

Chickensplash! Exploring the health concerns of washing raw chicken

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ABSTRACT

The Food and Drug Administration recommends against washing raw chicken due to the risk of transferring dangerous food-borne pathogens through splashed drops of water. Many cooks continue to wash raw chicken despite this warning, however, and there is a lack of scientific research assessing the extent of microbial transmission in splashed droplets. Here, we use large agar plates to confirm that bacteria can be transferred from the surface of raw chicken through splashing. We also identify and create a phylogenetic tree of the bacteria present on the chicken and the bacteria transferred during splashing. While no food-borne pathogens were identified, we note that organisms in the same genera as pathogens were transferred from the chicken surface through these droplets. Additionally, we show that faucet height, flow type, and surface stiffness play a role in splash height and distance. Using high-speed imaging to explore splashing causes, we find that increasing faucet height leads to a flow instability that can increase splashing. Furthermore, splashing from soft materials such as chicken can create a divot in the surface, leading to splashing under flow conditions that would not splash on a curved, hard surface. Thus, we conclude that washing raw chicken does risk pathogen transfer and cross-contamination through droplet ejection, and that changing washing conditions can increase or decrease the risk of splashing.

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INTRODUCTION

Food poisoning and cross-contamination are serious issues for home cooks. According to the Center for Disease Control and Prevention (CDC), *Salmonella* traced to chicken caused 8 outbreaks, 307 illnesses, 42 hospitalizations, and 1 death in the United States in 2016.¹ Between 2009 and 2015, 12% (3114 illnesses) of all outbreaks in the United States caused by a single food source were traced to chicken.² In an effort to reduce cross-contamination and food borne illness at home, the CDC,³ the United States Food and Drug Administration (FDA),⁴ the National Health Service (NHS),⁵ and the Department of Agriculture (USDA)⁶ all currently recommend against

washing raw chicken prior to cooking due to the risk of microbial transfer through splashed water droplets. Despite this guidance, many home cooks continue to wash their chicken before cooking.⁷ It is still an unanswered question whether or not the resulting splashed droplets provide a significant vector for microbial transfer.

In 2013, the USDA launched a campaign against washing raw chicken based on a 2003 study directed at understanding how far water droplets could travel outside of a sink during a rinse;⁸ the study used red food dye in water to assess the distance that ejecta could travel, but no microbiological analysis was performed to determine if viable microbes were present in these droplets. Furthermore, microbial

studies of splatter outside the sink from raw chicken were not found in the literature, though the danger of splashing has been reported and debated in the media.^{9–11} Splashing from sink-trap reservoirs in hospital sinks has been known to cause infection in patients,¹² but the full extent of danger from kitchen sink contamination through droplets is not known.

Extending the work in the 2003 study, we combine techniques from microbiology with fluid experiments to show that viable populations of bacteria can be transferred from the surface of raw chicken to the rest of the kitchen through droplets ejected during washing. Culturable levels of bacteria in the same genera as known pathogens will grow on media where the droplets land. This provides the first experimental evidence that washing raw chickens can lead to significant cross-contamination in a home kitchen. Many home cooks will continue to rinse their poultry before use, however, and the relevant question then becomes how to minimize the splatter risk associated with washing.

Single water droplets splashing from a hard surface have been extensively studied in recent literature.^{13–21} This is in contrast to the literature on droplet formation and ejection from an aerated fluid stream, common in sink faucets, impinging on soft materials, such as a chicken. We see that aerated and non-aerated flows initiate splashing and jetting distinct from each other as part of an exploration of different splash parameters, such as the faucet height, the flow rate, and the aeration level. Furthermore, we find that the geometry and material properties of the surface in conjunction with the flow rate and distance between the faucet and the splash surface appear to have the largest effect on splashing, and thus contamination. These results suggest that the risk of transferring pathogens while washing could be minimized by tailoring the washing conditions to minimize splashing.

EXPERIMENTAL SETUP

Chickensplash setup

Sterile water system

The setup for the splash trials is shown in Fig. 1. An autoclavable Nalgene carboy was used to hold the water, and a pipe system was developed to control the flow rate using gravity. The flow rate at various faucet heights was tuned to obtain flows of roughly 45 and 75 mlps in aerated and laminar conditions, respectively. Tuning was done by adjusting the height and diameter of the pipe shown in Fig. 1 to increase the total column height of the water. The flow rate was

measured using a graduated cylinder and a timer. The flow rates were within 5 mlps of the desired flow rate during testing. A standard junior-sized aerator (~ 14 mm inner diameter) from the lab sink was used for all splash trials. A smaller diameter for laminar flows was used to keep the height needed to initiate pearling flow within a reasonable range for the experiments (60 cm). The faucet height at each flow rate was controlled by increasing or decreasing the height of the Nalgene container, accomplished mainly by stacking objects and adjusting shelf height. Heights were within ± 5 cm. Black plastic garbage bags held up by a wooden frame were used to contain splashing and allow for easy sterilization between trials. The black also provided a contrasting background when videos were taken.

Chicken preparation and swab collection

Eight chicken breasts without skin were purchased from a local grocery store. Two brands were chosen, so that there were four breasts from each brand. The top, side, and bottom of the chicken breast were swabbed with sterile, individually wrapped with cotton swabs (Puratin), and then stored in microcentrifuge tubes at -80°C until further analysis (see “swab and filter processing” below). To make the bacterial contamination on the breasts more uniform for later trials, all chicken breasts were placed in an autoclave tray and mixed by hand to enhance the transfer of bacteria after swabbing. 100–500 ml of autoclaved water was added to enhance bacterial transfer. The breasts were then stored at 4°C for approximately 65 h. This process was repeated for eight chicken thighs with skin during later trials.

Plate preparation

Large baking sheets (18×24 in.²) were used to collect splash drops. Tryptic soy agar (TSA) purchased from VWR (Radnor, PA, USA) was prepared and poured according to the directions on the package, with 40 g of agar for every 1 l of distilled water. Each plate requires approximately 1 l of agar. Plates were covered by a second backing sheet and sealed by taping all edges with either masking tape or autoclave tape. Plates were stored in 4°C prior to experiments.

Trials

For each trial, the water system was set to the proper height and flow rate. Heights were checked prior to each trial. The system was then sanitized using fresh, 10% bleach by volume. The chicken breast or thigh was placed on a black platform, inside a shallow autoclave

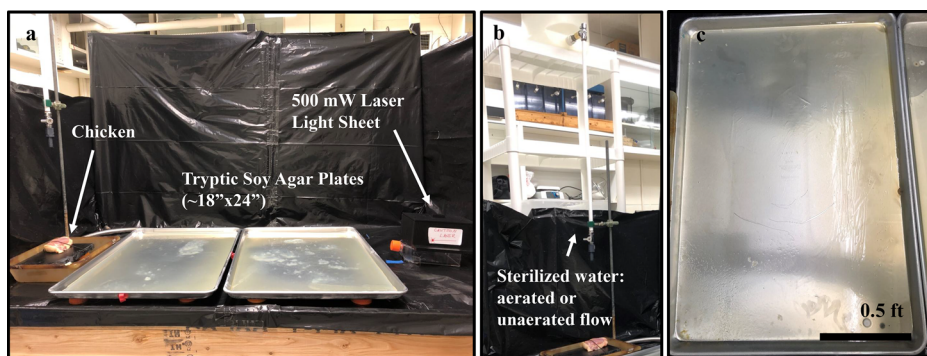


FIG. 1. Splashing experimental setup. (a) Horizontal view of the splashing setup with a splash platform, chicken, an overflow container, and two agar plates. The 500 mW laser light sheet is positioned to illuminate this plane as well. (b) Sterile flow setup with two spigots and a Nalgene container. (c) The control agar plate shows minimal microbial growth when water was splashed from a flat platform without raw chicken under the same flow rate conditions.

tray, as shown in Fig. 1. The chicken piece was held on with a rubber band in the figure, but we later found the chicken did not move during trials. Therefore, in trials done for microbial analysis, the chicken was set on the platform, but not secured. Each chicken piece was positioned, so that the thick, smooth part of the chicken was under the faucet. After the chicken was positioned, both agar plates were placed side by side on the platform as shown. The water was turned on for 20 s and then turned off. Videos were taken on an iPhone for documentation and later comparison. Trials at 15 cm had minimal splashing, so a single plate was used to collect splash drops for those trials.

Fluid and splash analysis

Tracking splash trajectory

We start by placing a high-speed camera (VEO 710L, Phantom) 2–3 m away from the splash setup, so that the entire splash trajectory (5 m) will fit in the frame. We then create a light sheet running horizontal to the camera. In early trials, we used two flood lights covered with cardboard or plastic sheets, and in later trials, we use a 500 mW laser with a Thorlabs plano-concave cylindrical lens ($f = -7.7$). The laser and lens were held in a custom-made 3D printed container, which is shown in Fig. 2 below. We then turn off the overhead lights and videotaped the splash trial at 400 frames per second.

We then run the video through a MATLAB²² particle tracking script based on a MathWorks documentation²³ and overlay the video frames to map splash trajectory. The particle tracking script works by first using a background subtraction model combined with blob analysis to identify the moving objects in each frame. The code then determines which detections correspond to the same object over time based on motion. Generally, detections are identified in the first frame, and then a Kalman filter predicts where the detection will occur in the next frame. The filter compares detections in the second frame to the expected locations of detections in the first frame and determines the likelihood of each detection in the first frame corresponding to the detection in the second. If the detections are within a threshold, the detections are assigned to a track. In this case, one track corresponds to the total motion of one droplet over time. The number of training frames and the size thresholds used to detect and eliminate particles



FIG. 2. Container and lens used to secure the laser and create a light sheet.

were adapted slightly in this documentation, but the otherwise the function and process were left the same. The tracking software was mainly utilized to help distinguish between drops and background noise in the video.

Splash close-ups and overlays

For close up videos used to analyze stream breakup and splashing, the same Phantom camera was placed between 0.5 and 1 m away from the splash point, and videos were taken at 1500 fps. Floodlights were used to illuminate the surface. A cue ball was used to simulate a solid surface, and a stress ball was used to study splashing from a soft surface. Trials were run for 15–30 s. Splash droplets were easier to see in the close-up videos, so the particle tracking software was not used here. Images were processed in MATLAB to make them binary, and then the video frames were overlaid. All videos used in Figs. 8–11 below can be found on Dryad here: <https://doi.org/10.5061/dryad.4xgxd25bz>.

Non-sterile water system

Since a sterile water system is not necessary for the fluid analysis trials, we use a standard lab water system for the fluid flow. Tubing was connected to the lab faucet and was held at a specified height above the splash surface using an O-ring stand and standard clamps. Heights for pearling and non-pearling flow were determined by increasing faucet height until the slow-motion videos showed flow breakup consistent with pearling. Flow rates were set by rotating the faucet to the same point and then confirmed for each trial by filling a graduated cylinder for 5 s immediately after the video. All flow rates were within ± 5 mlps of listed values.

Microbiology sample collection

Plates

After the water was turned off, the lids were placed back onto both baking sheets, and re-secured with masking tape. The plates were labeled and incubated at room temperature (20–25 °C). Images of the growth on each plate were taken after 48 h and one week. Colonies that appeared different visually at these time intervals were collected using plastic inoculating loops and streaked onto TSA plates with 1% glucose by weight. All plates were incubated at 37 °C for 24 h.

After 24 h, individual colonies were isolated from the plates and smeared across fresh plates (TSA agar + 1% glucose). These plates were incubated in 37 °C for another 24 h. After incubation, frozen stock solution was prepared, and all isolates were stored at –80 °C in 25% glycerol (Sigma-Aldrich, Prague, Czech Republic) with 75% Brain Heart Infusion (Oxoid, Hampshire, United Kingdom).

Water and filters

The water collected in the shallow autoclave tray was poured through a funnel into a 1 l pyrex bottle. The funnel was sanitized between uses using 10% bleach and allowed to dry before being used on the next trial. Following splash trials, a house vacuum line was used to pull 100 ml from each bottle through 0.22 μ m, 47 mm diameter filters (MilliporeSigma, Burlington, MA, USA). Three filters were created from each splash trial. Two filters were placed on TSA plates with 1% glucose and grown at 37 °C for 48 h for colony counting. All filters

were found to be >100 CFU/100 ml. One filter was placed in a falcon tube using forceps and frozen at -80°C for later use. See “swab and filter processing” for information about further analysis.

Microbiology sample analysis

Plates

We were unable to amplify directly from culture stocks, potentially due to inhibition from glycerol or other additives. As a result, colonies were propagated on TSA plates.

To replat, microcentrifuge tubes were either thawed for one minute in a 37°C water bath or placed into a microcentrifuge hot plate and checked about every minute until thawed. $30\ \mu\text{l}$ of the solution was then pipetted onto 12 cm TSA plates. Two to five shaker beads were added to the plates and shaken to spread out the sample. The small plates were then left in 37°C for approximately 24 h, or until growth was observed.

DNA was extracted from individual colonies using the One-Tube Tissue DNA Extraction Kit (Bio Basic, Ontario, Canada) according to the manufacturer's instructions, but at one-tenth the recommended volumes. Full-length 16S rRNA genes were amplified using the 27F (5'-AGAGTTTGATCCTGGCTCAG-3') and 1492R (5'-GGTTA CCTTGTACGACTT-3') primer pair with the OneTaq Hot Start PCR 2X master mix (New England Biolabs, Ipswich, MA, USA) according to the manufacturer's instructions in $25\ \mu\text{l}$ reactions with $0.2\ \mu\text{M}$ primers and 5–10 ng of template DNA. No template and positive control samples were included in the polymerase chain reaction (PCR) preparations. PCR amplification was performed using a Mastercycler ProS thermocycler (Eppendorf) with the program: 94°C for 1 min, followed by 30 cycles of 94°C for 20 s, 58°C for 15 s, 68°C for 40 s, and a final extension at 68°C for 5 min. The PCR products were assessed using agarose gel electrophoresis, and positive amplicons were sent for bi-directional Sanger sequencing analysis (Functional Biosciences, Madison, WI, USA) using the same set of primers. Sequences were deposited in GenBank (NCBI) under accession numbers OM070120–OM070334.

Filters and swabs

A DNeasy PowerWater Kit (Qiagen, Hilden, Germany) was used to isolate DNA from the filters and swabs. For filters, all steps were followed as outlined in the Power Water Kit Handbook, except that the filters were frozen at -80°C for a few months before isolation. We also note that a fast prep bead beater (MP Biomedicals, FastPrep 24 Bead Beater, Irvine, CA) for 5 ml bead tubes was not available, so tubes were strapped to the fast prep bead beater with tape, rather than placed in the tube holders. All tubes appeared thoroughly mixed after this process.

The collection process for the swabs was described above in “chicken preparation and swab collection.” To isolate DNA, each swab was placed in a bead beater tube. The stem was broken off as needed to seal the bead tubes closed. Steps 4–23 were then followed as described in the QIAGEN Power Water Kit Handbook, except again being frozen at -80°C and being strapped onto the bead beater as described for the filters above.

A DNA Clean and Concentrate Kit (Zymo Research, Irvine, CA, USA) was used to concentrate DNA from both filters and swabs. As noted, some concentrations were still too low for PCR analysis.

However, some samples were successful, and those results are reported below.

We prepared samples from extracted filters and swabs for Illumina MiSeq sequencing using a two-step PCR method. The bacterial and archaeal community was amplified with the “universal” primers F515A (GTGYCAGCMGCCGCGGTAA) and R806B (GGACTACNVGGGTWTCTAAT). PCR was performed in $25\ \mu\text{l}$ reactions using Phusion Hot Start II High Fidelity polymerase (ThermoFisher) with the following thermocycler program: Initial denaturation at 98°C for 30 s, 22 cycles of denaturation at 98°C for 15 s, annealing at 58°C for 15 s, extension at 72°C for 20 s, and a final extension at 72°C for 5 min. Residual salts, primers, and potential dimers were removed using the Ampure XP bead clean up with a 1:1.3 sample:bead ratio. Sequences from the first PCR were barcoded using the Nextera kit (Illumina) in a second $25\ \mu\text{l}$ PCR reaction using Phusion Hot Start II High Fidelity polymerase with an annealing temperature of 55°C for ten cycles. PCR2 products were purified using beads as above, and we quantified the amplicons with fluorometry using the Quant-It dsDNA kit (Invitrogen) and combined individual samples at equimolar concentrations. The final multiplexed library was quantified using the Qubit HS dsDNA kit (Invitrogen) and adjusted to 9 pM for sequencing. Paired-end 300 bp sequences were generated using the Illumina MiSeq platform with 10% PhiX added to increase sequence diversity. Sequences were deposited into the Short Read Archive (NCBI) under BioProject ID PRJNA808656.

Forward and reverse sequences were merged using USEARCH.²⁴ Due to the full-length overlap of the forward and reverse reads for all targeted genes, we increased the number of allowed mismatches to 20. Primers were trimmed, and merged sequences were quality filtered to remove any sequence with an expected error rate greater than 1.0. Zero-radius operational taxonomic units (ZOTUs) were identified using UNOISE3²⁵ pipeline, which performs error correction of sequences to separate biological sequences from those generated from PCR or sequencing errors. Representative sequences for ZOTUs were classified using SINTAX against two databases: the Ribosomal Database Project (RDP) database for 16S (v18)²⁶ and the Genome Taxonomy Database (GTDB) SSU database (v202)²⁷ with plastid out-group 16S sequences to identify eukaryotes. Sequences identified as eukaryotic or with low certainty at the domain level were removed from downstream analysis. To generate phylogenetic trees, environmental 16S sequences were aligned to full-length reference bacteria + archaea sequences using mafft (v7).²⁸ A maximum likelihood phylogeny was constructed using RAXML (v8)²⁹ with the GTR + GAMMA model. Due to the large number of sequences, we included only Zotus with a relative abundance greater than 0.1% in any sample. The resulting phylogeny was annotated using the Interactive Tree of Life.³⁰

RESULTS AND DISCUSSION

Bacterial transfer from the surface of raw chicken

To determine if splashing can transfer bacteria from the chicken surface to nearby counters, we developed a sterilized flow system and placed tryptic soy agar plates nearby to collect splashed droplets (see Experimental Setup, *Chickensplash setup: Sterile water system*). The system is shown in Figs. 1(a) and 1(b). To verify whether the system is sterile, we perform a control by splashing sterilized water from the splash platform without raw chicken. We observe minimal signs of

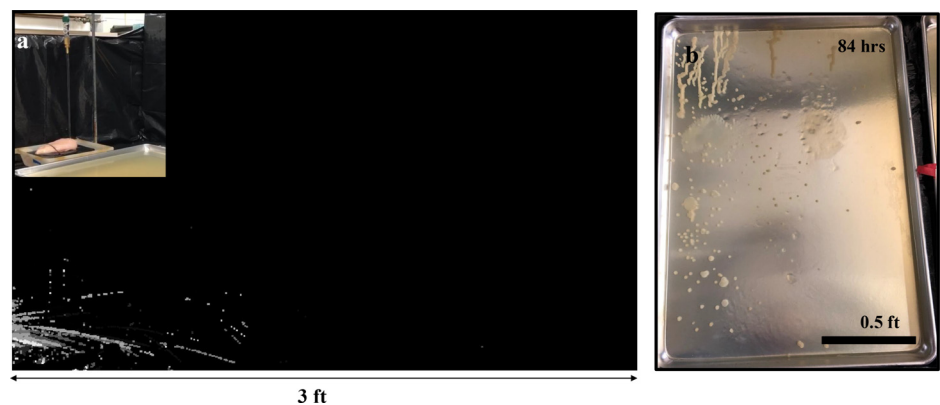


FIG. 3. Bacterial transfer through splash drops. (a) Overlay of all video frames taken by a high-speed camera during splashing. Faucet was 35 cm above the chicken surface, and the flow rate was 100 mlps. Total video time was 30 s. (b) Bacterial growth on a tryptic soy agar plate 84 h after the splash trial.

TABLE I. Values tested for each variable in chickensplash experiments.

Variable	Values	
Chicken type	With skin	Without skin
Flow rate	45 mlps	75 mlps
Faucet height	15 cm	40 cm
Flow type	Un aerated	Aerated

microbial growth after two weeks in 20 °C [Fig. 1(c)], and as such, we assume the system is sterile.

We then place raw chicken on the splashing platform and splash water at 100 mlps from the chicken surface for 30 s. The faucet was positioned 35 cm above the chicken surface. We also video record the trial using a high-speed camera, track the splashed droplets using MATLAB particle tracking software, and overlay the video frames (see Experimental Setup, *Tracking and splash trajectory*). The overlay allows us to visualize the trajectory of the splash drops and

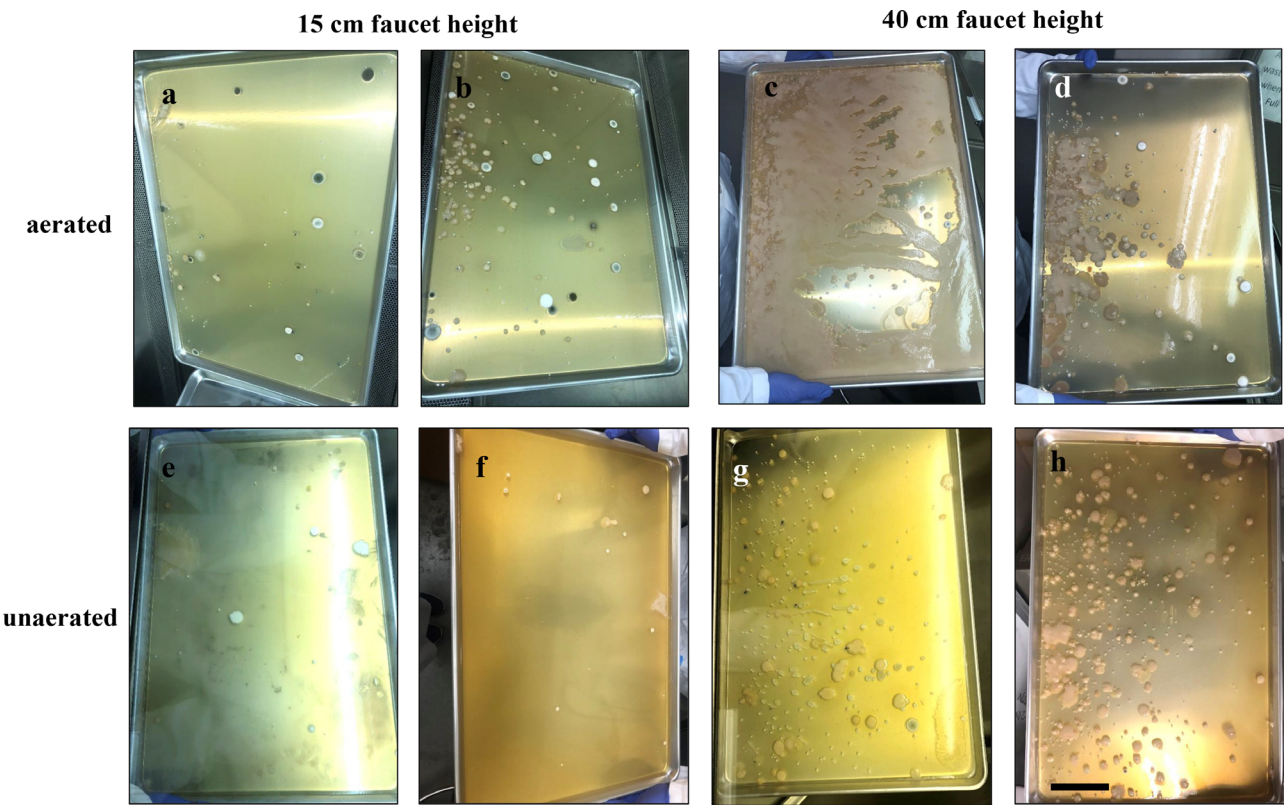


FIG. 4. Images of large agar plates one week after no skin splash experiments show the difference in splashing and splash transfer between 15 and 40 cm faucet heights. (a) Aerated, 75 mlps, 15 cm faucet height. (b) Aerated, 45 mlps, 15 cm faucet height. (c) Aerated, 75 mlps, 40 cm faucet height. (d) Aerated, 45 mlps, 40 cm faucet height. (e) Un aerated, 75 mlps, 15 cm faucet height. (f) Un aerated, 45 mlps, 15 cm faucet height. (g) Un aerated, 75 mlps, 40 cm faucet height. (h) Un aerated, 45 mlps, 40 cm faucet height. Scale bar is 4 in.

approximate the location drops landed on the agar plate [Fig. 3(a)]. After incubating the plates for 84 h at 20 °C, we observe bacterial growth on the plates and find the growth locations closely match the approximate landing location of the drops [Fig. 3(b)]. From this, we conclude that bacteria can be transferred through splashing from the surface of raw chicken.

We note that cooks could wash chicken under a wide variety of faucet conditions, which is likely to impact the amount of splashing that takes place. To determine how washing conditions affect splashing and bacterial transfer, we repeat the splash trial under varied faucet heights, flow types, and flow rates using chicken with and without skin. We chose two options for each characteristic listed, resulting in 16 total trials. A summary of the parameters varied is shown in Table I.

After performing splash trials under each of the conditions listed, we cover the agar plates and incubate them, taking pictures and colony samples at 72 h and at one week (see Experimental Setup, *Microbial sample collection: Plates*). Based on the growth, we note two observations. First, faucet height appears to play the largest role in splash transfer. In both chicken with skin and chicken without skin, the agar plates with a 40 cm faucet height appear to have significantly more area covered in bacterial growth than plates with a 15 cm faucet height.

Second, the flow rate may play a role in aerated flows but does not appear to play a role in unaerated flows. With both skin and no skin, the aerated trials at lower flow appear to have less splash transfer

than the aerated trials at higher flow. We note that this is only a visual observation of growth, not a quantification of bacterial transfer. Thus, the flow rate could result in more bacteria transferred per drop, but a similar amount of splashing.

Furthermore, the aerated trial with skin at a 15 cm faucet height appears to have similar splash transfer at both flow rates, meaning increased splash transfer in aerated flows from a higher flow rate is only true in three of four cases. Thus, observed differences between aerated flow rates may be a result of other factors in the trials. We did not identify any significant differences in bacterial transfer based on the flow type or skin/no skin cases. These factors may play a role when paired with different flow rates or faucet heights than those used here, but we leave this to further study.

We see evidence that the distance between the faucet and the chicken surface impacts bacterial transfer at flow conditions seen in a kitchen sink; however, other factors such as the flow rate, faucet type, and faucet height make little difference under the flow conditions studied. The growth on all agar plates three days after the trials is shown in Figs. 4 and 5.

Fluid analysis

To understand what causes splashing under various flow conditions, we video record faucet flow and splashing using a high-speed

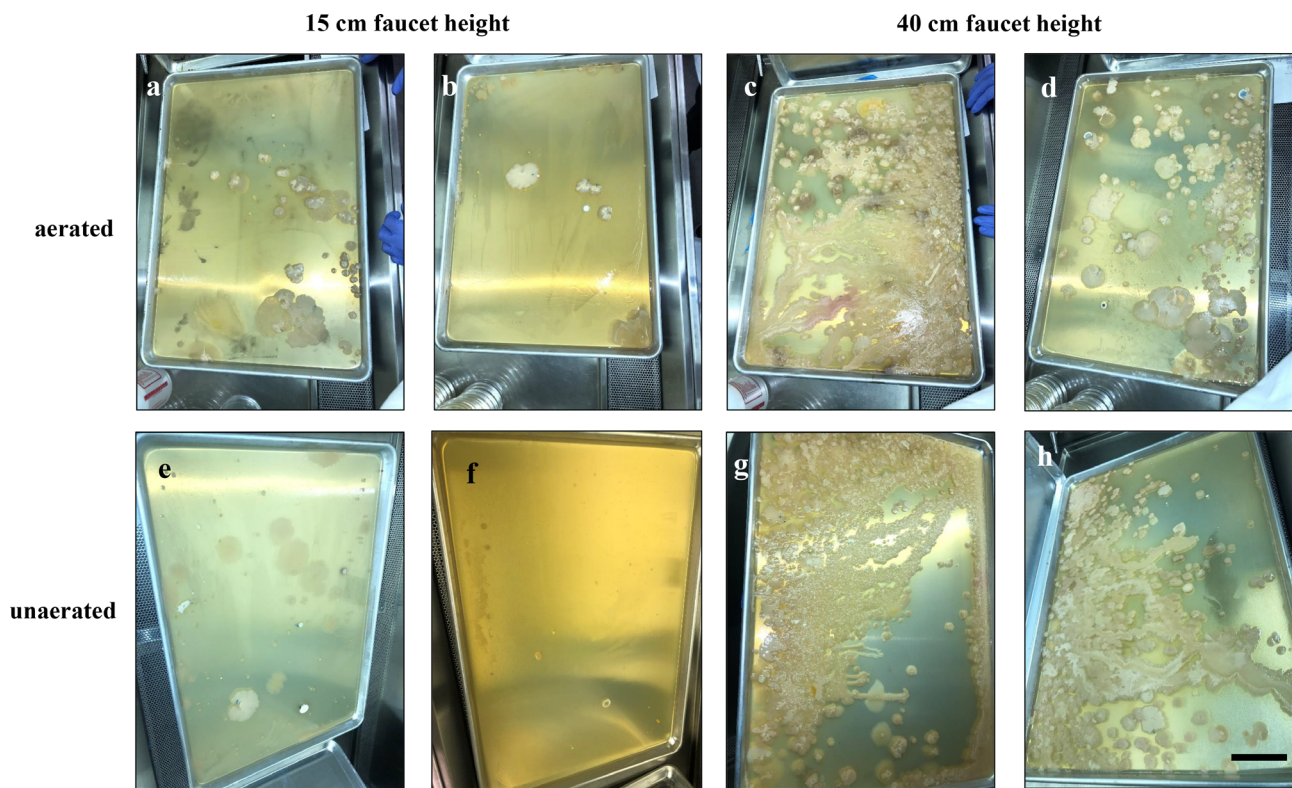


FIG. 5. Images of large agar plates one week after with skin splash experiments show the difference in splashing and splash transfer between 15 and 40 cm faucet heights. (a) Aerated, 75 mlps, 15 cm faucet height. (b) Aerated, 45 mlps, 15 cm faucet height. (c) Aerated, 75 mlps, 40 cm faucet height. (d) Aerated, 45 mlps, 40 cm faucet height. (e) Unaerated, 75 mlps, 15 cm faucet height. (f) Unaerated, 45 mlps, 15 cm faucet height. (g) Unaerated, 75 mlps, 40 cm faucet height. (h) Unaerated, 45 mlps, 40 cm faucet height. Scale bar is 4 in.

camera (see Experimental Setup, *Fluid and splash analysis: Splash close-ups and overlays*). Under many flow conditions, the Plateau-Rayleigh instability will break the fluid stream into individual droplets, which is expected to occur when the stream length reaches roughly 3.13 times the diameter of flow. We will refer to this as pearling throughout the rest of the paper. We find that pearling and jetting play the largest role in splashing under the studied faucet conditions. As such, we start by looking at the impact of faucet height in the un-aerated flow. As expected, there is a critical height where the laminar stream breaks apart into pearling flow [Figs. 6(a) and 6(b)]. We note that once this flow breakup begins to occur, splashing becomes much more prominent. To correlate pearling flow and increased splashing more directly, we again use the high-speed camera set up to measure splash trajectories. Using a cue ball as the splash surface, we vary height and the flow rate and then measure the height of the highest splash trajectory in the trial. We find, as before, that flow rate has

minimal impact on splashing, but that faucet height impacts splash height significantly [Fig. 6(c)].

At first, we assumed pearling was the main source of splashing in our trials, as splash coverage was closely related to faucet height. However, a wider diameter outlet was used for these experiments to accommodate the sterile water system, so the water stream was not fully broken up at the 40 cm height. As such, the faucet flow used in the experiments with chicken did not splash much from a cue ball at 40 cm. However, significant splashing does occur during the chicken splash trials based on the agar plates. To explore this difference, a deformable stress ball was used as a smooth analogue to the soft chicken for further flow experiments. The high-speed videos of the same faucet flow splashing from a hard surface (cue ball) and a soft material (stress ball) are compared in Fig. 7. The faucet height was set to 40 cm, as minimal splashing was seen at 15 cm. Aside from a transient splash on initial contact between the fluid jet and the cue ball, the cue ball had minimal splashing while the stress ball

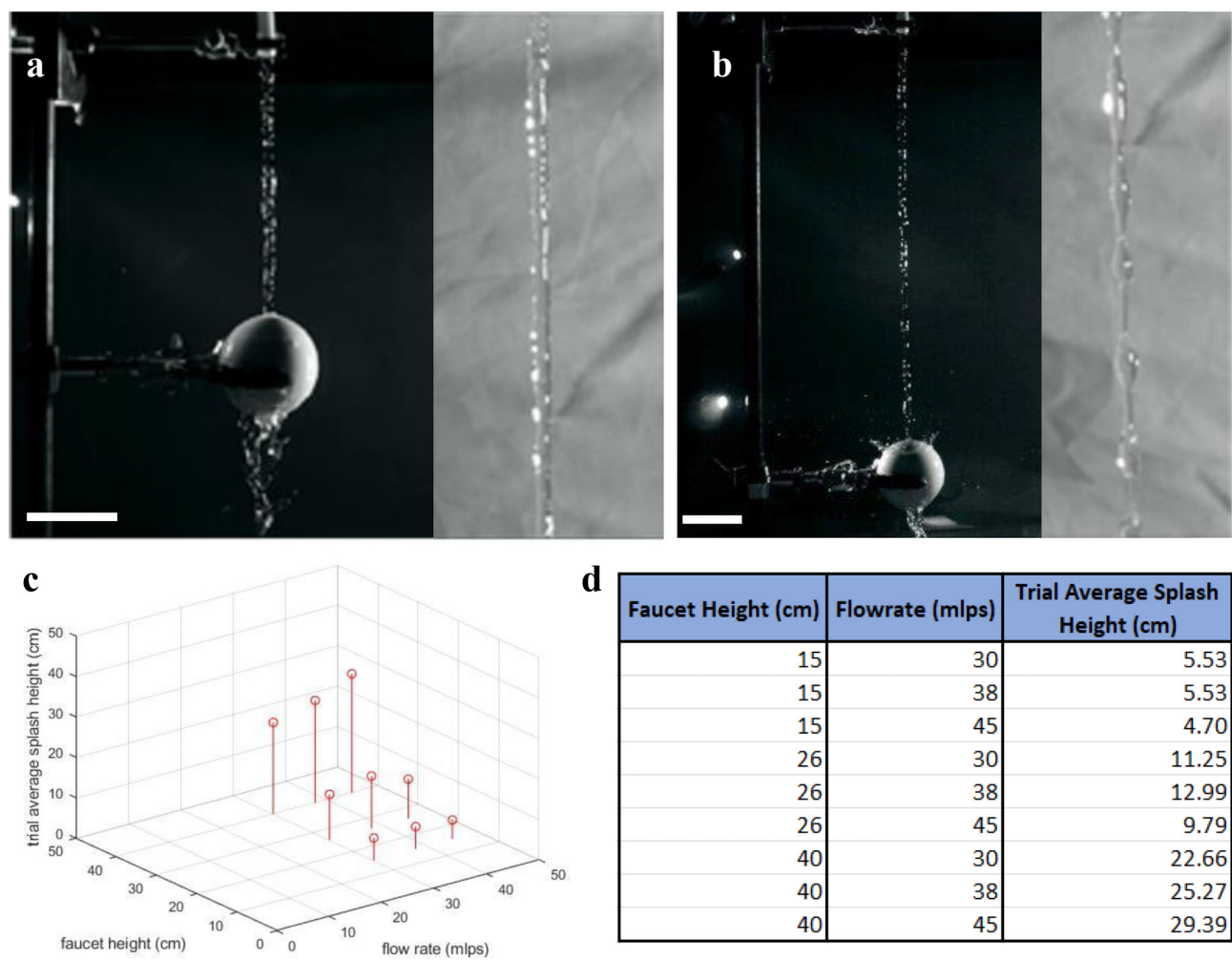


FIG. 6. Pearling flow increases splashing. (a) At a lower faucet height, the flow remains connected, and there is minimal splashing. (b) At a 40 cm faucet height, the flow breaks into the pearling regime and begins to splash. (c) Measuring the highest splash height at various faucet heights and flow rates shows that faucet height has a high impact on splash height, while the flow rate has minimal effect. (d) Data points shown in (c). Scale bars 2 in.

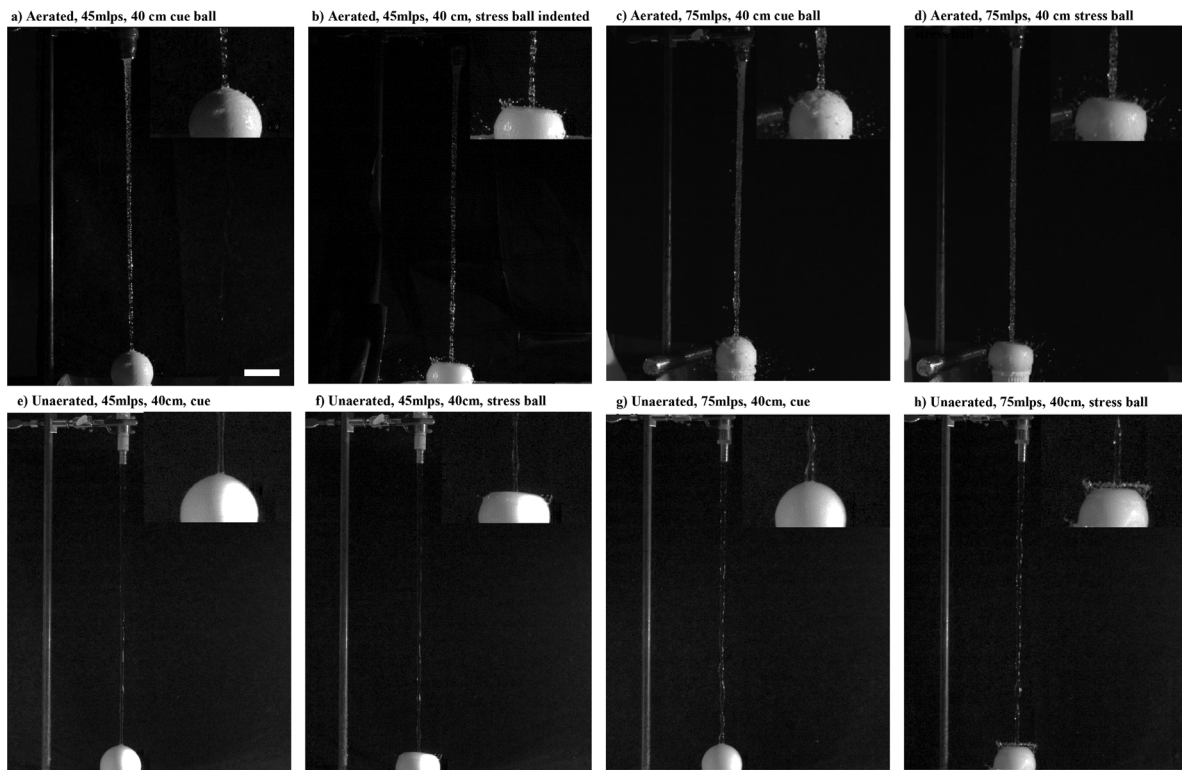


FIG. 7. Splashing conditions used for the chickensplash trials at a 40 cm faucet height. (a) Aerated, 45 mlps, cue ball. (b) Aerated, 45 mlps, stress ball pre-indented. (c) Aerated, 75 mlps, cue ball. (d) Aerated, 75 mlps, stress ball. (e) Unaerated, 45 mlps, cue ball. (f) Unaerated, 45 mlps, stress ball pre-indented. (g) Unaerated, 75 mlps, cue ball. (h) Unaerated, 75 mlps, stress ball. Scale bar is 2 in.

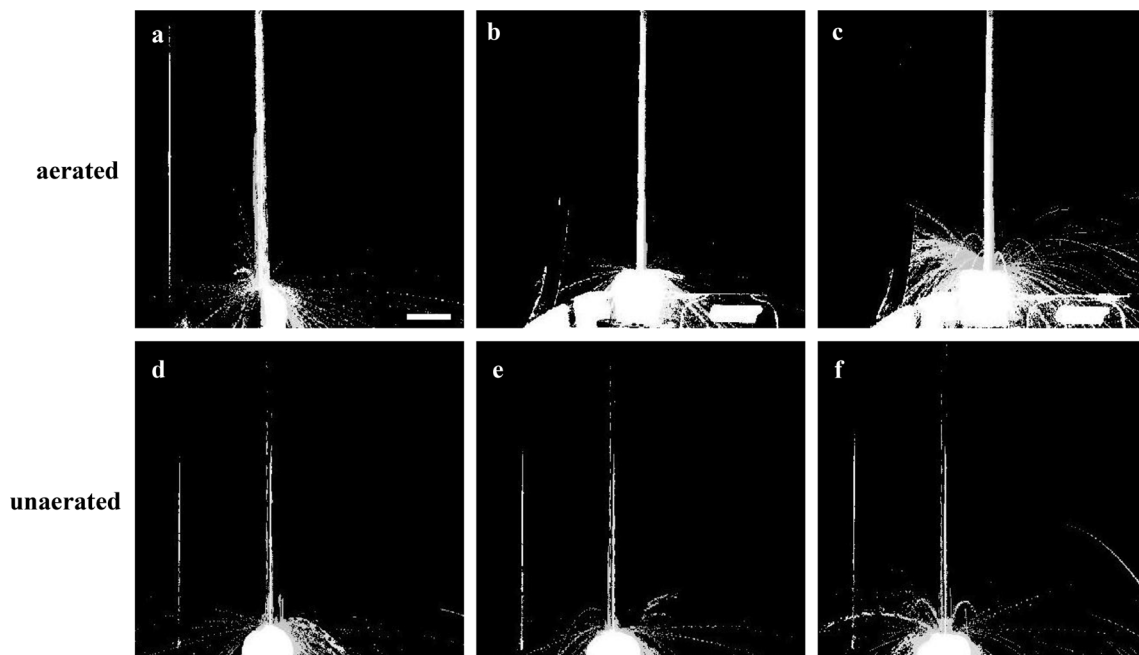


FIG. 8. The effect of surface indent on splashing. (a)–(c) Aerated flows splashing from (a) a cue ball, (b) a stress ball with a slight indent, and (c) a stress ball with a larger indent added manually before the trial. (d)–(f) Laminar flows splashing from (d) a cue ball, (e) a stress ball with a slight indent, and (f) a stress ball with a larger indent added manually before the trial. All trials performed at a 40 cm faucet height and 45 mlps flow rate. Images shown are an overlay of a high-speed video taken at 1500 fps. Scale bar is 2 in.

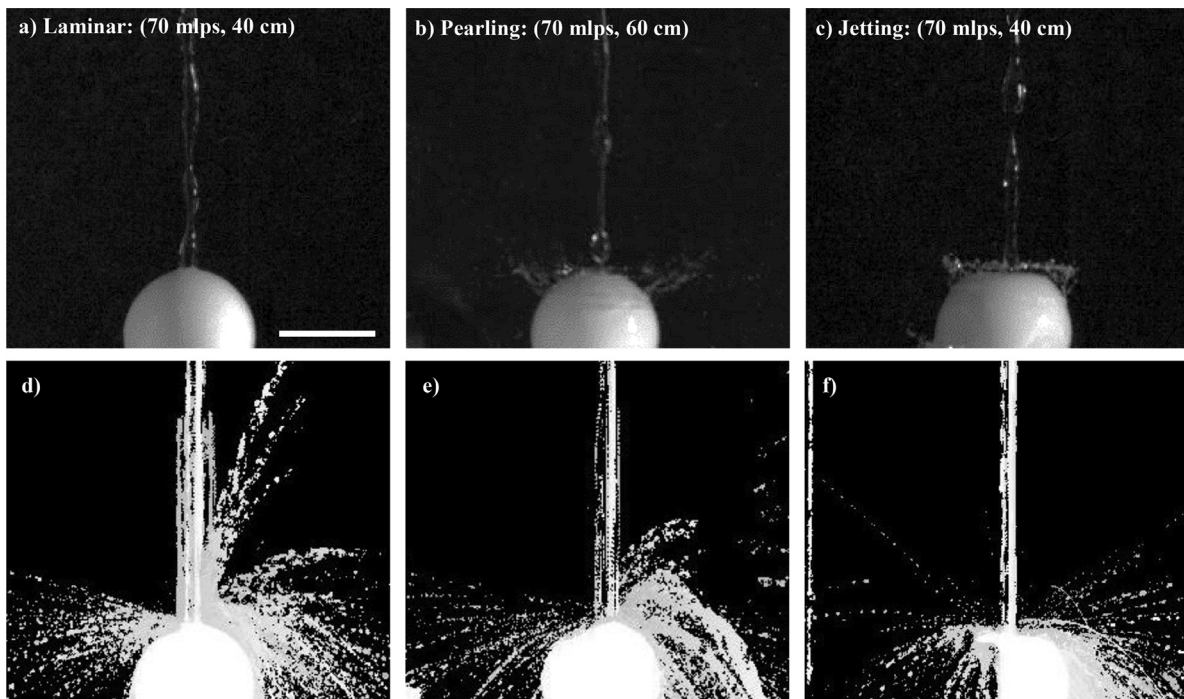


FIG. 9. Splash conditions in laminar flows with a flow rate of 70 mlps. (a) When the faucet is 40 cm from the chicken surface, the flow is laminar. There is a large initial splash, but minimal additional splashing (see Fig. 11). (b) At 60 cm, the flow begins to pearl, and the splash arcs upward from the cue ball. (c) At a 40 cm faucet height on an indented surface, the laminar flow is launched off, and splashing is caused through jetting. (d)–(f) Overlaid frames from the videos of splashing parameters in images (a)–(c), respectively, show differences in overall splash trajectory in each case. Scale bar is 2 in.

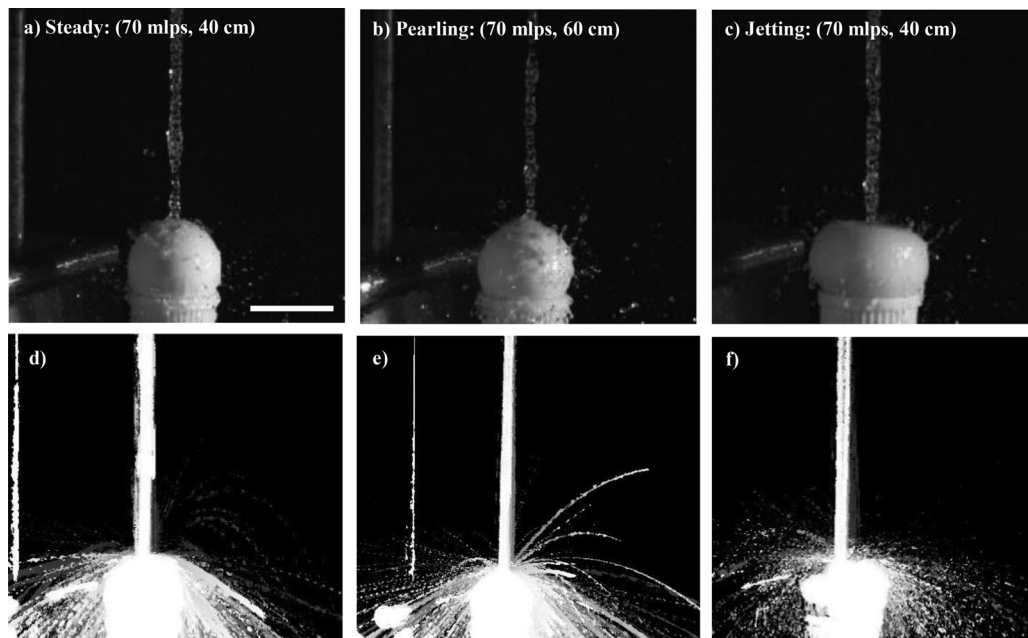


FIG. 10. Splash conditions in aerated flows with a flow rate of 70 mlps. (a) When the faucet is 40 cm from the chicken surface, the flow is laminar and has minimal splashing. (b) At 60 cm, the flow begins to pearl, and the splash arcs upward from the cue ball. (c) At a 40 cm faucet height on an indented surface, the flow is launched off and splashing is caused through jetting. (d)–(f) Overlaid frames from the videos of splashing parameters in images (a)–(c), respectively, show differences in overall splash trajectory in each case. Scale bar is 2 in.

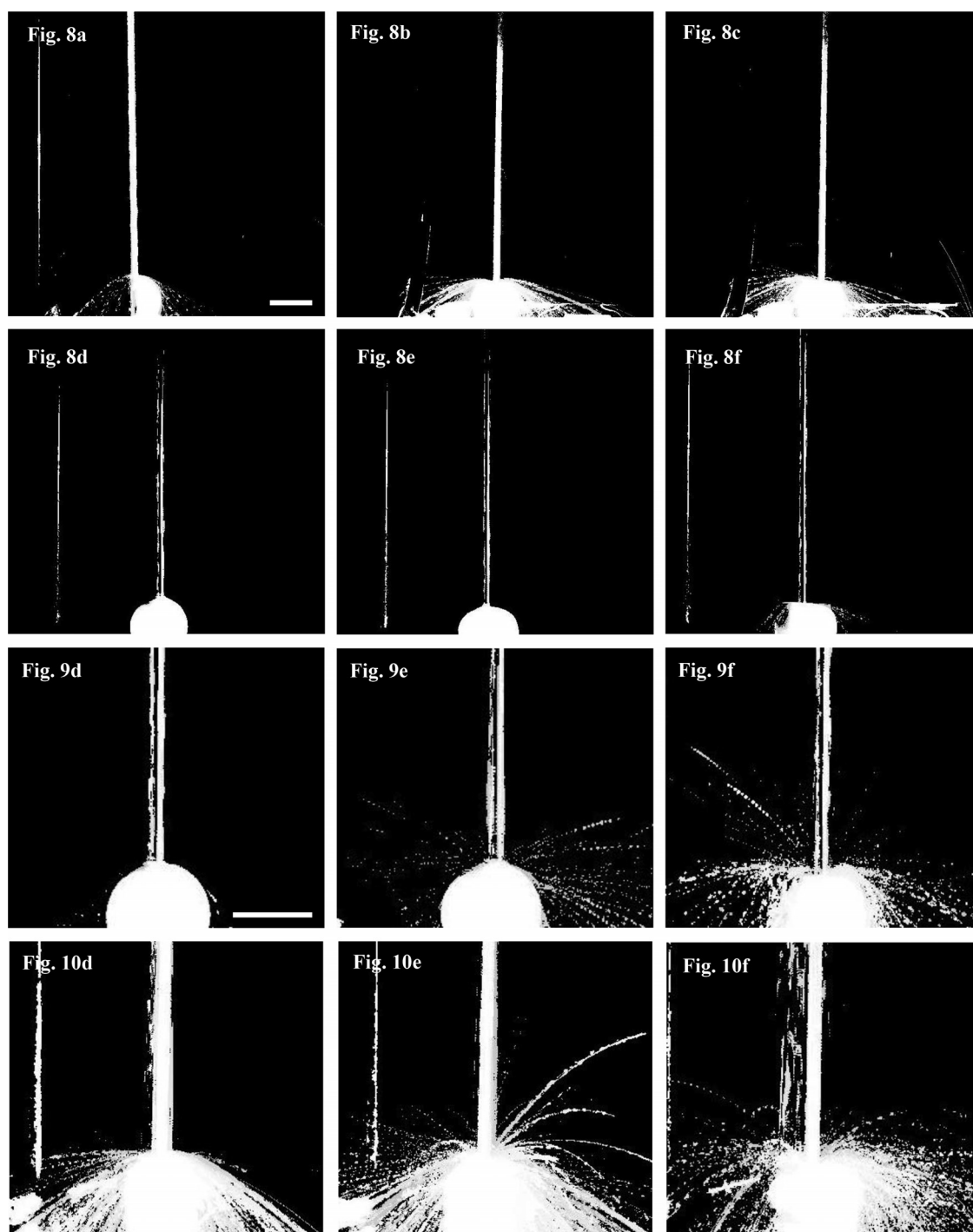


FIG. 11. Splash overlays without initial splash. All images show the overlay of the image referenced in the upper left corner of each image with the initial splash cropped out of the video.

splashed regularly. Thus, we suggest that surface geometry also plays a major role in splashing by increasing or decreasing jetting.

Increased splashing from a soft surface makes intuitive sense. The divot creates a concave shape that launches the water stream off the

surface, analogously to holding a spoon under the faucet. Notably, however, the stream must carry enough force to create a large enough divot on the soft material for this to be the case. When the indent was manually increased, splashing increased [Figs. 8(b), 8(c), 8(e), and 8(f)].

Additionally, pearling does still appear to play an important role in droplet ejection from the stress ball. While a full breakup of the flow did not occur in any of the cases shown in Fig. 8, the formation of a crown and the ejection of drops from the crown in the indented stress ball case always seemed to occur when the convex portion of the flow (wider diameter) hit the surface of the stress ball. This observation is also somewhat supported by the minimal splashing seen at the 15 cm faucet height. In this case, it seems that either the fluid stream did not carry enough force to indent the chicken to the point of splashing or the flow was still laminar.

Pearling and jetting appear as significant mechanisms in droplet ejection for both laminar and aerated faucet conditions (Figs. 9 and 10). Aerated flows are expected to behave differently from non-aerated flows due to the interactions between the air bubbles and the fluid stream; this difference can be seen in Fig. 8 and by comparing Fig. 9 with Fig. 10. It is important to note that the aerator used in this study had a greater diameter than the unaerated flow, so the flow streams are not an exact comparison. See Experimental Setup, *Chickensplash setup: Sterile water system* section for discussion on diameters chosen.

With that noted, we expect that the aerated flow will require a higher faucet height to fully break apart into a pearling flow than the 60 cm used for the laminar case. However, at 60 cm, the aerated stream appears to be in the early stages of breaking up into droplets, similar to those seen in the 40 cm laminar flow stream [Figs. 10(a) and 10(b)]. In general, the characteristics of multi-fluid faucet streams have not been

well-characterized, so this opens a door for future general research into multi-flow streams.

Finally, we note that initial splash plays a major role in the droplet transfer that occurs throughout the trials. Figure 11 shows the same information seen in Figs. 8–10, but with the initial splash removed. The decreased splashing in all images shows the importance of gradually initiating flow when washing raw chicken.

Nevertheless, cooks who wish to wash their chicken should know that increasing faucet height plays the largest role in splashing. Care and common sense should be used to balance the trade-off between blocking splash drops by placing poultry lower in the sink and increasing splashing by the corresponding increase in faucet height. Furthermore, a great deal of splashing can occur from the initial impact between the fluid stream and the chicken. This can be controlled through carefully initiating washing.

Microbial analysis

Above, we found that bacteria can be transferred from the chicken surface, but not all bacteria were from taxonomic groups known to be harmful to humans. To assess the risk of transferring harmful bacteria when washing chicken, we also collected microbial samples from the chicken surface, the run-off water, and the tryptic soy agar plates (see Experimental Setup, *Microbial Sample Collection: Plates, Filters, and Swabs*). We analyzed these samples and compared

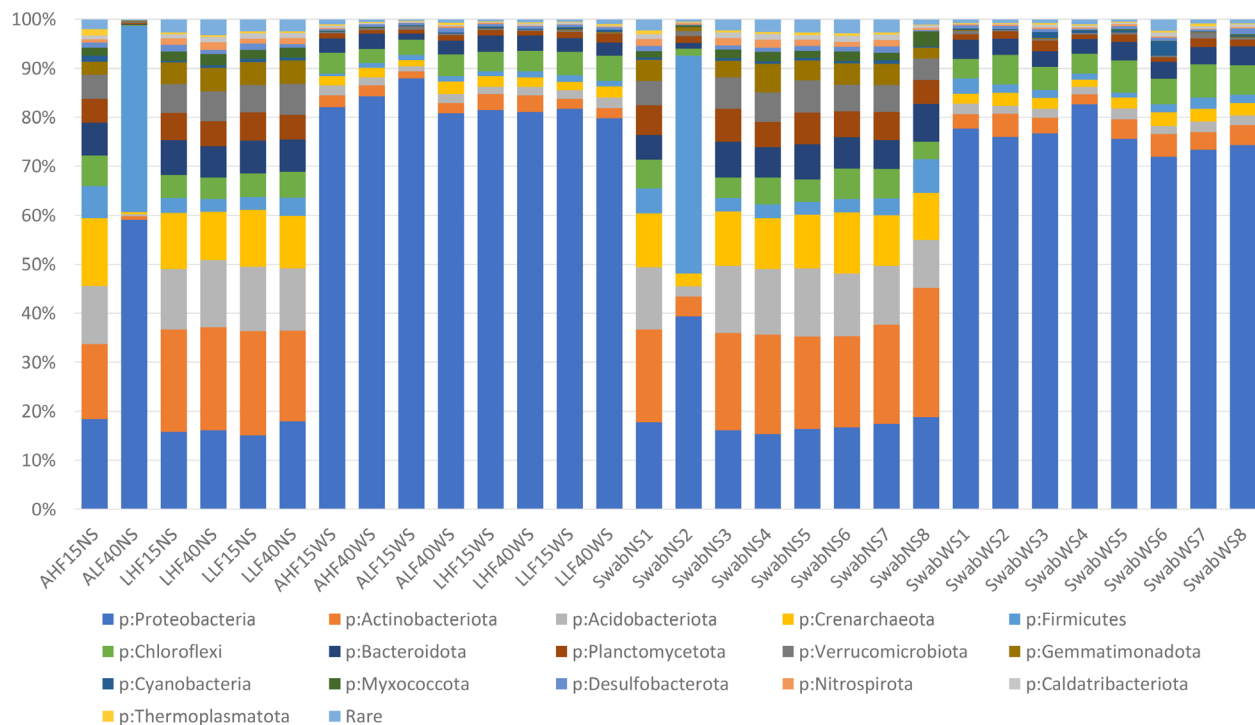


FIG. 12. The relative abundance of bacteria and archaea phyla detected on swabs and filtered water from samples with and without skin. Samples with and without skin were highly dissimilar, whereas we detected a high level of similarity between filtered water and swabs within these two groups (with and without skin). The first eight x-axis labels represent data from the filters and can be interpreted as follows: flowtype (A = aerated/L = unaerated), flow rate (HF = 75 mlps/LF = 45 mlps), faucet height (15 = 15 cm/40 = 40 cm), and chicken type (NS = no skin, WS = with skin). For example, AHF15NS is the aerated trial at high flow (HF = 75 mlps), a 15 cm faucet height, and chicken with no skin (NS). The last eight x-axis labels represent data from swabs of the chicken surface. NS = no skin and WS = with skin.

bacteria found on the tryptic soy agar plates to bacteria found on the chicken surface and in the run-off water. Phylogenetic markers such as the 16S rRNA gene are known to be poor indicators of pathogenic organisms due to the conserved nature of this marker across pathogenic and nonpathogenic strains but can be used to determine both the diversity of organisms present of surfaces and identify genera that contain potential and putative pathogens.

The culture-based analysis identified several genera that contain known human pathogens, including *Aeromonas*, *Pseudomonas*, *Serratia*, and *Staphylococcus*. Of these, *Aeromonas* is perhaps the most concerning, given that the majority of species identified to date are human pathogens, though they cause relatively treatable diseases, such as bacterial gastroenteritis.³¹

High throughput sequencing revealed a highly diverse community of both bacteria and archaea (Fig. 12). Like many microbial communities, the distribution of organisms was highly skewed, such that 25 zero-radius operational taxonomic units (ZOTUs) comprised over 50% of the total sequences generated. The high dominance of these particular taxa, which were primarily species in the Proteobacteria

and included *Ralstonia*, *Curvibacter*, *Pseudomonas*, *Carnobacterium*, *Afipia*, *Acidibacter*, *Delftia*, *Thermogemmatospira*, *Arthrosira*, *Yersinia*, *Cutibacterium*, and *Bacillus* and the archaeal genera *Candidatus Nitrocosmicus* and *C. Nitrososphaera*, suggests that while high numbers of bacteria and archaea are transferred to chicken during processing and other activities, the growth of these groups is likely favored on chicken surfaces. In comparing the experimental factors that led to differences in microbial composition, the strongest driver of community differences was the presence or absence of skin. Swabs from the surface of the chicken and filtered water samples taken after splashing experiments were similar in composition, indicating that a high proportion of the organisms present on the surface of raw chicken are dislodged by water flows. A small subset of the organisms identified through high throughput sequencing was subsequently cultured (Fig. 13); however, the culture conditions represent a small range of conditions under which bacteria and archaea are able to grow (e.g., relatively high levels of nutrients and higher temperatures). In residential kitchens, it is likely that a wider range of organisms present could establish on nearby surfaces, such as kitchen sponges, but this requires additional experiments.

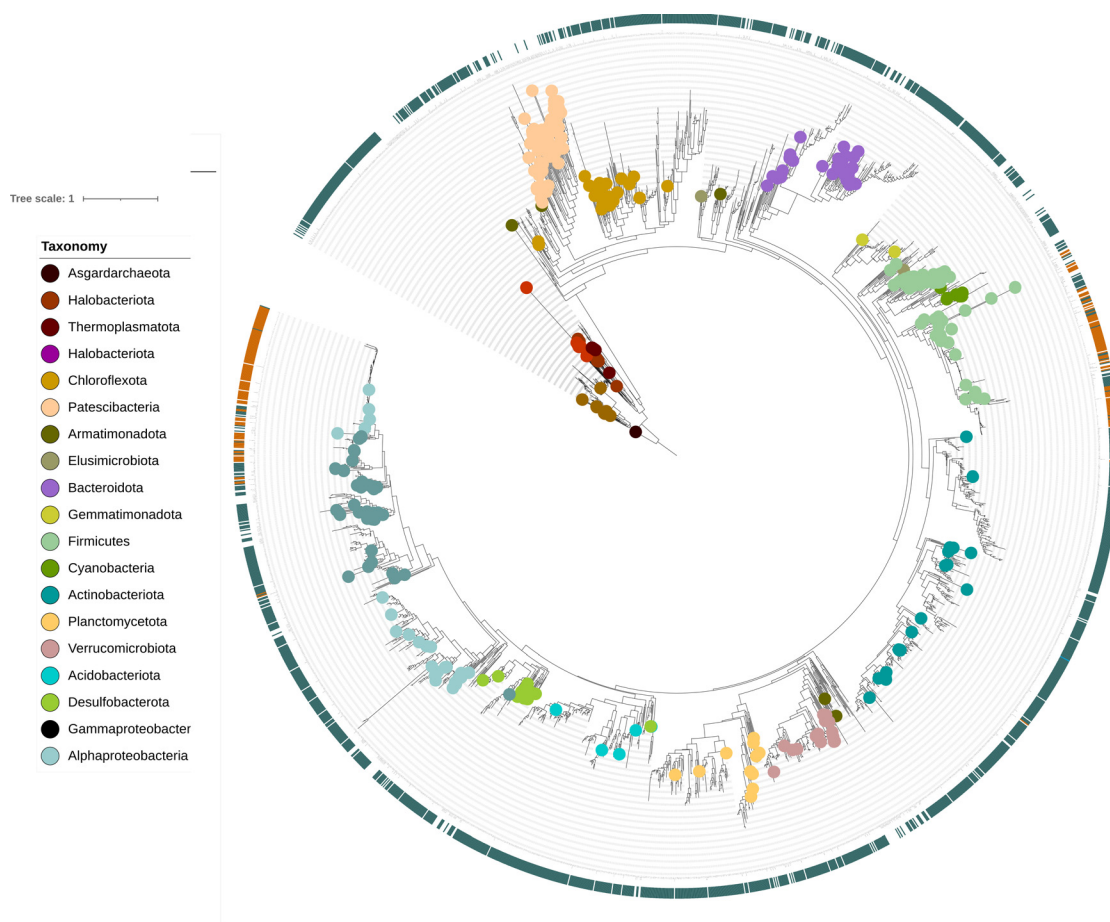


FIG. 13. A phylogeny of identified reference sequences (colored circles) with environmental sequences (outer ring). The phylogenetic breadth of microbes detected in chicken surfaces, both from swabs and filtered water (blue outer ring), included both bacterial and archaeal taxa and was greater than the organisms cultured from TSA plates (orange outer ring) at 32 °C, which were concentrated primarily in the Proteobacteria (Alphaproteobacteria and Gammaproteobacteria) and the Firmicutes. This indicates that the diversity of culturable organisms present on chicken surfaces may be greater than detected in this study, given the wider range of environmental conditions present in residential kitchens.

CONCLUSIONS

Washing chicken in the sink under standard kitchen faucet conditions can eject droplets containing culturable levels of pathogens throughout the kitchen. Regardless of this risk for cross-contamination, many home cooks are likely to continue rinsing their chicken before cooking and consumption. By varying the flow rate and the height from the faucet to the chicken's surface, careful washing can minimize the likelihood of splatter and cross-contamination. Slowly initiating flow from the faucet near the chicken can reduce the initial fluid on surface impact and the corresponding droplet ejection. However, sustained flow impinging on a soft material can actually create structure in the material that leads to jetting flows, and the possibility of more ejecta with a long wash.

Our preliminary exploration of water streams creating droplets after impact with soft materials revealed a host of interesting areas for further study such as the effect of aeration on stream and splashing behavior and the relative effectiveness in removing surface contamination between these flows for different flow conditions. We see an interesting interplay between droplet formation and jetting as the soft material deforms in response to different flow conditions. A whole host of interesting phenomena related to the interplay between aerated and non-aerated fluids interacting with soft biologically relevant materials remain to be studied.

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AUTHOR DECLARATIONS

Conflict of Interest

The authors have no conflicts to disclose.

DATA AVAILABILITY

The data that support the findings of this study are available from the corresponding authors upon reasonable request.

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