

**The Effects of Electrostatic Polarization Ultra-Violet Light Filters on the
Bioaerosols of a Commercial Broiler Processing Plant Hang Room**

by

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Abstract

Poultry processing hang rooms are recognized as potentially one of the dirtiest areas of a first processing plant. Little research focuses on the amount of debris in the air of a hang room and its potential effects on cross contamination further down the processing line. In order to determine the bioaerosols in the hang room of a particular processing plant hang room, three electrostatic polarization light filters utilizing ultraviolet light were mounted on three different walls of the hang room. Over a period of 24 sampling days, the filters were turned on or off and air and settle plate samples were taken of the air in the room. Bacteria tested for were *Enterobacteriaceae* and *Salmonella* spp. Other factors including relative humidity, temperature and wind speed were also taken inside and outside the hang room. Furthermore, number of workers in the room and number of fans on were also noted. Samples were collected every 0, 3, 6, and 9 hours into the processing shift, taken back to the laboratory, incubated and enumerated. Results showed a low initial count at the 0 hour of sampling, but then significantly increased from 3-9 hours. In conclusion, it would be beneficial to find a filtering system that could withstand the load applied to it by a commercial broiler processing plant.

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I. INTRODUCTION

The hang room is thought to be the most biologically contaminated area of a broiler processing plant. It is also a potential source for cross-contamination further down the processing line; however little research has been conducted to determine if there is a way of limiting or controlling the airborne particulates within the hang room. Cundith et al. (2002a) used an electrostatic polarization Ultra-violet light germicidal air filtration systems in the processing floor of a red meat processing plant to control *Micrococcus luteus* and *Serratia marcescens*, and the apparatus reduced levels of these bacteria by 90-92%. In another trial, the units reduced indigenous airborne bacteria and molds from the ambient air by 62-77%. While this system performed well under red meat processing conditions' results cannot be extrapolated to a poultry hang room because bioaerosol loads differ greatly. Another filtration system, electrostatic space chargers (ESCS) were used in hatching cabinets to eliminate *Salmonella* and *Enterobacteriaceae*, though they did not significantly decrease the amounts of *Salmonella* in the cabinet (Mitchell et al., 2002).

II. REVIEW OF LITERATURE

The hang room and receiving docks of a poultry processing plant are considered to be heavily contaminated with dust, dirt, feathers and microorganisms (Dickens and Vaughn, 1981). As the birds are hung on the shackles, more dust and debris from the birds themselves are loosened and disperse around the room. Research has shown that highly contaminated air in the hanging area can potentially contribute pathogens and spoilage organisms further down the poultry processing line (Kang and Frank, 1989; Franco et al., 1995).

Bacteria of Concern

Enterobacteriaceae

Enterobacteriaceae include organisms such as *Escherichia coli*, *Shigella*, *Salmonella*, *Yersinia*, *Klebsiella*, *Enterobacter*, *Serratia*, *Proteus*, *Providencia*, *Edwardsiella*, *Citrobacter*. They are Gram-negative, non-spore-forming rods and some are equipped with peritrichous flagella. They are facultatively anaerobic and ferment glucose. In developed countries infections occur outside of the intestines due to the overuse of antibiotics, immunosuppressive and cytotoxic agents. Enteric organisms are the most common causes of urinary tract infections (UTI). They are also the predominant etiologic agents in cases of endogenous systemic infections and nosocomial infections. They can be isolated from feces, urine, blood, wounds, pulmonary aspirates, and

cerebrospinal fluid. Treatment is difficult because of drug resistance and also because of the presence of underlying serious diseases or impaired host defenses.

All of these mentioned are significant causes of serious infection in humans. The *Enterobacteriaceae* family is commonly used to indicate fecal contamination (Miranda et al, 2007). *E. coli* counts are also used to evaluate sanitation procedures in plants. Plants are required to test for generic *E. coli* as a means to verify process control. A plant may fall into one of three categories; acceptable (100 cfu/ml or less), marginal (over 100, but not more than 1000 cfu/ml), or unacceptable (over 1000 cfu/ml) (FSIS, 2009a).

According to Paterson (2006) many of these organisms are becoming increasingly resistant to today's antimicrobials. The number of antimicrobial resistant *Enterobacteriaceae* isolates can change depending upon the type of animal production system used, i.e. organic vs. conventional (Miranda et al., 2007). It has been shown that *E. coli* is of concern to poultry because it is indigenous to poultry fecal matter. Currently, standards in the U.S. demand zero tolerance for contamination of carcasses with fecal matter. Required sampling occurs at the end of the chiller or drip line or at the last readily accessible point prior to packaging. In broiler processing a whole bird rinse is used to collect samples, though sponging is used for turkeys. Thirteen samples must be taken over a period of time; they are not collected all at once to ensure process control for the establishment.

Salmonella

As a member of the *Enterobacteriaceae* family, *Salmonella* is a Gram-negative facultative rod. It's size is approximately 0.5µm by 2 to 3 µm. Some motile strains are

equipped with peritrichous flagella. Optimum growth temperature for *Salmonella* is 37°C. *Salmonella* infection leads to gastroenteritis. One to ten cells are needed for an infective dose with symptoms appearing in 8 - 72 h after infection. Abdominal pain, diarrhea and occasionally fever are symptoms of the infection. Within 5 - 7 days the disease relieves itself in healthy adults without the need for medication. If medical attention is needed, often intravenous fluids are administered for dehydration. If the infection spreads to the intestines, antibiotics such as ampicillin, gentamicin, trimethoprim/sulfamethoxazole or ciprofloxacin may be used (CDC, 2005).

Salmonella enterica serovar Typhmurium (ST) is a leading cause of foodborne gastroenteritis in humans and is most commonly associated with ingesting improperly handled broiler chicken meat (Fasina, 2008). The Food Safety Inspection Service (FSIS) published a Federal Register Notice, *Salmonella Verification Sample Result Reporting: Agency Policy and Use in Public Health Protection* (71 FR 9772), which helped to describe sampling practices that were to be used in the poultry industry. Under the Pathogen Reduction/Hazard Analysis and Critical Control Point (PR/HACCP) final rule, performance standards for handling *Salmonella* in broiler plants were established. A sample set of 51 carcasses is taken, if 12 or less test positive the plant is deemed to be in process control. Plants that have two consecutive sets testing less than 50% positive (6 carcasses) they are placed in category 1. These plants are then tested less often for *Salmonella*. Plants that meet or go above 50% of the performance standard without exceeding the standard fall into category 2. If a plant fails the standard they are placed in category 3. Plants that fall into the latter 2 categories are subject to more frequent inspections by FSIS. In the second quarter calendar year (CY) 2009 *Salmonella* report,

82% of all broiler plants eligible for federal *Salmonella* testing were in Category 1, 16% of broiler plants were in Category 2, and 2% were in category 3. This compares to first quarter CY2006 results: 35% (66 plants) in Category 1, 51% in Category 2, and 12% in Category 3 (FSIS, 2009b).

A survey of U.S. poultry plants discovered a 3 - 4% prevalence of *Salmonella*-positive birds entering facilities and a 35% occurrence exiting (Lillard, 1989). Another study performed by Jones et al. (1991) found that 33% of samples collected from live haul trucks were found positive for *Salmonella* and 21.4% of the processed whole carcasses were positive. Controlling *Salmonella* is a multi-step operation because there are a number of sources that contribute to salmonella contamination (Bailey et al., 2001). Sources of *Salmonella* include the chicks, feed, rodents, wild birds, insects, transportation, environment and the processing plant itself. Many conclude that the hatchery is where *Salmonella* first comes into contact with the chicks due to their susceptibility, but others suggest that the grow-out phase of a broiler's development is a more likely source due to the fact that microbial serotypes taken from the processing plant are more similar to the grow out environment versus the hatchery (Bailey et al., 2001).

Multiple intervention strategies throughout the broiler process are needed for *Salmonella* control. Once the birds are done with grow-out, other factors influence *Salmonella* occurrences during processing. These are prolonged crating (Rigby and Petit, 1980), transport of animals to the plant (Stern et al., 1995), and cross-contamination during processing (Mead et al., 1994). The National Research Council (1987) found "there is conclusive evidence that microorganisms pathogenic to humans (such as

Salmonella and *Campylobacter*) are present on poultry at the time of slaughter and at retail.”

Poultry processing plant

There are several sites of concern throughout the processing plant that contribute to microbial contamination. The scald tanks, chill tanks, debone and cut up lines have been reported to transfer bacteria from one carcass to another. Potential for airborne *Salmonella* cross-contamination was performed by Bailey et al. (2001) at six processing plants. They sampled the pre- and post-transport coops, pre- and post-scald, and pre- and post-chill water. The results from this study indicated a minimum of 2.0% recovery from plant C to a high of 9.6% recovery from plant B. It should be noted that there was no *Salmonella* recovered from the pre-chill water.

In another study (Jeffrey et al., 2001) performed at a squab processing plant, pre-slaughter, pre-evisceration and post-evisceration stages were sampled for *Salmonella*. There was an increase in *Salmonella* counts from pre-slaughter to post-processing.

Control Methods

UV light

Ultraviolet (UV) light can be used as a germicidal disinfectant. Typically the wavelength for UV processing ranges from 100 to 400 nm. This range can be subdivided even further into UVA (315 - 400 nm), UVB (280 - 315), UVC (200 - 280 nm) and vacuum UV (100 - 200 nm). The range dedicated to germicidal disinfection is the UVC because it effectively inactivates bacteria and viruses (FDA, 2009). UVA light is attributed to the changes in skin pigments that are common with tanning. The wavelength produced by UVB light burns the skin and eventually leads to cancer. The absorption of

UV light into the bacterial cell mutates the DNA and results in a sigmoidal curve, which infers S-shaped, of microbial population reduction. Combining UV light with other powerful oxidizing agents such as ozone has been shown to be effective in reducing bacterial level in juice. It is also used to disinfect water supplies and food contact surfaces.

It was noted by Seo et al. (2000) that an accumulation of dust around the UV light could potentially limit the degree of output over time. This occurs because when UV light moves over a surface it cleans where the light reaches. When there is a dust build-up around the light, less light can actually make contact with the surface in question, thus giving a less thorough kill.

In terms of UV light as a factor for eliminating bacteria, Wallner-Pendleton et al. (1994), determined that a reduction of up to 61% *Salmonella* Typhimurium could be reached on the surface of broiler breast halves when exposed. It is widely known that reducing any potential for contamination of meat will potentially lengthen the shelf life of the product. However, their research determined that the UV light did reduce *Salmonella* on the surfaces of the meat, but did not actually improve shelf life. This determination was reached because both treated and non-treated products developed a surface slime, foul odor and yellow discoloration. It has been shown that *Salmonella* organisms were reduced by three logs when exposed to UV light on agar (Bank et al., 1991). The lack of reaching a >99% kill reported by Bank et al. (1991) study as well as results from the Wallner-Pendleton (1994) study leads to speculation that the surfaces of the agar and meat were not smooth, since UV light must reach the entire surface area.

UV light absorption is not dependant on pressure, temperature or pH of the medium it's disinfecting. If there are many crevices to the medium, the light cannot penetrate, therefore all surfaces will not be disinfected. Using UV light in conjunction with other disinfecting agents is recommended to improve kill on surfaces. Further research is needed to determine the pathogens that are most resistant to UV light and to develop validation methods to ensure microbiological effectiveness (FDA, 2009).

Eletrostatic space charging systems

The electrostatic space charging system (ESCS) is another way of reducing airborne particulates. However it can become less effective over time if not kept free of debris, such as dust. In the case of most electrostatic space chargers, a strong negative electrostatic charge is transferred to the airborne dust and microorganisms. After the dust is charged it is then collected in grounded metal trays. These trays may contain soapy water to help contain the particles. Many ESCS studies deal with their usage in broiler grow-out areas or areas where live birds are housed. Electrostatic space charging systems have also been utilized in red meat processing plants. An ESCS has many beneficial properties. It does not contaminate the product with toxic products like ozone or formaldehyde. It is also cost effective, suitable for large spaces and is capable of filling spaces similar to that of a disinfecting gas (Seo et al., 2000).

Electrostatic space charging systems differ from germicidal air purification systems in that they positively charge the particles and their effectiveness lessens with the load size (Cundith et al., 2002a). In one study done by Mitchell et al. (2002) similar filters using electrostatic space chargers (ESCS) were used to reduce *Salmonella*, *Enterobacteriaceae* (ENT) and total aerobic bacteria (TPC) in a poultry hatching cabinet.

Hatching cabinets have an influx of airborne fluff and dust that are the principal sources of *Salmonella*. Air sampling for this study was done with settle plates inverted on top of the exhaust stream for 15 seconds. In the cabinets the filters did not show a significant reduction in *Salmonella*. Although, in similar studies by other researchers an ESCS reduced the amount of airborne *Salmonella* (Gast, et al., 1999; Holt et al., 1999). Gast found that *Salmonella* contamination in dust samples could be limited with the use of the ESCS in grounded chick cabinets. A grounded chick cabinet is a typical commercial cabinet that is grounded so that the electrostatic pulse does not leave the cabinet. Upstream and downstream currents of air were tested in conjunction with a negative air ionizer to reduce *S. Enteritidis*. Since the cabinets were grounded, the cabinet itself was used to attract the charged ions. A 77.7% reduction in airborne dust concentrations was realized (Gast et al., 1999). Previous research performed by Mitchell et al. (1998) suggests that the reduction of dust particles and airborne bacteria, such as aerobic bacteria and *Enterobacteriaceae*, is also possible in hatching cabinets. In addition, the airborne transmission of *S. Enteritidis* is more likely to decrease in the experimental setting (Gast et al., 1999). The most significant difference between the Gast (1999) and Mitchell et al. (2002) studies was the sampling method. Gast used 1 second intervals for 1 hour using a TSI DustTrak² in order to determine the airborne dust concentration. A TSI DustTrak² is a laser-based instrument that measures concentrations up to 200 mg/m³. Mitchell used a 10 minute interval just inside the cabinet with a TSI DustTrak³ which measures dust concentrations up to 100 mg/m³.

Using a biosafety cabinet and an electrostatic space charge system, Seo et al. (2000) evaluated the bactericidal effect of high levels of negative ions on *Salmonella*

Enteritidis. They concluded that the reduction was based on the idea that the dust particles, which were attached to the *Salmonella* organisms, were trapped by the ESCS (Seo et al., 2000). The dust could potentially increase the size of the organism and make it more likely to be trapped. They also hypothesized that the ESCS would be effective at reducing airborne particulates in other areas of the processing plant. Since the ESCS has the potential to service the entire room like a gas but is not toxic, it would potentially be effective elsewhere in a plant, microbiology lab or animal product or isolation rooms. This has yet to be tested in an extreme location, which could potentially overload the system in terms of dust and airborne bacteria.

In another study, an ESCS was successful at destroying 99.8% of mixed bacterial biofilms on stainless steel tables (Arnold and Mitchell, 2002). A small chamber with an ESCS was used to treat the mixed bacterial populations that were grown on steel coupons (1 x 4 cm). The charge was attached to the coupon at the base of the chamber. This approach could benefit in processing plants since most equipment is made of stainless steel.

ESCS and UV light

Cundith et al. (2002a) studied the effectiveness of ESCS in conjunction with the UV light components to kill airborne mold and aerobic bacteria. There were three filtration system sampling treatments (1) filters only, (2) filter and electricity and (3) filters with the electricity and UV light. One portion of the study was performed in a cold storage room and the other portion of it was performed in a processing room. The filtration system (filter, electricity and UV light) effectively removed at least 70% of the

airborne mold and up to 92% of bacteria (*Micrococcus luteus* and *M. marcescens*) that passed through the unit (Cundith et al., 2002a).

Another study was performed by Cundith et al. (2002b) evaluating duct-mounted air cleaners and germicidal air purification console units and their effectiveness in a retail sales room, meat processing room, aging cooler and chill cooler. The duct-mounted cleaners were only used in the sales room and used electrostatic polarization to filter the air. The units that included electrostatic polarization were customized to fit the heating ventilation and air conditioner unit intakes. In this study the number of duct-mounted console units used and their interaction did not have any effect on airborne bacterial levels in the processing during normal production, but did reduce mold counts when three or more filters were utilized. In the sales room where the air duct system was used, no reduction seen in bacterial counts.

Electrostatic Precipitation

St. George and Feddes (1995) investigated whether or not the use of electrostatic precipitation could be used to reduce airborne dust in a environmentally controlled area for swine. Electrostatic precipitation ionizes particles in the air and collects them on a charged surface, which may be positively or negatively charged, and distributes them onto an agar or glass surface (Kang and Frank, 1988). Many electrostatic precipitators are made for the collection of microorganisms, but are seldom used because of their complexity (Wolf et al., 1959). Through the process of ionization, ozone and nitrogen which are potentially toxic to microorganisms are produced (Kang and Frank, 1988). In this study, dust was collected on a plate which is referred to as a dust layer. It is important to note that three different air speeds were used to conduct this research (0.55 m/s, 0.76

m/s, and 0.95 m/s). No significant effect was found between the air speeds for collection efficiency. However, the use of a recirculation duct in conjunction with the electrostatic precipitator did prove to be an effective method of dust removal from the swine facility.

Bioaerosols

An aerosol can be defined as a suspension of microscopic solid or liquid particles in air or gas (Kang and Frank, 1988). Bioaerosols cause a potential risk when associated with cross-contamination as well as farm workers health. Potentially, bioaerosols include bacteria, yeasts, molds, viruses and pollen (Kang and Frank, 1988). They may originate from undetected contaminations on surfaces as well as the birds themselves (Ellerbroek, 1997). The particulate may come from the bedding materials, foodstuff and microorganism in the environment (Nielsen and Breum, 1995). Kang and Frank (1988) provided a series of descriptive characteristics for aerosol movement. Bioaerosols range in size from 0.5 to 50 μm , and their size directly affects their aerodynamic performance. The movement of aerosols results from a combination of physical influences that include the Brownian motion, electrical gradient, gravitational field, inertial force, electromagnetic radiation, particle density, thermal gradients, hygroscopicity and humidity. When sampling for aerosols, some of these effects benefit their collection, such as gravitational field, inertial force and thermal gradients. With microbiological aerosols, humidity has been known to govern where, how and in what quantities the particles reach their destinations. Vegetative cells are often stressed from aerosolization, and then further stressed from collection procedures and growth techniques.

Sampling methods for bioaerosols

Several sampling techniques are available for recovery of bioaerosols. Basically the same methods are used to collect bioaerosols as are used to collect dust and other airborne particulate. The sedimentation method relies on the force of gravity and air currents to deposit particles on an agar surface (Kang and Frank, 1988). When using this technique, results are obtained as CFU or particles/min for an exposure time of 15 min or less (Kang and Frank, 1988). If more air is forced over the apparatus a higher count of microorganisms may be found, thus a shorter time may be used.

Another sampling technique is the impinger method. When air is dispersed through a liquid which contains additives, such as proteins, the particles in the air are trapped. The fluid is then diluted and plated using a membrane filtration plating technique (Kang and Frank, 1988). Impingement methods are very useful if sampling for particles that are 1 μ m or larger and collected with high jet velocities. The apparatus is easy to break and viability loss may occur due to the amount of shear force being placed on the particles (Kang and Frank, 1988). Impingers also destroy vegetative cells and may result in overestimating counts of bacteria (Radmore and Luck, 1984).

Impaction methods can be broken down into two different types of collection, slit and sieve, but they both use an air jet to force particles down onto an agar or coated surface (Kang and Frank, 1988). Slit samplers have a tapered slit, as the name suggests. A jet stream of air is vacuumed onto an agar plate. The agar plate is usually rotated so particles are distributed evenly (Kang and Frank, 1988). The sieve sampler draws air through a large number of evenly spaced holes drilled into a metal plate. This sampler can be a single stage or a multistage. The multistage has a stack of 2, 6, or 8 stacked

sieves each with continuously smaller holes (Kang and Frank, 1988). Typically, when collecting at sites where particles are in larger numbers, this sampler will allow for more than one particle to enter. This error may be corrected by sampling for smaller amounts of time. This method of bioaerosol sampling is useful because of its high number of particle recovery, lowered sampling stress and lack of manipulation required after collection (Kang and Frank, 1988). When air is sampled with an Anderson multistage sampler, bacterial counts are higher in all environments sampled compared to the slit sampler (Kang and Frank, 1988). When sampling a swine barn and classroom, Curtis et al. (1978), found that an Anderson 8-stage sampler recovered higher numbers of airborne bacterial colony-forming particles than a 2-stage disposable air sampler. Though air sampling requires special equipment, it can be a more convenient and time-saving method as opposed to carcass sampling (Rahkio and Korkeala, 1996). Sampling air instead of carcasses allows the individual to test for microbes but not stop or disturb the line (Rahkio and Korkeala, 1996). Also, when selecting air samplers there is no obvious choice. Although, a multistage sampler is most efficient at viable particle recovery, it is not best for taking repeated sampling on a routine basis. Most samplers are adequate for determining air quality, but may not obtain the smallest viable particles (Kang and Frank, 1988).

The filtration method is also widely used in the collection of aerosols because of its cost and ease in handling. They consist of an air filtration apparatus made of cellulose fiber, sodium alginate, glass fiber, gelatin membrane filter or synthetic membrane filters mounted and connected to a vacuum. The filter is then agitated in an appropriate solution and then either incubated on an agar surface or assayed with bacteriological techniques

(Kang and Frank, 1988). This method causes stress to vegetative cells because of its tendency to dehydrate the cell during sampling (Favero et al., 1984).

Centrifugal sampling methods use force to propel the aerosol particles onto a collection plate, but do not generate a high velocity jet flow and sometimes do not generate a high enough force to drive smaller particles onto the agar (Kang and Frank, 1988). This type of sampling method would not be conducive to areas of the plant that may be overcome with microorganisms because bacteria aerosols may be very small and range from 0.5 μm to greater than 100 μm (Lutgring et al., 1997).

Environment

The enumeration of microbial populace in food processing plants has been recorded since at least 1934. In a study performed by Olson and Hammer (1934), settle plates were used in dairy plants to measure for the number of bacteria, yeasts and mold. Throughout the research performed over the last 80 years, it has been determined that there are considerable similarities in the types of bacteria, regardless of the type of processing plant, the amount of bacteria found per plant differs greatly on the area tested, and that worker activity influences the counts (Heldman, 1974). The highest counts of *Enterobacteriaceae* are found at the reception area of a processing plant, while the lowest numbers were found in the debone area (Ellerbroek, 1994). It was also found that the highest number of Gram-positive bacteria, such as *Staphylococcus*, were distributed off the skin and feathers of the birds, most likely in the hang room as well (Ellerbroek, 1994).

The effect an environment has on the quality or safety of a product depends greatly on the exposure time to that environment and the quality of housing environment

(Heldman, 1974). The human health aspect from contamination of the product, shelf life of the product and the economic impact are problems associated with airborne contamination in regards to food safety. According to a study performed by Heldman (1974), microbial particles generated in one room, are likely to be transported to another through opening in walls connecting the two. This possibility increases the chances of cross-contamination and forces one to try and control the generation of bioaerosols as early in the process as possible.

Drains at food processing facilities were flooded at 10 minute intervals with rinse water and it demonstrated that the number of particles observed decreased with the number of times the drain was put to use (Heldman, 1974). Many areas of food processing plants may become backed up and are continuously drained and clogged with water.

Heldman (1974) also examined the ventilation systems and found that the higher the ventilation air flow rates were, the higher the population of airborne microbial contamination and organic matter. This being true, when the source of contamination is located in close proximity to the sampling location, the microbial populations there increase rapidly (Heldman, 1974). Burmester and Witter (1972) indicated that filters placed at the ventilation inlets into a room can effectively reduce viral particles.

Rahkio and Korkeala (1997) found airborne bacteria to be important factors in carcass contamination. They also agree that bioaerosols and the movement of workers throughout the processing area can directly affect the contamination of carcasses. Their research was conducted in a back splitting and weigh area of a beef and pork slaughter plant and utilized Anderson impaction air samplers with a sampling time of 4 minutes.

They also studied the direction of air flow thru the plant in relation to worker movement and bioaerosol contamination of carcasses (Rahkio and Korkeala, 1997). Results indicated that there were significantly higher counts of contamination on both beef and pork carcasses in areas where workers moved in and out of different parts of the processing line (Rahkio and Korkeala, 1997). It can be concluded that movement by the workers stirs up air, and therefore bioaerosols, assists in the contamination of the carcasses. It has also been proven that the movement of workers from clean parts of the plant to dirty and back is related to higher levels of carcass contamination (Rahiko and Korkeala, 1997). To reduce contamination by air, it is imperative that adequate separation of the dirty and clean parts of the plant be in place. Air conduction systems that do not function properly can devastate the separation of the clean and dirty sections of the poultry slaughtering process Ellerbroek (1994).

Lutgring et al. (1997) sampled bioaerosols in the shackling, picking, evisceration, post chiller, cut-up, portion packaging and whole bird packaging areas of a poultry slaughtering plant using the impaction method. They found that the most common contamination in aerosol samples were bacteria. The shackle room, otherwise known as the hang room, was found to be the most heavily contaminated with bioaerosols. Though previous studies quantified microorganisms present on the product or product surfaces, very little work has been done to characterize the processing environments in regard to bioaerosols (Lutgring et al., 1997). In the hang room of a plant, airborne bacteria found were predominately Gram-negative bacteria including *Escherichia coli* (Lenhart et al., 1982). The authors found 6.5×10^5 CFU/m³ in the morning sampling compared to 9.7×10^3 and 6.5×10^3 in the afternoon sampling (Lenhart et al., 1982). These results indicate

that concentrations of airborne bacteria are elevated in the afternoon, when it is presumed to be warmer temperatures than the morning. A similar study by Kotula and Kinner (1964) also saw an elevation in bioaerosol numbers in the shacking area of the plant from the morning to the afternoon sampling. They reported that the hang room, shackling area and holding areas of a processing plant are potentially the greatest risk for bioaerosol concentrations. According to Lutgring, et al. (1997) 62% of bacilli isolates from the hang room were *E. coli*. Bioaerosol concentrations are always higher in the hang room and picking areas so it is most critical to control airflow from these places (Lutgring, et al. 1997).

Conclusion

The hang room of a broiler processing facility has been greatly overlooked in previous examinations of processing plants. Most other areas in the plant have been investigated for potential cross contamination sources although little attention was given to the hang room.

The objectives of this study were to determine whether or not the bioaerosols in the hang room of a broiler processing plant could be eliminated or reduced by utilizing UV light in conjunction with electrostatic polarization as a kill agent. It was also to determine if other factors in the interior and the exterior of the hang room had any effect on the intensity of bioaerosols within the hang room.

III. MATERIALS AND METHODS

Samples $n = 1728$, (3 medias X 2 sampling methods X 3 replications X 4 sample times X 24 trips) were taken from a poultry processing plant hang room. The hang room dimensions were 3.35 m wide X 4.88 m tall X 9.14 m long. Light filtrations units developed by Environmental Dynamics Group (Environmental Dynamics Group, Princeton, NJ, 08542) were mounted on three of the four walls of the enclosed hang room at approximately 1.52 m high. Each filter sat approximately 3.81cm from the wall, with the intake facing outwards (Figure 1). The maintenance team at the plant designed mountings for the filtering units. The first filter, position 1 (Figure 2), was placed to the right of the hang line closest to the door leading to the loading dock. This location was chosen so that the sample would be gathered where the first bird was hung on the line. The second filter, position 2 (Figure 3) was mounted on the wall approximately 0.91m behind the hang line workers and approximately 0.61m beneath three wall-mounted fans. Its placement was approximately in the center of the hang room. The location was chosen because it was the middle of the room and hang line. The third filter, position 3 (Figure 4), was placed behind the hang line, facing the workers and sat approximately 0.91m from them. This filter's placement was the dirtiest part of the hang room. The following pictures are the actual sites of the three filters.



Figure 2: Position 1. Filter was positioned beside a fan, directly to the right of the first hang crew worker.

Figure 3: Position 2. Filter was positioned beneath three fans, directly behind the hang crew line.



Figure 4: Position 3. Filter was positioned behind shackle line, to the right of the bird ramp.

Twenty-four sampling trips were taken to the plant, 12 with the units turned on and 12 without the units. Before the start of the shift the units were hung on the walls and the fans and ultra violet lights were turned on 10 minutes before the processing shift started for the sample days with the units turned on. On days when no filters were used, the sampling times, locations and procedures did not change. The filters however were not installed for those sampling periods.

Samples were taken at the three locations at the start of shift (0h), mid morning (3h), early afternoon (6h) and afternoon (9h). Sampling took place on Tuesdays, Wednesdays and Thursdays to avoid the cleanest and dirtiest work days.

Three different types of media were used to collect air samples; they were Violet Red Bile Agar (VRBA, Acumedia Manufactures, Inc., Baltimore, MD), which is a solid agar that uses lactose fermentation as an indicator of *E. coli* and coliforms. A more selective media, Violet Red Bile Glucose Agar (VRBGA, Acumedia Manufactures, Inc., Baltimore, MD) similar to VRBA, but with the addition of glucose. Xylose lysine tergitol 4 (XLT-4, Acumedia Manufactures, Inc., Baltimore, MD) was used to select for *Salmonella*. Samples were taken for each of the media in duplicate at each site using the pbi air sampler SAS Super (International PBI S.p.A., Milano, Italy, 20153) using an air sample time of 20 s (Figure 5).



Figure 5: Bioaerosol air sampling with pbi air sampler SAS Super.

This amount of time for air sampling was determined from a pretrial run of several different times in the hang room on all three media used in the trial. Air samples were taken to observe the amount of bacteria floating in the air, while settle plates were used to determine what bacteria was actually settling on equipment, carcasses, workers etc. Before the first sample and after each sample thereafter, samplers were cleaned with a 70% ethanol solution and wiped dry. A 70% solution was used because it has been shown to be the most effective for a through kill as opposed to 100% ethanol which may shock the bacteria but not kill it (OEHS, 2009). Settle plates were held flat at the sample site and left uncovered for 30 s (Figure 6).



Figure 6: Bioaerosol settle sampling.

The amount of times used for settle plate sampling was also determined by a pretrial run of several different times in the hang room on all three media used in the trial. A total of four plates of each medium (two settle, two air) per sampling site were collected making a total of 36 plates per sampling time. Prior to their use, the plates were contained in a sanitized ice chest with ice packs and then placed in a designated sanitized second ice chest for the transportation back to the laboratory. When transported back, the plates were inverted and stored in an incubator at 37°C.

The VRBA and VRBGA plates were incubated for 24 h at 37°C in a Fisher Isotemp incubator (Fisher Lab, Equipment division, Indiana, PA) and the numbers of colonies were recorded. The XLT-4 plates were incubated for 48 h in the same Fisher Isotemp incubator and black colonies were counted at 24 h and again at 48 h to count additional black colonies as described by the USDA (1998).

Weather condition sampling

Relative humidity (n = 192), wind speed (n = 192) and temperature (n = 192) were taken both inside and outside the hang room. Samples were taken at 0, 3, 6 and 9 h starting at the beginning of the first shift. Temperature (C) and relative humidity (%) readings were taken using a Mannix LAM 880D digital thermometer/hyrdometer (Mannix, Plainfield, IL, 60544). Wind speed (m/s) was measured using a Fisher Scientific Anemometer (Fisher Scientific, Pittsburgh, PA, 15275). Samples collected inside were taken behind the line of workers in the middle of the room, while outside samples were taken to the right of the loading area (Figures 7 and 8).



Figure 7: Wind speed sampling inside the hang room.



Figure 8: Temperature/ relative humidity sampling inside the hang room.

Statistical Analysis

A 2 (filter on vs. filter off) X 4 (0, 3, 6 and 9 h) X 3 (filter position 1, 2, or 3) factorial arrangement of a completely randomized design was used. Data were analyzed using a general linear model in SAS (SAS Institute, Inc. Cary, NC) to determine all main and interaction treatment effects. Means were separated using Fisher's protected least significant differences. A correlation procedure was used to determine if there were any correlations between the bacteria and environmental factors. A stepwise procedure to determine regression was used to determine the best multiple linear regression model for predicting each bacteria type.

IV. RESULTS AND DISCUSSION

The objectives of this research were to determine whether or not the bioaerosols in the hang room of a broiler processing plant could be eliminated or controlled by utilizing UV light in conjunction with electrostatic polarization as a kill agent.

When examining the bacteria levels for the three different positions of the filters in conjunction with air sampling, *Enterobacteriace* (Lactose) levels were not affected ($P > 0.05$) by position (Figure 9). The highest recorded log for the bacteria was 1.53 log CFU/50L of air. *Enterobacteriace* (Glucose) levels were also not affected ($P > 0.05$) by the filter location, its highest count recorded was 1.47 log CFU/50L of air for position 3. Isolation of *Salmonella* was significantly low in counts sampled for at all positions, but were not different from one another when comparing position location within the bacteria. Lutgring et al. (1997) found that shackling room counts were 100- 1,000-fold higher than outside concentrations, and were the highest inside the plant. They also found counts in the hang room that were in excess of 6 logs cfu, where as this research saw no more than 3 logs cfu. This may be attributed to the different sampling methods and also the bacteria types sampled for.

The lack of variation in bacteria from position to position was unexpected due to the anticipated wind movement in the room and filter placement. The initial hypothesis was that at the position of filter 3 would have had a significantly higher amount of

Enterobacteriaceae. *Salmonella* were very low, but that is attributed to the overall low *Salmonella* count throughout sampling and the plant's low incidence of *Salmonella*.

However position 3 had a significantly higher level for *Enterobacteriaceae* (Lactose) (1.87 log CFU) (Figure 10). Positions 1 and 2 *Enterobacteriaceae* (Lactose) captured on settle plates were not significantly different *Enterobacteriaceae* (Glucose) numbers followed the same trend as *Enterobacteriaceae* (Lactose). *Salmonella* were significantly low ($P > 0.05$) at position 3.

The differences between position 3 and the other two positions were expected due to the location of the filter. It was located behind the bird hang line and facing the workers. Every time they placed a bird in the shackles debris could be seen visually coming off the bird in the direction of the unit. In addition to the birds, there were fans located behind the line of workers blowing towards the filter unit. This air flow problem was also reported by Cundith (2002b), although he implemented an additional two filtration units to compensate. No other filter had the same or more air traffic directed at it than filter 3. The lack of *Salmonella* found in settle plates was consistent with air plates. This plant showed no records indicating they had a *Salmonella* problem so the results were expected. Rahkio and Korkeala (1997) found similar bacterial counts ranging from 1.21 to 3.08 log CFU in processing plants of pork and beef. The low counts found in the hang room were not expected due to the amount of visible debris in the air. A potential basis for these counts may be the sampling location. Samples taken from the beef and pork plants were taken approximately 1m from the carcasses, whereas the poultry samples were dispersed throughout the room.

The filters had no impact on airborne *Enterobacteriace* (Lactose) during sampling hours 0, 3, and 9 ($P > 0.05$) (Figure 11). Although, in hour 6 the filter on was significantly higher ($P > 0.05$) from the hour 6 filter off, the difference in reality is extremely small. This change is potentially due to build-up over the first shift. When the shift changes the counts decrease to preslaughter sampling levels. The increase at hour 6 with the filter on for *Enterobacteriace* (Lactose) may be attributed to the workers in the hang room. The first set of workers were at the end of their shift, thus the dirtiest they would have been all day. The filter may not have been able to compete with what the workers had accumulated on their clothing during the work shift and become overloaded. This overloading was not seen in previous studies performed with ESCS with and without an UV light component (Gast et. al., 1999 and Cundith et al., 2002a). Riemensnider (1966) also found that the workers were contributing to the bioaerosols in the room of a dairy plant. Since the filters were not tested on the basis of being overloaded, the elevated level may also be attributed to a higher prevalence of bacteria.

In contrast, *Enterobacteriace* (Glucose) levels were not significantly different ($P > 0.05$) when evaluated on filter status against the sampling hours for air samples over the course of 9 h (Figure 12). At 6 and 9 h sampling periods with the filters on *Enterobacteriace* (Lactose) were not significantly lower than the same sampling time with no filter (Figure 13). When comparing filter on vs. filter off over time for *Enterobacteriace* (Lactose) the first and second sampling times exhibited signs of the filters working ($P > 0.05$). The filters may have become over loaded by that 6th hour, leading to the lack of difference. In addition, the reduction by 1 log was not reached by Cundith et al. (2002b) until 4 units were utilized after at least 12h.

Enterobacteriace (Glucose) levels via settle sampling were not affected ($P > 0.05$) by filter presence (Figure 14). At 9 h a decrease is seen when compared to 3 h. This may be attributed to the eventual shift change between hours 6 and 9. Hour 9 samples were not significantly different from hour 0 suggesting that the change of workers could be responsible for limiting the bacteria in the hang room.

Use of the filter had no significant impact on airborne *Salmonella* over time. The same results were seen when analyzing *Salmonella* for potential surface contamination. Figure 15 shows the amount of *Salmonella* per trip for filter on versus filter off. Two peaks are seen with the filter not engaged, though the increase in counts does not exceed 0.20 log CFU. No trends are seen for either filter on or filter off. It may be concluded that the *Salmonella* spp. occur in peaks due to dirtier flocks passing through the hang room.

Table 1 describes the minimum, maximum, and standard deviation for the environmental data. Note that the temperature inside as well as outside the hang room minimums and maximums differ only slightly. The same applies to the relative humidity data. In contrast the outside wind-speed versus the inside wind-speed maximums differ a great deal. The large increase in the inside wind-speed is contributed to the fans inside the hang room. The side sheds range from 0 to 1, that data was taken in quarters whereas the back sheds were measured based on the number of truckloads. Many variables minimums are 0, this is explained by sampling at 0 h. Several times, the fans and people were not on or in place by the time birds were put on the conveyer belt. When evaluating whether or not a correlation existed between the environmental data and each bacteria type only small correlations were noticed (Table 2). For both *Enterobacteriace* airborne bacteria and bacteria that have the potential to contaminate processing surfaces, position was

moderately correlated ($P < 0.01$). This was to be expected because position 3 is located in a highly contaminated area. A low correlation existed ($P > 0.05$) for both *Enterobacteriace* fermenters for settle plate and the wind speed outside the room. Few other correlations were significant, but were not highly correlated.

Another objective was to determine if other factors within or outside the hang room had any effect on the intensity of bioaerosols within the hang room. When assessing the air, settle and combined plate counts for microbial levels *Enterobacteriace* (Lactose) the air plates had no significant model ($P > 0.05$) (Table 3). The settle plates had 7.41% variation accounted for by environmental factors among microbial counts ($P < 0.04$). This variation was attributed to the trip, outside wind speed, and number of workers in the hang room. When combining the settle and air plates for an evaluation, trip, inside temperature, outside wind speed, the number of workers and the left side shed contributed to 4.99% of the variation in microbial counts ($P < 0.001$).

For *Enterobacteriace* (Glucose) the variation among bacterial counts was attributed to the inside relative humidity, the door position and the right side shed and accounted for 5.38% ($P < 0.003$). The settle plates for the bacteria had no significant model ($P > 0.05$). When combining the *Enterobacteriace* (Glucose) counts for air and settle plates, 7.11% of the variation in the counts was credited to the outside temperature, inside and outside wind speed, number of fans, number of workers, the left side shed and the right back side shed ($P < 0.001$). The inside wind speed, door position, number of workers, lights, both the left and right side shed and the right back shed contributed 20.58% of the difference in airborne *Salmonella* ($P < 0.001$). The inside wind speed, door position and the number of workers inside the hang room contributed 13.51% variation in

the *Salmonella* on settle plates ($P < 0.001$). *Salmonella*, as a whole, were affected by the trip, and both the left and right side sheds for 3.65% of the variation and a ($P < 0.001$).

The reoccurrence of outside wind speed in the stepwise procedure reinforces the idea of debris flowing into the hang room and increasing the variation in microbial numbers. The number of workers is also an important factor to examine. Workers and outside wind speed are coupled together for both airborne *Enterobacteriace* (Lactose) and *Salmonella* positive. The vacuuming of air onto the plate was probably effected by the air flow around the workers. The larger the number of workers and the faster the air increased the amount of bacteria moving about the room. The airborne *Salmonella* positive were also heavily reliant on air speed. It is conceivable that the movement of air in the side and back sheds towards the hang room and into the open door with a higher number of workers is the reason for the extreme amount of variation in airborne *Salmonella* counts. The lights were on in the hang room causing the birds to become more mobile and releasing an increased number of fragments into the air, indicates the large variation in bacterial counts.

Conclusion

Overall, the filtration units seemed to be overcome with bioaerosols early on in the sampling period. From this research study, it is unknown if additional filter units could have potentially helped to reduce the amount of bacteria in the air. It is conceivable to believe if a dirtier flock was brought to the plant that the filters helped to reduce the amounts to the levels a regular clean flock would have carried. It is also possible that more than 3 filters would have provided enough control to limit the bioaerosols.

It is interesting to note that neither the airborne *Enterobacteriace* (Lactose) or the *Enerobacteriace* (Glucose) that would potentially settle on surfaces were not impacted by the environmental factors. It may be concluded that the lactose fermenters, such as *E.coli*, are lighter and able to disperse in the air, while the glucose fermenters may be more of a contamination problem.

It would also be of interest to identify what the exhaust fans located near position three are removing from the room in terms of bacteria. The air changes per hour could be determined and analyzed to predict what is being forced back into the loading dock and potentially back onto the new birds coming into the hang room.

The implications behind this research offer suggestions for changes in this particular hang room. Although these changes would affect this hang room, it is important to note that not every hang room is constructed the same and therefore results could vary. Furthermore, although a great deal of visible debris was evident, the actual bacterial contamination was low, especially when consideration for the amount of impaction air samplers sampled for. Out of the 149, 512 L of air in the hang room, only 50 L of air was sampled each time. A more desirable air flow moving out of the hang room would potentially lower the microbial counts. Moving the fan currently located behind the employees could possibly deter the air from moving outside, while still performing its duties of cooling off the employees. Changes outside the hang room would also help render air flow problems. If the back sheds were not directly facing the loading dock, but turned sideways the added barrier of the wall could possibly assist in changing the direction of the airflow from the shed. It may also assist in further cooling

the birds with the direction change. Furthermore, shutting the door leading to the loading dock would greatly reduce the amount of air brought in from the outside.

If possible a change of clothing may also help to lower the counts. In between the 3rd and 6th hour of the shift acquiring different disposable coveralls may help to reduce the amount of bioaerosols in the room as well and on the employees. If personal clothing is worn, changing the shirt in between the break could potentially reduce contamination levels.

Implementing more than three filtration systems in the hang room could decrease the load each filter took on. This could extend their efficiency to the full 9 hours, perhaps the full 2 shifts. Further research would be needed to estimate the proper number of units that would be needed.

Table 1. Minimum, Maximum, Mean, and Standard Deviation for each environmental factor^a.

Variables	Min	Max	Mean	Std. Dev.
Time	0	9	4.515	3.355
Tempi	18	34	29.46	8.984
Tempo	17.6	34.2	29.37	8.722
Spdi	0.200	12.5	2.199	1.703
Spdo	0	2	0.257	0.335
Rhi	28.2	88.4	66.019	14.785
Rho	24.4	88.1	66.837	14.437
Door	0	1	0.036	0.188
Numfans	0	7	4.599	1.125
Worker	0	9	7.603	1.336
Light	0	1	0.097	0.297
Water	0	1	0.818	0.387
Ssleft	0	1	0.692	0.422
Ssright	0	1	0.621	0.452
Bsleft	0	3	0.231	0.611
Bsright	0	8	3.30	1.936
<i>Entero.</i> (Lac)	0	3.0	1.190	0.714
<i>Entero.</i> (Glu)	0	3.0	1.143	0.656
<i>Salm.</i> Pos.	0	0.889	0.022	0.106

^a indicates environmental factors time, tempi = temperature inside, tempo= temperature outside, spdi= wind speed inside, spdo= wind speed outside, Rhi= relative humidity inside, Rho=relative humidity outside, door= door position, numfans= number of fans activated in the hang room, worker= number of workers inside the hang room, light=lights on or off, water= water presence on the floor, ssleft= side shed left, ssright= side shed right, bsleft= back shed left, bsright= back shed right, *Entero.*(Lac) = *Enterbacteriace* (Lactose), *Entero.* (Glu)= *Enterobacteriace* (Glucose) *Salm.* Pos.= *Salmonella* positives.

Table 2. Environmental factors^a correlated with bacterial types

Variables	Air Plates			Settle Plates		
	Entero. (Lac)	Entero. (Glu)	<i>Salm.</i>	Entero. (Lac)	Entero (Glu)	<i>Salm.</i>
Position	0.531**	0.518**	0.049	0.610**	0.662**	0.141*
Time	0.015	-0.006	-0.125*	-0.075	-0.049	-0.094
Tempi	-0.063	0.005	-0.044	0.016	0.008	0.015
Tempo	-0.073	-0.005	-0.048	0.005	-0.023	0.022
Spdi	0.019	0.103	-0.019	0.0686	0.110	-0.027
Spdo	0.058	0.145*	0.037	0.208*	0.181*	-0.003
Rhi	-0.061	-0.030	0.098	0.098	0.070	0.042
Rho	-0.057	-0.022	0.097	0.083	0.083	0.037
Door	0.002	-0.002	-0.025	0.097	0.124	-0.015
Numfans	-0.032	0.113	-0.021	0.054	0.127*	-0.006
Worker	0.072	0.020	0.039	0.044	0.048	0.022
Light	0.035	-0.031	-0.043	-0.052	-0.080	-0.075
Water	-0.016	0.008	-0.010	0.013	0.046	0.001
Ssleft	-0.070	-0.143*	0.030	-0.070	-0.015	0.052
Ssright	-0.084	-0.068	0.090	0.051	-0.009	0.136*
Bsleft	0.064	0.001	-0.050	-0.046	-0.040	-0.004
Bsright	0.033	0.023	-0.084	-0.061	-0.124	-0.085

^a indicates environmental factors time, position= position of filter, temp_i = temperature inside, temp_o= temperature outside, sp_{di}= wind speed inside, sp_{do}= wind speed outside, R_{hi}= relative humidity inside, R_{ho}=relative humidity outside, door= door position, numfans= number of fans activated in the hang room, worker= number of workers inside the hang room, light=lights on or off, water= water presence on the floor, ssleft= side shed left, ssright= side shed right, bsleft= back shed left, bsright= back shed right, *Entero.*(Lac) = *Enterbacteriace* (Lactose), *Entero.* (Glu)= *Enterobacteriace* (Glucose) *Salm.* Pos.= *Salmonella* positives.

* indicates $P > 0.05$

** indicates $P > 0.01$

Table 3. A Stepwise comparison of the percent of variation in microbial levels accounted for by environmental factors.

Bacteria	R²	P value	Variables
<i>Enterobacteriace (Lac)</i>			
Air	Not significant		
Settle	0.074	0.004	Trip Wind speed outside Number of workers
All	0.049	0.002	Trip Temperature inside Wind speed outside Number of workers Side shed left
<i>Enterobacteriace (Glu)</i>			
Air	0.053	0.039	Relative humidity inside Door open/closed Side shed right
Settle	Not significant		
All	0.071	<0.001	Temperature outside Wind speed inside Wind speed outside Number of fans Number of workers Side shed left Back shed right
<i>Salmonella</i>			
Air	0.205	<0.001	Wind Speed inside Door Number of workers Lights Side shed left Side shed right Back shed right
Settle	0.135	<0.001	Wind speed inside Door Number of workers
All	0.036	0.004	Trip Side shed left Side shed right

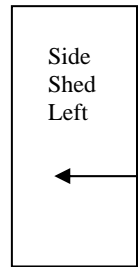
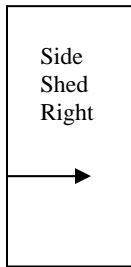
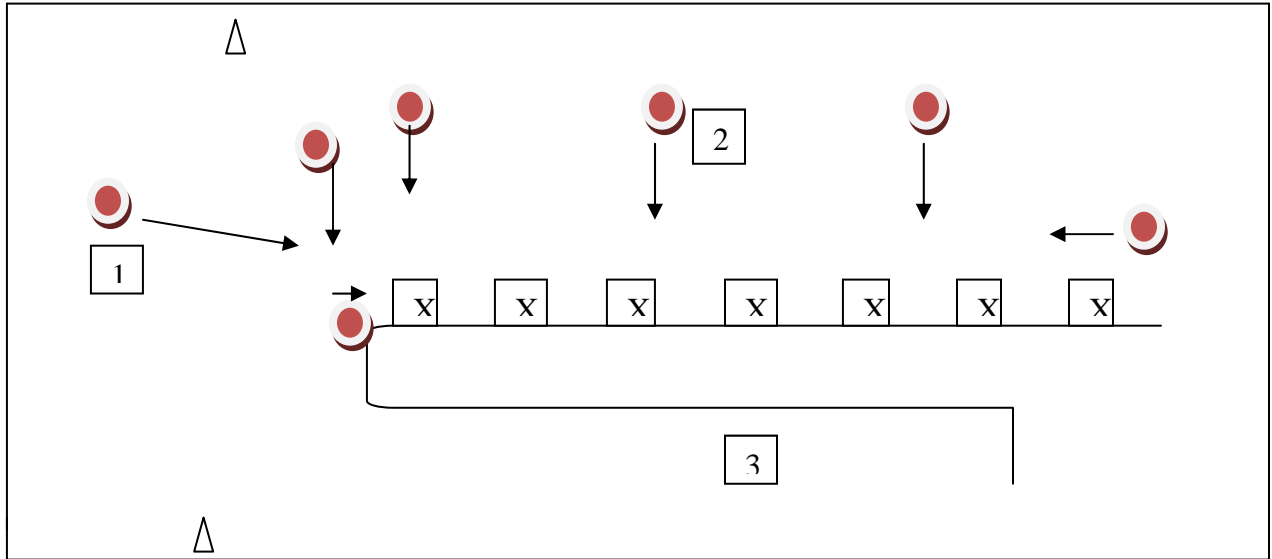




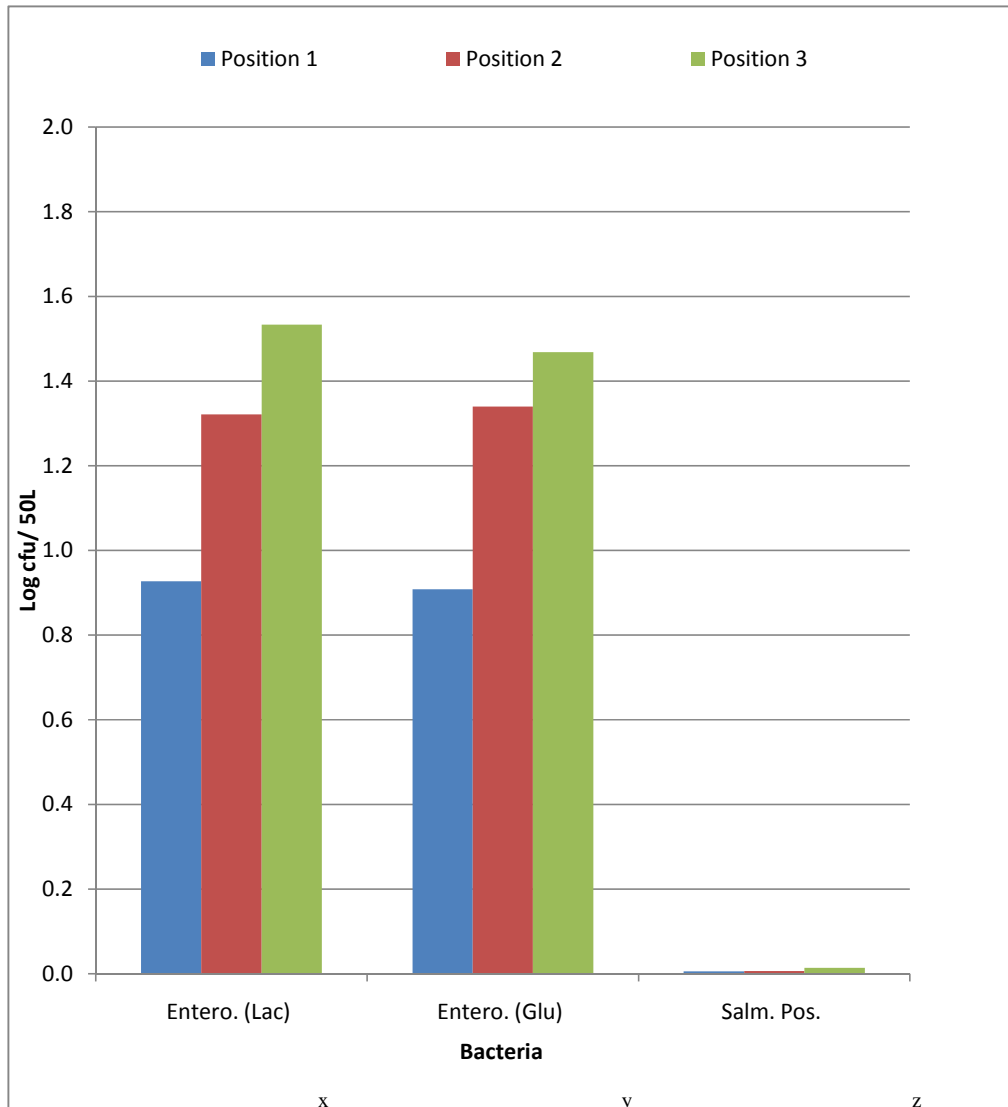


Figure 8- A schematic aerial view of the hang room of the processing plant.

-  indicates the location of the doors
-  indicates the location of the filter(s)
-  indicates the location of the fan(s)
-  indicates the location of a worker on the line

 indicates direction of air flow

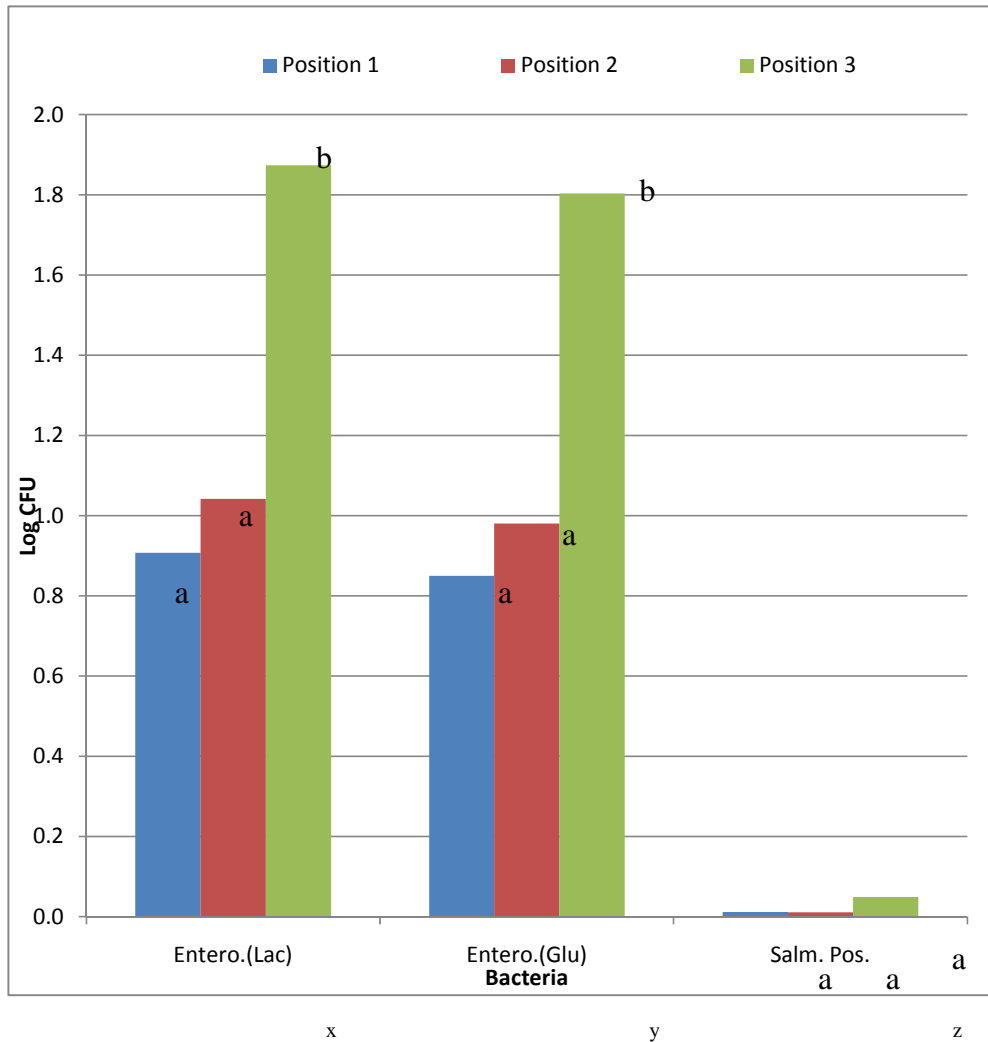


^x Entero. (Lac) indicates *Enterobacteriace* (Lactose)

^y Entero. (Glu) indicates *Enterobacteriace* (Glucose)

^z Salm. Pos. indicates *Salmonella*

Figure 9. LS means for bacteria versus position of filter for impact air samples. The positions represent the different locations of the filters within the hang room. Bacteria is counted in log CFU/ 50L of air. $P > 0.05$, no significance was found within each bacteria per position.



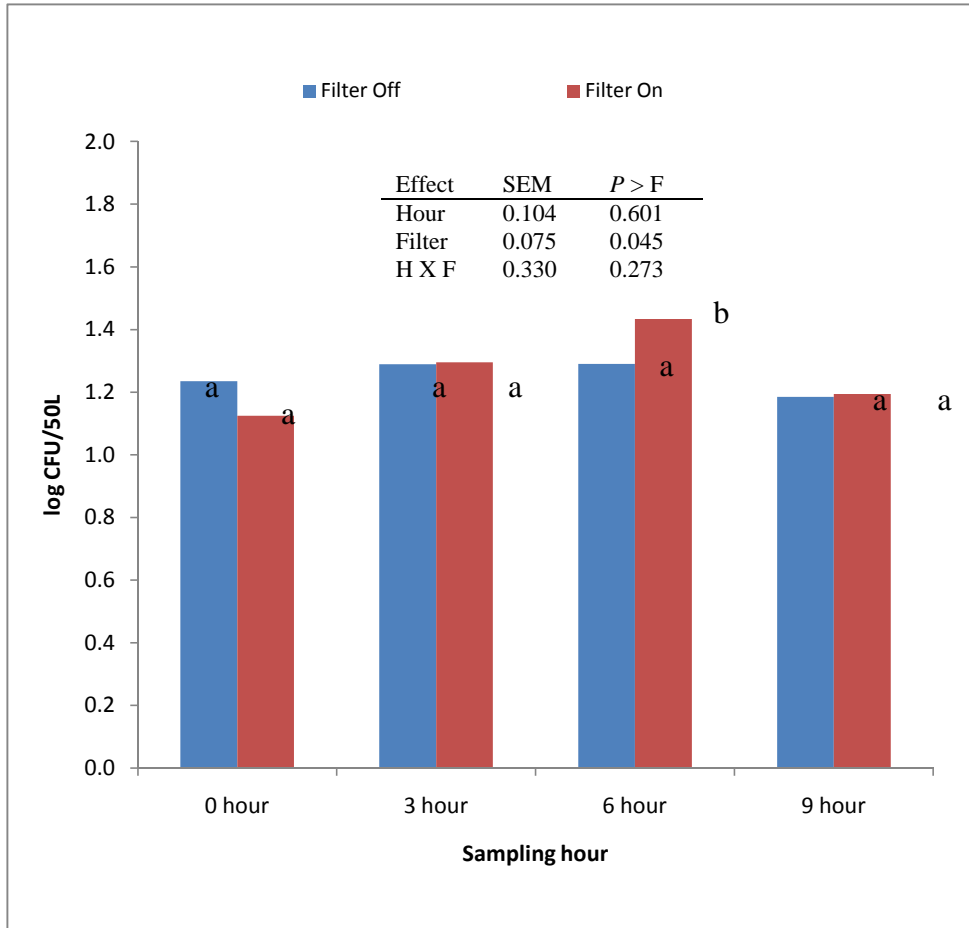
^xEntero. (Lac) indicates *Enterobacteriace* (Lactose)

^y Entero. (Glu) indicates *Enterobacteriace* (Glucose)

^z Salm. Pos. indicates *Salmonella*

Columns within a bacteria type with no common letter are significantly different.

Figure 10. LS means for filter versus position for settle plate sampling. The position indicates the location of the filters placed inside the hang room. P<0.05.



Columns with no common letter for filter main effect differ significantly $P > 0.05$
 Figure 11. Effect of filter on *Enterobacteriaceae* (Lactose) impaction air sampling over time. Counts are comparable between and within the hours. Bacteria is counted as log CFU/50L of air.

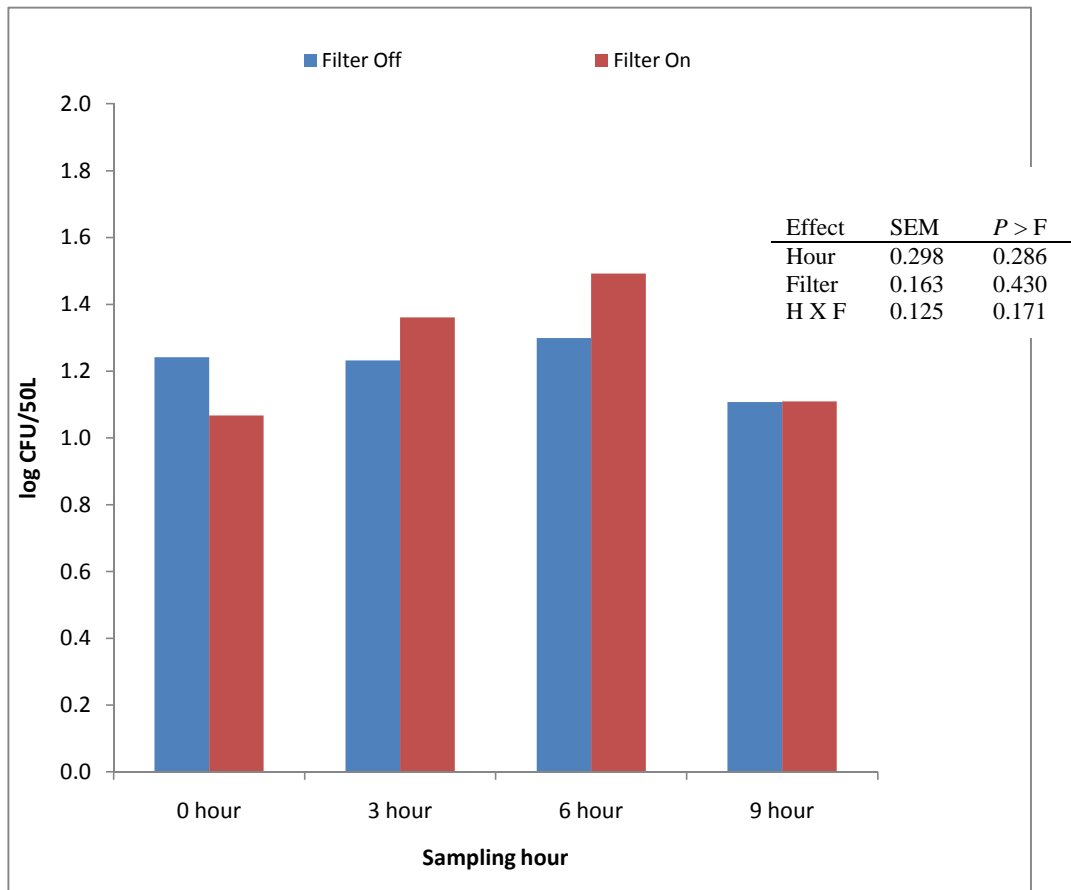
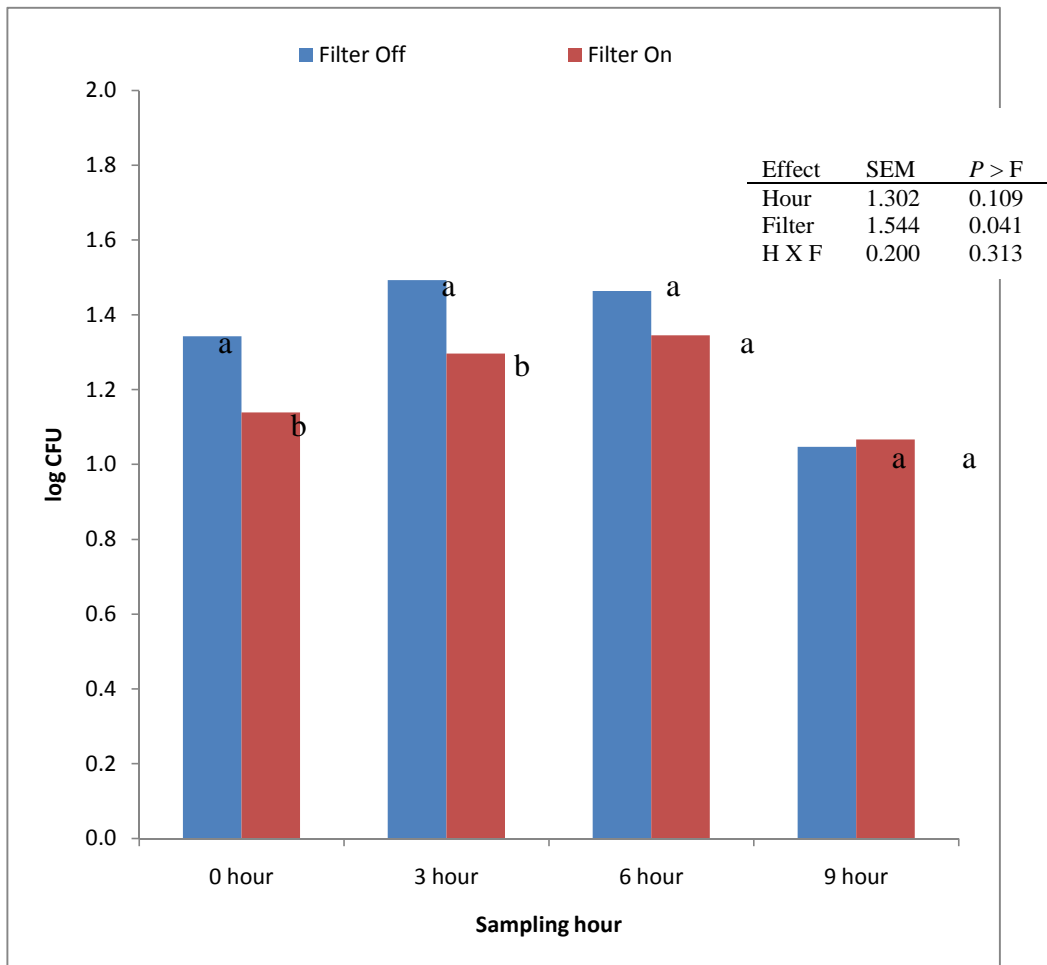
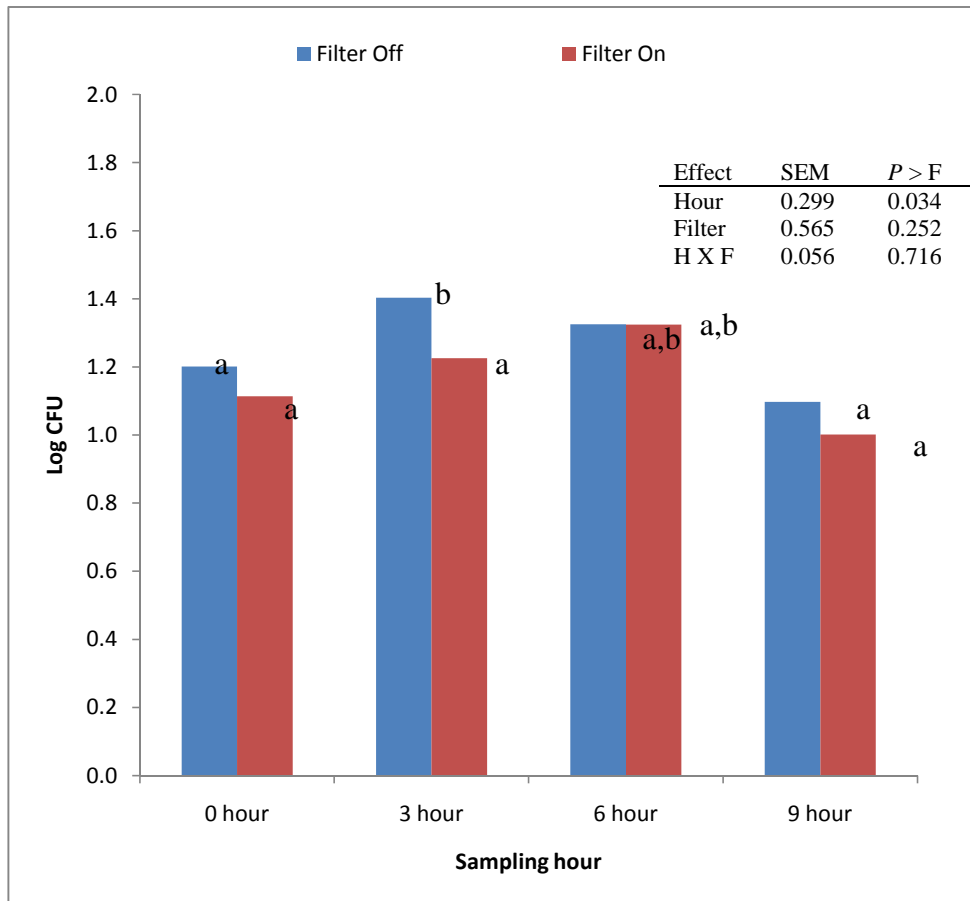


Figure 12. Effect of filter on *Enterobacteriaceae* (Glucose) impaction air sampling over time. Counts are comparable between and within the hours. Bacteria is counted as log CFU/50L of air. No significant differences exist.



Difference letters in hour between filter on and filter off are significantly different. Figure 13. Effect of filter on *Enterobacteriaceae* (Lactose) settle sampling over time. Counts are comparable between and within the hours. Bacteria is counted as log CFU.



Different letter within each hour indicate significant differences.

Figure 14. Effect of filter on *Enterobacteriaceae* (Glucose) settle sampling over time.

Counts are comparable between and within the hours. Bacteria are counted as log CFU.

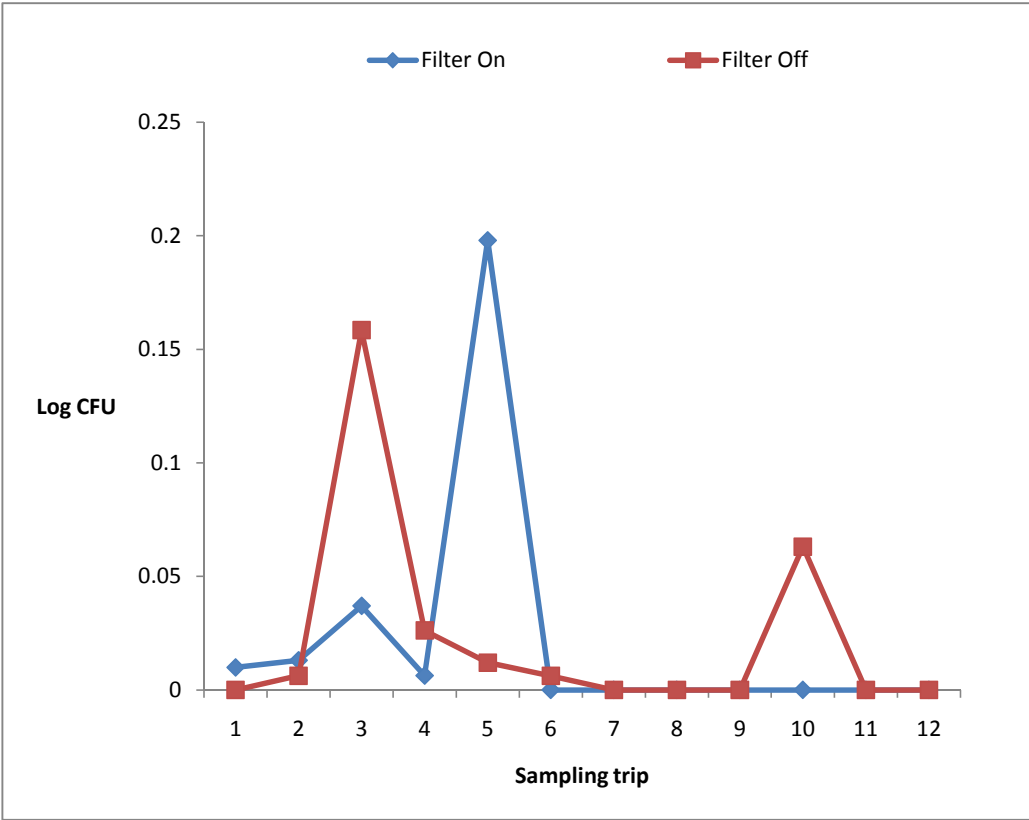


Figure 15
Combined log CFU *Salmonella* for both impaction air and settle plate sampling per day for filter on and filter off.

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