The Abundance of *Escherichia* coli in Local, Organic Poultry vs Commercially Produced Brand

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Introduction:

Poultry, when undercooked, is instrumental in transmitting foodborne pathogens. Many studies have shown that bacteria including *E. coli,* are often present in fresh poultry from commercially produced brands (Todd, E. C. 1997). *Escherichia* coli, was first identified in 1885 by a German pediatrician, Theodor Escherich (Neill, M. *et al.* 1994). *E. coli* is most commonly found in the intestines of humans and other warm-blooded animals. *E. coli* is a facultative anaerobe and is known to ferment lactose. It is an essential part of a healthy individuals gut microflora. *E. coli* is beneficial when it is balanced with other microflora in the gut and intestines. If there is too much of the bacteria present it can cause disease. Pathogens can cause mild to moderate self-limiting gastroenteritis, invasive diseases as well as more severe complications (Zhao, Cuiwei, *et al*.2001). The most common mode of *E. coli* contamination is by consuming food of animal origin.

Approximately 48 million cases of foodborne illness are diagnosed each year. Of these cases 270,000 were caused by *E. coli* (Burden of Foodborne Illness, 2018). Around 3,000 Americans die from foodborne illness every year (FoodSafety, 2018). No study has specifically compared the abundance of *E. coli* in local, organic, or non-organic commercially produced poultry meat. Research has shown if *E. coli* is present in a sample then it is reasonable to assume that more harmful bacteria might also be present.

The local, organic meat used in this project was processed naturally. No added chemicals or steps were taken to sterilize the meat after butchering. The USDA and FDA provided an exemption for small-scale on-farm poultry producers. This poultry exemption does not require the poultry to be harvested or processed at a USDA inspected facility (On Farm Poultry, 2019). This means that some small poultry operations are exempt from bird-by-bird inspection and the presence of USDA Food Safety Inspection Services (FSIS) inspectors during the slaughter and processing of their poultry. Commercial chicken processing plants have several antibacterial methods that they implement. One method is a chlorine dip. This simply means that the chicken was rinsed with chlorinated water. Just as chlorine helps make drinking water safe, it can help remove potentially harmful bacteria from raw chicken. Safe levels of chlorinated water (or chlorine washes) have been deemed safe and efficacious by the U.S. Department of Agriculture (USDA), and the U.S. Food and Drug Administration (FDA). Peracetic acid is another way poultry can be cleaned. Peracetic acid is an organic compound made of vinegar and hydrogen peroxide. It is permitted for use in poultry products that are labeled as "organic". Cetylpyridinium chloride or CPC is an antiseptic that kills bacteria and other microorganisms. It is commonly found in toothpaste, mouthwash and nasal sprays. CPC has been approved for use on poultry and is also often used. Combining all of the previously mentioned information**, I hypothesize that there will be *E. coli* present in both the commercial and local farm raised poultry meat. The commercial product has greater regulations concerning butchering and sterilizing the meat. I believe that the commercial meat will have lower bacteria counts**

than the local small farm product. Also, I believe that since the local, organic product hasn't been raised with antibiotics that the commercially produced product will have a higher chance of acquiring antibiotic resistance.

Experimental Design:

Pilot Study:

Before the main research was performed, a pilot study was done to test various concentrations of meat to buffer. The results of the pilot study concluded that 10g of meat and 20mL of PBS was the optimal ratio for testing.

Poultry Samples:

Sunrise Farms in Stuarts Draft, Virginia was the supplier of the local organic meat. The commercially produced meat was purchased from Martins in Staunton, Virginia. The meat was cut with a disposable scalpel blade and weighed on a digital scale.

Homogenizing:

A stomacher was not present at the lab, in its place a blender was used to homogenize the meat. To the blender 10 grams of meat sample and 20 mL of PBS were added. The chicken and PBS mixture were blended for 2 minutes in a sterile blender and allowed to rest for 1 minute. The solution was poured into a sterile conical tube with appropriate label. The large pieces that were not fully blended were discarded and the blender was sterilized again with lab grade alcohol. This process was repeated until all testing samples were blended. 10 samples were tested from both the local, organic, and commercially produced poultry.

Plating the Samples:

The samples were plated on nutrient agar and Charm Peel plates by pipetting 1 mL on to each plate. On the nutrient agar plate, a sterile rake was used to create a lawn of the solution. To the lawn, four antibiotic discs were added (Penicillin, Novobiocin, Streptomycin and Amoxicillin). The antibiotic resistance was tested by using the Kirby Bauer disc diffusion method. The plates were incubated for 24 hours. The 1mL on the Charm Peel plate was pipetted directly into the center of the plate and allowed to spread for 30 seconds before replacing the peeled portion. The plates were labeled and placed in a 37°C incubator for 24 hours.

Observing the Plates:

On the peel plates *E.coli* appeared as a blue or purple dot, while the coliforms appeared as red dots. All individual dots on the plate were counted. The results were recorded in terms of CFU and the plates were placed into the refrigerator for storage. The zone of inhibition for the antibiotics resistance test was measured in mm.

***Results**:*

There were more *E.coli* present in the commercially produced meat samples than in the local, organic samples. (Figure 2) When averaged there were 35.8 counts of *E.coli* for the commercially produced meat and 20.9 counts for the local, organic samples. A t-test was used on this data to determine that the results were statistically significant (p value 0.0133). When the counts for coliforms were averaged there were 4 for the commercially produced samples and 4.2 for the local, organic samples. A t-test was performed, and the data were not statistically significant (p value of 0.310). (Figure 1) The data from the antibiotic resistance test shows that there is a significant difference in the zone of inhibition for Penicillin and Novobiocin treatments (0.053,0.034). (Figure 3) However, for the other two antibiotics, Ampicillin and Streptomycin, there wasn't a statistically significant difference (0.280,0.321). By factoring in the standards for the zone of inhibition, Ampicillin was resistant in both the organic and commercial samples. Penicillin was also resistant in both categories. For Streptomycin, the organic samples exhibited resistance while the commercial samples were intermediate. Both samples showed resistance to Novobiocin.

The data from this experiment provides insight into the microbial abundance in raw poultry. The results show that the abundance of coliforms was very similar between commercially produced and local, organic poultry. Coliforms are gram-negative rod-shaped bacteria that are normally present in the intestine or digestive tract of animals including humans. Coliforms are also found in waste from plant and soil material. If coliforms are present in the meat, that would indicate that there was contamination at some point in the processing of the poultry.

***Conclusion**:*

There were significantly higher numbers of *E.coli* found in the commercially produced product as compared to the local, organic product. After researching the process of how both poultry brands are produced, a few conclusions can be made. The local, organic chicken supplier, Sunrise Farms, is exempt by the FDA under Virginia law 90-492. They are not required to test the chicken after slaughter or to sterilize the chicken. Small operations selling below a certain sales threshold and primarily into direct markets are qualified for the exemption. Sunrise Farm's method of preparing the poultry is to rinse it with water and then place it in the freezer. This practice does seem to work just as well as the sterilizing step in the commercial factories. The greater number of *E.coli* found in the commercially produced meat could possibly be a result of transportation. The local, organic meat was processed and stored on site while the commercially produced meat had to be transported from the factory to the store. During transport, the temperature has the potential to vary thus increasing the opportunity for bacteria to grow.

The n values for testing the antibiotics was 3. This is generally low for this type of test. Testing the antibiotics was an added step in this experiment. The goal of this was to enhance the study and contribute to the bacterial data. Penicillin and Novobiocin were found to have a significant higher level of antibiotic resistance in the commercially produced product than in the local, organic product.

This study adds to the pre-existing knowledge on poultry meat. It confirms that there are bacteria present in all meat no matter how it was raised or processed. Therefore, it is important to

understand how to handle meat in accordance with safety regulations to prevent harmful bacteria that are present on the meat from spreading.

References:

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