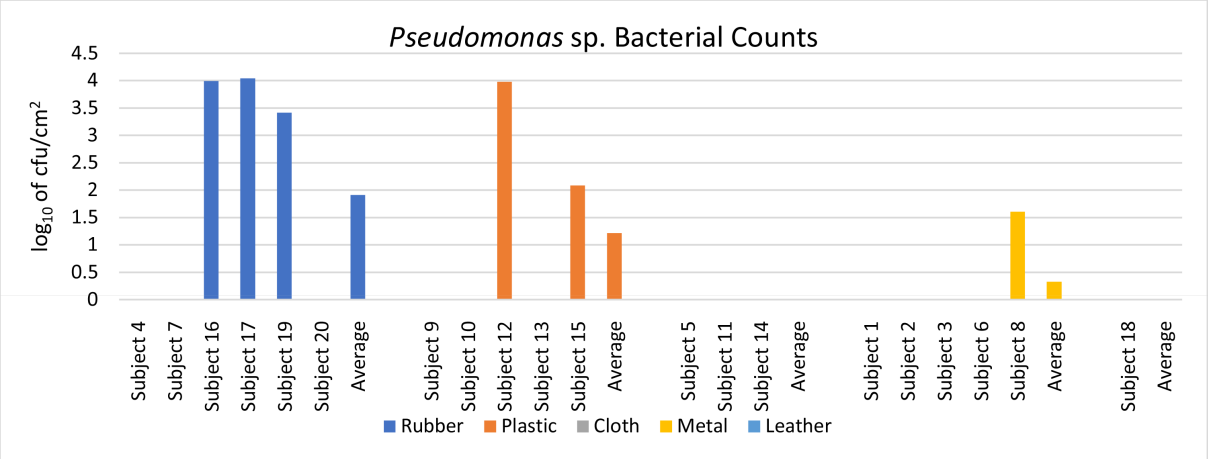


Figure 4. Prevalence of *Pseudomonas* bacteria ($\log_{10}$ cfu/cm^2) on 20 randomly recruited subjects grouped according to texture and type of wristband.

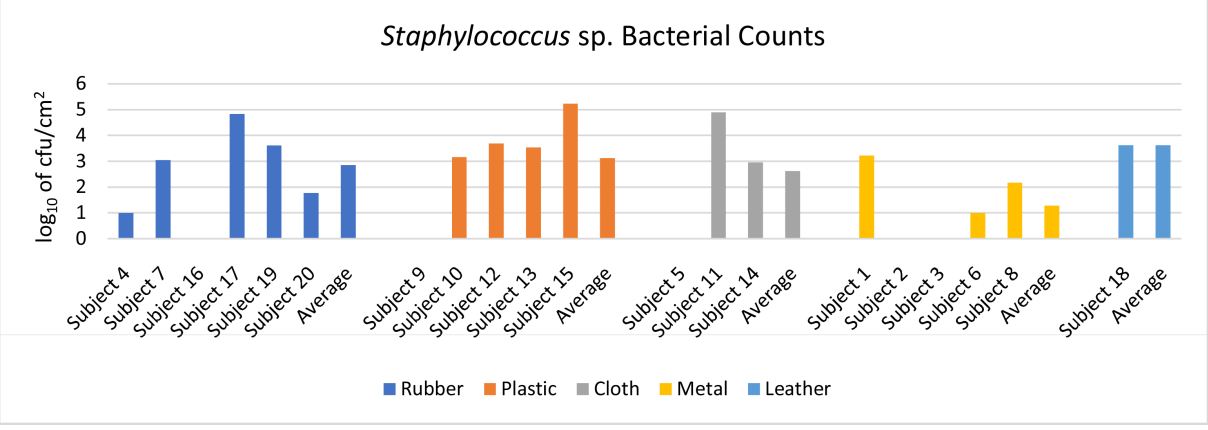


Figure 5. Population of enteric bacteria ($\log_{10}$ cfu/cm^2) from different types of wristbands from 20 randomly recruited subjects grouped according to wristband material.

bacteria. Wristbands made of rubber, plastic and to a lesser extent, metals harbored the most enteric organisms.

Overall Microbiology of the Wristband: Figure 6 presents the relative density of various bacteria assayed and the total heterotrophic counts in the context of gender and wristband matrix and gender of the subjects. Total heterotrophic bacteria counts associated with wristbands on male subjects (N = 12) were higher (4.045 $\log_{10}$ cfu/cm^2) than that of females (N = 8) at 3.422. There were no significant gender differences in the distribution and occurrence of the bacteria groups.

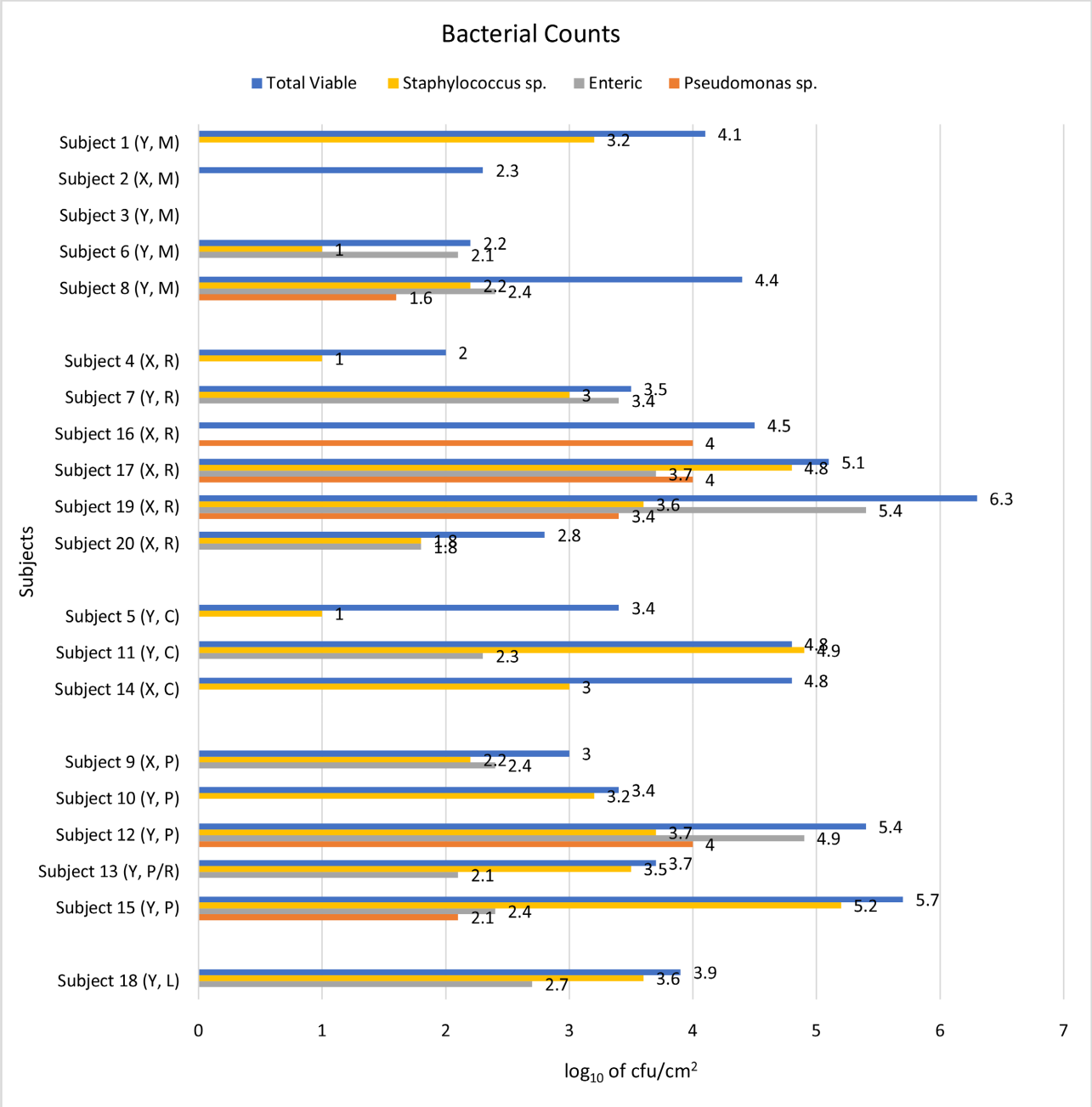


Figure 6. Total counts of bacteria ($\log_{10}$ cfu/cm^2) for Subjects 1 through 20. Gender of subject (X for female, Y for male) and wristband material (M for metal, R for rubber, C for cloth, P for plastic, and L for leather) are denoted in the parentheses.

3.2. Screening of Household Hygiene Products for Antibacterial Efficacy

3.2.1. In-Use Assay for Disinfecting Wristbands

Custom-made disinfectants patches measuring 1 cm^2 were prepared for Lysol spray, 70% alcohol and apple cider and placed on the duplicate representatives of the cloth, metal, plastic, rubber and leather wristbands. Patches of phosphate buffered saline were used as control. After 0, 30 and 120 seconds of exposure, the patches were assayed for viable bacteria load.

Across wristband materials, all tested disinfectants showed effectiveness where Lysol Disinfectant Spray, 70% Ethanol, and Heinz Apple Cider Vinegar reduced viable bacterial counts to 0 cfu/mL within 30 seconds on all except plastic wristbands. A longer exposure time of 120 seconds was required to achieve the same kill rate for the plastic wristbands.

For plastic wristbands, none of the disinfectants exerted a significant reduction in bacteria load in the first 30 seconds (Figure 7). At 120 seconds of exposure time, all the bacteria were killed, emphasizing the need for increased contact time for proper disinfection of rough, porous and adhesive surfaces. For all the other wristband materials, a greater than 3 log reduction was accomplished in 30 seconds (Figure 7).

3.2.2. Efficacy of Household Hygiene Product against Reference Bacteria Strains

To validate the results obtained from direct testing on the wristbands, we examined

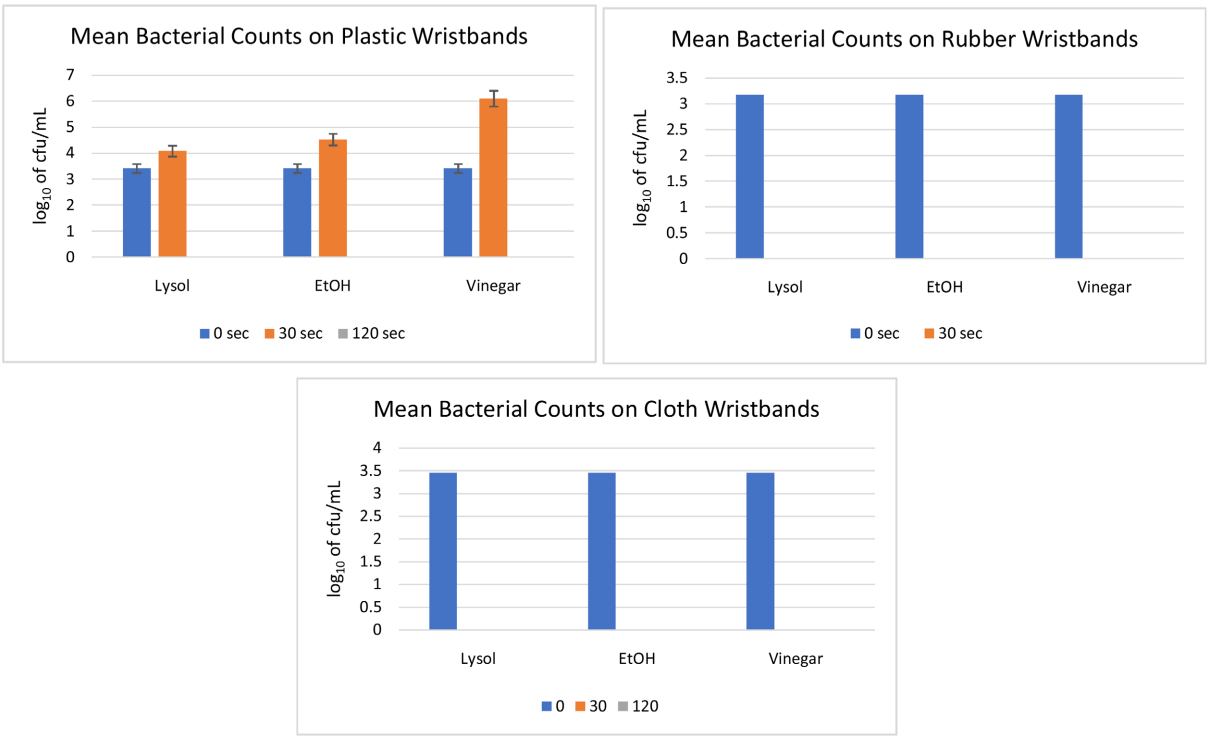


Figure 7. Mean viable heterotrophic bacterial counts (log_{10} cfu/mL) on various wristbands treated with Lysol, EtOH and Vinegar over time showed greater than 3 log bacteria reduction in all but plastic bands within 30 seconds.

the potency of the cleaning agents employed on known standard referenced bacteria. The disinfectants again showed high inactivation rate. Within 30 seconds, viable *Escherichia coli* (strain 7001) and *P. aeruginosa* counts (strain 10,662) were reduced from 10 - 100 million cfu/mL to 0 cfu/mL for all three disinfectants. Only Lysol and EtOH exerted similar kill rates for the *S. aureus* (6538). The Apple Cider Vinegar appeared to only induce a 1.5 log-reduction in *S. aureus* viable counts after 300 seconds (Figure 8). Curiously, no viable bacteria were obtained after 30 seconds, yet colony forming units were retrieved after the 120 seconds and 300 seconds exposure times. Is viable non-cultivable mode a transient resistance mechanism to vinegar by staphylococci?

4. Discussion

4.1. Identified Bacteria

Bacteria identified in the initial study were *Staphylococci* and *Pseudomonads* and Enterobacteriaceae (enteric bacteria). Staphylococcal bacteria likely included *Staphylococcus epidermidis*, a common resident of the skin, identified through the MSA assay, microscopy and catalase. *S. epidermidis* is a member of the commensal or mutualistic microbiota of humans. It prevents colonization by dangerous related pathogenic bacteria. *S. epidermidis* induces host production of antimicrobial proteins (AMPs), which selectively act against harmful bacteria

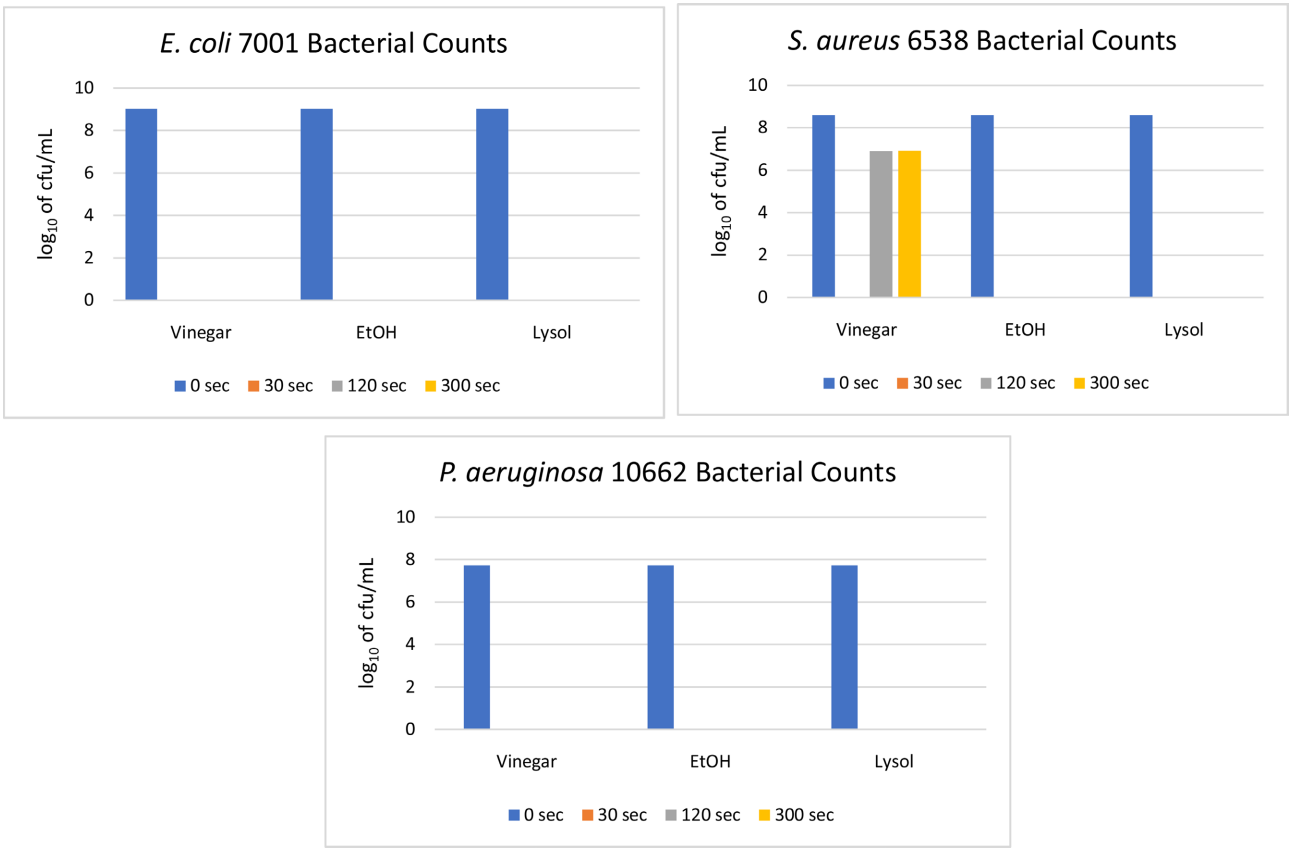


Figure 8. Average counts of *E. coli*, *Staph aureus* and *P. aeruginosa* ($\log_{10}$ cfu/mL) exposed to Lysol, EtOH and Vinegar over time.

like *S. aureus* [10]. However, *S. epidermidis* also acts as a significant opportunistic pathogen, frequently spread through the use of medical indwelling devices [11]. In the United States alone it is estimated that $2 billion is spent annually on blood-related *S. epidermidis* infections [12]. *S. aureus*, which was found on Sample 17, can pose a significant threat to human health under certain conditions. *S. aureus* has been shown to infect the cardiovascular system where its coagulase enzyme leads to blood-clotting issues, causing infective endocarditis or thrombophlebitis and, eventually, organ failure and death [13]. The highest staphylococcal bacterial counts were found on the plastic wristbands, followed by cloth, rubber, and leather which are common band materials.

Pseudomonas species, including *P. aeruginosa*, is a common skin resident, Gram-negative, aerobic bacteria. *P. aeruginosa* also acts as an opportunistic pathogen, targeting cancer and cystic fibrosis patients, invading burnt tissues, and causing a variety of health complications, including urinary tract infections (UTIs) [14]. This is due to its production of leucocidins which reduces polymorphonuclear leukocyte (PMNs) and other virulent factors [15]. *P. aeruginosa* was identified on the plastic and rubber wristbands, but not on the cloth, metal, and leather materials.

The family Enterobacteriaceae, specifically the genera Enterobacter, Klebsiella and Escherichia, consist of gram-negative, facultative bacteria found in the intestines, but have been reported to cause health problems in extraintestinal environments [16]. *Enterobacteriaceae* appeared on the Mac assay as pink, mucoid colonies. The findings included the identification of *Escherichia coli*, which most commonly begins infection through fecal-oral transmission. *E. coli* can also cause health problems including gastroenteritis and UTIs [17]. *Klebsiella* infections may also pose health concerns, including pneumonia and UTIs [14]. Our results found the highest counts of these bacteria on the rubber and plastic wristbands.

Gender and Microbial Load on Wristbands

In our study, wristbands were obtained from 12 males and 8 females. Although this sample size is not large enough to draw definitive conclusions, trends indicated greater cfu/person for wristbands from women: 267,470 vs 72,622 for males. However, this data is strongly skewed by Subject 19 (possibly contaminated by animal products), which showed a count almost four times larger than the next highest, Subject 15. When this outlier is excluded, the overall trend for total heterotrophic cfu/person was lower for females than males (**Figure 6**).

In terms of activity, 7 subjects were working at a news station, 6 were firefighters, 4 were at the gym, and 3 were working with animals at a veterinary office. The largest counts of fecal bacteria (such as *E. coli*) and total bacteria were observed from Subject 19, a veterinarian's wristband, while the largest count of *Pseudomonas* bacteria was observed on Subject 17, who was at the gym. Interestingly, the wristband from Subject 17 also displayed signs of *S. aureus*. Subject 15, another gym-goer, showed the highest staphylococcal counts. These results

emphasize the necessity of wristband sanitation after working with animals or engaging in rigorous activity at the gym or at home.

4.2. Effect of Wristband Materials

Samples were obtained from rubber, plastic, metal, cloth, and leather wristbands. The highest total counts were found on rubber and plastic wristbands, while the lowest was from metal and rubber wristbands. Plastic and rubber wristbands may provide a more appropriate environment for bacterial growth; many plastics have been shown to act as substrates for *S. aureus*, *S. epidermidis*, *P. aeruginosa*, and *E. coli* [18]. Porous and static surfaces tend to attract and be colonized by bacteria.

Metal wristbands overall did not show high bacterial counts; the lowest was from Sample 3, a gold wristband, with 0 cfu/cm^2. This material's anti-bacterial properties may come from its ability to create nanocages that fragment bacterial DNA and perforate membranes, thus inactivating bacteria [19]. Conversely, *S. epidermidis* shows enhanced bio-adhesion on stainless steel than on a biological skin model substrate, although *S. aureus* showed the opposite result [20]. Several studies [21] [22] [23] have shown that the material composition of a surface determines its wettability and surface tension, affecting the potency of different disinfectants. Bacteria adhesion to surfaces varies greatly between strains and gram reactions of species; and directly affects the efficacy of disinfection [21]. Adhesion is determined by surface roughness, hydrophobicity and surface free energy. Previous studies [22] have reported a positive correlation between surface hydrophobicity (surface wettability) and adhesion of *Staphylococcus aureus* and *epidermidis*. Tight adhesion to hydrophobic surface could explain the longer exposure time needed to inactivate bacteria on plastic wristbands (**Figure 7**) as well as the relatively high prevalence of commensals on rubber and plastic wristbands which are synthetic hydrophobic polymers.

Disinfectants employed in the follow-up study on the plastic wristband reduced bacterial counts to 0 after two minutes of application which is within prescribed exposure recommendations.

Lysol Disinfectant Spray, 70% Ethanol, and Heinz Apple Cider Vinegar were tested on the wristbands of different materials. PBS was used as a control because it lacks antimicrobial properties and prevents osmotic lysis unrelated to chemical disinfectants. All disinfectants tested reduced bacterial counts to 0 cfu/mL within 30 seconds for cloth and rubber wristbands, whereas a full 2-minute exposure was required for apple cider vinegar (ACV) to reduce bacterial counts to 0 cfu/mL on the plastic wristbands. The similar trends of Lysol Disinfectant Spray and 70% ethanol in this study may be because the most significant active ingredient in Lysol Disinfectant Spray is also 79% ethanol plus other reducing agents. They inactivate bacteria by dissolving the membrane, disrupting cellular respiration and denaturing proteins. Apple cider vinegar showed different trends, generally as being less effective in this study and on the

surfaces examined. ACV is also known to disrupt the cell membrane integrity, metabolism, and nuclear material of *E. coli* and *S. aureus* (Yagnik *et al.*, 2018) [4] and can inhibit the growth of antibiotic strains of bacteria, such as MRSA [24]. Notwithstanding the outcome of any disinfection exercise is a product of the strain, surface physiochemical attributes, bacteria density and biofilm state of the organisms.

The recommended usage time for Lysol Disinfectant Spray is 5 minutes for the bacteria we identified, and the United States Center for Disease Control (CDC) found that 70% ethanol is effective against *P. aeruginosa*, *E. coli*, and *S. aureus* after 10 seconds [25].

4.3. Application and Significance

The COVID-19 pandemic places special emphasis on regular sanitation and disinfecting of surfaces that come into contact with body mucous membranes (*i.e.*, eyes, nose, and mouth). Oftentimes, basic sanitization and disinfection of surfaces are not practiced as a result of perceived difficulties and ignorance of their processes; most notably, types of disinfectants that may be used, cost of disinfectants, recommended usage times, and concerns over chemicals involved in popular disinfectants. Common household disinfectants, which can be obtained easily at low costs, include Lysol Disinfectant Spray, 70% ethanol, and ACV. ACV was chosen in this study due to its reputation as a natural or organic disinfectant. Our results indicate that these household disinfectants, even at usage times below their recommended, have significant disinfecting properties. The recommended usage times of disinfectants are still advised.

As these results indicate some sanitary effect for all three disinfectants for the two-minute periods on all four materials, their use may contribute significantly to COVID-19 and other communicable disease safety measures. Other potential forms of bacterial transmission and facilitation of infection, such as earbuds or cell phones, should be similarly studied. A special emphasis on this method of study in relation to healthcare workers and hospital environments should also be pursued, as many of these bacteria pose significant threats to the health of immunocompromised patients. Although the disinfectants showed maximum potency after 2 minutes, increasing the exposure time during personal hygiene is recommended.

5. Conclusion

Wristbands, often worn daily without routine cleaning, may accumulate potentially pathogenic bacteria. However, the quantity and taxonomy of bacteria found on the wristbands in this experiment show that there is a need for regular and popular sanitation of these surfaces. Generally, it was found that rubber and plastic wristbands had higher bacterial counts, while metal ones, especially gold and silver, had little to no bacteria. Bacteria found were common skin residents, of the genera *Staphylococcus* and *Pseudomonas*, and intestinal symbionts, like of

the genera *Escherichia*. The ability of many of these bacteria to significantly affect the health of immunocompromised hosts indicates a special need for healthcare workers and others in hospital environments to regularly sanitize these surfaces. Common household disinfectants, such as Lysol Disinfectant Spray, 70% Ethanol, and Heinz Apple Cider Vinegar all proved at least somewhat effective on all materials (rubber, plastic, cloth, and metal), although antibacterial efficacy was significantly increased at two minutes compared to thirty seconds.

Author Contributions

JM, BW and DD were undergraduate researchers who assisted cfu determinations, sample analysis and drafting of manuscript; KD and BS were graduate researchers—direct supervision of undergrads and experimentation. MC conceived the project and sampled all the subjects. NE is the senior author—data interpretation, editing manuscript.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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