Rapid Detection of *Salmonella* in Fresh Meat Using Computer Vision and Digital Imaging

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ABSTRACT

The rapid detection of foodborne pathogens is critical for improving food safety and reducing the risk of contamination in meat products. This study evaluated the detection and survival of Salmonella Typhimurium in ground chicken meat using advanced digital imaging and computer vision approaches alongside conventional microbiological analysis. Four treatment groups were investigated: Salmonella Typhimurium culture only (control), ground chicken inoculated with Salmonella Typhimurium, inoculated ground chicken treated with 1% vinegar solution, and inoculated ground chicken treated with sterile Buffered Peptone Water (BPW). Samples were prepared under sterile conditions, serially diluted, and plated on Tryptic Soy Agar (TSA) and Xylose Lysine Deoxycholate (XLD) agar for microbial enumeration. Fluorescence-based digital imaging was employed to visualize microbial presence and assess cellular morphology within the meat matrix. Statistical analyses were conducted using the Kruskal–Wallis nonparametric test followed by Dunn's pairwise comparisons at a significance level of $p < 0.05$. Results demonstrated significant differences in microbial counts among treatment groups in the primary analysis (Kruskal–Wallis $\chi^2 = 9.684$, $p = 0.0215$). Dunn's post-hoc test revealed significant differences between the control culture and all meat treatment groups, indicating that the meat matrix and antimicrobial intervention influenced bacterial survival. The robustness median test further confirmed significant differences among groups (Pearson $\chi^2 = 11.0769$, $p = 0.011$). However, a secondary analysis showed no significant differences among selected treatment groups (Kruskal–Wallis $\chi^2 = 2.604$, $p = 0.272$), suggesting variability in treatment effects under certain conditions. Digital imaging successfully visualized microbial distribution and morphological characteristics, supporting rapid and non-destructive assessment of contamination. The findings demonstrate the potential of integrating computer vision, digital imaging, and conventional microbiological techniques for enhanced detection of Salmonella Typhimurium in fresh meat. This combined approach offers a promising tool for real-time monitoring, quality assurance, and improved food safety management in meat processing systems.

Keywords: *Salmonella Typhimurium, computer vision, digital imaging, fluorescence imaging, food safety, antimicrobial treatment, ground chicken meat*

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INTRODUCTION

Foodborne illnesses remain a major public health concern in the United States and worldwide, with it contributing to mortality, economic loss each year, and even death. The top five leading causes of this type of illness are *Escherichia coli*, *Listeria monocytogenes*, *Campylobacter jejuni*, *Staphylococcus aureus*, and *Salmonella spp (CDC, 2025)*. In just the United States of America alone there are millions of cases of illnesses and death that point towards pathogenic microorganisms (CDC, 2025). The affected populations are vulnerable young children, elderly adults, pregnant women, and immunocompromised individuals. These pathogens cause millions of reports annually on hospitalizations, short and long-term causes or repercussions. An estimation of 2.3 percent of all cute foodborne illnesses develops long-term illnesses (Alum *et al*., 2016). Creating a comparison of conventional and advanced imaging systems helps to identify the differences between the traditional way of detecting and ways to improve the system. Traditional microbiological detection methods can be time-consuming, labor-intensive, and destructive to the sample (Wang *et al*., 2021). As a result, there is an urgent need for faster, highly accurate detection strategies that can improve food safety, reduce disease transmission, and protect public health (Quintela *et al*., 2022).

Among these pathogens bacterial contamination of meat products remains a persistent challenge of cross-contamination risks and limitation in existing detection methods. Conventional microbiological techniques are used for pathogen detection such as culture-based assays which consist of sample collection, inoculation, incubation and these methods are time-consuming. Identifying each sample individually requires precision and as deep understanding of the bacteria and the proper use of the microscope. Food safety monitoring and the reduction of foodborne disease transmission require a faster, highly accurate, and less time-consuming detection approach.

Developments in imaging technologies and computer vision systems offer such promising results compared to traditional methods of detection. Computer vision techniques help to enhance the analysis part of detecting, creating ways to classify, and interpret what you are seeing. This technique could potentially help improve the efficiency and accuracy when detecting pathogens in meat. Combing imaging technologies and computer vision into a program where it helps to provide real- time images of the bacteria embedded in the meat matrix.

Salmonella species are a dominated and consistently reported group of bacteria that causes a tremendous number of foodborne illnesses. Now, according to the CDC there is an estimated of 1.35 million *Salmonella* infections that occur every year in the United States (CDC, 2024). *Salmonella Typhimurium* is one of the leading serovar that is responsible for frequent number of outbreaks that is associated with fresh meat products. The infection first starts as gastroenteritis which is the stomach flu on the severe side this pathogen can invade past the gastrointestinal tract and cause other life-threatening conditions and illnesses.

In this study, inoculated fresh meat have been used to create controlled *Salmonella* contamination conditions for accurate evaluation specifically *Salmonella Typimurium*. Inoculation allows for precise simulation of real-world contamination patterns and ensures experimental consistency. The usage of Olympus ApexView APX100 combined with the traditional microscopy provides a novel approach to analyze surface and structural changes in contaminated poultry meat that may not be detectable by the naked eye. Computer vision systems utilize advanced algorithms to process digital images, identify any possible contamination patterns, and classify microbial presence with high accuracy (Nnachi *et al*., 2022). When integrated with imaging technology, these systems offer the potential for real-time, non-invasive detection of *Salmonella* in any meat products. Therefore, this research bridges the fields of food microbiology, imaging technology, and artificial intelligence by evaluating *Salmonella spp.* contamination in inoculated fresh meat using imaging and computer vision techniques. The study contributes to improve food safety monitoring systems, reduce the burden of foodborne illness, and support the development of more efficient detection technologies for the meat industry and public health agencies.

Literature review

Salmonella

This pathogenic bacterium that primarily causes gastrointestinal illness in humans (Patel *et al*., 2024). As reported by the CDC, previously stated of 1.35 million *Salmonella* infections each year that result in roughly 26,500 hospitalizations, and 420 deaths that happen annually (CDC, 2024). This bacterium is commonly associate with poultry, beef, eggs and other food products which make it a huge health concern in food processing systems. It can survive through various stages of food production, processing, and even being distributing to retail stores. This helps to highlight the desperate need for a more reliable detection method to reduce contamination and protect the public (Rahman *et al*., 2024). *Salmonella* strains in meats which include poultry and beef products cause a tremendous public health concern nationwide. Studies have shown that *Salmonella* can persist in processing plants which have a reoccurring event of contamination. In the study of Wang *et al*. (2022) the authors characterized *Salmonella* strains and environmental microorganisms which were

Salmonella enterica serovar Typhimurium, *Montevideo*, and *Cerro*. These bacterial organisms were isolated from beef processing plant floor drains of the hotbox and storage cooler area. As you can see *Salmonella* can be introduced to meat through various avenues like the farm, slaughterhouse, transportation, processing, and retail markets.) This bacterium can reoccur onto equipment, animals, personnel, processing environments which make it challenging to identify the source. The persistent increase in the bacteria causes a higher risk of foodborne illness, which can lead to hospitalization or even death if not treated promptly.

Outbreaks in poultry are associated with contaminated poultry eggs where it is categorized in two sections of horizontal and vertical transmission being the main routes of infections in chicken. In Australia, *Salmonella Typhimurium* is a primary serovar that contaminates the external egg which creates a tremendous number of problems like it can be transferred to surfaces, hands, and other foods.

While, *Salmonella Enteritidis* in North America and Europe have the egg-borne transmission because it carries the pathogen inside where it can be transmitted to humans. Microbial contamination contributes to a great range of foodborne illnesses. It mainly occurs on the surface of all kinds of meat but can lead to spoilage, microbial growth and can be transformed into a foodborne pathogen.

This is a significant concern for the consumers, meat and retail industry. Meat contamination can come in different forms like soil, water, air, processing equipment and even humans but ultimately the main source is the animal themselves. An example would be during slaughter the carcass of the animal can be contaminated by feces, equipment, human hands and even the air. Microorganisms are one of the essentials of cross-contamination which can come from equipment used, workers and any insects in the processing plant. Post-processing stages represent a critical point in the meat supply chain, as microbial contamination can occur during storage and distribution. Inadequate temperature control, poor handling, and cross-contamination from surfaces or packaging increase the risk of pathogen growth. Therefore, strict adherence to food safety measures such as cold chain management, hygienic handling, and antimicrobial interventions is essential to minimize contamination and ensure consumer safety. This is the reason that cold storage is an important factor in the process of after the processing stage because it helps to reduce the microbial growth on the meat. This research highlights the sources of bacterial contamination in fresh meat during processing and retail stages. The impact that the bacteria and pathogens have on meat quality and human health can cause meat deterioration some visual aspects are texture changes, gas production, discoloration and off odors. The health risks that are associated with microbial contamination are foodborne illnesses, toxin production, and antibiotic resistance. Toxin production is one of the bacteria called clostridium perfringens where mold produces toxins that persist even after cooking your food. Antibiotic resistance is when meat contaminants carry these antibiotic-resistant genes which can later be transferred to consumers through food consumption.

Fresh Meat and Microbial Techniques

Fresh meat is very susceptible to contamination when strict hygiene, sterilization, sanitation, and meat-handling practices are not enforced. The contamination process can occur at any stage of production, including on the farm, at slaughterhouses, during meat processing, storage, distribution, retail markets, and household food preparation (Nnachi *et al*., 2022). Raw beef and pork are frequently linked in with *Salmonella* outbreaks due to improper temperature control, cross-contamination, inadequate cooking and cooking temperatures. Fresh meat, with its nutrient-rich environment and high moisture content, creates an ideal habitat for bacteria to thrive and multiply. The essential property of fresh meat includes high water activity of 0.89-0.99, near neutral post-mortem pH of 5.5-6.5 which creates favorable conditions for microbial survival and spread of bacteria (R.A. Lawrie, 1966). Intrinsic factors of this would be the water activity and pH level and nutrient density. Extrinsic factors are storage temperature, oxygen exposure to the fresh meat, and packaging process. It primarily serves as a reservoir for *Salmonella*, making it a critical area for research in improving detection methods within this food category.

There are two different methods to detect microorganisms in meat and evaluate meat which is conventional and the rapid advanced methods (Patel *et al*., 2024). The conventional method or destructive testing consists of altering and destroying the sample. Rapid advanced method or non-destructive allows the scientist or researcher to observe and retain the properties of the bacteria to detect and study the state of it. Conventional methods, which are destroying samples in the preparation stage would include, culture-based count methods, selective enrichment procedures, biochemical assays and immunological techniques such as enzyme-linked immunosorbent assay (ELISA). Government approved low initial threshold of *Salmonella* in contaminated meat there should be selective enrichments steps to help increase pathogen levels prior to selective plating and any biochemical confirmation tests. The methods are labor intensive and meticulous to operate this type of equipment (Hong-Ju He, 2015). While culture-based confirmation of *Salmonella* requires 3-5 days due to the tedious process that needs to be done.

Rapid advanced methods may allow for detection and the evaluation of bacteria in the matrix of meat. This could help to preserve the sample consistency.

There was a study where the authors targeted invA gene using a loop-mediated isothermal amplification (LAMP) which successfully heightened the sensitivity of eight different serotypes of *Salmonella* (Wang *et al*., 2022). Some examples of this advanced detection equipment are infrared spectroscopy, fluorescence spectroscopy and computer vision systems. While these advanced methods require some sort of training, it offers a substantial reduction in detection time, enhanced effectiveness, and a decrease in the need for extensive sample preparation. Microbial identification executes a pivotal role in monitoring bacterial contamination in any food system. In conventional microbial assessments there are culture-based methods, molecular techniques etc. which require long periods of incubation and will ultimately destroy those samples (Hong-Ju He *et al*., 2015). The limitations include the rapid identification of pathogens, the availability of on-site equipment for detection, and the ability to conduct real-time monitoring in industrial food production environments. The limitations include the rapid identification of pathogens, the availability of on-site equipment for detection, and the ability to conduct real-time monitoring in industrial food production environments. Another constraint would be to detect and identify multiple samples all at once instead of doing everything individually. There is emerging technology to help resolve these challenges by exploring rapid and automated equipment for food safety and microbial detection as well as techniques. In large-scale industrial meat production, where diverse meat types are rapidly moved through processing lines, the inability to conduct real-time microbial detection substantially increases the risk of widespread contamination events and subsequent large-scale recalls (Shi *et al*., 2021).Fresh meat contains pseudomonas spp., lactic acid bacteria and Enterobacteriaceae which is why it is necessary to isolate *Salmonella* from these other different microbial populations (Hong-Ju He *et al*., 2015). It naturally harbors a diverse microbial population which can be acquired from different stages of the whole process (Wang *et al*., 2021). Lactic acid bacteria and members of the Enterobacteriaceae family are primarily associated with spoilage rather than pathogenicity but just the mere presence of these bacteria impacts the detection of pathogens (Quintela *et al*., 2022). During the non-selective media process background microorganisms may outgrow *Salmonella* or injured *Salmonella* cells that reduce efficiency. Also, *non-Salmonella Enterobacteriaceae* family can produce colony morphology that is like *Salmonella* on selective media which increases the potential of false positives. Having these complex microbial ecosystems on the presence of meat makes it difficult to distinguish pathogenic contamination.

Non-destructive Imaging Methods

Hyperspectral imaging systems are one of the microscopy systems that are increasingly being investigated to be included for food safety applications (Shi *et al*., 2021; Eady *et al*., 2019). The techniques include the use of spectral signatures to detect any surface-level contamination without having the needs to do any physical sampling. An example of this would be where hyperspectral imaging has been demonstrated in success of identifying *Salmonella* and *Escherichia coli* contamination on meat (Patel *et al*.; He & Sun, 2015). These non-destructive technologies could help routine monitoring in meat processing facilities. These findings help to detect contamination early, with the potential of reducing the risk of foodborne illnesses. Despite these advantages hyperspectral imaging has it does have limitations onto what the machine can do. The limitations consist of the high cost of the equipment, and it needs to have complex data processing systems which can limit how accessible it can be (Quintela *et al*., 2022). Next, the complexity of the spectral data analysis where it requires advanced mathematical processes and variability. The variations in meat compositions like fat and moisture content can interfere with spectral readings. While hyperspectral imaging system holds a great advantage compared to a traditional method there is a need for further research in the terms of affordability and integration of a more routine food safety workflow.

Raman & Fluorescence Microscopy

These recent advances in microscopy and imaging have opened new doors for there to be real-time microbial evaluation. Traditional microscopy has been useful for general morphological identification, but it lacks the resolution and sensitivity that is needed for detecting at an early stage of contamination (Murphy & Davidson, 2012). While fluorescence microscopy has confocal laser scanning and electron microscopy has significantly improved the visualization of microbial structures of bacteria. These new developed methods have helped researchers to being able to differentiate between living to dead cells and monitor pathogen behaviors. Fluorescence imaging has been successful when applied in food safety specifically in detecting contamination on meat surfaces. With the fluorescence imaging combined with deep learning algorithms it can automatically identify fecal contamination on meat carcasses with high accuracy (Gorji *et al*., 2022). A similar detection method developed a handheld fluorescence imaging system which is capable to detect and disinfect contaminated surfaces which goes to highlight in-field applications and real-time imaging (Sueker *et al*., 2021). Another study revealed that fluorescence imaging could distinguish between healthy and contaminated poultry carcasses.

Raman microscopy helps to enhance the detection capabilities even further by providing through a molecular level. Unlike a fluorescence method where it requires staining and labeling where Raman microscopy is label free for identifying bacteria. A study emphasized how

effective the spectroscopy is in evaluating microbial contamination in food products (He and Sun, 2015). Raman spectroscopy pathogen detection helps to demonstrate the integration of machine learning which helps to improve the accuracy and speed of detecting pathogens (Rahman *et al*., 2024). Demonstrating these approaches allow for having rapid identification of bacteria based on their unique fingerprints.

Olympus Apex View Bench top Fluorescence Microscope APX100

This advanced imaging system provides a benchtop fluorescence microscope which makes it quick and simple to operate. It has prominent optics, artificial intelligence, and high-quality image data to help with any research you are needing to get done (Shibata & Kojima, 2024). It uses slides, dishes and well plates for imaging applications where it can multichannel and use time lapse. As for this piece of equipment, it has the features of autofocusing, different types of magnifications, and images saved for any future reference. These advancements align with the trends in imaging technology where artificial intelligence is used to reduce the user being dependent variability and improve accuracy (Gorji *et al*., 2022). Practical standpoint, the APX100 provides efficiency compared to the conventional methods. The machine uses automated focusing, data processing, and image acquisition which helps to reduce the time for analysis and human error (Shibata & Kojima, 2024). These are important factors in food safety applications where having rapid and accurate detection is crucial. Unlike other advanced imaging systems, the APX100 it requires technical infrastructure and investment into this piece of equipment.

Application of Imaging System

Despite having the promise of imaging technologies there are few studies that directly compare conventional light microscopes with a more advanced microscope such as the APX100. Existing literature is divided which the research being so heavily focused on high tech applications than the all-inclusive food safety integration. This is a gap where there is a need for side-by-side evaluation of the more advanced imaging systems to the more traditional systems. Traditional methods like biochemical assays and culture-based detection are heavily used due to its reliability and how standardized it is (Fagerberg and Avens, 1976).

In comparison, advanced imaging technologies provide substantially faster detection capabilities and enable real-time microbial analysis. Studies show that these imaging-based methods can achieve and improve detection to traditional techniques. Additionally, with non-destructive imaging which allows a more continuous monitoring without destroying the sample it makes it more valuable in industry settings. There is an opportunity for a more practical implementation of these technologies but requires the evaluation of cost-effectiveness, adaptability, integrating into real-world meat science labs and existing workflows. A review that emphasized rapid detection technologies which are advancing and improving operational equipment to be of use in food safety systems but have limitations that are hindering it (Quintela *et al*., 2022). Comparative studies show that both traditional and advanced methods that determine if they are suitable in real-world applications.

MATERIALS AND METHODS

Experimental Design and Treatments

This study was designed to evaluate the detection and survival of *Salmonella Typhimurium* in a meat matrix under different treatment conditions. Four experimental groups were established:

1. *Salmonella Typhimurium* culture only (baseline control)
2. Ground chicken inoculated with *Salmonella Typhimurium*
3. Ground chicken inoculated with *Salmonella Typhimurium* with antimicrobial treatment (1% vinegar solution)
4. Ground chicken inoculated with *Salmonella Typhimurium* with sterile Buffered Peptone Water (BPW) as a liquid control

Twenty-five grams (25 g) of ground chicken were aseptically weighed and placed into sterile stomacher bags. For inoculated treatments, *Salmonella Typhimurium* was added and allowed to attach for 15 minutes at room temperature prior to further processing. All procedures were conducted under sterile laboratory conditions to prevent cross-contamination.

Preparation of Salmonella Typhimurium

Salmonella Typhimurium was obtained from the College of Veterinary Medicine laboratory at Tuskegee University. The culture was transferred into Tryptic Soy Broth (TSB) and vortexed to ensure homogeneity. The inoculated broth was incubated at 37°C for 18–24 hours under aerobic conditions to achieve active bacterial growth prior to experimental use. The bacterial suspension was standardized to a known concentration prior to inoculation.

Serial Dilutions

Serial dilutions were performed using sterile Buffered Peptone Water (BPW) to obtain appropriate bacterial concentrations for plating on Tryptic Soy Agar (TSA) and Xylose Lysine Deoxycholate (XLD) agar. Six sterile tubes

containing 9 mL BPW each were prepared and labeled from 10^{-1} to 10^{-6}. Twenty-five grams (25 g) of sample were homogenized with 225 mL sterile BPW using a Stomacher 400 Circulator Lab Blender at 230 rpm for 1 minute, producing a 10^{-1} dilution. 1 mL aliquots were transferred into 9 mL BPW sequentially to achieve 10-fold serial dilutions up to 10^{-6}. Each dilution was vortexed thoroughly before subsequent transfer to ensure uniform bacterial distribution.

Treatment Applications

For antimicrobial treatment, a 1% (v/v) vinegar solution (acetic acid-based) was applied evenly to inoculated meat samples and allowed to react for 5 minutes prior to dilution. Control samples received sterile Buffered Peptone Water (BPW) instead of antimicrobial treatment to account for dilution and handling effects. Baseline samples consisted of *Salmonella Typhimurium* culture without meat matrix exposure.

Microbial and Statistical Analysis

Aliquots (100 µL) from appropriate dilutions were spread-plated onto:

(a) Tryptic Soy Agar (TSA) for total viable counts
(b) Xylose Lysine Deoxycholate (XLD) agar for selective isolation of *Salmonella*

Plates were incubated at 37°C for 24–48 hours, inverted during incubation.
Colonies were counted using plates containing 25–250 colonies, and results were expressed as CFU/g using the standard plate count method:

$$CFU/g = \frac{\text{Colonies} \times \text{Dilution Factor}}{\text{Volume Plated (mL)}}$$

Statistical analysis was performed using SPSS (figure out the version).

The Kruskal–Wallis test was used to determine significant differences among treatment groups, followed by Dunn's post-hoc test for pairwise comparisons.
Statistical significance was defined at $p < 0.05$. A robustness check was performed to valiadta consistency of results.

Kruskal-Wallis H statistic: $H - \frac{12}{N(N+1)} \sum_{i-1}^{k} \frac{R_i^2}{n_i} - 3(N+1)$

where N= total number of observations, k= number of groups,

$n_i = size\ of\ group\ i$, $R_i = sum\ of\ ranks\ for\ group\ i$

RESULTS AND DISCUSSION

Treatment 1: *Salmonella* Only (Baseline)

The baseline concentration of *Salmonella Typhimurium* was determined using serial dilutions of cultures grown in Tryptic Soy Broth (TSB) and plated on Tryptic Soy Agar (TSA) and Xyline Lysine Deoxycholate (XLD) agar. Colony counts obtained from plates within the acceptable range of 25- 250 colonies (Figure 1). The results demonstrated a clear and expected trend in which colony counts decreased with increasing dilution. This confirmed that the serial dilution procedure was performed accurately and that bacterial populations were evenly distributed through proper vortexing prior to each transfer. The consistency between duplicate plates further validated the reliability of the plating technique. These findings are consistent with the standard microbiological procedures where predictable linear decreases in colony counts under a controlled dilution (Wang *et al*., 2021). Having this consistency in duplicate plates further confirms the reliability of the plating procedure. Statistical analysis using the Kruskal–Wallis test showed significant differences among dilution groups ($p < 0.05$), confirming that the dilution factor had a measurable and predictable effect on colony counts (Table 1).

Dunn's pairwise comparisons further supported this, showing that lower dilutions (higher concentrations) differed significantly from higher dilutions (Table 2). From a biological perspective, this treatment established a critical reference point for all subsequent comparisons. Since no external factors such as meat matrix or treatment solutions were present, the observed counts reflect the true viability and growth potential of the bacterial culture under sterile laboratory conditions. This baseline is essential for interpreting whether reductions observed in later treatments are due to environmental interference or antimicrobial effects (Table 3).

When *Salmonella Typhimurium* was inoculated into ground chicken and homogenized with peptone water, a noticeable change in bacterial recovery was observed compared to the baseline. Although colonies were still recoverable across all dilutions, the overall counts were slightly reduced, particularly at higher dilutions. This reduction can be attributed to the nature of the meat matrix. Ground chicken contains proteins, fats, and naturally occurring microflora that can interfere with bacterial recovery in several ways. First, bacterial cells may attach to meat particles which could make them less accessible for plating. Second, the physical structure of the meat can protect or trap bacteria, reducing their distribution in the homogenate. Third, background microorganisms may compete with *Salmonella* on agar plates, particularly on non-selective media such as Tryptic Soy Agar (TSA). The reduction of the counts in the meat matrixes are consistent with how complex food

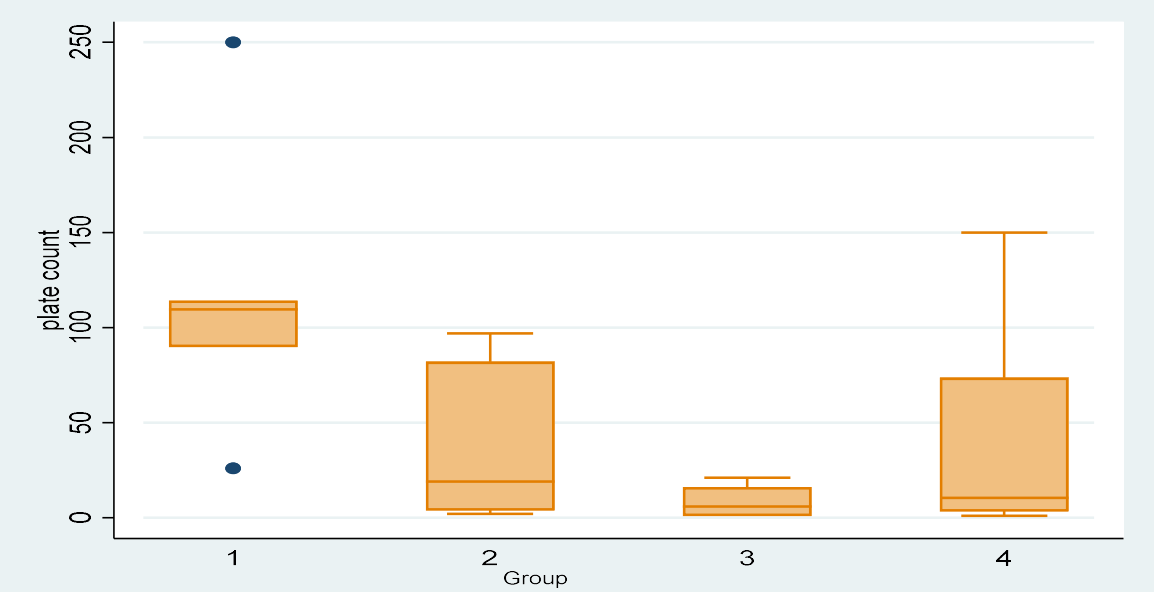

Figure 1: *Box plot showing plate count by group*

Table 1. *Kruskal–Wallis equality-of-populations rank test (A).*

Group	Observation	Rank sum
1	6	118
2	6	68
3	4	25.5
4	8	88.5

chi2(3) = 9.659; Probability (p) = 0.0217, chi2(3) with ties = 9.684; Probability (p)= 0.0215
Decision: In both cases, we reject the null hypothesis of equality of median values by groups.

Table 2. *Dunn's pairwise comparison of plate count by groups.*

Col. Mean/ Row Mean	1	2	3
2	2.04		
	0.02**		
3	2.92	1.09	
	0.00***	0.14	
4	2.26	0.07	-1.08
	0.01***	0.47	0.14

Group 1 vs 2; 1 vs 3; 1 vs 4 are statistically significant at 5%, 1%, and 1% significance level, respectively

Table 3: *Robustness check for median test.*

Greater than the median	Group				
	1	2	3	4	Total
No	0	4	4	5	13
Yes	6	2	0	3	11
Total	6	6	4	8	24

Pearson chi2(3) = 11.0769 Pr. = 0.011
Robustness check indicates that we reject the hypothesis that equality of median plate count by groups.

matrices are and pathogen recovery due to the proteins and lipids which interfere with the microflora (Quintela *et al*., 2022). Statistical analysis (Kruskal–Wallis test) indicated that differences among replicates were not

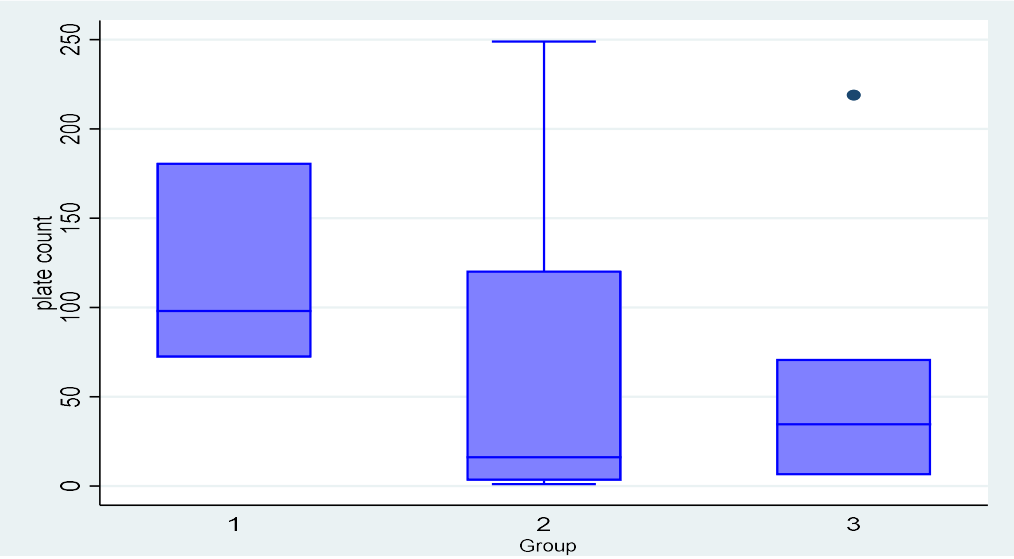

***Figure** 2: Box plot for plate count by groups (B)*

***Table 3.** Kruskal–Wallis equality-of-populations rank test (B).*

Group	Observation	Rank Sum
1	3	39
2	8	60
3	6	54

chi2(2) = 2.588; Prob = 0.2741
chi2(2) with ties = 2.604; Prob.= 0.272
In both cases, we fail to reject the null hypothesis indicating median values by groups are not different.

***Table 4:** Dunn's pairwise comparison of plate count by groups (B) Dunn's Pairwise Comparison of plate count by group (No adjustment.*

Col. Mean/ Row Mean	1	2
2	1.613751 0.05333	
3	1.123672 0.1306	-0.551712 0.2906

strongly significant (p > 0.05), suggesting that while the meat matrix had some effect, it did not drastically alter bacterial populations (Figure 2). However, trends observed in Dunn's comparisons suggested a slight reduction in detectable bacteria compared to the baseline. Importantly, the results confirm that *Salmonella* can still be reliably detected in meat, providing a valid foundation for evaluating treatment effects.

The addition of the antimicrobial treatment to the *Salmonella*-inoculated meat resulted in a substantial reduction in bacterial counts across all dilutions. Plates from treated samples consistently showed fewer colonies compared to both the baseline (Treatment 1) and the untreated meat samples (Treatment 2). This reduction indicates that the treatment was effective in inhibiting or inactivating *Salmonella Typhimurium*. The effectiveness of the treatment was observed across multiple dilutions, suggesting that its impact was not limited to a specific concentration range (Table 4). These results help to align with previous research that demonstrate how organic acid-based treatments such as acetic acid interrupt bacteria and reduce microbial viability (Gorji *et al*., 2022). This observation goes to show how the reduction across multiple dilutions suggests more of a consistent antimicrobial treatment. Statistical analysis confirmed these observations, with significant differences detected between treated and untreated groups (p < 0.05). Dunn's pairwise comparisons showed that treated samples differed significantly from both the baseline and meat-only treatments, reinforcing the conclusion that the treatment had a measurable antimicrobial effect (Table 5). From a practical standpoint, these results

demonstrate the potential application of the treatment in reducing *Salmonella* contamination in poultry products. The ability to significantly lower bacterial counts highlights its relevance for food safety interventions.

The control treatment, in which sterile buffered peptone water was added instead of the antimicrobial solution, showed colony counts like those observed in the meat-only treatment (Treatment 2). This indicates that the addition of liquid and the handling procedures themselves did not significantly affect bacterial survival. The similarity between Treatments 2 and 4 confirms that any reductions observed in Treatment 3 were due to the antimicrobial properties of the treatment rather than dilution, mixing, or mechanical effects from the stomacher process.

This examination is consistent with previous studies showing how neutral buffer solutions primarily only function as diluents for microbiological assays (Shi *et al*., 2021). The similarity observed in treatments 2 and 4 reinforces the conclusion that the reductions observed in treatment 3 are attributable to the antimicrobial properties of vinegar. Statistical analysis further supported this conclusion, as no significant differences were observed between the control and untreated meat groups. This control is critical for validating the experimental design. Without the control treatment, it would not be possible to determine whether reductions in bacterial counts were caused by the treatment or by external procedural factors.

Comparison and Interpretation

The study aimed to compare visualization accuracy, speed, and image clarity between traditional microscopy and advanced imaging systems. It also evaluated the cell morphology of *Salmonella* in the meat matrix using advanced microscopy and demonstrated the potential of advanced imaging for real-time food safety monitoring in poultry products. To achieve these objectives, ground chicken samples were inoculated with *Salmonella Typhimurium* and analyzed using advanced imaging techniques. Four treatment conditions were evaluated: treatment one *Salmonella* culture only, treatment two ground chicken + Salmonella culture, treatment three was ground chicken + *Salmonella* culture + antimicrobial treatment, lastly treatment four was ground chicken + *Salmonella* culture + water (control). Statistical analysis of median microbial counts was conducted to test the null hypothesis that the bacterial populations across groups were equal.

The comparison of treatments revealed clear trends for all treatments. Treatment one had the most high and consistent bacterial counts that were observed. Treatment two showed that there was a slight reduction in bacteria. Treatment three there was a significant difference in reduction that was observed in the bacterial counts which confirms the effectiveness of the antimicrobial treatment. For treatment four it showed similarities like to treatment two. Advanced imaging systems have evolved the field of microscopy by significantly helping to enhance visualization, accuracy, speed, and image clarity (Rahman *et al*., 2024). These systems have demonstrated remarkable capabilities in capturing cell morphology. This directly addresses Objective 1 and Objective 2 by showcasing how advanced imaging systems can help to improve detection capabilities and clearer visualization of bacterial morphology. Furthermore, the results underscore the potential of advanced imaging for real-time food safety monitoring which is objective 3. The faster detection and accurate visualization facilitated earlier interventions in poultry processing, thereby reducing the risk of contaminated products reaching consumers. Statistical analysis further supports these conclusions. The median microbial counts exhibited substantial variations across different treatment conditions, with significant differences observed between detection methods. This indicates that the choice of imaging technology can have a profound impact on both the recovery and assessment of *Salmonella* (Shibata & Kojima, 2024).

Implications for Food Safety

The findings emphasize that advanced imaging systems offer significant advantages for detecting microbial contamination in food matrices. Beyond demonstrating the antimicrobial effectiveness of treatment solutions, the study provides evidence that these systems can enhance accuracy, speed, and clarity in routine food safety monitoring (Gorji *et al*., 2022). By integrating advanced imaging technologies, poultry processing facilities could benefit from faster detection, improved pathogen identification, and better-informed interventions, ultimately contributing to safer food products (Rahman *et al*., 2024).

Paper Highlights

Salmonella Typhimurium was successfully detected in all experimental treatments using TSA and XLD selective media. Serial dilution produced consistent colony reduction patterns. The 1% antimicrobial vinegar treatment significantly reduced Salmonella Typhimurium populations inoculated in the ground chicken samples. Results demonstrated that organic acid treatments can effectively reduce Salmonella contamination in ground chicken. Kruskal-Wallis and Dunn's test confirmed significant differences at $p < 0.05$. These findings contribute to the understanding of Salmonella in meat processing and its response to antimicrobial treatments.

CONCLUSION

This study demonstrates that advanced imaging systems significantly enhance the detection of *Salmonella*

Typhimurium in ground chicken compared to conventional microscopy. The baseline analysis confirmed the reliability of bacterial cultures and plating techniques, while the presence of the meat matrix introduced minor reductions in recoverable bacteria due to possible physical and biological interactions. Results helped to confirm the antimicrobial treatment which effectively reduced the Salmonella populations. Together, these findings suggest that advanced imaging could serve as a valuable tool for food safety testing, which could bridge the gap between laboratory precision and practical application in processing environments. Despite these promising findings, there were several limitations identified. The system requires such careful calibration, limitations in cost, operator expertise to ensure accurate visualizations and consistent results. Additionally, the small sample size and the system's performance with larger or more complex food matrices has not yet been fully evaluated. Future research should focus on formal training programs or workshops to guide users in proper operation, optimizations for larger, more diverse sample types, evaluation of cost-effective alternatives or more affordable imaging systems. Additionally, integration with antimicrobial treatments and real-time or continuous monitoring in industrial settings could help to further improve food safety applications.

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