Developments in Marine and Freshwater Toxins Have Global Impact

Several activities, many of which have a global impact, are developing for the Marine and Freshwater Toxins Community (currently at nearly 200 members) and Task Force, which met during the AOAC Annual Meeting in Philadelphia, Pennsylvania, USA, on September 13, 2009. Attendees represented Canada, Chile, China, Italy, Japan, The Netherlands, Spain, the United Kingdom, and the United States, with diverse backgrounds from academia, the seafood industry, federal governments, the European Union (EU), test kit companies, and instrument manufacturers. Chaired by **Jonathan Deeds** of the U.S. Food and Drug Administration's Center for Food Safety and Applied Nutrition (FDA/CFSAN; College Park, Maryland, USA), the meeting focused on current and potential community/task force activities (update on toxins, validation, workshops), European regulations impacting the community, and much more.

Attendees at a workshop in 2009 examine an enzymatic method. The Marine and Freshwater Toxins Community continues to partner with vendors, stakeholders, the AOAC Pacific Northwest Section, the Washington State Public Health Laboratory, and other communities on training events

Toxins Update

Ostreopsis

Task Force member and Topic Advisor **Aurelia Tubaro,** University of Trieste (Italy), provided an update on toxins produced by *Ostreopsis,* explaining that this species of microalgae has caused toxic aerosols and human illness along the Mediterranean coasts of Italy, France, and Spain. The associated toxins, related in structure to the well-known marine toxin, palytoxin, are now the object of study by many marine toxin researchers including Tubaro. The potential health threat posed by *Ostreopsis* is underlined by the high toxicity of palytoxins, and the severity of the *Ostreopsis* problem became clear following a 2005 incident where more than 200 people in Genoa, Italy, were admitted to emergency rooms and some hospitalized. In the Genoa incident, the presence of *Ostreopsis* ovata in seawater was reported, as well as that of a putative palytoxin and new palytoxin anolog, ovatoxin-A. The Genoa incident and other *Ostreopsis*-associated exposures over the years did not result from ingesting food. Concerning the seafood safety risk however, Tubaro pointed out that there have been recent reports of Mediterranean shellfish being contaminated with palytoxin-like compounds, and that in tropical areas, consumption of other types of seafoods contaminated with palytoxins has resulted in fatalities. There are only limited toxicological and epidemiological data available for these toxins, complicating the protection of human health with regulatory limits. Multiple detection methods, biological and chemical, have been developed for the palytoxins and related compounds, and biosensors are also in development. However, none of these methods have been validated.

Okadaic Acid in U.S. Shellfish

Deeds described last year's *Dinophysis* bloom, which led to contamination of U.S. shellfish by okadaic acid off the coast of Texas in the Gulf of Mexico. Early detection of the *Dinophysis* prebloom, using **Lisa Campbell's** (Texas A&M University) Imaging Flow Cytobot with subsequent shellfish sampling by Texas state officials and toxin confirmation by FDA, allowed for rapid closure of impacted shellfish harvest areas preventing any human illnesses. Okadaic acid was the only diarrhetic shellfish toxin found. The closure event, the first in U.S. history due to confirmed toxins, lasted approximately 1 month. Deeds discussed the nature and extent of the bloom, the levels of okadaic acid found in the shellfish, and detection methodology applied (LC-MS/MS).

European Regulations

Further Reductions in Animal Use

Ana Gago-Martínez (University of Vigo, Spain) outlined important new changes coming in Europe's regulation of lipophilic marine toxins. It

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has been mandated that EU members move away from use of the lipophilic toxins mouse bioassay to chemical methods starting with the validation of an LC-MS/MS method for lipophilic toxins. In addition, the European Community Reference Laboratory for Marine Biotoxins (CRLMB) will play a leading role in the validation efforts and is formulating plans for studies, as well as training sessions and proficiency studies. Information exchange is valuable in these initiatives, which require a high level of international and cooperative efforts, both within and outside of Europe. New Zealand's Cawthron Institute, for example, already has extensive experience monitoring multiple marine toxins by LC-MS/MS, including multiple lipophilic toxins.

These efforts follow some already impressive accomplishments in the United Kingdom, which have reduced animal usage in toxin monitoring. The United Kingdom is applying a refined and automated version of an LC-precolumn oxidation method for the saxitoxins (*Official Method*℠ **2005.06**) to completely replace the paralytic shellfish poisoning (PSP) mouse bioassay (*Official Method*℠ **959.08**) in the monitoring of UK mussels. At the task force meeting and at the seminar session on September 16, 2009, "Marine and Freshwater Toxins: Moving Ahead in the Real World via Validation and Implementation," which he chaired, **Andrew Turner** of Center for Environment, Fisheries & Aquaculture Science (Cefas; Weymouth, UK) described the performance of the modified method and also ongoing (shellfish) matrix extension work. Of particular interest were the improvements in efficiency and throughput afforded by automation of the method using commercially available hardware.

Lorna Murray of Food Standards Authority Scotland described the regulatory aspects of the UK efforts from a federal stakeholder's perspective. In addition to the analytical aspects, sample collection and handling impacts the quality of data collected on the distribution of toxins, as well as other shellfish contaminants.

European Food Safety Authority Opinions

Hans van Egmond (National Institute of Public Health and the Environment; RIVM) discussed several European Food Safety Authority (EFSA) Opinions on marine biotoxins, pub-

China Section Engages Local and International Members and with AOAC Analytical Communities

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method be submitted to the Official Methods Board, and plans are underway for the preparation of a collaborative study involving an international team of laboratories.

China and Pacific Northwest Sections Collaborate on "Marine and Freshwater Toxins: Biosensors and Biochemical Tools" Symposium

Members of the China and Pacific Northwest Sections collaborated on organizing the symposium, "Marine and Freshwater Toxins: Biosensors and Biochemical Tools," which was held on September 16, 2009. Cochaired by Bao; **James Hungerford,** FDA/ORA; and **Hans P. van Egmond,** National Institute of Public Health and the Environment (RIVM; The Netherlands), the symposium examined the needs for biosensors and biochemical tools, strategies for their development, and their performance in the field.

Biosensors and biochemical tools for marine and freshwater toxins have generated extensive interest and remain the subject of much research. These tools could advance seafood safety, and form the basis for primary monitoring methods if they prove rugged enough and compare favorably with existing officially approved and accepted methods.

Talk about AOAC and CNCA

Speakers included **Benjamin Suárez-Isla,** University of Chile; **Stacey Etheridge,** FDA/CFSAN; **Atsushi Yoshino,** Tropical Technology Center Ltd. (Japan), who focused on developing, evaluating, and applying these assays in marine and freshwater toxins analysis.

About 30 scientists participated in the symposium and provided positive feedback.

Conclusion

Through the Annual Meeting and other ongoing activities, the China Section is experiencing a renewed energy. "My goal as president of the Section is for Chinese scientists to become involved with the Section and solve their analytical problems with the help of AOAC," said Bao.

For more information about the China Section, visit the China Section Web site at www.aoacchina.org. For information on the 2010 China Section Meeting, contact Lei Bao at baoleiqd@yahoo.com.cn. ■

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lished in 2008 and 2009, which could have, if codified into EU Directives, a significant impact on stakeholders and others. Among the potential impacts include much more conservative action levels (for example, as much as 11 times lower quarantine levels in the case of the saxitoxins) that would force a major shift in monitoring. Community members agreed that if some of the EFSA risk assessments would lead to lower regulatory limits, this might have serious implications for the shellfish industry, applicability of detection methodology, and increasing resources requirements for those regulatory stakeholders responsible for monitoring.

Validation Activities

In fall 2009, an interlaboratory study of a receptor binding assay for saxitoxins in shellfish was initiated by **Frances van Dolah** of National Oceanic Atmospheric Administration (NOAA). Study materials have been distributed to most laboratories, with a few laboratories delayed due to shipping restrictions.

The Canadian Food Inspection Agency (CFIA), with **Jeffrey van de Riet** as Study Director, is pursuing an *Official Method of Analysis*℠ with a collaborative study of its post-column oxidation (PCOX) HPLC method for saxitoxins. The Interstate Shellfish Sanitation Conference (ISSC), at its 2009 biannual meeting, recently examined and approved the PCOX method at ISSC Level IV. Further, at the AOAC Annual Meeting, several voting members of the Marine and Freshwater Toxins Community recommended pursuit of an *Official Method of Analysis*℠ (collaborative) study.

In addition, Tropical Technology Center Ltd. (TTC; Okinawa, Japan), an AOAC Organizational Affiliate (OA), is pursing *Performance Tested Method*℠ status for a proprietary method, a rapid test for the okadaic acids based on inhibition of recombinant PP2A enzyme. Early supporting data is encouraging. **Atsushi Yoshino** (TTC) presented details of the single-laboratory validation of the rapid test kit at the symposium, "Marine and Freshwater Toxins: Biosensors and Biochemical Tools."

Training and Workshops

In partnership with the AOAC Pacific Northwest Section, the Washington State Public Health Laboratory, and other AOAC analytical communities, the Marine and Freshwater Toxins Community and Task Force in recent years has been providing training, including on newly approved *Official Methods of Analysis*℠, commercially available rapid tests, and instrumentation. A special workshop on seafood contaminants will be held in February 2010 in Seattle, Washington, USA (see sidebar). Training opportunities in other regions of the world are in development.

Honors

Yasumoto to Be Honored in Spain

Takeshi Yasumoto (Okinawa Science and Technology Promotion Center) will be awarded with the distinction of *Doctor Honoris Causa* by the University of Vigo, Spain, on January 28, 2010. The award recognizes his many achievements and leadership in the marine toxins field. In addition, on January 29, 2010, a 1-day seminar will be held entitled "Marine Biotoxins Analysis and Toxicology."

The aim of these events, organized by Gago-Martínez and **José A. Rodríguez Vázquez** (University of Vigo, Spain), is to gain new insights into the field of marine biotoxins research by bringing together world experts on both analysis and toxicology. For more information on registration and accommodations, contact Ana Gago-Martínez at anagago@uvigo.es.

Gago-Martínez Appointed Director of the EC's CRLMB

Gago-Martínez, professor of Analytical Chemistry at the Department of Analytical and Food Science of the Faculty of Chemistry at the University of Vigo, Spain, has been appointed director of the European Community CRLMB. In addition to her many accomplishments in the marine toxins field, Gago-Martínez is a long-time AOAC member and has made significant contributions to the Marine and Freshwater Toxins Task Force. In 2005, she organized the first international Joint Symposium and Task Force Meeting, cochairing with Task Force Chair **James Hungerford** of FDA, which was instrumental in bolstering European participation.

For more information, contact James Hungerford at James_Hungerford@hotmail.com. Not a member of your local Section? Contact Liz Cribbin, program manager, Sections Program, at lcribbin@aoac.org and join now. For more information, visit the AOAC Web site at www.aoac.org and click on "Sections." ■

Rapid Test Laboratory Workshop for Seafood Contaminants

The Marine and Freshwater Toxins Community and the Washington State Public Health Laboratory are holding a Rapid Test Laboratory Workshop for Seafood Contaminants on February 24-26, 2010, in Seattle, Washington, USA. Emphasis will be on detection of histamine, antibiotics, and melamine as well as shellfish toxins.

The workshop, organized with Pacific Fisheries Technologists (PFT) by the AOAC Pacific Northwest Section, the Washington State Public Health Laboratory, and the Marine and Freshwater Toxins Community with participating test kit vendors, will be held at the Washington State Public Health Laboratory following the 61st PFT conference on February 21-24, 2010, at the Edgewater Hotel in Seattle.

Transportation from the hotel to the laboratory, as well as on-site lunches, is included in the $300 course fee ($100 for students). Registration for the laboratory course, which is separate from the PFT conference, will be available (check or money order) at the AOAC Pacific Northwest Section Web site www.aoacpacnw.com and by link from the PFT site at http://pftfish.net/. ■