Chicken in the Real World: Comparison between bacteria found on major brands of chicken

Researchers: Dyaan Malik[1], Derick Pillaga[2]

Mentor: Dr. Marianne Williams[3]

[1]Forest Hills High School, [2] Benjamin N. Cardozo High School, [3] Queensborough Community College

Abstract

From 1995 to 2016, the estimated pounds of chicken eaten in America increased from 28.0 to 93 pounds per capita ("Per Capita Consumption of Poultry and Livestock, 1965 to Estimated 2016, in Pounds" 2016). Research has also shown that foodborne illness such as Salmonellosis approximately affects one million people in the United States each year ("https://www.cdc.gov/salmonella/," 2016). This research is in attempt to answer the question whether chicken companies are telling the truth about raising and handling their chicken products. Specifically, are there antibiotic resistant bacteria on chicken products raised antibiotic free. Also more generally, are there differences in the bacteria found on chicken raised antibiotic free, chicken raised antibiotic free with organic feed, chicken raised cage free, from those fed traditional feed. The experiment conducted used four selective media which are EMB plates, MSA plates, Sheeps Blood Agar and Mueller Hinton plates with antibiotic discs to select for certain pathogens and bacteria. In addition, swabs from each chicken were cultured on a generalized growth medium TSA. Once bacteria was cultured DNA Barcoding steps were taken to distinguish the different species found on the chickens. Results obtained from these methods were that sixteen different species of bacteria were discovered through the four different brands of chickens some of which are pathogenic to humans. In addition, the antibiotic sensitivity test indicated that all four chickens carried multidrug resistant Staphylococcus aureus. The S. aureus was resistant to ampicillin, penicillin, and cefixime. However Chicken B, a brand that claimed to raise their chickens antibiotic free with organic feed also demonstrated resistance to colistin and oxacillin. . These findings suggest that the chicken the public consumes from these companies is not safe unless handled properly and that the companies may have mislead the public about their chickens when implying that antibiotic free means that the bacteria found on the chicken might not have been exposed to antibiotics at all. Our results indicate that that may not be the case and that further testing is necessary. Many questions remain.

Introduction

•Eaten by most people in the United States, associated with many cultures, chickens (Gallus gallus domesticus) are one of the most widely distributed poultry across the world, raised and served daily.

•From 1995 to 2016, the estimated pounds of chicken eaten in America increased from 28.0 to 93 pounds per capita ("Per Capita Consumption of Poultry and Livestock, 1965 to Estimated 2016, in Pounds" 2016).

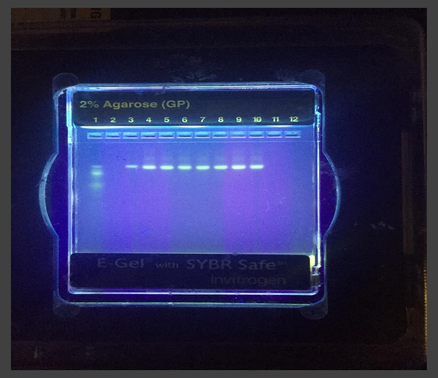
•Most companies have specific practices to meet USDA and FDA criteria. Perdue is a well known chicken brand which doesn't use hormones or steroids to affect growth rate or size. Perdue also has a "No Antibiotics Ever" campaign resulting two-thirds of their products having this label as of 2016 (https://www.perdue.com/connect-with-us/faqs/,"2016).

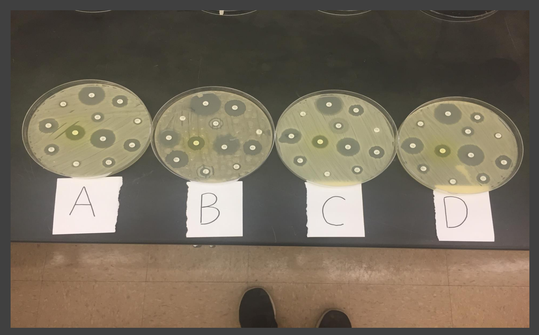
•Coleman is a well known organic brand that uses an all-vegetarian diet and the chickens are free-range ("http://www.colemannatural.com/frequently-asked-questions/," 2016).

•The experiment being conducted aims to see whether claims made by chicken companies on the feed and the antibiotic use on their chicken were true. Simultaneously, the experiment tests whether proper handling of chicken has been taken due to the fact that if bacteria are present, it can be clearly shown that the chickens are not being taken care of properly.

Materials & Methods

For the experiment there were certain methods and precautions taken. To collect bacteria researchers obtained four oven-stuffer, fresh roaster, uncooked whole chickens. One for each type of brand of chicken. All chickens were bought from one store so that preservation and handling of the chickens are controlled for. After obtaining the chickens sterile cotton swabs dipped in distilled water then were used to swab the cavity and the skin of the chicken. Once swabbing was complete the swabs were spread on 4 different media. One swab was placed on a TSA plate which is a general growth medium for bacteria while other swabs will be used on different plates that are selective for certain bacteria. Since prior research showed that E. coli, and Staphylococcus species are commonly found in the bacteria of chicken, selective media for these specific bacteria will also be used. Selective media includes MSA plate for *Staphylococcus aureus*, SBA plate for pathogens, EMB plate for *E. coli* and Mueller-Hinton plates with 12 different antibiotic filter discs to test for antibiotic resistant bacteria. The 12 antibiotics being used were penicillin, amoxicillin, neomycin, cefixime, piperacillin, colistin, vancomycin, polymyxin b, tobramycin, oxacillin, nitrofurantoin and ciprofloxacin. Bacterial colonies will be selected to isolate DNA from. Afterwards the DNA was amplified through PCR using a primer for the 16s ribosomal subunit which is specifically used for Barcoding of bacteria ("DNA Barcoding Protocol: Isolating DNA," 2014). Once DNA had been amplified gel electrophoresis was used to see if DNA was selected. Afterward DNA was sequenced and DNA Subway was used to blast samples and see the data collected.

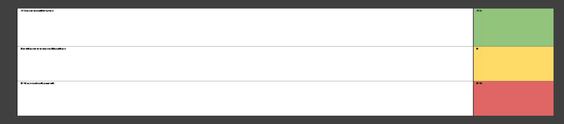




The gel electrophoresis proved that DNA has been extracted from the cavity and the skin from all chickens and that DNA Barcoding was successful. Furthermore, Mueller-Hinton plates results showed that each chicken was resistant to one or more of the twelve different antibiotics. Based on the diameter of the colonies, we were able to determine if the bacteria was resistant, intermediate, or sensitive to the antibiotic.

Results

Antibiotics	Chicken A	Chicken B	Chicken C	Chicken D
CLP 5-Ciprofloxacin 5 µg	30 mm = S	36 mm = S	22 mm = S	32 mm = S
PIP 100-Piperacillin 100 µg	18 mm = I	25 mm = S	18 mm = I	16 mm = R
CL 10-Colistin 10 µg	11 mm = S	0 mm = R	0 mm = R	9 mm = I
Va 30-Vancomycin 30 µg	18 mm = S	22 mm = S	18 mm = S	19 mm = S
PB 300-Polymyxin B 300 units	14 mm = S	9 mm = I	12 mm = S	15 mm = S
P2-Penicillin 2 units	10 mm = ?	11 mm = ?	8 mm = ?	10 mm = ?
Am 10-Ampicillin 10 µg	16 mm = R	25 mm = R	14 mm = R	14 mm = R
N 30-Neomycin 30 µg	22 mm = S	20 mm = S	22 mm = S	25 mm = S
CPM 5- Cefixime 5 µg	9 mm = R	0 mm = R	0 mm = R	9 mm = R
F/M 300-Nitrofurantoin 300 µg	22 mm = S	19 mm = S	19 mm = S	17 mm = S
OX 1-Oxacillin 1 µg	14 mm = S	8 mm = R	11 mm = I	13 mm = S
NN 10-Tobramycin 10 µg	28 mm = S	25 mm = S	27 mm = S	30 mm = S



•Barcoding results demonstrated various species of bacteria, but most notable are *Staphylococcus epidermidis, Aeromonas salmonicida, Pseudomonas poae* and *Proteus mirabilis.*

•In addition, results obtained from the antibiotic sensitivity test displayed that all four chickens contained *Staphylococcus aureus* that were resistant to ampicillin, cefixime and penicillin. These antibiotics are specifically known to target the cell walls of bacteria. Additionally, Chicken B also exhibited resistance to colistin and oxacillin.

Discussion

•The resistance demonstrated by all chickens leads to the conclusion that the chickens may have been exposed to the antibiotics beforehand or they were in contact with other bacteria that do have the resistance gene. This conclusion is due to the fact that colistin is a last resort antibiotic for gram-negative bacteria, however, within the experiment *Staphylococcus aureus*, a gram-positive bacterium has shown resistance to colistin. Ultimately, demonstrating that bacteria can exchange genes for resistance to antibiotics.

•Furthermore, one possibility for the resistance within Chicken B can be due to the fact that the company feeds their chicken probiotics. As stated on their website when they claim, "advanced nutrition to address growth rates, natural light in chicken houses, and herbal products and probiotics" ("https://www.coleman.com/," 2016). Even though the company thinks that by feeding the chickens positive bacteria, is helping them it may in fact be the cause for the resistance to these antibiotics.

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