Can Phage replace Chemicals in Post Harvest Poultry Processing?
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1 Executive Summary

Salmonella continues to be a leading cause of foodborne illness despite significant resources of time money and effort put forth by industry to combat its presence. In addition, pressure is mounting to reduce the use of harsh chemicals due to worker safety concerns. These opposing pressures have the potential to put the industry between a rock and a hard place. Phages are a natural, safe and effective alternative that can be part of the solution. Phage technology has already been used in the US food industry for over 10 years to combat Listeria and new developments have made it possible to combat Salmonella as well. A newly published scientific study (1) found that PhageGuard S, a new treatment for Salmonella reduction on food products, is more effective than lactic acid (LA) and peracetic acid (PAA) in reducing salmonella in beef trim. The findings are of particular interest to meat and poultry processors looking to minimize the use of chemicals in their production processes and protect worker safety. These results challenge the perceived effectiveness of traditional chemical interventions for foodborne pathogen control and highlight phages as a superior and more successful alternative for enhanced consumer safety.
2 Introduction

For years now, the poultry industry has been struggling to limit the presence of *Salmonella*. According to USDA regulations, no more than 7.5 percent of the chicken carcasses coming from a chicken plant can test positive for *Salmonella* and the majority of poultry slaughter plant have no problems meeting this standard. However industry is well aware that the more the carcass is disassembled, the more *salmonella* levels increase. Raw food products, including meat and poultry, are not sterile. In an effort to make them and the environment in which they are processed as safe as possible, the industry uses a variety of chemicals to help reduce pathogen loads and prevent food borne illnesses.

When the USDA proposed in 2012 its regulation to “modernize the poultry inspection system,” consumer groups and food safety organizations warned that the industry would rely more and more on chemical sanitizers.

To stay within the new USDA performance standards the industry is not only introducing new antimicrobial chemicals, but is also changing the volume, process and number of times by which chemicals are applied to food products. Chemicals that were previously only used in dip tanks on the slaughtering side where few workers were present are now being used in multiple open dip tanks throughout the processing side of poultry plants.

Recently the pressure on industry has been mounting in regard to their use of chemicals and that they are failing to protect workers from exposure to any number of chemicals including but not limited to chlorine, peracetic acid, bromine, hydrogen peroxide and acetic acid.
3 Trends

3.1 Salmonellosis in the US

According to the Centers for Disease Control and Prevention (CDC), it is estimated that *Salmonella* causes 1.2 million illnesses, 24,000 hospitalizations, and 450 deaths every year in the U.S. (CDC, 2018). In 2012 the CDC reported that between 2000 and 2008, *Salmonella* was the leading foodborne pathogen causing the largest number of foodborne illness related deaths. Due to this significant public health concern, the U.S. Department of Agriculture’s (USDA) Food Safety and Inspection Service (FSIS) released the *Salmonella* Action Plan, a combination of comprehensive strategies to improve robust food safety systems to reduce *Salmonella* contamination in meat and poultry products (USDA-FSIS, 2013).

Poultry products are the most frequently implicated sources of human *Salmonella* infections in the U.S. Furthermore, increased consumption of meat and poultry has increased the potential for exposure to *Salmonella* enterica. While advances have been made in reducing the prevalence and frequency of *Salmonella* contamination during poultry processing, there is mounting pressure on commercial growers to reduce or eliminate these human pathogens in preharvest production facilities.

Several factors contribute to *Salmonella* colonization in commercial poultry, including the serovar and the infectious dose. Raw poultry contaminated with *Salmonella* can therefore cause illness if the poultry meat consumed is undercooked or contamination from the raw poultry is transferred to cooked poultry or other food that is ready-to-eat. Cross contamination between raw and ready-to-eat food is a particular concern, as only small numbers of the bacteria are needed to cause human illness (2).
3.2 Regulatory Pressure Continues from FSIS

Even though food companies in North America have a large number of chemical interventions in place to reduce Salmonella it continues to be a major cause for foodborne illnesses. Recently FSIS began online posting of individual establishments’ category status for Salmonella performance standards for poultry carcasses, with parts standards expected to follow in June 2018. Giblets are currently being monitored and are expected to follow soon. The public display of category ranking influences both domestic and export sales, increasing the pressure on industry to meet or exceed USDA’s published standards (USDA-FSIS 2018).

According to FSIS:

- **Category 1. Consistent Process Control:** Achieving 50 percent or less of the Salmonella or Campylobacter performance standard
- **Category 2. Variable Process Control:** Meeting the Salmonella or Campylobacter performance standard but have results greater than 50 percent of the maximum allowable percent positive
- **Category 3. Highly Variable Process Control:** Exceeding the Salmonella or Campylobacter performance standard

July 1, 2016: Performance standards implemented for chicken parts (breast, wing, leg) and comminuted poultry

- 15.4% for Salmonella (8/52)  
  - Category 1: < 7.7%
  - Category 2: > 7.7%; < 15.4%
  - Category 3: > 15.4%

- 7.7% for Campylobacter (4/52)  
  - Category 1: < 3.85%
  - Category 2: > 3.85; < 7.7%
  - Category 3: > 7.7%
January 2018 reporting showed that approximately 40% of US slaughter facilities were in category 1 or 3 against the parts standards. Ground products and giblets will prove to be even more challenging.

3.3 pressure from consumer activists
Seattle attorney Bill Marler, who makes his living suing companies when their food makes people sick, say it’s not good enough. “The standard is, it’s still OK to have a pathogen on your product that can sicken and kill your customers. And as long as that’s the way it is, we’re always going to limp from outbreak to outbreak to outbreak,” he says.

Marler says the USDA should take the same stand against *salmonella* that it did against another dangerous microbe: disease-causing *E. coli*. When the USDA declared these *E. coli* bacteria illegal adulterants in food, the meat industry complained, but it also found new ways to prevent them from poisoning people.

“It used to be 90 percent of my law firm’s revenue, and now it’s nearly zero. It’s a success story,” says Marler.

Eliminating *salmonella* altogether would be difficult — it’s much more common in...
the environment than disease-causing *E. coli*.
So for now, the USDA is pressuring companies to reduce *salmonella* contamination, but it’s not requiring chicken meat to be completely *salmonella*-free (3).

### 3.4 OSHA Cracking Down on Peracetic Acid Exposure

As an antimicrobial agent, PAA is broadly effective against a wide range of microorganisms; it disrupts bonds in proteins and enzymes and interferes with cell membrane transportation through the rupture of cell walls, oxidizing essential enzymes and impairing vital biochemical pathways.

The properties of PAA that make it a chemical antimicrobial intervention also cause challenges with worker safety.

Specifically, the National Institute for Occupational Safety and Health (NIOSH) Registry of Toxic Effects of Chemical Substances identifies PAA as a primary irritant, known tumorigenic and mutagen.

The New Jersey Department of Health and Senior Services Occupational Health Service released a study on the health effects of PAA exposure. The study also found that PAA is very irritating to the skin, eyes, nose, throat, and lungs, with the potential for causing permanent scarring of the skin, cornea, and throat. Higher exposures in the short term can also cause pulmonary edema as well as liver and kidney effects.

Industry is beginning to feel pressure by workers and inspectors related to health hazards associated with the use of harsh chemicals, especially peracetic acid. The regulatory environment concerning Peracetic acid and other chemicals is changing. In 2014, the internationally recognized association, American Conference of Governmental Industrial Hygienists (ACGIH), released a 15 minute Short Term Exposure Limit (STEL) for Peracetic acid of 0.4 ppm. March 2017, The National Institute for Occupational Safety and Health (NIOSH) of the Centers for Disease Control and Prevention (CDC) has proposed an Immediately Dangerous to Life and Health (IDLH) level of 0.55 ppm. Due to lack of sufficient data, NIOSH has requested more information from industry stakeholders regarding health risks to workers associated with occupational exposure to Peracetic acid with the intention of establishing a final IDLH and Recommended Exposure Limit (REL).
Acute exposure to peracetic acid is irritating to the eyes, respiratory tract, and skin. Peracetic acid is a strong sensory irritant considered to be more potent than acetic acid or hydrogen peroxide.

New disinfection systems using a variety of other antimicrobials besides peracetic acid and chlorine (e.g., ozone, bromine compounds, lactic acid, citric acid, chlorine dioxide, ..) continue to be approved by the U.S. Department of Agriculture (USDA). As these systems are introduced to the market and implemented by industry facilities, it should be noted that USDA approval of such systems falls under their food safety directive for purposes of killing bacteria. USDA approval does not speak to potential occupational exposure concerns for workers in facilities where such systems are installed and operated. Therefore, safety and health professionals in the industry should be aware of the potential for such occupational exposures and act to prevent such exposures accordingly (3). These chemicals may also have significant effects on equipment & concrete. They can cause protein denaturation resulting in difficulties in manufacturing of further process products.

### 3.5 Pressure from workers Unions

Meat and poultry slaughter and processing is one of the most hazardous industries in the United States. The US Government Accountability Office (GAO) was asked to review federal efforts to help ensure meat and poultry worker safety and health (US-GAO 2017).

Marc Perrone, international president of the United Food and Commercial Workers (UFCW) Union stated in December 2017 that the GAO report “confirms the U.S. Department of Agriculture doesn’t consider worker safety when allowing new and dangerous chemicals to be used in poultry plants, and that OSHA can’t or won’t adequately protect poultry workers from injury” (UFCW).

### 3.6 Conclusion

Combined, these pressures along with providing safe food to consumers highlights the importance of finding new and innovative approaches to reduce or eliminate *Salmonella* in fresh meats.
4 Phage

4.1 Phage
Bacteriophages ("phages") are the most abundant micro-organisms in the biosphere. They are naturally present in significant numbers in water and foods of various origins. Phages are harmless to humans, animals, and plants. Humans are routinely exposed to phages at high levels through food and water without adverse effect. Phages use bacteria for their multiplication. Via this mechanism, phages contribute to environmental homeostasis, the situation wherein none of the bacterial species in a biosphere becomes dominant. Every species of bacteria is thought to be the host for at least one phage type. Several phages exist that are able to recognize and lyse (kill) a number of different bacterial strains within one species; these have a ‘broad spectrum’ or a wide host range.

Bacteriophages can be regarded as natural enemies of bacteria, and therefore are logical candidates for targeted control of food borne bacterial pathogens like *Salmonella*.

Important attributes of bacteriophages include:
• they kill only bacterial target cells (no impact on plant or animal cells);
• they do not cross species or genus boundaries; therefore they will not affect desired bacteria in foods (e.g., starter cultures for cheese and sausages), and commensals in the gastrointestinal tract, or accompanying bacterial flora in the environment;
• they are composed entirely of proteins and DNA, so their breakdown products consist exclusively of amino acids and nucleotides, both of which are present in abundance in food products.
Safety
With respect to their potential application for the biocontrol of undesired pathogens in foods, feeds, and related environments, it should be considered that phages are the most abundant micro-organisms in our environment, and are present in significant numbers in water and foods of various origins, in particular fermented foods (reviewed by Sulakvelidze and Barrow, 2005). On fresh and processed dairy and meat products, more than 10^8 viable phages per gram are often present (Kennedy and Bitton, 1987). It is a fact that phages are routinely consumed with our food in high numbers. Moreover, phages are also normal commensals of humans and animals, and are especially abundant in the gastrointestinal tract (Furuse, 1987; Breitbart, 2003).

In conclusion, bacteriophages are known to be harmless for all other organisms and are species-specific.

Phages have been successfully used for over 10 years in the food industry.
Over the past 10 years, many US companies have used *Listeria* phages to eliminate *listeria* and reduce risk in in RTE meats, cold smoked fish and soft cheese applications as well as an environmental tool to take out biofilms.
4.2 PhageGuard S

PhageGuard S is a water based phage solution which contains two *Salmonella*-specific bacteriophages, Fo1a and S16 and is characterized by its broad spectrum toward *Salmonella* strains. PhageGuard S kills all *Salmonella* serovars including those that are antibiotic resistant and USDA’s top 20 most virulent. PhageGuard S is approved for use by both FDA and USDA as a processing aid.

PhageGuard S infect all Serovars:
- Effective against “Top 20” + more
- Effective against all Antibiotic resistant strains:
- Phages “don’t care” about presence of antibiotic resistance genes as they use a different attack vector
- Thus far no resistant strain has been identified

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PhageGuard S is an organic and natural antimicrobial intervention which kills *Salmonella*. PhageGuard S is tasteless and odorless, it has no impact on the organoleptic properties of the treated product and there is no risk in regard to workers safety. By applying PhageGuard S on fresh poultry or meat pre-grinding or pre-packaging, *Salmonella* is reduced by up to 2 logs or 99%. PhageGuard S is an effective anti-*Salmonella* hurdle during processing of fresh meats, leading to safer products.

In both laboratory and factory trials PhageGuard S has shown to outperform common chemicals such as PAA and CPC.

The phages in PhageGuard S are very specific for the genus *Salmonella*, and therefore cannot affect or influence the natural bacterial flora of a food or raw material used to produce food or feed.

**Regulatory**

PhageGuard S is for both USDA and FDA GRAS (GRN000468). Processing aid approvals for USDA appear in 7120.10. It is further approved as a processing aid in Canada, Australia, Israel and others. It is organic certified (OMRI USA and SKAL EU), Halal and Kosher.
5.1 PhageGuard S outperforms peracetic acid (PAA)

Organic acids including lactic and peracetic have a limited effect on reduction of *Salmonella* in ground meat in large part because the levels of organic acid required to provide >1 log kill cause damage to the protein integrity of the raw materials before grinding. Phages however give a >1 log kill and have no effect on the quality of the meat.

Yeh et al., from the University of Nevada published a paper in Meat Science (1) where the effect of PhageGuard S on trimmings was compared with 400 ppm PAA and 5% LA.

A 1% Phage solution applied pre-grinding gave more than 1 log reduction (94%), where both chemicals gave less than 0.4 log or 59% reduction.
5.2 Scientific efficacy data
Studies have confirmed that by applying PhageGuard S on fresh meat pre-grinding or pre-packaging, *Salmonella* is reduced by up to 99%. In a typical lab study performed by Certified Labs (2015) Skin-on, Bone-in chicken thighs were contaminated with *Salmonella enterica* and then treated with 1% PhageGuard S. Efficacy was within minutes and increases over time up to 24 hrs after application.

5.3 Industrial Trials
Multiple trials in US poultry plants have shown that PhageGuard S can be easily applied and gives significant reduction in *Salmonella* positives.
Giblets and Parts
Naturally contaminated Livers and Gizzards were treated with 1% PhageGuard S or tap water (control) at 4C and subsequently crust frozen for 24 hours. The samples were rinsed and tested following the USDA method. Lab work shows a 1.3 log reduction on livers and a 1 log reduction on hearts.

PhageGuard efficacy has been proven in multiple plant trials. The number of Salmonella positives reduced significantly. Number of positives for livers came down from 17% to zero; skinless necks from 34% to zero; skin on necks from 100% to 10%. For trimmings the number of positives was reduced by approximately 50%.
**Turkey Backs & Wings**

Turkey backs and wings were processed through a commercially available dip tank auger system using PhageGuard S and then ground in a POSS separator. All chemical interventions had no effect while PhageGuard S reduced the number of positives up to 100%.

PhageGuard efficacy has been proven in multiple plant trials. The number of *Salmonella* positives reduced significantly. Number of positives for livers came down from 17% to zero; skinless necks from 34% to zero; skin on necks from 100% to 10%. For trimmings the number of positives was reduced by approximately 50%.
Conclusion
The US Poultry Industry faces many challenges in regard to producing safe foods in a safe environment and phage can help replace or reduce the use of chemicals in post-harvest poultry processing.
PhageGuard S is a natural and organic antimicrobial intervention that kills *Salmonella* by using the natural enemy of the bacteria. It is a safe and powerful tool with many benefits including lack of impact on flavor, odor, texture or protein denaturation of finished products. In addition Phage is worker and environmentally friendly.
PhageGuard S is an effective anti-*Salmonella* hurdle during processing of fresh meats, leading to safer products without compromising worker safety.
PhageGuard S is not a silver bullet nor can it replace all other chemicals or organic acids used in poultry slaughter. However when used efficiently and effectively on finished products it brings great value.

Contact Micreos to discuss what PhageGuard S can do in your poultry operation:

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