The effect of FMB11™ and Avicorr probiotics on *Salmonella* incidence in commercial tom turkeys

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Abstract

This study examined the effect of probiotic treatment on *Salmonella* incidence in commercial tom turkeys in grow finish barns. Two probiotics were tested and administered through the drinking water. Both probiotics were started at the recommended production stage and given at their specified doses. Ten barns were tested overall, with five barns on the first probiotic and five barns on the second probiotic. Initial incidence of *Salmonella* was determined with drag swabs when the turkeys were about 17 weeks of age. Barns were assigned a particular treatment on the basis where five sites were used, each site having two barns, each barn receiving a different probiotic. Initial *Salmonella* incidence was equal for each probiotic. After the turkeys were treated and marketed, post-market drag swabs were taken with the same procedure as the initial drag swabs. Post-market *Salmonella* incidence was determined for each probiotic. While there was not a statistically significant difference in *Salmonella* incidence between each probiotic, there was a statistically significant reduction in *Salmonella* incidence overall. Results indicated a 30% reduction in *Salmonella* incidence after treatment with a probiotic. Furthermore, the data comparing each probiotic against each other, while not statistically significant due to low sample size, does seem to favor one probiotic over the other. With additional research, this could potentially become statistically significant. According to the results of this study, overall use of either probiotic does reduce the incidence of *Salmonella* in commercial market age heavy tom turkeys. However, there were uncontrollable factors that may have had an effect on the results. Weather, humidity, and litter conditions were observably inconsistent during pre-swab and post-swab analysis, which may have affected the survival of *Salmonella*. Furthermore, due to an infectious disease, several barns required antibiotic treatment during this study; removal of their data was not statistically significant.

Introduction

Foodborne pathogens are estimated to cause approximately 76 million illnesses, 325,000 hospitalizations, and 5,200 deaths in the United States annually (US CDC, 2010). The cost of these illness causing pathogens is unknown; however, it is estimated that medical costs and lost wages due to Salmonellosis, caused by *Salmonella*, incur over one billion dollars per year (US CDC, 2010).

Salmonella are microscopic organisms that can be transferred from the feces of humans or animals to other humans or animals. It has been previous thought that poultry and poultry products were the main source of *Salmonella* contamination; however produce-associated outbreaks have been increasingly prevalent (Hanning et al, 2009). From 2002 – 2003, there were 31 produce-associated Salmonella outbreaks and only 29 poultry-associated outbreaks (Hanning et al, 2009). The National Salmonella Surveillance System is conducted through public health laboratories and the National Notifiable Diseases Surveillance System (NNDSS). This information is then used to identify trends over time. Such trends conclude that *Salmonella* Enteritidis cases rose noticeably between 1980 and 1995, but showed a 34% decline between 1995 and 2006 (US CDC, 2010). Since 1997, *Salmonella* Typhimurium has been the most prevalent serotype, but also showed a 28% decline between 1996 and 2006 (US CDC, 2010). Currently, there are at least five antimicrobial agents to which *Salmonella* Typhimurium is resistant (US CDC, 2010). *Salmonella* Newport incidences have increased considerably since 1995 and present the third most frequent serotype (US CDC, 2010). Newport strains are also resistant to at least seven antimicrobial agents (US CDC, 2010).

According to a press release from the Center of Disease Control (CDC), the incidences of the most common foodborne illnesses have reached an "improvement plateau" (Division of Media Relations, 2009). These findings were collected in 2008 by the Foodborne Disease Active Surveillance Network (FoodNet), a cooperative effort of the CDC, United States Department of Agriculture's Food Safety and Inspection Service (FSIS), United States Food and Drug Administration, and ten state sites. FoodNet monitors reports of foodborne cases and performs epidemiologic studies to help health officials understand the impact these diseases have in the United States. Every year, data are collected and compared to the previous three years, as well as the first three years the data base was started, 1996-1998. From this report, it was concluded that *Salmonella* has shown the least improvement with the incidence of infections staying around 14 to 16 cases per 100,000 persons per year since the surveillance began (Division of Media Relations, 2009). Foodborne pathogens, especially *Salmonella*, pose a significant challenge to the food industry to provide a safe, disease free food supply. In order to reduce food safety problems, the government administration proposed $1.09 billion in their 2008 budget for the USDA's FSIS, which was $22 million more than 2007 (Roos, 2008). The budget also collected new fees worth $96 million per year from all the food-processing plants that it inspects (Roos, 2008). Regarding *Salmonella*, the budget included goals for the FSIS to raise the percentage of broiler chicken plants that achieve "Category 1" status, which means having a maximum of 10 percent of tested product samples that are positive for *Salmonella* (Roos, 2008).

Most research effort is targeting the improvement of post slaughter sanitation to facilitate safer meat. Despite the effort, statistics show that foodborne pathogens still pose a major threat to food safety, as well as environmental, human, and pre-slaughter animal safety. Fecal shedding of pathogens is linked to carcass contamination, therefore the grow-out and pre-slaughter stages are becoming more important in reducing incidences of contamination (Anderson *et al*, 2004). Focusing on pre-slaughter treatments against these pathogens, such as *Salmonella*, may further reduce the incidence of contamination as well as help other issues concerning the environment and safety of humans and animals.

The poultry industry has historically used growth-promoting antibiotics in the context of the competitive exclusion principle, otherwise known as colonization resistance or bacterial interference (Mountzouris, 2007). Current research utilizes probiotics as an alternative to growth promoting antibiotics (Mountzouris, 2007). By lowering populations of *Salmonella* in the gastrointestinal (GI) tracts of turkeys, shedding of this pathogen should be decreased during times of increased stress, such as transport. By using two different probiotics, used at different stages of production, this study monitored the difference in *Salmonella* incidence between treated flocks of each probiotic.

Materials and Methods

We focused on all types of *Salmonella* and the survival response to each probiotic. Our main goal was to determine if probiotic treatment would decrease the incidence of *Salmonella*, and if so, we determined the percentage decrease of positive results.

Prior to beginning treatment, 17 barns were drag swab tested for *Salmonella*. The swabs were taken on eight separate sterile surgical booties. Before swabbing, the booties were placed in a Ziploc® bag with approximately 10 milliliters of sterile saline solution to moisten the booties. The swabs were taken in the pattern described by Figure 1. At the first "X," the first pair of booties was placed over the boots. At the second "X," they were removed and properly placed into Whirl-Paks® according to protocol. A second pair of booties was placed over the boots. This procedure repeated

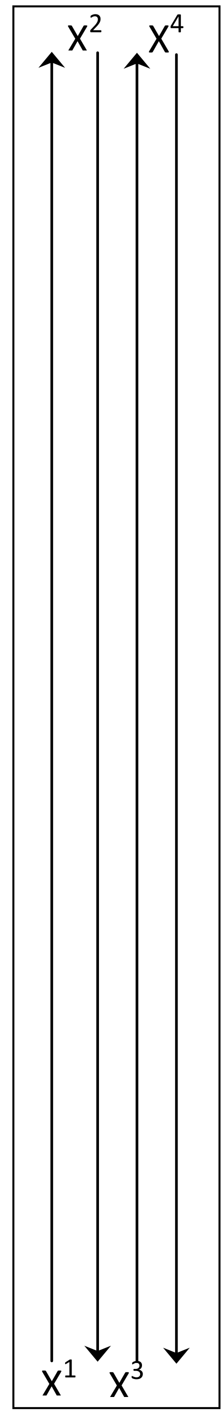

Figure 1.

itself for the next two “X’s” until 8 bootie swabs were taken. As biosecurity is a priority when entering the farms, coveralls, hairnets, gloves, and plastic boots were donned before entering the barn. Upon entering the barn, a footbath was used and a second pair of boots was placed over the first. The booties were then placed over both pairs of plastic boots. This helped to prevent any contamination from outside the barn that would skew the swab results. After each bootie had been swabbed, it was placed in a sterile Whirl-Pak® bag with approximately 10 milliliters of peptone water. The samples were kept cool until they could be transported to an out of state laboratory. Of the 17 barns tested, ten were chosen based off of initial incidence of *Salmonella*. Eight barns had 100% initial positive incidence and two barns had 75% initial positive incidence. Each treatment was designated to one of two barns per farm, with equal initial incidence of *Salmonella* among both treatments for its set of barns.

Ten turkey flocks were monitored between 18 weeks of age until market. There were two probiotic treatments used. One treatment, FMB11™ contained live *Lactobacillus fermentum, Lactobacillus helveticus, Lactobacillus paracasei, Lactobacillus saliverius,* and *Pediococcus parvulus*. This treatment began approximately 18 weeks of age, which was about two weeks before marketing. FMB11™ treatment was preceded by Optimizer, a water acidifier used for 24 hours prior to FMB11™ treatment to reduce biofilm in the water lines and enhance delivery of beneficial microflora of the probiotic. Five of these flocks received the probiotic FMB11™ with Optimizer. On day one, the grower was instructed to shut chlorine treatment off in the morning and to start running the allotted ten gallons of Optimizer solution. Eighteen week old turkeys will consume ten gallons of stock solution, so one gallon of Optimizer concentrate was mixed with five gallons water to make stock solution. This solution would run for one day and be refilled as needed. On day two of treatment, the turkeys were pulled off the Optimizer solution in the morning and were immediately given the FMB11™ probiotic. The turkeys also drank ten gallons stock solution of treatment per day. One bottle of FMB11™ concentrate powder was initially mixed in one gallon of water with a half pack of vaccine stabilizer. Then half a gallon of this solution was mixed with two packs of vaccine stabilizer to five gallons of water to make the final stock solution. The turkeys were kept off chlorine and the FMB11™ solution was refilled as needed through day two. On day three of treatment, the turkeys were pulled off FMB11™ treatment after they consumed ten gallons. Producers were instructed to begin running chlorine when probiotic treatment stopped. Water consumption levels were monitored to make sure there was not a decrease from normal consumption.

The other five of these flocks received the second treatment, Avicorr. Avicorr contains dried *Propionibacterium freudenreichii* fermentation product, and treatment began six days prior to market and lasted four days. On the night before the first day of treatment, the grower was instructed to shut off chlorine treatment. On day one of treatment, the turkeys were started on ten gallons of Avicorr stock solution. Turkeys will consume ten gallons of stock solution at six days preslaughter, so one jar of Avicorr concentrate was mixed with five gallons water and two packs vaccine stabilizer to make the stock solution. This solution was made twice per day in order for the turkeys to consume ten gallons. On days two, three, and four, the turkeys were given ten gallons of Avicorr stock solution per day while being kept off chlorine water treatment. On day five, the turkeys were taken off probiotic treatment and producers were instructed to resume chlorine water treatment. Producers were instructed to begin running chlorine when probiotic treatment stopped. Water consumption levels were monitored to make sure there was not a decrease from normal consumption.

Results

Overall initial *Salmonella* incidence was equal for each probiotic. After the turkeys were treated and sent to market, post-market drag swabs were taken with the same procedure as the initial drag swabs. Post treatment *Salmonella* incidence was determined based on each probiotic. While there was not a statistically significant difference in *Salmonella* incidence between each probiotic, there was a statistically significant reduction in *Salmonella* incidence in post-treatment of either probiotic. Results indicated a 30% reduction in *Salmonella* incidence after treatment with a probiotic. Statistical significance was based off of a P-value <0.5.

	Farm	Pre-Treatment + (out of 8 swabs)	Post-Treatment + (out of 8 swabs)	Pre-Treatment Infection Rate (%)	Post-Treatment Infection Rate (%)	Percent Decrease
FMB11™	Farm A	8	7	100	87.5	12.5
	Farm B	8	8	100	100	0
	Farm C	8	8	100	100	0
	Farm D	6	2	75	25	50
	Farm E	8	4	100	50	50
	Average	**7.600**	**5.800**	**95.000**	**72.500**	**22.500**
	Farm	**Pre-Treatment + (out of 8 swabs)**	**Post-Treatment + (out of 8 swabs)**	**Pre-Treatment Infection Rate (%)**	**Post-Treatment Infection Rate (%)**	**Percent Decrease**
Avicorr	Farm A	6	1	75	12.5	62.5
	Farm B	8	8	100	100	0
	Farm C	8	3	100	37.5	62.5
	Farm D	8	3	100	37.5	62.5
	Farm E	8	8	100	100	0
	Average	**7.600**	**4.600**	**95.000**	**57.500**	**37.500**

Figure 2. Treatment chart indicating pre- and post-treatment results. Highlighted rows designate flocks that required antibiotic treatment during the trial.

Farm	Pre-Treatment + (out of 8 swabs)	Post-Treatment + (out of 8 swabs)	Pre-Treatment Infection Rate	Post-Treatment Infection Rate	Percent Decrease
Farm A	8	7	100	87.5	12.5
Farm B	8	8	100	100	0
Farm C	8	8	100	100	0
Farm D	6	2	75	25	50
Farm E	8	4	100	50	50
Farm A	6	1	75	12.5	62.5
Farm B	8	8	100	100	0
Farm C	8	3	100	37.5	62.5
Farm D	8	3	100	37.5	62.5
Farm E	8	8	100	100	0
Average	**7.600**	**5.200**	**95.000**	**65.000**	**30.000**
Farm	**Pre-Treatment + (out of 8 swabs)**	**Post-Treatment + (out of 8 swabs)**	**Pre-Treatment Infection Rate**	**Post-Treatment Infection Rate**	**Percent Decrease**
Farm A	8	7	100	87.5	12.5
Farm B	8	8	100	100	0
Farm D	6	2	75	25	50
Farm E	8	4	100	50	50
Farm A	6	1	75	12.5	62.5
Farm B	8	8	100	100	0
Farm D	8	3	100	37.5	62.5
Average	**7.429**	**4.714**	**92.857**	**58.929**	**33.929**

Figure 3. Treatment chart indicating use of either probiotic. Highlighted rows designate flocks that required antibiotic treatment. The second set of numbers compiles all the flocks that did not require antibiotic treatment.

Discussion

While every effort was made to keep tests as consistent as possible, there were uncontrollable factors that may have had an effect on the results. Weather, humidity, and litter conditions were not the same during pre-swab and post-swab analysis, which may have affected the surviving capabilities of *Salmonella*. Weather conditions during pre-treatment swabs were hot and humid. Temperatures ranged from 60°F to 98°F with an average temperature of 80°F. Humidity ranged from 58% to 98% with an average humidity of 86%. Post-treatment weather conditions were considerably favorable. Temperatures ranged from 61°F to 90°F with an average temperature of 76°F. Humidity ranged from 36% to 82% with an average humidity of 52%. Pre-treatment litter analysis described litter conditions as dry, sandy, good, semi-dry, fair, poor, wet, and swampy. Post-market litter analysis described litter conditions as dry, dusty, and very few damp areas. Although several barns required antibiotic treatment during this study, removal of their data was not statistically significant.

Conclusion

The data comparing each probiotic against each other, while not statistically significant due to low sample size, seems to favor one probiotic over the other. With additional research, this could potentially become statistically significant. According to the results of this study, overall use of either probiotic does reduce the incidence of *Salmonella* in commercial tom turkeys.

Implications

With tightening government regulations on incidence of *Salmonella* in processing plants, and the movement away from sub-therapeutic antibiotic use; turkey producers are forced to seek alternate methods of controlling disease and pathogens that cause foodborne illnesses. Focus is now be concentrated the use of probiotics during grow-out. Similar research to that in this trial will be crucial in determining new methods.

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