Project report submitted in Microsoft Word version. Do not provide a pdf version.

Project Leader (PI): Dr. Rama Nannapaneni

Co-PI(s): Chander Sharma and Aaron Kiess

Collaborator(s):

Objective(s):

Objective 1: Determine the cross-resistance of acid-, alkali-, salt-, cold- and oxidative-stress resistant phenotypes of *L. monocytogenes* against commonly used antibiotics.

Objective 2: Determine the cross-resistance of acid-, alkali-, salt-, cold- and oxidative-stress resistant phenotypes of *Salmonella* against commonly used antibiotics.

Milestones for FY 2016-17: Fully met

Cross resistance to antibiotics in highly virulent *Listeria monocytogenes* and *Salmonella* serotypes adapted to sub-lethal acid-, alkali- and oxidative-stress environments.

1. Homologous stress adaptation and antibiotic resistance of *Listeria monocytogenes* following exposure to sublethal chlorine concentrations has been determined.

2. Homologous stress adaptation and antibiotic resistance of *Salmonella* Typhimurium following exposure to sublethal chlorine concentrations has been determined.

Progress Report:

Listeria monocytogenes and *Salmonella* Typhimurium was screened for susceptibility to different antibiotics after exposure to increasing sublethal concentrations of chlorine. The antibiotic resistance patterns of the adapted (rugose and smooth) and control cells were determined for *Salmonella* Typhimurium. The results showed that there was no difference in the antibiotic susceptibility patterns of the adapted smooth cells when compared to the control cells, for any of the antibiotics tested in *Salmonella* Typhimurium. However, the adapted rugose cells showed a slight reduction (≤ 2mm) in susceptibility to streptomycin, nalidixic acid, ciprofloxacin, and ceftriaxone when compared to adapted smooth and control. A change in MIC (antibiotic susceptibility) was observed for the adapted cells (rugose and smooth), which exhibited resistance to amoxicillin. In addition, a slight increase in MIC was observed for adapted rugose cells against all the antibiotics tested except ciprofloxacin compared to adapted smooth and non-adapted control cells.

Accomplishments:

Both *Listeria monocytogenes* and *Salmonella* serotypes demonstrated an acquired tolerance to chlorine with the adapted cells growing in concentrations above the minimum inhibitory concentrations. Chlorine induced a morphological change to the rugose variant in *Salmonella* serotypes. The possibility of cross-adaptation to antibiotics was evaluated after exposure to sublethal chlorine stress in *L.*

monocytogenes and *Salmonella* according to the Clinical & Laboratory Standards Institute (CLSI) guidelines. The average zones of inhibition by disk diffusion assay for chlorine adapted *Listeria monocytogenes* cells was decreased by 0.5 – 2.2 mm compared to control cells against all antibiotics tested. Also, MIC values doubled for four antibiotics tested against chlorine adapted *L. monocytogenes* cells compared to control. Chlorine exposed cells showed reduced susceptibility against some of the antibiotics tested. The findings suggest that continuous exposure to sublethal chlorine concentration could induce changes in homologues and heterologous stress adaptation *L. monocytogenes* and *Salmonella* cells.

Significant Activities that Support Special Target Populations: (100 words or less)

The findings on the morphological changes in *Salmonella s*erotypes after continuous exposure to sublethal concentrations of chlorine have been presented at the Poultry Science Association Meetings in 2016 and at the International Poultry Scientific Forum meetings in 2017. Two journal articles have been submitted based on these findings which will be shared with food industry professionals working with sanitation in the poultry processing plants and in catfish processing plants.

Technology Transfer:

Number of new CRADAs: None

Number of active CRADAs: None

Number of new MTAs (providing only): None

Number of invention disclosures submitted: None

Patent Disclosure. None

US Patent Application, describe the number and title

0 Number of new germplasm releases: N/A

0 Number of new commercial licenses granted: N/A

0 Number of web sites managed: N/A

2 Number of non-peer reviewed presentations and proceedings

0 Number of newspaper articles and other presentations for non-science audiences

0 Number of other technology

International Cooperation / Collaboration:

None

Publications:

Obe T, Nannapaneni R, Sharma C, Kiess A. 2017. Homologous stress adaptation, antibiotic resistance, and biofilm forming ability of *Salmonella enterica* serovar Heidelberg ATCC8326 on different food-contact surfaces following exposure to sublethal chlorine concentrations. Poultry Science (submitted May 29, 2017).

Obe T, Sharma C, Nannapaneni R, Kiess A. 2017. Rugose morphotype of *Salmonella enterica* serovar Typhimurium ATCC14028 exhibits chlorine resistance and strong biofilm forming ability. Food Microbiology (submitted May 29, 2017).

Presentations:

Obe T., R. Nannapaneni and C.S. Sharma. 2016. Development of rugose morphotype of Salmonella Typhimurium following exposure to sub-inhibitory chlorine concentrations that exhibit chlorine resistance and strong biofilm forming ability. 2016 Annual Meeting of Poultry Science Association at New Orleans July 11-14, 2016.

Obe T, Nannapaneni R, Sharma C. 2017. Homologous stress adaptation, antibiotic resistance, and biofilm formation of Salmonella Heidelberg on different food-contact surfaces following exposure to sub-lethal chlorine concentrations. Poster presented at the International Poultry Scientific Forum Georgia World Congress Center, Atlanta, Georgia, January 30-31, 2017

Please attach a photo or figure with a brief explanation to showcase the achievement of your project that will be placed on our food safety research website

Formation of rugose morphotype in *Salmonella* serotypes after continuous exposure to sublethal concentrations of chlorine: (a) Smooth colonies; (b) Rugose colonies