Minimizing Pathogens on Everyday Dishes With an Autonomous Dishwasher

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While pathogens in food multiply exponentially with time in room temperature, soiled dishes wait for hours in kitchens until dishwashers are full, leaving behind substantial pathogen traces on clean dishes, which leads to dysbiosis of the microbiota and serious illnesses such as inflammatory bowel disease, diabetes and colorectal cancer. We propose here to study the effect of immediate cleaning of dishes with an autonomous robot dishwasher–called dishcare–we invented and developed to clean and sanitize soiled dishes individually and immediately without waiting for a large number of dishes to accumulate. With Americans using around a billion dishes every day, minimizing pathogen replication at such scale would constitute a significant impact on public health, while raising awareness on not creating environments for pathogens to replicate freely, both for staying healthy and for discouraging the prospect of allowing more harmful mutants to emerge.

Pathogens replicate exponentially on soiled dishes at room temperature.

The sight of dirty dishes piling up in a kitchen sink is an eyesore for many. Yet the kitchen sink continues to remain as the easiest place to drop soiled dishes after a meal or a drink. In many kitchens, soiled dishes wait for hours–sometimes days–before they are cleaned. A bigger cause of concern than a mere eyesore is that microorganisms multiply exponentially in food residues on soiled dishes. With soiled dishes in kitchen sinks being at room temperature and food residues acting as growth media, microorganisms find themselves in an unhindered environment for growth. According to the U.S. Department of Agriculture, when food is left out for too long at room temperature, bacteria such as *Staphylococcus aureus*, *Salmonella Enteritidis*, *Escherichia coli O157:H7*, and *Campylobacter* are known to grow to dangerous levels that can cause illness [22]. Bacteria double in as little as 20 minutes in the range of temperatures between 40°F and 140°F. At that doubling rate, a single bacteria could exponentially grow to over 2 million in 7 hours and potentially to over 68 billion in 12 hours if there is sufficient food residue to support growth. No wonder soiled dishes stink before they are cleaned.

Dishwashers make soiled dishes wait and enable pathogen replication.

Regular dishwashers are designed for washing a large number of dishes at once. An average dishwasher has a capacity of over 12 place-settings—over 130 dishes including silverware. However, dishes tend to accumulate gradually throughout the day. Depending on the number of people and dish usage levels, it can take anywhere

between a few hours to days before a full load of dishes have accumulated. People find it wasteful to run their dishwashers before they are fully loaded. For good reason, because an average dishwasher cycle consumes over 1 kWh of energy, 3 gallons of water, and a dose of detergent. As a result, soiled dishes wait for hours or days until many of them have accumulated to justify running the dishwasher. According to the U.S. Energy Information Administration, 85% of American households with dishwashers do not run their dishwashers every day [23]. And 15% of American households with dishwashers run their dishwashers once a week. At this scale of wait times, soiled dishes are typically



Figure A.1: Dishwashers do not kill all pathogens on dishes. A) swab sampling of pre-rinsed dishes before dishwashing [21]; B) LB-Agar plates showing bacterial colonies before (left) and after (right) cleaning with a 2-hour heated dry cycle, even when soiled dishes are pre-rinsed [21]; C) Biofilm on dishwasher rubber seals comprising a variety of pathogens [1].

brimming with microorganisms by the time they get their chance to get cleaned.

Clean dishes from dishwashers have pathogens.

Dishwashers are not autoclaves. Dishwashers do not operate at the temperature necessary to kill all microbes on soiled dishes. While autoclaves operate at 250°F, dishwashers operate at 140°F. Bacteria such as *Pseudomonas, Escherichia, Acinetobacter* and fungi such as *Exophiala sp, Candida, Cryptococcus, Rhodotorula, Aspergillus, Magnusiomyces, Fusarium, Penicillium* have been found in dishwashers [1, 2]. Clean dishes from dishwashers have been found to host over 100 bacterial colonies after they are cleaned in regular dishwashers [24]. Clean dishes from dishwashers that carried more bacterial colonies were also the dishes that were categorized as more soiled prior to cleaning. This suggests that the number of microbes on dishes after cleaning is a function of the number of microbes on dishes before cleaning. To get clean dishes with fewer microbes, dishwashers will need to start with soiled dishes with fewer microbes. Advocating for reducing the number of microbes prior to cleaning dishes by asking Americans to dramatically reduce wait times for soiled dishes would be an effort in vain due to the fundamental batch mode design of regular dishwashers and deeply rooted social behaviors around using them.

Pathogens in food cause dysbiosis of the microbiota.

As dishes are the articles that make final contact with food before people eat, pathogens on dishes have a direct pathway to infect people. When the immune system is compromised, these microbes disrupt the gut microbiome and cause dysbiosis of the microbiota, potentially leading to shorter-term illnesses such as diarrhea and chronic illnesses such as inflammatory bowel disease, obesity, allergic disorders, diabetes, autism, and colorectal cancer [3, 4, 5, 25].

Americans use 1 Billion dishes a day.

Americans use around a billion dishes every day to consume food and drinks. Around the globe, this number expands to around 14 trillion dishes in a year. Minimizing pathogen replication at such a massive scale constitutes a significant benefit to public health.

Uncontrolled replication of pathogens could create more harmful mutants.

This study will also raise awareness on the importance of not creating environments where bacteria can replicate freely. Stopping such uncontrolled bacterial replication is essential not only to stay healthy but also to avoid the possibility of bacteria in food potentially mutating into more harmful strains. According to the U.S.

Food and Drug Administration, the incidence of mutators among isolates of pathogenic *Escherichia coli* and *Salmonella enterica* is over 1% [26].

Innovation

Dishcare is an autonomous dishwasher.

Over the last couple of years, we have invented and developed an autonomous robot dishwasher called dishcare. People can drop soiled dishes in dishcare like they would in a kitchen sink. Dishcare picks soiled dishes up, cleans with targeted pressure jets, and puts the clean dishes away. Unlike a regular dishwasher, dishcare cleans dishes individually with a robot arm that perceives dishes using artificial



Figure B.1: Dishcare is an autonomous dishwasher. A) Dishcare has a soiled cabinet (middle) where soiled dishes are dropped and a wash module (bottom) where dishes are cleaned and sanitized. Dishcare also has a clean cabinet (top) where clean dishes are organized. A robot located behind the cabinets picks dishes individually from the soiled cabinet, cleans and sanitizes them in the wash module, and organizes them in the clean cabinet; B) Dishcare with front panels removed; C) Dishcare with soiled cabinet open for dropping dishes; D) Dishcare with clean cabinet open for picking clean dishes.

intelligence. Dishcare's technology is backed by 7 pending patents in the U.S. Patent Office.

Dishcare is powered by robotics and artificial intelligence.

Dishcare has two cabinets, one for soiled and another for clean dishes. The soiled cabinet is where soiled dishes are dropped. And the clean cabinet is where clean dishes can be found. Behind the cabinets, dishcare has a robot arm designed to handle multiple dish types, such as mugs, bowls, plates, cups, spoons, forks, knives, and chopsticks. The arm is informed by perception and planning modules that use a camera to see and understand dishes. Under the two cabinets, dishcare has a wash module with pressure nozzles and a camera to see and target dirt on dishes with water jets. When a soiled dish is dropped in dishcare, its arm picks the dish, cleans the dish in its wash module, and organizes the dish in its clean cabinet. And when a number of soiled dishes are dropped at once, dishcare picks, cleans, and organizes each dish individually.

Dishcare cleans soiled dishes immediately without making them wait.

As dishcare cleans one dish at a time, it does not wait until a large number of dishes have accumulated. Dishcare cleans soiled dishes immediately after they are dropped off, without allowing them to act as growth media for pathogens.

Dishcare sees dirt on dishes and targets them with water jets.

To clean a dish, dishcare's wash module sprays soap on the dish, targets dirt on the dish with a recycled water jet, and rinses the dish with a fresh water jet. Dishcare targets dirt on dishes by precisely coordinating the camera in its wash module with its arm. When the wash module sees a region on a dish with stubborn dirt, it informs the arm to move the dish to a position where the stubborn dirt is right under the recycled water jet. For spotless cleaning of every side of the dish, dishcare's robot arm moves and rotates the dish, while the soap spray and the water jets turn on and off at different times. After rinsing, the arm shakes off water droplets from

the dish for faster drying. As dishcare can see and target dirt on dishes, there won't be food residue on clean dishes anymore. So, there would be no more running the dishwasher twice or hand washing of clean dishes right out of the dishwasher. Also, as dishcare can target dirt, it is fast. A few seconds to get a dish cleaned. Not hours of spraying as in a dishwasher. Dishcare's ability to target dirt also allows it to save water and energy. By using more resources for soiled dish regions and less resources for cleaner dish regions, dishcare is able to use resources thoughtfully.

Dishcare will sanitize dishes with UV-C light.

After cleaning a dish with soap and water, dishcare will sanitize each dish with UV-C light to kill any residual microorganisms present on dishes. According to the U. S. Food and Drug

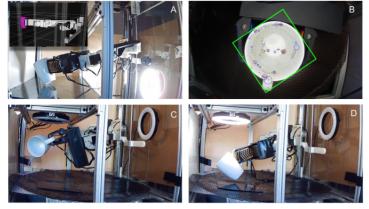


Figure B.2: Dishcare prototype autonomously picks up soiled dishes individually from its soiled cabinet, cleans them in its wash module, and organizes them in its clean cabinet. A) Dishcare's robot arm autonomously picking up a soiled mug from its soiled cabinet using camera based artificial intelligence; B) Dishcare's wash module seeing a soiled mug with its camera and targeting dirt (blue circles) on the mug. C) Dishcare's wash module cleaning the mug with pressure jet sprays; D) Dishcare's wash module irradiating the mug with an illumination module.

Administration, UV-C has effectively been used for decades to reduce the spread of bacteria, and has been shown to destroy the outer protein coating of the SARS-Coronavirus [27]. Similar to targeted cleaning, dishcare can contain UV-C only on a dish without spreading UV-C to regions outside the dish, thus maximizing UV-C intensity and antimicrobial efficacy.

Dishcare takes the chore out of doing the dishes.

People can drop one or more soiled dishes in dishcare, just like how they would typically drop them in their kitchen sink. No more arranging dishes in racks carefully, as the robot arm can pick dishes up however they might be arranged in the soiled cabinet. For instance, a mug standing right-side up is fine, but so is a tilted one. Actually, a fallen mug is alright too. People also won't need to put dishes away, as the robot arm organizes dishes for them. With dishcare, people won't have to bend forward with dishes in their hands. Dishcare's clean cabinet is located right under the countertop. And dishcare's soiled cabinet pulls out and rises to the level of the countertop. People also won't have to add detergent before every run. Dishcare draws a few drops of soap from a dish soap container, dilutes it, and sprays it precisely on dishes to eliminate waste. Replacing the soap container once a month or so would do.

Dishcare has the same size as a standard dishwasher.

Dishcare is designed to be a direct replacement for a standard 24" wide dishwasher for easy installation. Dishcare's first model is for offices. This model can clean mugs, bowls, plates, cups, spoons, forks, knives and chopsticks. Dishcare's second model is for homes. In addition to the office dish types, the home model can also care for pots, pans, ladles, and more.

Approach

Phase I overview: This proposal will study the effect of cleaning of dishes with dishcare when compared to a regular dishwasher.

Figure B.3: Dishcare prototype cleans a variety of dish types and conserves energy and water. A) plate cleaning; B) bowl cleaning; C) spoon cleaning; D) Dishcare's energy and water sensors measuring resource consumption while cleaning a mug. Dishcare's robot arm also picks up cups, forks, knives and chopsticks.

General Experimental Approach: Outlined below are the standard procedures to be used throughout this Phase I proposal.

Aim 1

Estimation of pathogens on clean dishes from a regular dishwasher.

<u>Null hypothesis</u>: The wait time for soiled dishes has no effect on the number of pathogens on clean dishes after cleaning with a regular dishwasher.

<u>Alternative hypothesis:</u> Increasing wait time for soiled dishes increases the number of pathogens on clean dishes after cleaning with a regular dishwasher.

Experimental Design

Strategy

Sterile dishes, autoclaved at 250°F for 30 minutes, will be used in all groups below. Food suspension will consist of a mixture of common food consumed by people, including meat, potatoes, pasta, rice, and dairy.

<u>Experimental group</u>: This will consist of autoclaved dishes coated with a food suspension and left waiting for t hours, where t will be set to be one of 0, 8, 12, 24 hours for each dish in the group. For each wait time t, d number of dishes will be prepared. Therefore, the experimental group will comprise 4d dishes in total. Dishes in this group will be cleaned in a regular dishwasher using a high temperature heated dry cycle. Two swab samples, first immediately before cleaning and second immediately after cleaning, will be obtained for each dish. Each swab sample will be used to innoculate 3 different types of agar plates. In total, there will be 24d agar plates for this group.

<u>Negative control group</u>: This will consist of autoclaved dishes without food suspension, left waiting for t hours, where t will be set to be one of 0, 8, 12, 24 hours for each dish in the group. For each wait time t, d number of dishes will be prepared. Therefore, the negative control group will also comprise 4d dishes in total. Dishes in this group will also be cleaned in a regular dishwasher using a high temperature heated dry cycle. Two swab samples, before and after cleaning, will be obtained for each dish. Each swab sample will be used to innoculate 3 different types of agar plates, resulting in 24d agar plates for this group.

<u>Positive control group</u>: This will consist of autoclaved dishes coated with a food suspension and left waiting for t hours, where t will be set to be one of 0, 8, 12, 24 hours for each dish in the group. For each wait time t, d number of dishes will be prepared. Therefore, the positive control group will also comprise 4d dishes in total. Dishes in this group will be hand washed using soap and hot water. Two swab samples, before and after cleaning, will be obtained for each dish. Each swab sample will be used to innoculate 3 different types of agar plates, resulting in 24d agar plates for this group.

<u>Total number of dishes and agar plates:</u> Including all groups, 12d dishes and 72d agar plates will be used for this experiment. Choosing d as 3, 36 dishes and 216 agar plates will be used for this experiment.

Methodology

<u>Selection of regular dishes and sterilization</u>: 8.5 inches, *Corelle* triple-layer strong glass, which is break and chip resistant, lightweight, and dishwasher safe are to be used. These will be autoclaved (250°F for 30 minutes).

<u>Preparation of food suspension</u>: Food suspension, consisting of a mixture of meat, potatoes, pasta, rice and dairy will be prepared using sterile water to form a thick paste-like suspension.

Inoculation of dishes with food: 2ml of this food suspension will be spread on the dishes with a sterile spreader and washed in a dishwasher, hand washed or incubated for t hours, and then washed, as indicated for the particular dish.

<u>Culturing the samples:</u> All the samples will be serially diluted up to X10⁻¹⁰. 2ml from each diluted sample will be used to inoculate on nutrient agar (NA) containing peptone 5.0g, HM peptone B# 1.5g, yeast extract 1.5g, sodium chloride 5.0g, and agar 15.0g in 1000mL of distilled water, through spread plate inoculation technique. However, since Nutrient Agar does not support the growth of a few microbes, we plan to inoculate the samples in other general media as well. This would include Tryptic Soy Agar (general media for most fastidious bacteria) and Potato Dextrose Agar (general media for growing most molds). The plates can then be incubated at 37°C or as required, for 24 hours or more until colonies are seen.

<u>Counting bacterial and fungal colonies</u>: The plates showing a better number of colonies are to be selected for counting and the dilution factor of that plate is to be noted down for the calculation of Colony Forming Units (CFU) per milliliter of the starting culture. An automated colony counting equipment will be used for counting colonies.

Analysis

Mean CFUs and their standard deviations from different wait times will be estimated for the experimental group and will be compared with those from the negative control group and the positive control group. Plots of CFUs will be visualized as a function of wait times for each of the three groups. We will attempt to use these results to either accept or reject our hypothesis. If the results are inconclusive, we will evaluate if increasing the value of d would help in achieving statistical significance.

Aim 2

Comparison of pathogens on clean dishes from a dishcare with a dishwasher

<u>Null hypothesis:</u> Cleaning soiled dishes with dishcare has no effect on the number of pathogens on clean dishes.

<u>Alternative hypothesis:</u> Cleaning soiled dishes with dishcare reduces pathogens on clean dishes.

Experimental Design Strategy

Sterile dishes, autoclaved at 250°F for 30 minutes, will be used in all groups below. Food suspension will consist of a mixture of common food consumed by people, including meat, potatoes, pasta, rice, and dairy.

<u>Experimental group</u>: This will consist of autoclaved dishes coated with a food suspension and left waiting for t hours, where t will be set to be one of 0, 8, 12, 24 hours for each dish in the group. For each wait time t, d number of dishes will be prepared. Therefore, the experimental group will comprise 4d dishes in total. Dishes in this group will be cleaned in dishcare. Two swab samples, first immediately before cleaning and second immediately after cleaning, will be obtained for each dish. Each swab sample will be used to innoculate 3 different types of agar plates. In total, there will be 24d agar plates for this group.

<u>Negative control group</u>: This will consist of autoclaved dishes without food suspension, left waiting for t hours, where t will be set to be one of 0, 8, 12, 24 hours for each dish in the group. For each wait time t, d number of dishes will be prepared. Therefore, the negative control group will also comprise 4d dishes in total. Dishes in this group will also be cleaned in dishcare. Two swab samples, before and after cleaning, will be obtained for each dish. Each swab sample will be used to innoculate 3 different types of agar plates, resulting in 24d agar plates for this group.

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Potential Problems and Contingency Plans

- 1. As dishcare is in development, dishcare prototypes may exhibit technical issues. To overcome this, we plan on building three dishcare prototype units for this study.
- 2. We do not exactly know what types of microbes we may encounter in this study. This is why we have proposed to use three different types of agar plates so we can grow common bacteria and fungi. If we suspect that there might be other species that are not able to grow in the media we have chosen, we will consider expanding our agar plate types further.
- 3. There is a chance of contamination of samples in the experimental group. However, we expect that any such contamination should be also observable in the rigorous controls we have chosen for the study.
- 4. Growing pathogens in growth media poses risk of contracting infections. We will follow rigorous microbiology lab practices, including using laminar flow hoods and autoclaving of used agar plates in autoclave bags prior to disposal.

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