## DISTRIBUTION AND PREVALENCE OF BACTERIA FOUND ON THE DOOR HANDLES OF OLIN HALL, DRAKE UNIVERSITY

Abstract: Fomites are inanimate objects that serve in the spread of infectious disease. Our hypothesis was that door handles may aid in the spread of microbes between individuals and that they may be a reservoir of microbial contamination. In our experiments, we assessed the prevalence of specifically the Gram negative bacteria that were found on door handles of Olin Hall. It was hypothesized that during times where the building was near its peak usage, a larger percentage of the bacteria sampled from the door handles of Olin Hall would be Gram negative. The results showed that of total microbial colonies observed, 49% were Gram negative bacteria. Further statistical analysis revealed an interesting observation and the data showed differences in prevalence of Gram negative bacteria found on individual doors.

**Introduction:** The spread of infectious diseases through hand contact has been an area of major concern. According to a study conducted by Itah et al, Gram (+) Staphylococcus aureus, and Gram (-) enteric bacteria such as Escherichia coli, Klebsiella species, Citrobacter species, was found to contaminate various contact surfaces including chairs, tables, windows, door handles, and many other common household fixtures (Itah 2004). This study showed the striking presence of pathogenic bacteria in nursery schools, day care centers, and other public places. The study also highlighted the variety of potential pathogens. According to Salton, Gram negative bacteria show virulent characteristics due to the presence of endotoxin in their outer membranes. Endotoxin causes activation of immune system cells such as macrophages, neutrophils, and B lymphocytes, which may lead to tissue necrosis and endotoxic shock. It has been further demonstrated that Gram negative bacteria have higher antibiotic resistance, partially due to the presence of an outer membrane (Thomson 2005). This outer membrane may prevent entry of antibiotics. These observations are important considerations that merited our study with door handles. The presence of these pathogenic bacteria on environmental surfaces such as door handles poses a potential risk to vulnerable, immune-compromised individuals. It has been shown that hard, non-porous surfaces, such as door handles, have the highest bacterial transfer rates to hands (Rusin 2002). In recent past a lot of effort has been invested in emphasized hand hygiene through hand wipes and hand sanitizers. According to a study by Stout et al, hand wipes with higher ethanol content are more effective in not only antimicrobial activity but also removal of endospores via the mechanical action (Stout 2010). Even though people are commonly aware of such practices, the possibility of inaccessibility or lack of use of these practices does occur. According to Hansen, up to 60% of adults do not wash their hands when appropriate (2002).

The present study was done to not only investigate the significance of bacterial growth in relation to peak building usage but also to investigate a hypothesized trend in bacteria based on their Gram staining characteristics. Moreover, it was hypothesized that the percentage of bacteria observed would be represented in greater proportions by Gram (-) bacteria during these peak times. We studied the door handles of Olin Hall at Drake University. Olin hall is a science building that numerous students and faculty use on a daily basis. The results of the study will help our understanding of the prevalence and nature of bacteria that are transmitted through door handles. It will also lead us to search for practical ways of controlling the growth and spread of infectious diseases thereby playing a better role as an engaged citizen.

**Materials and methods:** In this study the inner door handles of Olin hall were analyzed. Germ-C hand sanitizer, at 63% ethyl alcohol, was applied to the door handle and was used as a control for the experiment. The door handles were sampled every hour for seven hours on Monday February 7, 2011. Two samples were taken along with negative controls. The doors were swabbed on Mondays, March 7, 2011, and on March 21<sup>st</sup>, at peak building usage times as previously studied. Sterile cotton swabs were used to obtain specimens from door handles. These samples were then streaked on nutrient agar plates and incubated for a period of 48 hours at 37°C. After the incubation the number of resulting colonies was counted and characterized based on margin, elevation, and colony shape. Mean colony counts were calculated. The Gram-stain procedure was then used to assist in the separation of bacterial cells. Microscopy was then used to identify the cellular morphology and Gram staining characteristic.

**Results:** This experiment analyzed colony numbers sampled from doors along with cultural characteristics and cellular morphological characteristics. The negative controls showed no observed growth. An average number of colonies were calculated. Figure 2 illustrates the average number of colonies over time. The greatest number of colonies (19) was found at 12:00pm. The second greatest number of colonies was observed at 10:00am and 11:00am both with average colony counts of 15. After 12:00pm a decrease in the number of colonies was found after every hour subsequent. Further analysis was conducted to identify the number of Gram (-) bacteria found on the door handles during the peak times. The Gram stain identified that 49.0% of the bacteria found on the door handles were Gram negative (Figure 3).

Moayad Baadhaim, David Kusner, Humayun Ahmed, Chinh Dao (Mentor), Allan Trap (Mentor) Department of Biology, College of Arts & Sciences



Figure. 1 Shows the door handles of Olin Hall labeled from left to right with A, B, C, and D



Figure 3. Picture of the gram (-) bacteria found on door B. The bacteria shows a cocci shape and tetrad cell arrangement





Figure 4. Percent number of gram (-) bacteria found on all doors of Olin Hall.

Door	Number of Colonies	<b>(-)</b>
Α	295	of G
В	43	age
С	33	rcent
D	40	Рег



Figure 2. Average number of colonies swabbed from Olin hall inner door handles over a six hour period.



Figure 5. Picture of the gram (+) bacteria found on multiple door handles. This gram (+) is predicted to Staphylococcus aureus with cocci shape and staphyl cell arrangement.



Figure 6. Picture of the gram (+) bacteria found on door B. The bacteria shows a bacillus shape and staph cell arrangement



The Gram negative colonies had a yellow color, convex elevation, and smooth margin. Under the microscope it was identified as a Gram (-) cocci with tetrad arrangement (Figure 3). Another strand of Gram (+) was found on door B, the colonies had a white color, flat elevation, and smooth margins. Figure 6 shows a photo of the Gram (+) strand with a staphylobacillus arrangement.

**Discussion:** The results confirmed the first hypothesis that increased use of door handles at particular times of the day would result in increased amounts of microbes. However, our second hypothesis that majority of these bacteria would be Gram negative was rejected. These results imply that the majority of bacteria, transmitted through door handles, are Gram positive. There are several issues pertaining to the present study that require further discussion.

Our results suggest a relationship between the use of door handles and amount of bacteria observed on the door handles. During the early and late times of the day when few individuals are attending classes, there was less number of bacterial colonies present on the door handles. These results imply that the majority of bacteria found on door handles came from hand contact with the door handles. Hours 11am to 1pm showed a significantly higher amount of bacterial colonies with the 12pm sample being the maximum number of colonies. These results correlate with the increased number of classes schedule for those times. The majority of classes take place in Olin Hall between 11 and 1 pm.

Our results from the second hypothesis did not support the assumption that the majority of bacteria observed on door handles would be Gram negative. Out of 411 colonies sampled during the peak hour, 49% of the total colonies were Gram negative. This finding implies that the number of Gram negative colonies were roughly equal to number of Gram positive colonies. Nevertheless our results of gram negative bacterium accounting for 49% of the total colonies imply that Gram negative organisms can be transmitted through door handles. The common prevalence of Gram positive bacterium can be attributed to the presence of gram positive bacteria found on our skin. According to Cogen, Gram (+) *Staphylococcus epidermidis*, accounts for 90% of microbes that usually occupy a particular body site. Using colony morphology, cell shape, and arrangement we were able to speculate that *Staphylococcus aureus* was found on the door handle samples (Figure 5). Further genetic or biochemical tests are needed to confirm or reject our hypothesis. Future studies could involve data being collected over a seven-day period instead of just two.

The samples collected from the door handles are only a representation of those microbes that could satisfy their nutritional needs provided by nutrient agar. Thus our present study fails to provide exact representation of bacteria that were present at the time samples were collected. Other growth medium like EMB, MSA, blood agar plates could be used to further differentiate bacterial colonies. However, the results obtained reveal a general distribution of bacteria found on door handles, which have the capability to transmit disease.

Acknowledgement: We would like to thank Dr. Chinh Dao and Dr. Allan Trapp for their assistance, and guidance toward completing this study. In particular, Dr. Dao's recommendations and suggestions have been valuable and this study would not have been possible without his assistance.

ReferencesCogen AL, Nizet V, Gallo RL (2008) Skin microbiota: A source of disease or defence? Br J Dermatol 158:442–455Hansen TB, Knochel S. Image analysis method for evaluation of specific and non-specific hand contamination. J APPL Microbiol 2003;94(3):483-494.Itah, and Ben AE. "Incidence of Enteric Bacteria and Staphylococcus Aureus in Day Care Centers in Akwa Ibom State, Nigeria." The Southeast AsianJournal of Tropical Medicine and Public Health (2004): 202-09. Web.Rusin, P., S. Maxwell, and C. Gerba. 2002. Comparative surface-to-hand and fingertip-to-mouth transfer efficiency of gram-positive bacteria, gram-negative bacteria, and phage. J. Appl. Microb. 93:585-592.Salton, Milton R.J, and Kwang Shin Kim. "2." Ed. Samuel Baron. Medical Microbiology. 4th ed. PrintThomson JM, Bonomo RA The threat of antibiotic resistance in gram-negative pathogenic bacteria: beta-lactams in peril! Curr Opin Microbiol2005;8:518-24.

Stout, Yu VL, Rihs JD, and D'Antonio NN. "Revisiting the Hand Wipe versus Gel Rub Debate: Is a Higher-ethanol Content Hand Wipe More Effective than an Ethanol Gel Rub?" *American Journal of Infection Control* (2010): 678-82 Web.