ORIGINAL ARTICLE

Taking your work home with you: Potential risks of contaminated clothing and hair in the dental clinic and attitudes about infection control

Taylor Davidson,^{1*} Erica Lewandowski,^{1*} Meghan Smerecki,^{1*} Halee Stratton,^{1*} Jamal Alhabeil,² Michelle Wheater PhD,² Kathi Shepherd MS¹ and Eric S. Krukonis PhD^{2**}

- ¹ Department of Periodontology and Dental Hygiene, University of Detroit Mercy School of Dentistry, Detroit, MI 48208,
- ² Department of Biomedical and Diagnostic Sciences University of Detroit Mercy School of Dentistry, Detroit, MI 48208
- * These authors contributed equally to the work

Eric S. Krukonis, Associate Professor, Assistant Director of Research, Department of Biomedical and Diagnostic Sciences University of Detroit Mercy School of Dentistry, 2700 Martin Luther King Jr. Blvd RM444, Detroit, MI 48208. phone: 313-494-6851 Email: krukones@udmercy.edu

ABSTRACT

Background: Microbial contamination of clinic clothing is a potential source of infectious organisms spreading to the environment and susceptible people. The goal of this study was to educate dental professionals about the levels of bacterial contamination on clinic clothing and hair following dental clinic sessions.

Methods: Surveys of 30 dental and dental hygiene students assessed attitudes regarding microbial contamination on clinic clothing. Bacterial samples were isolated from a sterilized swatch of clinic clothing (scrubs) attached to the pants below the coat-line or to a hair band and processed for bacterial enumeration and identification.

Results: We found nearly all dental and dental hygiene students perform errands in their contaminated clinic clothing, but almost all felt they would be more likely to take better infection control precautions if they were aware of how much bacteria contaminate their clothing after a day in the clinic. Microbial analysis of swatches from scrubs showed a range from 250-60,000 bacteria/swatch (median=5,400), while hair samples contained 130-84,800 bacteria/swatch (median=19,300), including some potential pathogens like *Staphylococcus aureus* and *Enterococcus faecalis*.

Conclusion: These findings demonstrate the importance of changing out of clinic clothing and washing one's hair as soon as possible after a clinic session.

KEY WORDS

bacterial contamination, infection control, S. aureus, pathogens, clinic clothing

INTRODUCTION

Most dental professionals are aware of infection control in a dental office. However, many are unaware of the amount of bacteria that is transferred to their clinic clothing (scrubs) and hair during a day in the clinic. A study by Nordstrom et al found that 79% of unwashed operating room clothing (23/29) was contaminated with gram-positive cocci bacteria, 10% including Staphylococcus aureus [1]. In that same study 69% of clothing samples (20/29) contained gram-negative coliforms, in some cases including Escherichia coli [1]. In another study where nurses were provided with sterilized scrubs prior to a 12-hour shift, the average bacterial load was 1246 or 5795 bacteria/inch² for day and night shifts, respectively and 70% of scrub samples contained methicillin-resistant Staphylococcus aureus (MRSA) an important nosocomial pathogen [2, 3]. The importance of proper glove removal and hand washing in a clinical setting was demonstrated in a study by Munoz-Price et al where they

demonstrated that potential bacterial pathogens present on health care workers hands often leads to contamination of lab coats which can then serve as a source for recontamination [4].

Since contaminating bacteria remain on scrubs in hospital settings, it is likely that bacteria, including potential pathogens, are transferred to dental professionals' scrubs and hair after a day in clinic (hair coverings are not typically worn in the dental clinic). These microbes could then be a source of cross contamination to the environment since many dental professionals wear their scrubs home and launder them themselves. The purpose of this investigation was to improve the knowledge of dental professionals on the amount and potential species of bacteria that they are unknowingly bringing home with them after a day in clinic in order to prevent cross contamination to the environment.

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^{**}Corresponding Author:

MATERIALS AND METHODS

Survey of attitudes towards infection control
A survey was conducted by a judgmental sample of dental
and dental hygiene students (n=30) at the University of
Detroit Mercy School of Dentistry (IRB approval #1516-29).
The survey consisted of five questions: Three multiple-choice
questions to measure participants' infection control protocols
utilized after a day in clinic. Two questions in the Likert scale
format to measure awareness of bacterial cross contamination
of scrubs to the environment.

Microbial analysis

Autoclaved (sterile) scrub swatches (12 inch², 3" x 4") were pinned on clinic scrubs on the thigh area (n=12) or attached to a hair band (n=10) to collect bacteria during a typical clinic day. After one or two clinic sessions (3 hours/session) scrub swatches were submerged in 10ml of sterile phosphate buffered saline (PBS), minced with sterile scissors and gently vortexed to elute adhered bacteria from the fabric. 50mµ of bacteria were plated along with 10⁻¹,10⁻²,10⁻³ (ten-fold) dilutions onto Blood Agar Base (Oxoid CM0055) + 5% defibrinated sheep blood (BD 211947), 1 mg/ml Vitamin K (MP Biomedicals 102259) and 0.5 mg/ml hemin (ACROS Organics 345960050) and grown at 37°C in an anaerobic chamber (Coy Laboratory Products) for 48-72 hours to culture facultative and obligate anaerobic microbes often associated with dental procedures. Colonies were enumerated to determine the level of contamination. Numerous colonies with distinct colony morphologies were subjected to culturing, DNA isolation, PCR and DNA sequencing analysis to determine the species.

Distinguishing Streptococcal species

In one instance the DNA sequencing results could not distinguish Streptococcus mitis from S. pneumoniae. We employed the use of the optochin test to distinguish these two species. The undetermined Streptococcus strain was struck onto half of a Blood Agar Base plate (Oxoid CM0055) with 5% sheep blood (Hemostat DSB500) with 1 mg/ml Vitamin K (MP Biomedicals 102259) and 0.5 mg/ml hemin (ACROS Organics 345960050) while a lab isolate of Streptococcus sanguinis (in the Mitis group of oral streptococci) or *S. pneumoniae* was struck on the other half of the plate. Mitis streptococci are resistant to growth inhibition by optochin, while S. pneumoniae is sensitive to optochin. Optochin-impregnated disks were purchased from Fisher Scientific (Oxoid DD0001) and applied to each half of the plate with the use of sterile forceps. The plate was incubated at 37°C for 24 or 72 hours in a 5% CO₂ incubator. No observed zones of inhibition around the disk were noted for either S. sanguinis or the unknown strain (S. mitis, Table 1 and Fig. S1).

DNA sequencing analysis

Distinct bacterial species (based on colony morphology) were isolated on blood agar plates, re-struck to a fresh blood agar plate and a single colony was inoculated into a microcentrifuge tube containing $100 \text{mm} \, \mu$ of autoclaved ultrapure H_2O . The sample was boiled for 5 minutes then centrifuged at $13,000 \, \text{rpm}$

for 5 minutes in a microcentrifuge. 70mu of the lysate was moved to a fresh microcentrifuge tube and used as the DNA template for PCR. The 16S rRNA gene was amplified using the primers 16S 5' GAGAGTTTGATYMTGGCTCAG and 16S 3' GAAGGAGGTGWTCCARCCGCA. We performed 30 rounds of PCR amplification using an annealing temperature of 50°C and elongation time of 1 minute/cycle with Phusion High-Fidelity DNA Polymerase (Thermo Scientific F530S). After purification of the 1.5 kb PCR product using a Qiagen PCR clean-up kit, the PCR product was quantified using a Qubit spectrophotometer and sent for sequencing by Genewiz using the 16S 5' GCAACGCGAAGAACCTTACC 3' primer to read the V6 variable region or 5' CCAGACTCCTACGGGAGGCAG 3' to read the V3 variable region 16S [5]. 16S sequences were aligned to known bacteria 16S rRNA genes using BLAST (NCBI) and results are reported in Table 1. This allowed for identification of the types of bacteria that typically contaminate scrubs and hair after working in the dental clinic.

FIGURE S1: Discriminating between a streptococcal isolate as *S. mitis* or *S. pneumoniae*. The unknown streptococcal isolate was struck onto blood agar base with 5% sheep blood and an optochin disk was placed in the center of the streak. Cell were grown for 1 (A and B) or 3 days (C and D) in 5% CO₂ incubator at 37°C and the appearance of a zone of inhibition around the optochin is indicative of a *S. pneumoniae* isolate. A previously characterized lab isolate of *S. pneumoniae* was used as a positive control for sensitivity to optochin (A-D, top) and a previously acquired lab isolate of *S. sanguinis* (also in the Mitis Group of streptococci) was used a strain resistant to optochin (A and C, bottom). The unknown (B and D, bottom) grew similarly to *S. sanguinis* (resistant to optochin). Thus, based on DNA analysis of the 16S gene, this strain is *S. mitis*.

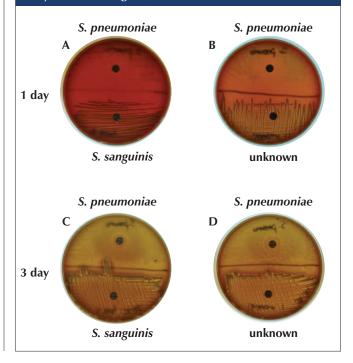


FIGURE 1: Survey questions reveal potential sources of environmental contamination from clinic clothing and hair after a day treating patients. 30 dental or dental hygiene students filled out a questionnaire assessing habits after a day treating patients in the clinic including whether students performed errands while still in clinic clothing, whether they felt it important to change out of clinic clothing as soon as they arrived at home and whether they washed their hair after a day treating patients in the clinic. The impact on changing behaviors upon knowing whether bacterial contaminants are brought home was also assessed.

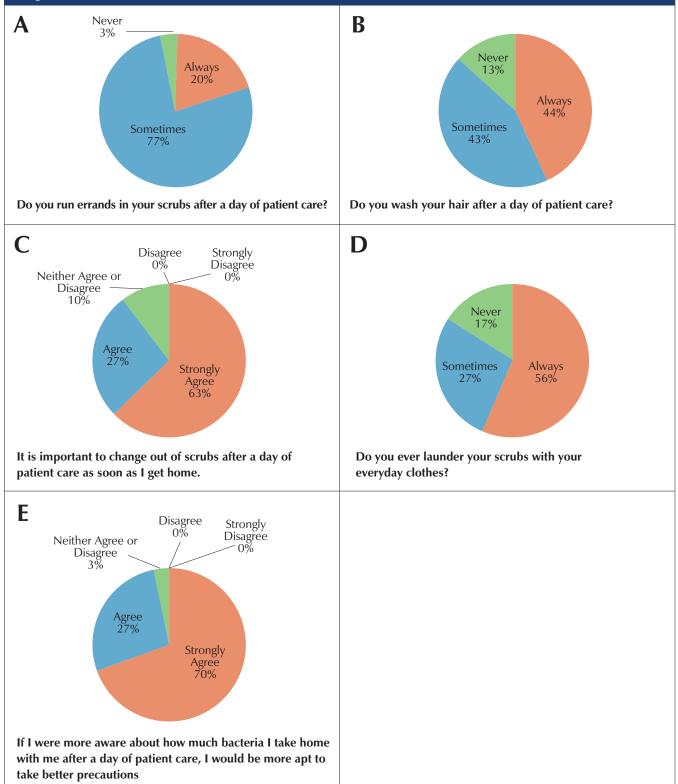
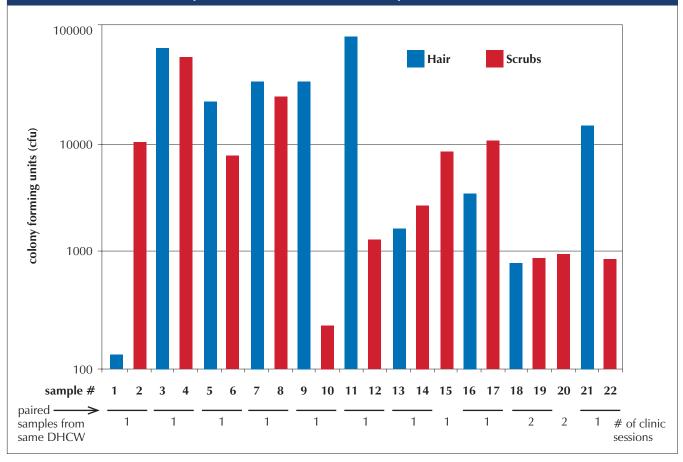


FIGURE 2: Levels of bacteria on scrub swatches from clothing and hair. Sterile swatches were removed from scrubs or hair-bands after 1 or 2 clinic sessions (3 or 6 hours) and minced in sterile PBS prior to plating on blood agar plates and grown for 48-72 hours at 37°C under anaerobic conditions. Colonies were counted and a total bacterial count was determined based on the dilution plated and the total volume of the sample.



RESULTS

Attitudes and awareness concerning infection control Based on our survey, 97% of dental healthcare workers (DHCWs) sometimes or always perform errands in their clinic scrubs after treating patients (Fig. 1A). Furthermore, only 44% of DHCWs always wash their hair after a clinic session (Fig. 1B). While most DHCWs (90%, Fig. 1C) recognized the importance of changing out of scrubs as soon as they get home, 83% sometimes or always wash their clinic clothing along with other garments (Fig. 1D). In some cases the DHCWs indicated they use cool water for washing clinic clothes to prevent the color from bleeding (personal communication). While laundry machines do not typically kill all microorganisms in a contaminated load, the use of cold water would result in higher levels of survival of some bacteria (and viruses) than using hot water with bleach [6-9]. Furthermore washing garments together can lead to transfer from one garment to another (crosscontamination, [10, 11]).

Encouragingly, 97% of DHCWs also indicated they would take better precautions if they were more aware of the bacteria they may acquire on their clothing and take home from the dental clinic (Fig. 1E).

Levels of contamination acquired in the dental clinic

To assess the level of bacteria that may travel home with
DHCWs on their clothing if they do not change out of scrubs
at work, 12-inch² (3" x 4") swatches of sterilized scrub material
were pinned onto the DHCWs clinic clothing just below the
clinic coat line. While clinic coats remain in the dental clinic and
are washed by a professional clinical clothing laundry service,
DHCWs often wear their underlying scrubs home and launder
them on their own (alone or mixed with other garments, Fig. 1D).
By placing a 12-inch² scrub swatch just below the clinic coat
line we obtained contaminants that might normally travel home
with the DHCWs. In addition, some DHCWs wore a hairband
with a sterile scrub swatch attached as well to capture bacteria
that may land on one's hair during dental procedures. Typically
DHCWs do not wear hair coverings in the clinic.

In 12 scrub swatches attached to clinic clothing, the level of bacterial contamination on the scrub swatch varied from 250-60,000 colonies/sample (median=5400; Fig. 2). Of the 10 samples from hair, the level of bacterial contamination on the swatch varied from 130-84,800 colonies/sample (median=19,300; Fig 2). Thus, both clothing and hair are potential sources of contamination after DHCWs leave the clinic for the day.

Specific bacteria associated with clinic samples
To identify specific bacterial species that contaminated scrub
swatches after a dental clinic session, single colonies from
bacteria plated on blood agar plates were isolated, re-struck
to fresh plates and lysed by boiling for DNA analysis. The
16S rRNA gene is typically used for bacterial identification by
sequencing the variable regions, which provide a unique DNA
sequence fingerprint for each species [5]. For our analysis, the
16S rRNA gene was amplified by PCR for each strain isolated
(41 total strains) and then subjected to DNA sequencing. Results
are provided in Table 1.

In multiple instances we identified *Staphylococci* and *Propionibacterium* species common on the skin and hair from both hair and scrubs samples (Table 1). In other cases we identified common oral bacteria such as *Streptococcus sanguinis, Streptococcus mitis, Veillonella parvula, Micrococcus species,* and *Granulicatella species* likely from contamination of scrubs during dental procedures (Table 1, Fig. S1). Potential pathogens like *Bacillus cereus, Enterococcus faecalis, Corynebacterium pseudodiphtheriticum* and *Dolosigranulum pigrum* were also identified, raising concerns about what species could be spread to the environment if one were to wear scrubs home from the clinic or not wash one's hair after a clinic session (Table 1). Finally, *Staphylococcus aureus,* a common nosocomial pathogen and major threat in the battle against multiple drug resistance [3] was also identified on scrubs (Table 1). A number

of the species isolated were β -hemolytic, indicating virulence factors capable of lysing host cells (Table 1).

DISCUSSION

The goal of this study was to make DHCWs aware of the numbers and types of bacteria they may be unwittingly bringing home to their families or to the community after a day of treating patients in the clinic. While clinic coats are worn to prevent the transmission of microbes from patients to clinic clothing, organisms may still get transmitted to other areas of the clothing (such as below the coat line) or to the DHCW's hair. It is encouraging that based on our survey 97% of DHCWs agreed or strongly agreed that if they were more aware about how many bacteria were taken home after a day of patient care, they would be more apt to take better precautions (Fig. 1E).

Based on the often large numbers of bacteria isolated on swatches attached to dental scrubs or DHCWs' hair (as high as 7000 bacteria/inch2), it is concerning that 97% of DHCWs in our survey sometimes or always perform errands on their way home from a clinic session in their scrubs (Fig. 1A) and only 63% strongly agree that it is important to change out of clinic clothing as soon as they get home from a day of treating patients (Fig. 1C). Finally, given that in >50% of our swatches from hair samples, the levels of bacterial contamination were >1,000 bacteria/inch2, the fact that only 44% always wash their hair after a day in the clinic was concerning (Fig. 1B).

Genus and Species	β-нІу	Times Isolated	Source	Reservoir
Bacillus cereus	+	1	Scrubs	Soil/Food
Bacillus thuringiensis	+	1	Scrubs	Soil/Pesticides
Corynebacterium pseudodiphtheriticum	-	1	Scrubs	URT
Dolosigranulum pigrum	-	1	Scrubs	URT
Enterococcus faecalis	-	1	Scrubs	GI Tract/Oral Cavity
Granulicatella sp.	-	1	Hair	Oral Cavity/GI Tract
Micrococcus sp.	-	2	Scrubs	Skin/Hair/Oral Cavity
Neisseria perflava	-	1	Scrubs	Oral Cavity/URT
Propionibacterium acnes	-	4	Hair and Scrubs	Skin/Hair
Propionibacterium avidum	+	1	Hair	Skin/Hair
Staphylococcus aureus	+	1	Scrubs	Skin/Hair/Nose
Staphylococcus capitis	-	4	Hair and Scrubs	Skin/Hair
Staphylococcus epidermidis	-	12	Hair and Scrubs	Skin/Hair
Staphylococcus hominis	-	4	Hair and Scrubs	Skin/Hair
Staphylococcus pasteuri or S. warner	-	1	Scrubs	Skin/Hair
Streptococcus sp. VT 162	-	1	Scrubs	Oral Cavity
Streptococcus mitis	-	2	Scrubs	Oral Cavity
Streptococcus sanguinis	-	1	Scrubs	Oral Cavity
Veillonella parvula	-	1	Scrubs	Oral Cavity

 β -Hly = β -hemolytic; URT=upper respiratory tract, GI=gastrointestinal tract

The large range in colonies isolated could be a result of the specific procedures taking place in clinic, the number of clinic sessions during which the sterilized scrub swatch was worn, the frequency with which a DHCW brushed their hands against their scrubs or hair or a variety of other factors. Although it should be noted we found no correlation between the number of clinic sessions attended and the levels of contamination (Fig. 2). Due to the quantity and types of species found on the samples the results clearly demonstrated that there is potential for crosscontamination from the dental clinic to the environment, including the transmission of pathogens (Fig. 2 and Table 1). A comparison of species found on the hair swatches and previous studies in the normal microbiota of hair [12, 13] showed many of the isolates we identified are normally present on human hair, yet some isolates (Granulicatella species) are not typically found on hair and may have been acquired while providing patient care (Table 1). Thus, human hair is not considered a sterile surface and a limitation of this study is that we did not document any transmission from DHCW hair or scrubs to patients. One may presume the risk of such transmission to be fairly low given the absence of documented cases of such transmission. However, previous reports have demonstrated potential pathogens such as S. aureus present in HCW's hair leading to the suggestion that head coverings be worn while performing certain medical procedures where the chance of cross-contamination or wound infections are present [14-16].

While most of the species found on the samples were common environmental microbes that can be found in soil or on the skin, a few samples revealed potential pathogens were present such as *Staphylococcus aureus*, *Bacillus cereus*, *Enterococcus faecalis*, *Corynebacterium pseudodiphtheriticum* [17] and *Dolosigranulum pigrum* ([18]; Table 1). It should be noted that in general healthcare workers have a higher risk of nasal carriage of *S. aureus* than the general population [19] although in a recent study dental students had a lower rate of carriage as compared to medical students [20].

We hope the findings presented in this work will highlight the issue of clothing contamination for DHCWs and help prevent cross contamination to the environment. There is a growing body of data implicating healthcare workers' uniforms as a potential reservoir of pathogenic organisms [1, 2, 4]). This study suggests the importance of using in-house laundry services at one's dental facility or at least being sure to change out of clinic clothing as soon as arriving at home as well as washing clinic clothing in hot water with bleach to facilitate decontamination. Additionally, if laundry service for scrubs is not provided by one's dental care facility, one may want to change out of scrubs before leaving work and carry the soiled items home separately. For those who don't routinely wash their hair after a clinic session, a head covering may also be advisable.

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