

Electrostatic Powder Coating Inoculation Methodology for Dry Sanitation Experimentation

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Abstract

Introduction: Modeling a replicable pathogen cross contamination event in dry processing environments remains challenging. However, simulating electrostatic powder accrual on equipment surfaces may provide an improved representation of cross contamination for dry cleaning and sanitation experimentation.

Purpose: The purpose of this study was to develop a standardized dry inoculation methodology using an electrostatic coating process for dry sanitation experimentation.

Methods: A wheat berry mixture (80/20 ratio of hard winter red and soft white wheat, 1000 g) was inoculated using a six-strain *Salmonella* cocktail, to model contamination of raw agricultural products, and equilibrated to $\sim 0.45 a_w$ in a controlled-humidity chamber for 48 ± 2 h. The wheat berries were fabricated into all-purpose flour using a KitchenAid[®] metal grain mill followed by mechanical sieving. The fabricated flour (~ 8.25 log CFU/g) was applied to #304 and #316L stainless steel coupons (32.18 cm²) using an electrostatic powder coating gun. Samples ($n=48$) were either coated (a positive control) or coated then brushed to visibly clean using a soft bristled brush. *Salmonella* was recovered by hand-massaging bagged coupons with buffered peptone water, which was serially diluted and enumerated on Modified Tryptic Soy Agar.

Results: Within each stainless steel grade, there were significant differences in remaining *Salmonella* populations between coated and brushed coupons ($p < 0.0001$). Coated coupons had 5.45 ± 0.37 log CFU/cm² (#304) and 5.65 ± 0.14 log CFU/cm² (#316L) (mean \pm 95% CI). Brush-cleaned coupons had 3.92 ± 0.36 log CFU/cm² (#304) and 4.10 ± 0.39 log CFU/cm² (#316L). Brush-cleaning reduced *Salmonella* populations on stainless steel surfaces by ~ 1.5 log.

Significance: The dry powder coating inoculation methodology was consistent and replicable at pilot scale. Brushing of contaminated surfaces until they appear visibly clean does not ensure microbial cleanliness.

Background

Recent outbreaks of *Salmonella* within flour products are of increased concern for public health (CDC, 2023). Research toward dry cleaning and sanitation methods using powdered food products is limited. Some methods use an appreciable amount of water during the inoculation process to adhere pathogens to relevant target surfaces (de Oliveira, et. al, 2010). These methods may not reflect actual cross contamination scenarios in dry processing environments, which occur mainly through the accrual of inoculated particles on equipment surfaces.

Determining an inoculation method for dry cleaning and sanitation research that models powder fabrication (wheat milling) will close a knowledge gap in dry sanitation research. This method must adhere enough populations on surfaces to be quantifiable after sanitation treatments. One potential method for modeling dust accrual uses an electrostatic powder coating gun. Electrostatic coating is used in manufacturing to apply powdered paints to electrically grounded metallic objects (Pilcher, 2001; Prasad, et. al, 2016). Food powders can be similarly applied to metallic food contact surfaces using this method, which may provide a novel inoculation strategy for further research.

Objectives

- Develop an electrostatic powder coating dry inoculation methodology for *Salmonella*
- Model dust accrual on equipment surfaces (cross contamination) in low-moisture food processing
- Adhere > 4 log CFU/cm² of *Salmonella* on “visibly cleaned” stainless steel surfaces

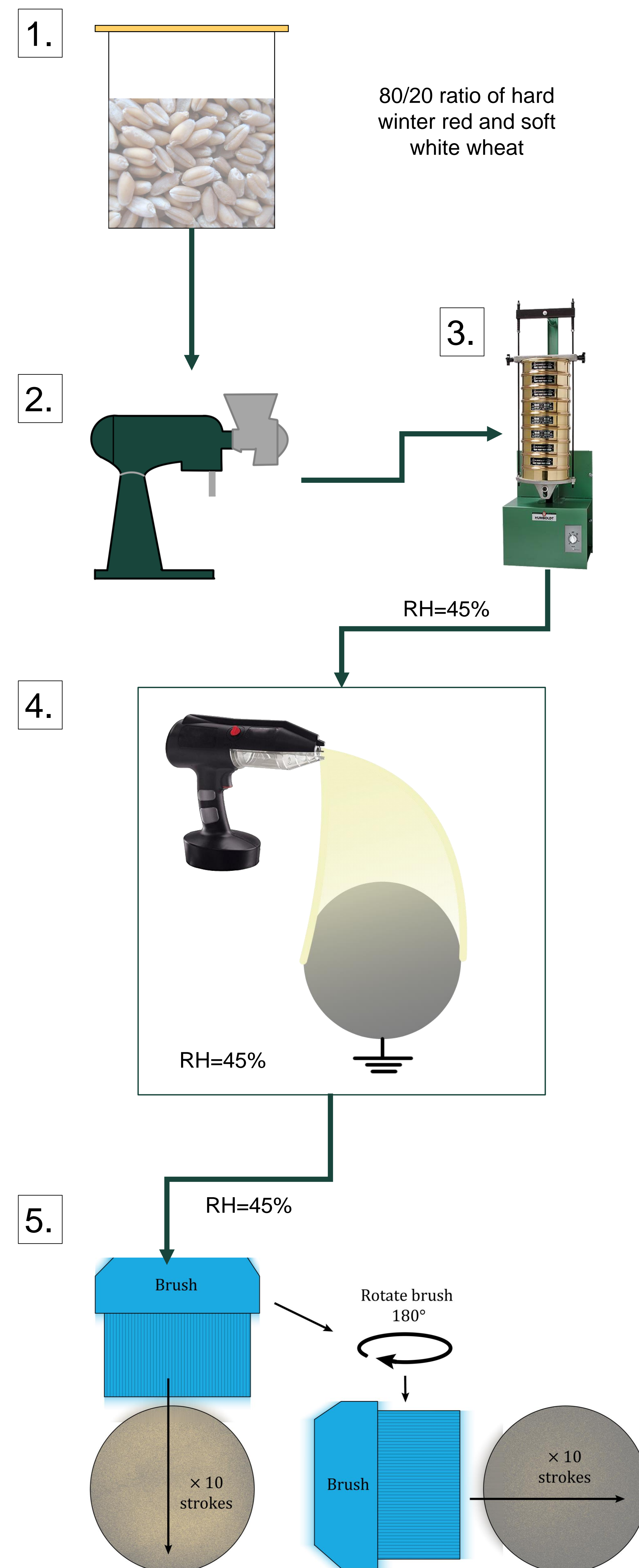
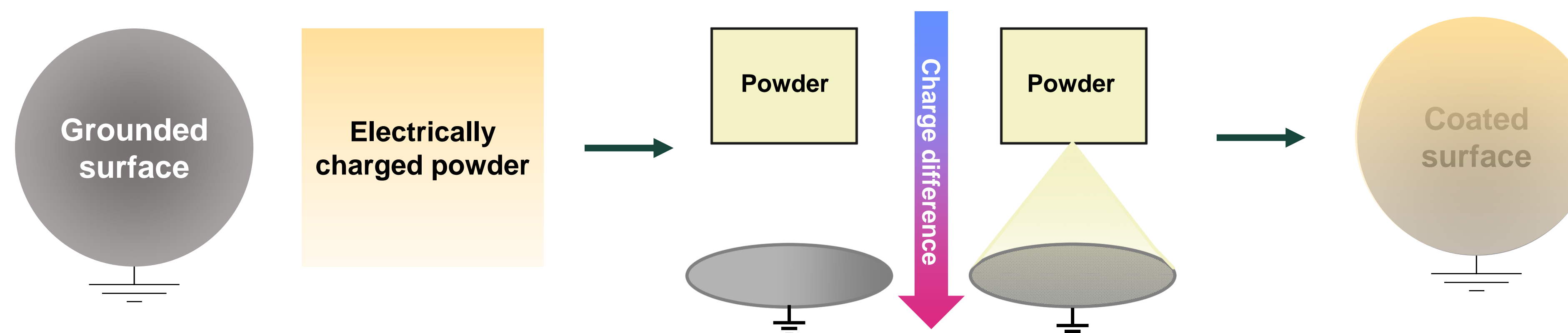
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Methods and Materials

Electrostatic Coating

- Adheres charged particles to electrically grounded surfaces
- Replicates cross-contamination of food contact surfaces through dust accrual
- Minimizes moisture added to the system in comparison to other inoculation methods



- 1. Inoculate wheat berries**
 - 1000 g of wheat berries inoculated to simulate in-line contamination during flour processing
 - 6-strain *Salmonella* cocktail (~ 11.4 log CFU/g)
 - Hand massaged and equilibrated to $a_w \sim 0.45$ for 48 ± 2 hr
- 2. Grind berries into wheat meal**
 - Grind berries using a stand mixer and grain milling attachment (~ 125 rpm, finest setting)
- 3. Sieve wheat meal into flour**
 - Sieve wheat meal particles to meet U.S. standards and regulations for flour
 - Use U.S. Standard No.'s 20, 70, 100, 200 mesh to generate an assortment of particle sizes
 - Discard particles that do not pass through No. 70 mesh (>212 μ m)
- 4. Contaminate stainless steel coupons**
 - Use a custom electrostatic coating apparatus in an equilibrated chamber (RH $\sim 45\%$)
 - Adhere ~ 0.1 g of inoculated flour to stainless steel surfaces (#304, #316L brushed finish)
 - Image and record coated samples using a trinocular microscope, 10x resolution
- 5. Brush coupons to “visibly clean”**
 - “Visibly clean” = no particles visible to the naked eye (Daeshel, et. al., 2023)
 - GOAL: ~ 4 log CFU/cm² remaining on cleaned surfaces
 - Brush coupon ten times from top to bottom
 - Rotate the brush, then brush times left to right
 - Assume **0.01 g** of particles remain on surface (1/10 of initial 0.1 g adhered)
 - Image cleaned samples using a trinocular microscope
- 6. Plate and enumerate samples**
 - Dilute coupons in 5 ml buffered peptone water
 - Serially dilute and plate on modified tryptic soy agar (MTSA)

Results

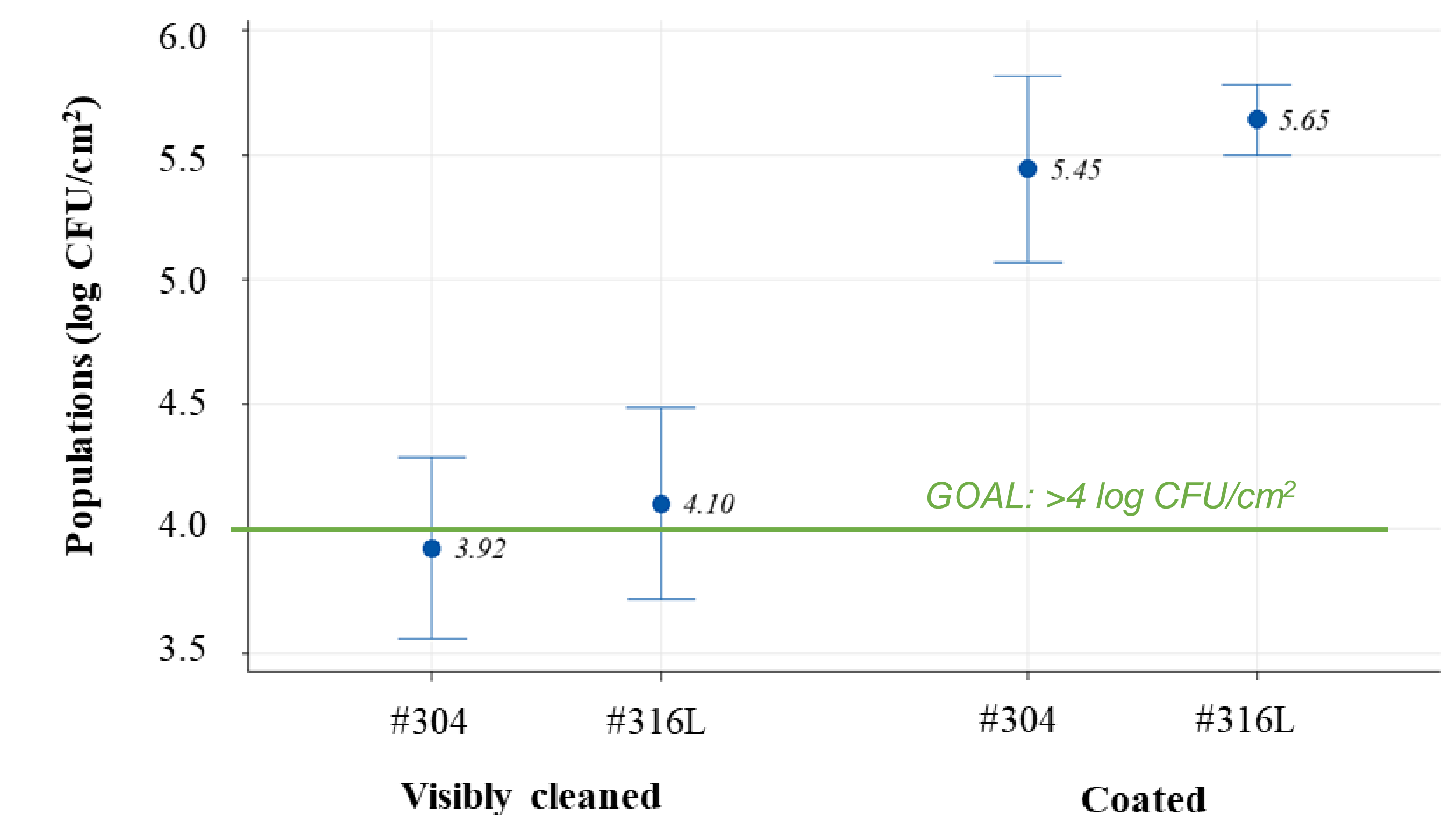
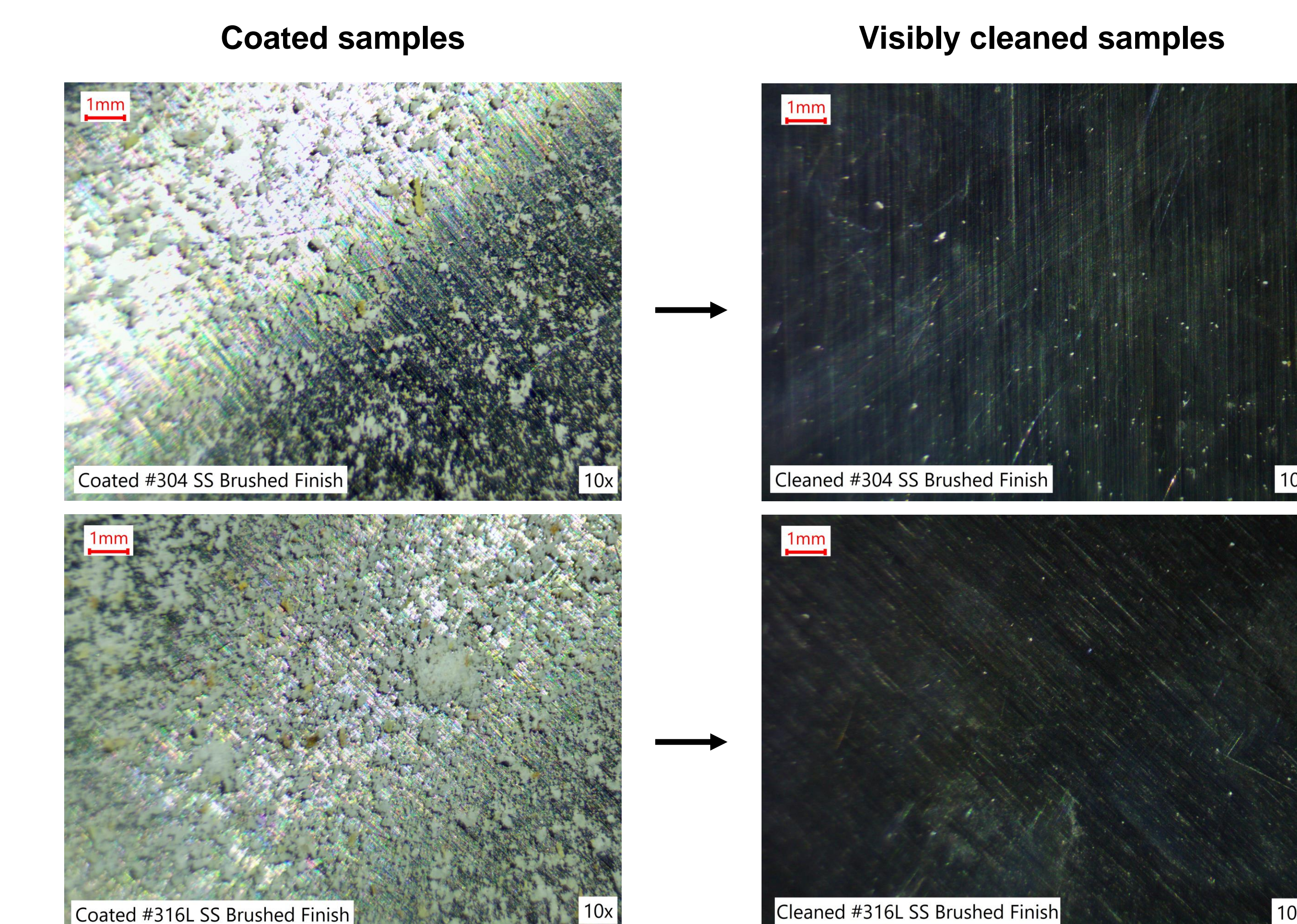


Figure: *Salmonella* populations on brushed and visibly cleaned SS coupons

- Significantly different *Salmonella* populations occurred between coated and visually clean coupons ($p < 0.001$)
- Grade ($p=0.22$) and finish ($p=0.48$) did not significantly impact inoculation levels

Imaging the Cleaning Process



QR Code: Video demonstration of the cleaning process under 10x resolution:



Conclusions

- **The inoculation methodology was consistent and replicable**
- Visibly cleaned samples had residual *Salmonella* populations > 4 log CFU/cm²
- Brushing removed most particles in the first pass (see video QR code) and appeared visibly clean to the naked eye
- Upon further inspection under a light source, residual particles on the surface were confirmed
- Visibly cleaned surfaces were not microbiologically clean

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