

# Marine and Freshwater Toxins Community Tackles Emerging Toxins, Validations, and Training

In 2011, for the Marine and Freshwater Toxins Community, two *Official Methods<sup>SM</sup>* were approved for paralytic shellfish toxins (PSTs), the second symposium and joint task force conference was held in Vigo, Spain (see July/August 2011 issue of ILM), presentations and task force meetings were held New Orleans, Louisiana, USA, at the AOAC Annual Meeting, and a task force co-chair was announced (see sidebar). Nature also intervened when the Pacific Northwest region of North America was hit with another type of shellfish poisoning.

## Receptor Binding Assay for PSTs Approved as OMA 2011.27

Following closely on the heels of another recently approved method for PSTs, the "PCOX" LC method OMA 2011.02 (see July/August 2011 issue of ILM), a receptor binding assay (RBA), interlaboratory validated by a National Oceanic Atmospheric Administration (NOAA) team led by **Frances van Dolah**, has now been approved as an *Official Method of Analysis<sup>SM</sup>*. The reviewing committee commented on the high quality of the study, which included comparative data with two other official methods and a variety of shellfish species from locations around the world.

Official methods for

PSTs are critically important because paralytic shellfish poisoning (PSP) can cause respiratory paralysis due to the potent neurotoxicity of the PSTs, or saxitoxins. Fatalities are not uncommon when recreational shellfish gatherers ignore "red tide" closures, or when monitoring programs are not in place. The RBA is the fourth *Official Method<sup>SM</sup>* for marine toxins to be approved (and the third *Official Method<sup>SM</sup>* for detecting PSTs) since the toxins task force was initiated.

Like the LC methods, the RBA is much more sensitive than the widely used mouse bioassay (MBA) and does not suffer from the matrix effects at low PST levels that plague the MBA. High throughput is also easily realized due to the parallel format of multichannel scintillation counters. The RBA is also unique among the newly approved methods in that the response is proportional to the overall toxicity of the sample tested, which means that it can respond to any of the numerous PSTs found in nature but does not require routine calibration with toxins other than the parent compound, saxitoxin. Although radionuclides are used in the method, radiation levels are lower than analogous radioimmunoassays already granted exemptions from special licensing. The RBA is expected to make major improvements in shell-



The waters of the Salish Sea where shellfish were harvested that caused DSP outbreaks in summer 2011. Upper arrow = Canadian shellfish harvest area; lower arrow = U.S. area.

fish safety, particularly in many developing countries where the International Atomic Energy Agency (IAEA) has provided training, instrumentation, and validation funds and NOAA has provided expertise, training courses, and the validation efforts.

## Diarrhetic Shellfish Poisoning Emerges in the Pacific Northwest

In summer 2011, the Pacific Northwest was impacted by diarrhetic shellfish poisoning (DSP), a nonfatal malady characterized by gastrointestinal distress and caused by a

suite of toxins referred to as the okadaic acids, or diarrhetic shellfish toxins. Over 60 illnesses were reported in British Columbia, Canada, and three became ill in Washington state, USA.

Although DSP is a global problem and outbreaks have occurred over the years in Asia, Europe, and South America, last summer's DSP outbreaks were the first documented in the Pacific Northwest and also the first in the United States. Although British Columbia had not previously had DSP outbreaks, this

(Continued on page 16.)

## Marine and Freshwater Toxins Community Tackles Emerging Toxins, Validations, and Training

Continued from page 15.

was the second outbreak in Canada; DSP was reported in Nova Scotia in the early 1990s, and the dinoflagellate *Dinophysis norvegica* was implicated as the toxin-producing organism.

The 2011 illnesses in the Northwest were traced to *Dinophysis acuminata*. The U.S. Food and Drug Administration (FDA) Gulf Coast Seafood Laboratory has assisted Washington State Department of Health by running samples by LC-MS/MS and found dinophysis toxin-1 (DTX-1) and esterified forms of DTX-1. Washington State is now working with NOAA's Northwest Fisheries Science Center and receiving methodology training from FDA to ramp-up DSP-specific

monitoring of shellfish growing areas to prevent future outbreaks of DSP. These efforts include both plankton monitoring and toxin detection. Similarly in Canada, the Canadian Food Inspection Agency (CFIA) Dartmouth Laboratory provides analytical support to British Columbia. Although DSP outbreaks have not occurred previously in the United States, Texas shellfish became contaminated with okadaic acid by *Dinophysis ovum* in early 2008. This resulted in the first shellfish bed closure in the United States due to both elevated concentrations of *Dinophysis* and DSP-contaminated shellfish. In this instance, early detection of the *Dinophysis* prevented human illness.

### Third Joint Symposium and AOAC Task Force Meeting: June 18–22, 2012

The AOAC Marine and Freshwater Toxins Task Force invites interested parties to a symposium, discussion, and laboratory training event on June 18–22, 2012, at the University of Puget Sound (UPS) in Tacoma, Washington, USA. The conference will address new developments, method validation efforts, and method implementation in the analysis of marine and freshwater toxins. The format of the conference includes discussion and roundtable sessions, and also a meeting of the Marine and Freshwater Toxins Task Force.

All seafood and freshwater toxins are of interest. However, since recent progress has been made on these toxins in validations and in test kit development, PSTs, ciguatoxins, and diarrhetic shellfish toxins will be emphasized, with the latter and other lipophilic toxins given a full day of coverage due to emergence of DSP in the United States, specifically the Pacific Northwest region.

Laboratory sessions will be offered on rapid tests for marine and freshwater toxins and histamine in fish. The conference will be held in parallel and at the same venue as the 2012 Pacific Northwest Section annual meeting (see page 10 of this issue), making additional sessions available on topics on other natural and manmade contaminants, and bioanalytical methods and microbiological methods like qPCR.

Reduced registration rates will be available to

AOAC members, students, and/or those attending the Pacific Northwest Section annual meeting. Details will soon be available at [www.aoac.org](http://www.aoac.org) and [www.aoacpacnw.com](http://www.aoacpacnw.com). For more information, contact **James Hungerford**, FDA and co-chair of the task force, at [James\\_Hungerford@hotmail.com](mailto:James_Hungerford@hotmail.com).

### Laboratory Training: LC-MS/MS Determination of Diarrhetic Shellfish Toxins and Other Lipophilic Toxins: June 25–26, 2012

In response to the U.S. outbreak and progress made in validations, the Marine and Freshwater Toxins Task Force will provide training on the LC-MS/MS determination of diarrhetic shellfish toxins and other lipophilic toxins. The training will be conducted at the Washington State Public Health Laboratory near Seattle, Washington, USA, on June 25–26, 2012. The training will follow a 5-day marine toxins conference in Tacoma, Washington, USA.

Several variations on the LC-MS/MS procedure, originally developed in Canada, have appeared in the literature. However a procedure validated in Europe and approved by the European Union will be taught in the course (and is currently under AOAC OMA review). Agilent will assist and provide in-kind sponsorship. Participants will perform extractions and work with the instrumentation. For this reason, space is limited and participants should contact Hungerford at [James\\_Hungerford@hotmail.com](mailto:James_Hungerford@hotmail.com) at the earliest convenience to reserve space. ■

## Gago-Martinez Becomes Task Force Co-Chair

**A**na Gago-Martinez, professor at the University of Vigo, Spain, and director, European Union Reference Laboratory on Marine Biotoxins (EURLMB), joins **James Hungerford**, U.S. Food and Drug Administration, as co-chair of the Marine and Freshwater Toxins Task Force. Gago-Martinez, an analytical chemist by training, is accomplished in phycotoxin research and has participated in AOAC activities for many years, including service as a founding and voting member of the task force, and as a reviewer of proposed *Official Methods*<sup>SM</sup>. She co-organized two international conferences for the task force in 2005 and 2011, and now serves on AOAC's Technical Programming Committee. ■



During the 2011 AOAC Annual Meeting, Russ Flowers (then-president), on behalf of AOAC, presented Ana Gago-Martinez with a Community Volunteer of the Year award for her efforts with the Marine and Freshwater Toxins Community.