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A clean sweep: Enhancing “flushing” as a dry sanitation strategy for low moisture food manufacturers

ABSTRACT

Cleaning and sanitation operations are essential for low moisture food processors to maintain hygienic conditions and reduce the risk of product cross-contamination. However, the introduction of water from typical wet sanitation methods into a low moisture food environment may in some cases increase risk by elevating microbial growth of environmental pathogens like *Salmonella*. Alternatively, dry cleaning methods such as brushing, scraping, and product flushing as well as dry sanitizing methods (formulated oil-flushes) can reduce microbial contaminants without the risk of moisture introduction, but are typically less effective than aqueous sanitizers at microbial inactivation. This trade-off in risk reduction between wet and dry cleaning/sanitation operations in a low moisture food environment is poorly understood in part because of a lack of understanding about the efficacy of dry cleaning and sanitation methods. In this article, we explore the advantages and limitations of dry flushing methods as a cleaning strategy based on the available research. Flushing typically uses dry food material as the physical force to dislodge cells and the vector to carry them out of the system. Flushing treatments may also act as a sanitation step by incorporating heat or chemicals in flushing materials such as oil.

Maintaining hygienic conditions in low moisture food environments is inherently challenging. Sanitation programs usually consist of both cleaning and sanitizing strategies. Cleaning removes food residue and potential pathogens from the system, which is then followed by a sanitization step to destroy the remaining pathogens. Bench-scale experiments typically show that conventional wet sanitation methods are more effective (i.e., result in more significant microbial reduction) than standard dry cleaning methods. However, introducing water into these environments generally increases the risk of pathogen harborage, growth, and dispersal. Many dry

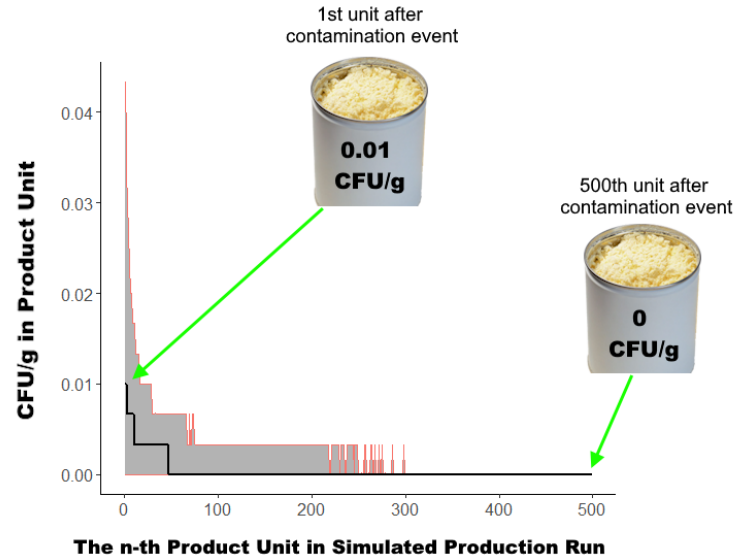
cleaning methods rely exclusively on physical removal. Methods such as brushing, wiping, scraping, and product flushing that “rub off” cells from surfaces. Flushing, sometimes called dry rinsing, purging, or push-through, is a cleaning strategy where dry food material is run through the processing line and serves as the physical force to dislodge cells and the vector to carry them out of the system. Additionally, some flushing treatments incorporate a sanitization element through heat or chemicals to inactivate pathogens.

The theoretical advantage of flushing is that it can be applied to a large surface area in a processing line without the need to disassemble equipment and introduce water. Despite being commonly used, limited published research exists on the efficacy of flushing as a cleaning intervention to remove microbial contaminants from a processing line (1), and the research that does exist is often specific to an individual piece of equipment. The efficacy of flushing is dependent on the hygienic design of the equipment (i.e., “dead zones” on the processing line where flushed material does not contact but may still be a harborage point for microorganisms), parameters of the flushing process, and flushing material (i.e., shear stress). The large diversity of commercial processing lines means that some operations, and even specific areas within the line of a given operation, will be better suited to flushing than others. For example, operations where product is simply recirculated, as is common for flour mills, would be cleaned less effectively through flushing. Economic and sustainability burdens must also be balanced due to the potentially large amounts of flushing material required for an effective intervention. This article is to summarize the available research on flushing as a dry cleaning intervention and identify areas for future research in this field.

Summary of prior flushing research: important considerations in evaluating efficacy

The Snyder lab has used simulation models to estimate the efficacy of product flushing after a *Salmonella* contamination event in a simplified, theoretical milk powder processing line (2). *Salmonella* transfer from a food contact surface into milk powder during a contact event was measured through benchtop experiments and used as input parameters for the model (3). The model was used to estimate the prevalence and concentration of contaminated milk powder products after a *Salmonella* contamination event. We found that simulated units produced immediately after a contamination event received the bulk of the *Salmonella* contamination, leaving little remaining to transfer into subsequent units (**Figure 1**).

Figure 1. The median (black line) and 5th/95th percentiles (grey shaded area) for the concentration of *Salmonella* in simulated milk powder product units produced directly after a 2-log CFU contamination event. The median amount of contamination dropped from 0.01 CFU/g to 0 CFU/g after producing 500 units (150 kg of milk powder).



We then modeled the impact of different dry sanitation interventions. After a 2-log CFU contamination event, the number of consumer-size milk powder units (300 g) contaminated with *Salmonella* was 72 [24, 96] (median [p5, p95] across 1,000 simulation iterations). The average concentration of *Salmonella* within contaminated units was -2.33-log CFU/g [-2.46, -1.86]. While wiping with a dry towel reduced the average number of contaminated units to 26 [12, 64], product flushing with 150 kg of milk powder reduced the number of contaminated units to 0 [0, 41]. These results suggest that product flushing may reduce product contamination after an equipment contamination event without the use of water. However, this model only considers a contamination event on a flat stainless steel surface measured through benchtop experiments. Transfer dynamics on a real processing line will likely differ within a niche caked with residual food product. Therefore, building a more representative model will require collaboration with industry partners to better define the surfaces, equipment, and niches that act as vectors of microbial contamination. Following this, model validation will be a critical step towards leveraging these tools to make decisions on risk tradeoff between different cleaning strategies with and without water.

Other flushing research has been conducted on the pilot scale. For example, Warren et al. (2018) have shown that flushing was an effective method for removing milk chocolate from equipment surfaces (4). In this study, authors ran liquid milk chocolate over a stainless steel pipe and butterfly valve, as well as through a pilot scale system, followed by milk-free dark chocolate. The authors evaluated dark chocolate for the presence of residual milk allergen after flushing and after the use of a silicone pig to purge chocolate from the pipe. While the initial dark chocolate samples off the line contained high levels of milk allergens, milk levels were below the limit of quantitation (2.5 ppm) for the ELISA assay after removing 13 to 15 kg of dark chocolate. Use of the pig dramatically reduced initial milk levels, but >18 kg of dark chocolate flushing was still needed to decrease milk levels below 2.5 ppm. Although these findings are specific to one model

system, it is an indication of the utility of flushing for reducing milk allergen residues from equipment surfaces.

One of the challenges in effectively flushing is ensuring consistent contact between the flushing material and all of the internal equipment surfaces. Therefore, hygienic design and assessment of the dry flushing process are essential and understudied factors. In many real-world systems, a myriad of niches reduce the efficacy of dry flush procedures. Investigating these niches within existing equipment and improving the hygienic design of new equipment are essential to improving the application of dry flush procedures. For example, in a previous study by Steinbrunner et al. (2021), the authors studied the fate of *Salmonella* and the surrogate *Enterococcus faecium* during pilot-scale spray drying of soy protein isolate (5). Their results show that while the spray drying process itself results in a modest reduction in viable counts (approximately 2 to 4-log) in both product and residual material accumulated in the dryer's collectors, survivors were also found in many niches within the interior of the dryer. While no dry cleaning intervention was applied in this study, this finding highlights the role of niches within equipment and the need for experimental assessments on the pilot scale rather than on coupon surfaces. A greater understanding of what interior locations within equipment represent "worst-case scenarios" for dry flushing would improve environmental monitoring efforts.

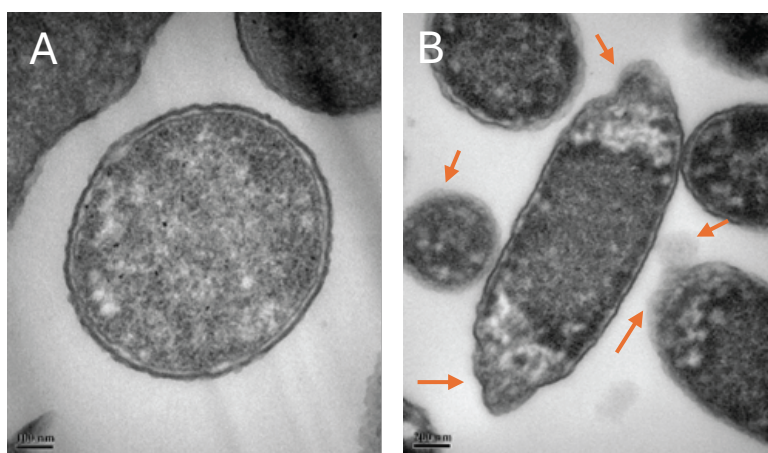
Finally, the physical properties of the material used in flushing influence food safety outcomes. Bench-scale experiments have revealed slight differences in transfer rates based on material type that may correspond to meaningful differences in outcomes when simulated across large production runs (6). Some industries use lactose crystals instead of milk powder in flushing because the increasing abrasiveness of the lactose increases the rate of removal of cells from surfaces. However, examples of less successful flushing materials have also been studied. For example, the study by Grasso et al. (2015) revealed significant limitations in hot oil flushes to inactivate *Salmonella* from peanut butter processing lines (7). Although the oil was 90°C, *Salmonella* survived this treatment and about 3-log CFU/mL oil was recovered. Additionally, surface swabbing recovered detectable *Salmonella* from the processing equipment surfaces. In this instance, the oil temperature did not provide a high level of thermal lethality because of the low water activity level of the oil medium. It must also be noted that the flushing material itself could contain microbial hazards. In a scenario where incoming ingredients are contaminated, flushing the line may spread contamination rather than remove it. Therefore, any risk analysis considering flushing as a cleaning treatment should also consider potential microbial hazards in the flushing material.

Formulated oils to enhance flushing

The McLandsborough laboratory evaluated the antimicrobial efficacy of formulated oils for enhanced destruction of vegetative bacteria, such as *Salmonella*, in oils used for hot oil flushes. The first approach was to acidify oils with organic acids. The rationale is that organic acids in oil

are present in the non-dissociated acid form and would likely make a highly effective antimicrobial. In Ghoshal et al. (2022) (8), after *Salmonella* cells were desiccated to 75% equilibrium relative humidity (ERH), they were treated with 500 mM acetic acid in oil. When combined with mild heat (45°C), a 4.4-log reduction was observed. Differential staining confocal laser scanning microscopy (CLSM) and transmission electron microscopy (TEM) (**Figure 2**), suggest the antimicrobial efficacy was due to cellular membrane damage. However, the decreased efficacy against cells dried to a lower humidity (33% ERH), indicated that acidified oils alone may not provide robust antimicrobial efficacy over a continuum of bacterial dryness. Furthermore, while a 4.4-log reduction was a promising antimicrobial efficacy, sanitizing agents for food processing are expected to show a 5-log decrease in target organisms.

Figure 2. Treatment of desiccated *Salmonella* Enteritidis with peanut oil (A) and peanut oil with 250 mM acetic acid for 15 min at 20°C. Arrows indicate areas of membrane damage, as indicated by ruffling and loss of definition.



In subsequent work, acidified oils were further formulated to enhance antimicrobial efficacy by adding a surfactant and a low water level, creating acidified water-in-oil (A-W/O) emulsions. The A-W/O emulsions were composed of all food-grade materials (peanut oil, acetic acid (200 mM), polyglycerol polyricinoleate n (PGPR (3% w/w), an emulsifier), and water (3% v/v)). The levels of surviving desiccated *Salmonella* and *Listeria monocytogenes* after a 30 min treatment with A-W/O emulsions were below a detection limit of 0.48-log using the Most Probable Number (MPN) method, creating an observed reduction of > 6.52-log MPN/coupon. This level of microbial destruction is similar to water-based sanitizing agents and uses lower amounts of water than typically used in solvent-based sanitizers (30%). Time-course studies demonstrated that the antimicrobial efficacy of 200 mM acetic acid A-W/O emulsions produced >6.52-log MPN/coupon reduction within 20 min, regardless of the cellular desiccation levels (33% or 75% ERH) and antimicrobial treatment temperatures (22°C or 45°C) tested (Figure 3a) (9). After treatment with A-W/O emulsions for 30 min, stainless steel chips were negative by enrichment. Scanning electron microscopy of the samples after treatment shows a sparse population of cells (assumed to be nonviable based on the enrichment results) and larger emulsion droplets on the stainless steel surface (Figure 3b).

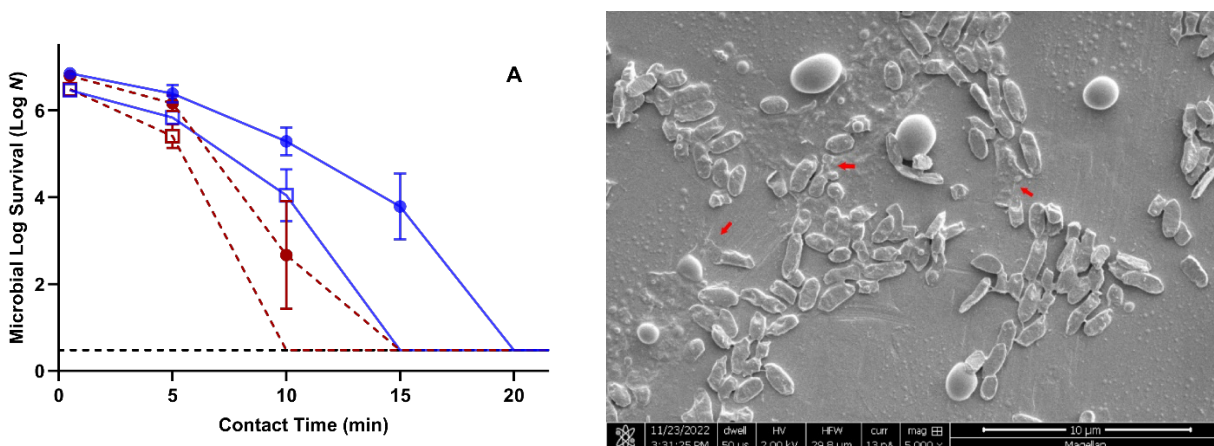


Figure 3. Efficacy of A-W/O emulsion against *S. Enteritidis*. (A) Survival after treatment over time. Cells were desiccated to 75% (closed circle) and 33% (open square) ERH were treated with acidified W/O emulsion (200 mM acetic acid, 3% w/w PGPR, 3% v/v distilled water) at 22°C (solid, blue line) or 45°C (dashed, red line). Per stainless-steel coupon, the inoculum level was 7-log CFU and the detection limit was 0.48-log MPN. The initial time point was 30 sec. Once samples reached below the detection limit (dotted, black line) and remained there upon subsequent incubation, the points were omitted from the graph. (B) Scanning electron micrograph of remaining cells after treatment with A-W/O emulsion for 30 min.

Controlled water dispersion in acidified oils (i.e., A-W/O emulsions) enhanced the antimicrobial efficacy by a pronounced margin (5 logs greater than acid alone) (9). The partition coefficient (K_{ow}) is an estimate calculated based on the equilibrium mole fractions of a substance between two immiscible liquids, usually octanol and water. With a log K_{ow} value of -0.17, acetic acid is more soluble in an aqueous phase than in an organic phase. Thus, creating A-W/O emulsions accelerates bacterial inactivation by allowing the partitioning of acetic acid from the continuous oil phase to the encapsulated water droplets, which carry the acid into the bacterial cytoplasm. Based on the results with reduced osmotic pressure formulations (9,10), it is hypothesized that the enhanced killing by A-W/O emulsions was due to a combination of acid, which causes cell membrane disruption (8), and the differential osmotic pressure created by the water in A-W/O emulsions, lysing the cells with a damaged membrane upon such osmotic downshift (9,10).

Further testing of contaminated peanut butter in a benchtop flow system confirmed that after an oil flush to remove bacterial-contaminated peanut butter, oil flush-cleaning has been validated on the bench scale at 60 °C since this is the temperature at which peanut butter transitions into a molten state and flows freely through tubes. A laminar oil flush (Reynolds number < 100) was applied to clean a tube/flow system loaded with peanut butter inoculated with *Salmonella* to a high level (approx. 9-log CFU/g), and the efficacy of cleaning exhibited a linear relation with

time (**Figure 2**). Subsequently, the cleaned system was subjected to a stagnant treatment with A-W/O at 60 °C. A two-step cleaning (3.6 min) and sanitizing (30 min) procedure reduced *Salmonella* contamination in the tubing to a level negative upon enrichment (11). *Enterococcus faecium* (NRRL B-2354) was tested in parallel and confirmed as a suitable nonpathogenic surrogate for *Salmonella* to validate oil sanitation in the food processing environment. Results to date indicate that A-W/O emulsions showed robust efficacy over various levels of physiological dryness and treatment temperature, had efficacy in a bench-top peanut butter system, and provided a level of antimicrobial efficacy (> 5-log kill) needed for sanitization. Pilot plant-level testing is needed for the future.

Dry flushing for monthly cleaning or when pathogens are detected is practiced (12) however, dry flushing alone is not considered antimicrobial (1). While oil sanitation has excellent potential to increase the frequency of sanitation in low-moisture food processing, there are still several barriers to implementation. However, this process is expensive due to the high amounts of oil needed at the industrial level, and the requirement to ship spent oil to a rendering facility for disposal is unlikely to recoup the entire raw ingredient cost. Furthermore, single-use oil on a large scale is not an environmentally sustainable practice. A logical next step for adoption is to determine if there are processes that will allow for safe oil reuse. Safe reuse of cleaning and sanitation oils will enable future design and implementation of oil-based clean-in-place (CIP) systems, allowing for more frequent cleaning of low-moisture food processing equipment, ultimately improving food safety.

Conclusion

In concept, flushing procedures can reduce the pathogen load on surfaces through physical removal, or thermal and chemical inactivation on vast surface areas in a processing line without the need for water. However, the efficacy of flushing is dependent on numerous factors such as the product being manufactured, the physical and chemical properties of the flushing material, the shear stress generated, and the hygienic design and niches within the processing line. Past research on flushing has elucidated some of these factors, but additional research is needed for improved implementation in the industry. First, more collaboration with industry partners is required to understand the niches in typical processing lines and equipment so that the microbial transfer and inactivation dynamics of these areas can be measured through benchtop and pilot scale work. This work will help define the types of equipment on which flushing will be effective while simultaneously informing the hygienic design of future processing equipment that is designed with dry cleaning in mind. This may also require replacement of some legacy equipment which is not well suited to flushing. Next, validation data for various flushing treatments should be generated at the pilot and industry scale. This will facilitate the development of models and tools that help optimize risk tradeoffs between flushing, other dry cleaning interventions, and wet sanitation methods. A combination of modeling frameworks that can compare tradeoffs in wet and dry sanitation, and novel dry sanitation technologies such as formulated oil flushing, will give the food

industry sector much-needed options for assessing and implementing risk reduction in their operations.

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This SIB was independently reviewed and approved by the IUFoST Scientific Council.

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