

## Food Safety 2

**W98 Efficacy of ultraviolet light systems for control of microorganisms in poultry and beef brine and marinade solutions.** K. L. Beers\*, P. E. Cook, C. W. Coleman, and A. L. Waldroup, *MCA Services, Rogers, AR*.

Brine and marinade solutions injected into raw beef and poultry products have increasingly become a food safety concern due to continuous injection and recycling of the solution, allowing for a significant microbial increase. This results in several food safety issues including transfer of microorganisms into the sterile muscle of the products and deterioration of the injection solution as microorganisms increase to unacceptable levels. Safe Foods Corporation conducted a series of 8 individual trials, either in a laboratory setting (MCA Services, Rogers, AR) or at USDA-inspected processing facilities, to investigate the effects of using commercially available UV light (UVL) systems, specifically, *FreshLight* models 210 and 220, for control of naturally occurring and inoculated pathogens in these solutions. The laboratory testing included inoculation of solutions with pathogenic bacteria including *Salmonella typhimurium*, *E. coli* O157:H7 and *Listeria innocua*. Processing plant trials involved evaluating solutions for naturally occurring organisms. The flow rate in the UVL systems ranged from 30 to 150 L/minute depending on the UVL model and application. In all 8 trials, regardless of the bacteria evaluated (naturally occurring organisms or inoculated pathogens), location of testing (laboratory or processing facility) or the UVL system utilized, the original level of microorganisms in the solutions was significantly reduced by >2 logs/mL (99%) to as much as 6 logs/mL (99.9999%) at standard plant operating conditions. In some trials, the UVL system eliminated all microorganisms from the solutions. In conclusion, the commercially available *FreshLight* 210 or 220 is fully capable of consistently controlling, and/or eliminating, microorganisms from poultry and beef brine and marinade solutions while providing the processor with an affordable means of significantly improving the safety and quality of their injected products.

**Key Words:** *FreshLight*, ultraviolet light, brines and marinades

**W99 Antimicrobial susceptibility profile of enterotoxigenic *Staphylococcus* sp. recovered from foodborne outbreaks in Minas Gerais state, Brazil, from 1998 to 2002.** J. F. Veras, C. F. A. M. Penna, M. R. Souza, M. O. Leite, L. M. Fonseca, L. S. Carmo, and M. M. O. P. Cerqueira\*, *Federal University of Minas Gerais state, Belo Horizonte, Minas Gerais, Brazil*.

The aim of the present study was to evaluate the antimicrobial susceptibility profile of the *Staphylococcus* sp recovered from 16 outbreaks involving milk and dairy products that took place from 1998 to 2002 in the State of Minas Gerais, Brazil. These outbreaks were investigated by Fundação Ezequiel Dias (FUNED), Brazil and in 16 food poisoning outbreaks, 14 (87.5%) were due to cheese consumption and 2 (12.5%) were due to raw milk intake. For the isolated food, the main agents were enterotoxigenic *Staphylococcus* sp and *Salmonella* sp, *Shigella* sp. and fecal coliforms. The average of *Staphylococcus* sp strains was  $8.6 \times 10^7$  CFU/g or mL (7.93 log CFU/g or mL). The enterotoxins detected isolately were SEB and SEC, and in association were SEA + SEB. For antimicrobial susceptibility profile of 152 samples recovered, at least 48 samples (31.57%) were resistant to one or more antimicrobial. It was observed that 18.42% (28) of these samples were resistant to penicillin G, 12.5% (19) to erythromycin, 7.9% (12) to tetracycline, 6.5% (10) to oxacillin, 5.92% (9) to cephalosporin, 3.28% to gentamycin, and same

percentage to chloramphenicol. *S. aureus* showed the highest percentage to antibiotic resistance.

**Key Words:** antimicrobial, susceptibility, enterotoxigenic *Staphylococcus* sp.

**W100 Occurrence and antimicrobial resistance of *Campylobacter jejuni* isolated from poultry carcasses commercialized at the Federal District area, in Brazil.** A. P. Santana\*, D. C. Ruy, H. M. Moura, and S. Perecmanis, *Universidade de Brasília, Brasília, DF, Brasil*,

Worldwide, campylobacteriosis is a disease that causes diarrhea in humans and poultry have been identified as a significant source of the *Campylobacter jejuni*. Frequently this disease is associated with the consumption of contaminated poultry meat or cross-contaminated with other foods. In Brazil, there are few researchers studying the occurrence of this microorganism and analyzing antimicrobial resistance in this type of food, however, not at the Federal District area. The aim of this work was to verify the occurrence of *C. jejuni* in cooled carcasses of poultry commercialized in the Federal District, as well as to analyze the antimicrobial resistance. From a total 101 samples of cooled carcasses were analyzed, 9 of them were acquired in popular markets without previous sanitary inspection and 92 samples with. Considering the 101 samples, *C. jejuni* was detected in 17.82% of samples (18 carcasses), all from markets with previous sanitary inspection. However, *C. jejuni* was not detected in any of the 9 samples from popular markets without sanitary inspection. In this study, a total of 18 strains were isolated from the carcasses but only 16 were successfully not cultivated in plates. The 16 strains of *C. jejuni* were submitted to the antimicrobial analysis to 8 drugs (nalidixic acid, streptomycin, gentamycin, erythromycin, amoxicillin, chloramphenicol, ciprofloxacin and tetracycline). All 16 (100%) strains of *C. jejuni* were resistant to ciprofloxacin, 15 (93.75%) were resistant simultaneously to nalidixic acid, streptomycin, tetracycline and gentamycin, 14 (87.5%) of strains were resistant to amoxicillin, 11 (68.75%) were resistant to erythromycin and the minor resistance observed was to chloramphenicol 6 (37.5%). Considering the presence of antimicrobials resistance, these results were similar to other countries, however superior in percentage of resistance, except for ciprofloxacin and nalidixic acid. These results suggest imperfections in many phases of processing of poultry carcasses and a possible problem of public health due the high resistance observed.

**Key Words:** resistance, *Campylobacter jejuni*, chicken meat

**W101 Antibacterial activity of trans-cinnamaldehyde, eugenol, carvacrol, and thymol on *Salmonella* Enteritidis and *Campylobacter jejuni* in chicken cecal contents *in vitro*.** A. Kollanoor Johny\*<sup>1</sup>, M. J. Darre<sup>1</sup>, M. I. Khan<sup>2</sup>, A. M. Donoghue<sup>3</sup>, D. J. Donoghue<sup>4</sup>, and K. Venkitanarayanan<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Connecticut, Storrs*, <sup>2</sup>*Department of Pathobiology and Veterinary Science, University of Connecticut, Storrs*, <sup>3</sup>*Poultry Production and Product Safety Research Unit, ARS, USDA, Fayetteville, AR*, <sup>4</sup>*Center for Excellence in Poultry Science, University of Arkansas, Fayetteville*.

*Salmonella* Enteritidis and *Campylobacter jejuni* are 2 major food-borne pathogens that are transmitted through poultry products. These pathogens colonize the chicken cecum leading to contamination of carcasses during slaughter and subsequent processing operations. We investigated the antimicrobial efficacy of 4 GRAS-status, plant-derived molecules

namely, trans-cinnamaldehyde (TC), eugenol (EUG), carvacrol (CAR) and thymol (THY) against *Salmonella* Enteritidis and *Campylobacter jejuni* in chicken cecal contents in vitro. The plant molecules were added at different concentrations (ranging from 10 to 75 mM for *S. Enteritidis* and 10 to 30 mM for *C. jejuni*) to autoclaved chicken cecal contents inoculated with  $\sim 7.0 \log_{10}$  CFU/mL of *S. Enteritidis* or *C. jejuni*. The pathogen populations in the cecal contents after 15 s, 8 h and 24 h of incubation at 39°C were determined. Duplicate samples of treatments and control were included, and the study was replicated 3 times. *C. jejuni* was more sensitive to all the molecules than *S. Enteritidis* ( $P < 0.05$ ). All molecules were highly bactericidal, with the lowest concentration of TC (10 mM) significantly reducing ( $P < 0.05$ ) *S. Enteritidis* populations by  $> 6.0 \log_{10}$  CFU/mL after 8 h and  $> 8.0 \log_{10}$  CFU/mL after 24 h of incubation. TC at 25 mM completely inactivated ( $P < 0.05$ ) *S. Enteritidis* by 8 h of incubation. On the other hand, TC at all tested concentrations (10, 20, and 30 mM) completely killed *C. jejuni* ( $P < 0.05$ ) after 8 and 24 h of incubation. CAR and EUG completely inactivated *S. Enteritidis* and *C. jejuni* at 50 and 75 mM and 20 and 30 mM, respectively. THY was also equally effective in killing both pathogens. The results indicate that these aforementioned molecules could potentially be used to reduce *S. Enteritidis* and *C. jejuni* in chicken ceca, however follow up in vivo studies are necessary.

**Key Words:** *Salmonella*, *Campylobacter*, antimicrobial, plant-derived molecules

**W102 Effects of dietary antimicrobials on fecal shedding of *Campylobacter*, *Salmonella*, and Shiga-toxin producing *Escherichia coli* in production swine.** J. E. Wells\*, N. Kalchayanand, E. D. Berry, and W. T. Oliver, USDA, ARS, US Meat Animal Research Center, Clay Center, NE.

Antimicrobials are used in swine diet to improve growth and reduce disease. Increasing pressure to remove antimicrobials from swine diets could alter efficiency, but little information is reported on the impact of feeding antimicrobials on zoonotic pathogen shedding. Barrows (n = 160) were sorted by weight into 2 treatments, and fed growing, grow-finishing, and finishing diets from age 10 to 14 wk, 14 to 18 wk, and 18 to 22 wk, respectively. For each feeding phase, diets were prepared without (A-) and with (A+) dietary antimicrobials (chlortetracycline, 10 to 18 wk; bacitracin, 18 to 22 wk). At wk 10, 14, 18, 19, 20, and 22, fecal swabs were collected from each animal for *Campylobacter* and *Salmonella* spp. and fecal grabs were collected from one-quarter of the piglets for Shiga-toxicogenic *Escherichia coli* and Shiga-toxin gene analyses. *Salmonella* was only observed early in the study at low prevalence and was not affected by treatment. *Campylobacter* was not found in piglets at wk 10, but prevalence increased over the study for both treatments. In the growing and grow-finishing phases (wk 14 and 18), *Campylobacter* was present in 23 and 7% ( $P = 0.04$ ) and *stx* genes were present in 25 and 17% ( $P = 0.05$ ) of samples from A- and A+ groups, respectively. In the finishing phase, time was an interaction with treatments. *Campylobacter* did not differ between treatments for pooled wk 19 and 20 (15 vs. 9.4%, A- and A+;  $P = 0.09$ ) but was different for wk 22 (19 vs. 38%, A- and A+;  $P = 0.007$ ). The prevalence of *stx* genes was not different between treatments for wk 19 (39 vs 26%, A- and A+;  $P = 0.06$ ), but by wk 20 and 22 *stx* gene prevalence was different (49 vs. 68%, A- and A+;  $P = 0.01$ ). Shiga-toxin producing *E. coli* serogroups O26, O103, and O145 were isolated from 7.2% of the samples with fewer positives found at end of trial ( $P > 0.1$ ). Diets with chlortetracycline reduced pathogen shedding, but switching to bacitracin at 18 wk of age increased pathogen shedding with time compared with the A- group.

**Key Words:** antibiotics, food safety, pathogens

**W103 Persistent effect of thymol and diphenyliodonium chloride against *Campylobacter coli* in vitro.** N. A. Krueger\*, R. C. Anderson, T. R. Callaway, T. S. Edrington, and D. J. Nisbet, USDA-ARS Southern Plains Agriculture Research Center, Food and Feed Safety Research Unit, College Station, TX.

*Campylobacter* are important foodborne pathogens that may colonize the gut of food producing animals. The objective of this experiment was to determine if a single administration of the purported deaminase inhibitors, thymol and diphenyliodonium chloride (DIC), each may effectively reduce *Campylobacter coli* concentrations during consecutive batch culture of mixed populations of swine gut bacteria. Gut bacteria present in freshly collected fecal material (0.3 g) were mixed with 150 mL anaerobic (100% N<sub>2</sub>) Bolton broth and inoculated with approximately 10<sup>6</sup> colony-forming units (CFU) of an overnight grown (in Bolton broth) *C. coli* culture. Ten-milliliter volumes were distributed in triplicate to crimp top tubes preloaded with 0.1 mL volumes of water, thymol or DIC to achieve a single initial treatment of either 0, 1.0 mM thymol or 0.01 mM DIC and were incubated at 39°C. After 8 h incubation, 1 mL was withdrawn from each tube and inoculated into a second series of tubes containing 9 mL fresh anaerobic Bolton broth but without treatment additions. The second tube series was then incubated for 8 h after which time the process was repeated to achieve the completion of 5 consecutive batch cultures. Log<sub>10</sub>transformations of *C. coli* CFU determined at the beginning and end of each culture series were subjected to a general ANOVA. Results revealed a treatment × time × series interaction ( $P < 0.0001$ ). Counts measured upon initiation of the first incubation series averaged  $5.73 \pm 0.05$  (SD) log<sub>10</sub> CFU mL<sup>-1</sup> but declined ( $P < 0.05$ ) more than  $3.4 \log_{10}$  CFU mL<sup>-1</sup> from this value by the end of the first culture series for thymol and DIC-treated cultures and never recovered thereafter; being below our detection limit ( $1.3 \log_{10}$  CFU mL<sup>-1</sup>) by the end of the third transfer series. Conversely, *C. coli* counts in nontreated cultures were not reduced from initial values until the end of the third cultures series, at which time *C. coli* counts were  $2.7 \log_{10}$  CFU mL<sup>-1</sup> lower ( $P < 0.05$ ) but always above our level of detection. Results demonstrate that a single initial treatment of thymol or DIC effectively and persistently reduced *C. coli* during anaerobic culture in vitro.

**Key Words:** *Campylobacter*, food safety, swine

**W104 Evaluating different gas delivery methods that create a microaerophilic environment for culturing *Campylobacter jejuni*.** M. D. Haines\*, K. N. Eberle, C. D. McDaniel, and A. S. Kiess, Mississippi State University, Mississippi State.

Most techniques used for culturing *Campylobacter* species require a microaerophilic gas atmosphere. Currently there are several different methods available to deliver the appropriate microaerophilic gas environment. The objective of this experiment was to evaluate *Campylobacter jejuni* growth using 3 different gas delivery methods (Anoxamat, Campy EZ Gas-Pak, and Zip-Lock bags). Approximately 50 to 100 *Campylobacter* cells were suspended in brucella broth and spread plated onto Campy-Cefex agar plates. Plates were placed into either Mart anaerobic canisters or zip-lock bags for culturing. The microaerophilic gas was delivered to the plates in the Mart anaerobic canisters by either the Anoxomat AN2CTS Mark II System or through the activation of 3 Campy EZ Gas-Paks. For plates being incubated in zip-lock bags, the microaerophilic gas was delivered by directly flushing the bag with a pre-mixed gas; once the bag was full it was sealed. The canisters and zip-lock bags were then placed into a low temperature incubator for 24 h, at 42°C. After the 24 h incubation period, plates were counted. The entire experiment was then repeated. The results indicated that no difference in colony counts existed between the methods evalu-

ated. Colonies on plates which had the gas delivered by the Campy EZ Gas Pak method were much smaller in size than colonies on plates that had the gas delivered by the other 2 methods. In conclusion, all 3 gas delivery methods were able to produce similar *Campylobacter* results between experimental runs. Additionally, the smaller colonies from the EZ Gas-Pak method could be a result of our media choice or the anaerobic chamber used. It is important to consider these issues when deciding on the appropriate microaerophilic gas delivery method to use for culturing *Campylobacter*.

**Key Words:** *Campylobacter*, microaerophilic, gas delivery

**W105 Aflatoxicosis in Haiti: Detection and detoxification strategies.** M. E. Filbert\* and D. L. Brown, *Cornell University, Ithaca, NY.*

Aflatoxins are carcinogenic, immunosuppressive agents produced by *Aspergillus* mold in crops. Haiti's climate facilitates fungal growth, threatening staple foods such as peanuts and maize, which are highly susceptible to aflatoxin contamination. The danger of aflatoxin poisoning is of concern in Haiti where food is scarce and consumption of the toxin is inevitable. Because of serious health risks, the maximum allowable level of aflatoxin in agricultural commodities (except milk) intended for human consumption is 20 µg aflatoxin/kg product. The overall objective of this research project was to determine the nature and extent of aflatoxicosis in a malnourished population. There are no food safety standards or regulations on any level of food production in Haiti. Our laboratory has not yet found Haitian peanuts from any source that did not exceed allowable aflatoxin limits. Initial peanut samples taken in northern Haiti in 2005 and analyzed using ELISA-aided fluorometry, averaged  $797.5 \pm 218.5$  µg/kg and ranged from 380 to 1567 µg/kg. In 2006, sampled peanuts from a public market found to average  $412.5 \pm 32.1$  µg/kg; peanuts from a farmer's stored supply averaged  $125 \pm 7.1$  µg/kg. A visibly unspoiled sample from a local farm was tested, averaging  $26.8 \pm 7.0$  µg/kg. Another post-sorting sample taken in early 2007 averaged  $0.20 \pm 0.10$  µg/kg, revealing the effectiveness of selection and sorting methods in the elimination of aflatoxin. Peanut butter samples collected in 2009 averaged  $236.4 \pm 196.7$  µg/kg and ranged in levels from 7.3 to 720 µg/kg, validating the need for implementation of sorting methods at every level of food production. An ongoing study using urine will assess aflatoxin catabolism in a malnourished population. Health risks can be reduced by determining contamination levels and understanding aflatoxin metabolism. The results of this study will provide the

information needed to meet the long-term goals of implementing food preparation procedures to remove aflatoxin as a barrier to the nutritional support and recovery of malnourished individuals.

**Key Words:** aflatoxin, peanuts, Haiti

**W106 Conjugated linoleic acid does not modify liver histology and hepatic triglyceride content in young pigs.** I. Fernandez-Figares\*<sup>1</sup>, A. Martin<sup>2</sup>, M. Lachica<sup>1</sup>, R. M. Nieto<sup>1</sup>, and J. F. Aguilera<sup>1</sup>, <sup>1</sup>CSIC, Spanish Research Council, Granada, Spain, <sup>2</sup>Servicio de Anatomia Patologica, HU Virgen de las Nieves, Granada, Spain.

Interest in feeding CLA to pigs has increased in the last decade as a result of its potential to improve growth parameters as well as to reduce body fat (J Anim Sci. 79:1821–8). Interestingly, in mice, adipose tissue reduction was accompanied by liver enlargement and steatosis (Diabetes. 49:1534–42). In pigs, however, there is no evidence of enhanced liver weight after CLA administration (J Anim Sci. 86:102–11). Therefore, the aim of the present work was to evaluate possible adverse effect of feeding CLA supplemented diets to pigs on liver fat content and histology. Twenty gilts (20 kg BW) were individually penned and fed at 95% ad libitum barley-soybean meal based diets (12% CP, 0.81% lysine and 14.8 MJ ME / kg DM) containing 1% CLA (60% CLA isomers, half *cis*-9, *trans*-11 and half *trans*-10, *cis*-12 in FFA form, BASF) or linoleic acid. At 50 kg, pigs were slaughtered and liver samples immediately frozen in liquid nitrogen and preserved at –80°C until analysis. Thawed liver samples were homogenized and TG extracted with 2:1 chloroform/methanol using the Folch method and dissolved in isopropanol. TG content was determined quantifying the glycerol content, using an enzymatic colorimetric assay. For histology, the livers were thawed and fixed in 10% buffered formaline. After routine processing, livers were embedded in paraffin, sectioned at 5 µm and stained with hematoxylin-eosin and reticulin stain. Triglycerides data were analyzed as one-way ANOVA in a completely randomized design with treatment as the fixed effect. Significance was set at  $P < 0.05$  and differences among means were determined using a Tukey's *t*-test. Overall, no differences in triglyceride content was encountered when CLA fed pigs were compared to control pigs (2.1 and 1.6 mg/g, respectively;  $P > 0.10$ ). In the histological study, all livers showed normal histology, without evidence of inflammatory changes, fibrosis or steatosis.

**Key Words:** CLA, pig liver, triglycerides