

# DEVELOPMENT OF A MULTI-CLASS CYANOBACTERIA AND ALGAL TOXIN DETECTION METHOD

*Cecilia Silva Sanchez, Nancy Denslow, and Stephen M. Roberts*

Center for Environmental & Human Toxicology, University of Florida, Gainesville, FL, USA.

A harmful algal bloom (HBA) is observed when algal colonies from either fresh or saline waters overgrow, releasing into the waterbodies toxic compounds that can adversely affect humans, fish, shellfish, marine mammals and birds. Among these toxic compounds, Microcystins (MC-x), nodularins (Nod), cylindrospermopsin (Cyl), anatoxin -a (Ana), b-N methylamino-L-alanine (BMAA), okadaic acid (OA), domoic acid (DA), and saxitoxin (Sax) are the main compounds identified from a large group of cyanobacteria and algal toxins. Due to their diverse structural and physiochemical properties, it is necessary to use different laboratory tests in order to identify all of them, making their monitoring very difficult and often cost prohibited. Our goal was to develop a multiclass analytical method for their simultaneous detection and quantitation by LC-MS/MS. A comprehensive panel of 12 microcystins, Ana, BMAA, Cyl, DA, Nod, OA, and Sax standard solutions were directly infused on a 6500 QTRAP to optimize each compound for a multiple reaction monitoring (MRM) experiment. A mixture of all the toxins was prepared, and different concentrations of standards solutions were injected to an Acclaimed Trinity P1 column. A gradient of water and acetonitrile acidified with 0.1% formic acid was used to separate them. So far, 9 out the 12 microcystins were detected with a limit of detection (LOD) ranging from 4 to 30 ppb ( $\mu\text{g/L}$ ), Ana LOD=2.5 ppb ( $\mu\text{g/L}$ ), BMAA LOD=80 ppb ( $\mu\text{g/L}$ ), Cyl LOD=1.9 ppb ( $\mu\text{g/L}$ ), DO LOD=1.5 ppb ( $\mu\text{g/L}$ ), Nod LOD=4.6 ppb ( $\mu\text{g/L}$ ), OA LOD=21 ppb ( $\mu\text{g/L}$ ), and Sax LOD=6.2 ppb ( $\mu\text{g/L}$ ). To our knowledge, this is the first analytical method to detect a multi-class panel of cyanobacteria and algal toxins with good sensitivity.

**PRESENTER BIO:** Dr. Silva-Sanchez is the Scientific Laboratory manager of the Analytical Toxicology Core laboratory (ATCL) at the Center for Environmental and Human Toxicology (CEHT) at the University of Florida. She has over 12 years of experience working in core facilities and specializes in mass spectrometry as an environmental toxicological analytical method.