Transmission of *Escherichia coli* Through Toilet Seats

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Abstract

Microbes are found on all kinds of surfaces, but not all are pathogenic. Bathrooms are perceived by the public as an important area in the transmission of pathogenic microbes, and the purpose of this project was to investigate the transmission of bacteria from toilet seats to skin. *Escherichia coli* strain K-12 was applied to the toilet seat and different methods were used to see which was the best way to prevent the transmission of bacteria to the skin. *E. coli* strain K-12 was identified in samples using the differential media, Eosin Methylene Blue agar. This experiment found that cleaning the toilet seat prior to use will be the most effective method for stopping transmission, and that using a paper barrier will not stop the transmission.

Introduction

The warm and humid environment typically found in bathrooms provides a perfect setting for the survival of microorganisms. Many of these pathogens, like *E. coli,* can survive on environmental surfaces for weeks or even months. Toilets can also play a role in transmission both through direct contact and through the generation of aerosols during flushing (Suen, et al., 2019; Johnson, et al., 2013). Previous studies have shown that flushing toilets produces a plume that can last for up to 8 minutes and seed the toilet seat with bacteria (Johnson, et al., 2013).

Eosin methylene blue (EMB) agar is a selective and differential medium used for the identification of Gram-negative fecal coliform bacteria. EMB agar selects for Gram-negative bacteria based on the presence of eosin and methylene blue dyes. It is differential based on the fermentation of lactose. Bacteria that ferment lactose in the medium form colored colonies (dark purple or black), while those that do not ferment lactose appear as colorless colonies. Colonies of *E. coli* will be a metallic green on EMB agar, making them easy to distinguish from other lactose fermenting colonies. EMB agar is used in water quality tests to distinguish coliforms and fecal coliforms that signal possible pathogenic microorganism contamination in water samples (Lal and Cheeptham, 2007; Johnson and Case, 2019).

This study investigated the transmission of *E. coli* from toilet sets in order to compare different ways of preventing transmission. It was hypothesized that if a paper barrier is used, it will not stop the transmission of *E. coli*.

Methods

The experiment was performed in a biological safety cabinet to decrease the risk of contamination. Prior to the experiment, the toilet seat was disinfected using Lysol disinfectant wipes and UV light. A toilet seat was marked to separate it into two sides. One side had a paper toilet seat cover over it and the other did not. Each side was also divided into a wet and dry area.

The nonpathogenic *E. coli* strain K12 was applied to the testing area and allowed to dry. A paper toilet seat cover was placed over half of the toilet seat, and distilled water was used to wet the “wet” testing areas. Using a gloved hand, a finger

was placed on the testing area and then pressed onto the corresponding EMB plate. A sterile swab saturated with distilled water was used to swab the finger. The swab was placed into nutrient broth. Each condition was repeated three times and gloves were changed after each replicate. Disinfectant wipes were used on the toilet seat according to manufacturer's protocol, which stated that the surface needs to dry for 10 minutes after application. The gloved finger and wet swabs were used to test these areas in the same manner as previously discussed. All media was incubated at 37ºC for 12 hours.

Results

Metallic green colonies, typical of *E. coli* growth, were observed on EMB plates corresponding to the paper wet (Figure 1) and no paper wet conditions. No growth was observed on the EMB plates for paper dry, no paper dry and disinfectant wipes. Turbidity was observed in the nutrient broth for the paper wet (Figure 1) and no paper wet conditions. No growth was observed in the nutrient broth for paper dry, no paper dry and disinfectant wipes.

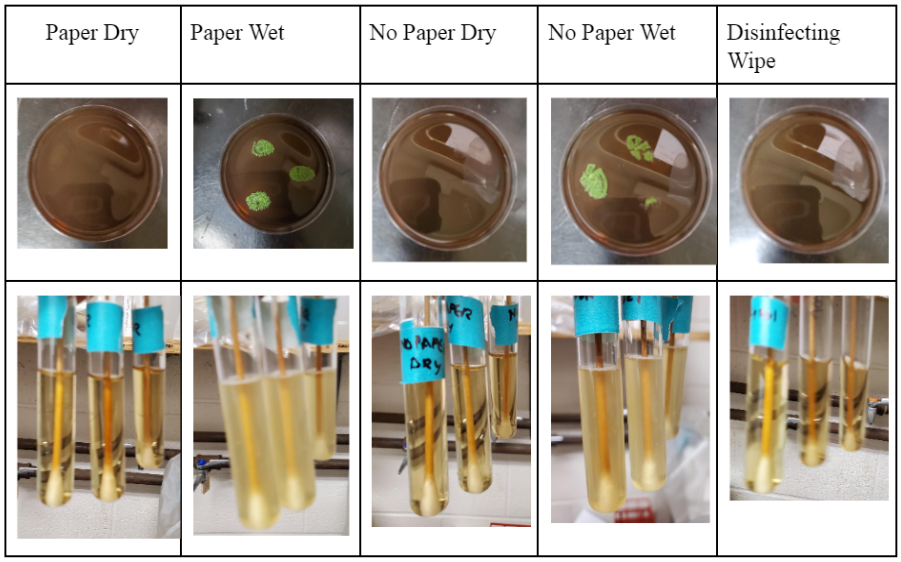

Figure 1. Showing results on EMB plate (top) and nutrient broth (bottom)

Discussion

This study demonstrated that *E. coli* was transferred from the toilet seat to the gloved finger, only when the seat or paper was wet. The hypothesis that the transmission of *E. coli* would not be prevented by the paper toilet seat cover was

supported. Further, a wet toilet seat could potentially be the cause of the transmission of pathogenic microbes found in human feces and vomit, and emphasizes the importance for handwashing after using the bathroom. In fact, *E. coli* can persist after a few days to a few years in water, soil, and habitats where generation could increase opportunities for transmission (Chiyo, et al., 2014).

Cleaning the toilet seat with a disinfectant wipe is better at stopping transmission than using a paper toilet seat cover. Previous studies have shown that disinfection is the most effective way to reduce the amount of microbial contamination on a toilet seat (Johnson, et al., 2013). Based on all of these studies, it would be more beneficial to have disinfectant in public restrooms than to have paper toilet seat covers to prevent the transmission of pathogenic microbes. This specific disinfectant's main ingredients are similar to many aerosol germicides found on the market today. Furthermore, the active ingredients in Lysol are full of Alkyl dimethyl benzyl ammonium chlorides, which share a similar ingredient list to Pledge (multi-surface disinfectant spray). Pledge was one of the most effective aerosol agents used in a study conducted by Turgeon, et al. (2016), which tested various aerosol germicides (Turgeon, et al., 2016).

The risk of infection resulting from transmission in a public restroom depends on the infectious dose of the microbes. Microbes with lower infectious dose, such as norovirus and *Shigella,* have a higher chance of causing infection compared to microbes with higher infectious doses (Gerhardts, et al., 2012).

In conclusion, this study demonstrated that if skin were to contact a wet toilet seat with or without a paper barrier, then transmission of microbes found on the seat would occur. The data also demonstrated that disinfection lowered the risk of transmission. In order to prevent the transmission
of microbes from a toilet seat, it is better to use a disinfecting wipe rather than use a paper toilet seat cover.

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