Whole Effluent Toxicity Testing Expert Committee Meeting Summary
Forum on Laboratory Accreditation, Milwaukee, WI, January 29, 2019

1. Welcome and Announcements

Michele and Chandra were the only committee members present in the room, but a number of members participated by teleconference. Michele did the presentation and moderated the discussion, with perhaps ten participants in the session. Three PT providers participated, from Sigma, Phenova and ERA.

An outline of the PowerPoint presentation used is included in Attachment 2, below, and the presentation itself will be posted to the conference website later.

2. Discussion During and After the Presentation

The meeting was informal, so that discussion typically occurred as the relevant slides were brought up on the screen.

Proficiency Testing

The first discussion began with slide 11, Purpose of PTs. As Michele reviewed the committee’s efforts to have PTs performed under consistent conditions, so that the results are comparable, one of the PT providers asked how PT data are used. The response was that in addition to reporting to Accreditation Bodies (ABs), the labs can use them for a self-evaluation, and that the data-based LC50 values are more meaningful for that purpose than the hypothetical NOEC values. Since, with the organisms being the detector, there can be no “true” value (unlike with Chemistry PTs), it is important to have the data sets as large as possible so that reliable statistical calculations can be made on the aggregated results, for pass/fail determinations that are meaningful comparisons with other reported data.

Another participant noted that PTs with purchased organisms (as opposed to those grown in-house) may have problems with either actually receiving the organisms or with getting the data about the organisms that are actually required by EPA for their use (e.g., supporting chemistry data for two weeks pre-shipment). This is a particular problem with non-routine (wild caught) organisms such as starfish and sea urchins, but also occurs with the more routine organisms. One commenter noted that it’s the adult organisms that are purchased but when the tests measure effects on gametes of those organisms, pre-catch/pre-shipment data may not be critical. One participant recommended trying to address this issue – the distinction between wild-caught and in-house, routine and non-routine species – in the revised standard.

DOC/IDOC

Michele noted early on in the presentation that the laboratory demonstration of capability (or competence, DOC) is clear and acceptable, but that the committee is struggling with acceptable DOC for individual analysts. The current expectation is that a new analyst must perform five Standard Reference Toxicant (SRT) tests, which can take up to a full year, and depending on the organism, can be costly in terms of purchasing as well as time, because some tests take more than a week to complete. ABs insist on both individual and lab DOCs.

Rami noted that IDOCs are the focus of the revised module, and that the WET committee is examining how various labs accomplish their IDOCs. The current variability across labs is large,
and burdensome to assessors, so that better standardization of IDOCs would benefit all by providing consistent training across the industry.

Much discussion within the committee has focused on dividing tests into discrete stages or tasks, so that an analyst could perform all tasks but at different times, in different tests, rather than performing each task in a single test sequentially, from start to finish. Another approach has been to adapt the concept of “work cell” from the 2003 NELAC Standard. This was dropped completely from the 2009 TNI Standard.

[NOTE: A search of the 2003 NELAC Standard provides:
1 – a definition of Work Cell as “a well-defined group of analysts that together perform the method analysis. The members of the group and their specific functions within the work cell must be fully documented” and
2 -- the following text about work cells in Chapter 6 (Quality Systems) Appendix C, page 5C-1:
PROCEDURE FOR DEMONSTRATION OF CAPABILITY
A demonstration of capability (DOC) must be made prior to using any test method, and at any time there is a change in instrument type, personnel or test method (see 5.5.4.2.2). Note: In laboratories with specialized “work cells” (a well-defined group of analysts that together perform the method analysis), the group as a unit must meet the above criteria and this demonstration must be fully documented.]

Chandra offered to share the FL DEP’s SOP that defines work cell with the committee. One participant recommended that the concept, work cell, might benefit from creation of a guidance document, rather than trying to incorporate a rigid definition into the standard itself.

Questions and Follow-Up Discussion Items

One participant asked if WET labs must follow ISO/IEC 17025, and the answer is yes, as it’s part of the Quality Systems module (V1M2) of the TNI Standard. While the quality control for supporting measurements in WET testing does not (and the committee believes, should not) have to be as stringent as the Chemistry module (V1M4) requires, if the laboratory needs accreditation for chemistry tests from its primary AB for reporting those data (for other purposes than support measurements), then yes, they would have to follow V1M4 anyway for those accredited, reportable data.

Another participant asked whether there is a minimum or “standard” data reporting package or data set, and the answer is no. There is nothing in the WET module (V1M7) about reporting, that is left up to the various state program requirements.

A comment was made with respect to reference toxicants, that if the same chemical and strength were used for all PT tests, the results would be more consistent. This led into an in-depth discussion of the WET committee’s history of efforts to improve the comparability of reported PT data.

A PT provider representative stated that the committee should be able to get PT data on request, by simply asking the PT providers, and others agreed. One participant asked if there was any value in surveying WET labs about what they are doing with PTs, and Rami noted that the original WET white paper (on the WET web page) made recommendations for “standard conditions” for PT samples.
Sharon Mertens, the TNI Board’s Past Chair, is a member of the Environmental Laboratory Advisory Board (ELAB) and offered a summary of the WET PT discussion from the Monday afternoon, January 28, ELAB meeting. Henry Liebovitz, past Chair of ELAB, is suggesting that “occasional” blind quality control samples (QCS) be performed and reported to the Wastewater program office to satisfy its demand for samples run “according to the permit” (the NPDES permit), and to let the PT samples be run with consistent parameters so that the PT results will be comparable.

Rami noted that different permits have different requirements (in each state but sometimes for each permit writer), so that running PTs “according to the permit” as has been EPA’s requirement thus far, means that each result is just that, one individual result. He questioned whether the people requiring this “per permit” approach have any awareness of what they are NOT getting, since there is no way to know whether a one-off result is accurate and reliable or not, because there is nothing to which it can be compared. While there was an extended discussion about this after the only meeting with ELAB, WET and EPA, we truly do not know if the Wastewater program people truly understand what they have, as PTs and DMR-QAs are run, now.

The current ELAB Chair had made a suggestion that the ELAB PT workgroup meet with representatives of the WET committee and the DMR-QA program to discuss this further. WET representatives, of course, stand ready to participate with ELAB’s efforts.

Then, Bob Wyeth rushed into the room to ask about the status of WET’s discussions on revising the Technical Director qualifications. As the now-former Chair of the Consensus Standards Development Executive Committee (CSDEC), he wanted to take WET’s language into the Quality Systems (QS) meeting that was underway concurrently. He received the printed paper copy of the latest draft that Lynn had, and hopefully, the feedback from that QS session will be shared at the next CSDEC meeting.

At this point, the session time was expired. Michele thanked everyone for their enthusiastic participation.

3. **Next Meeting**
The next teleconference meeting will be at **1 pm Eastern on February 20, 2019**. An agenda and documents will be sent before the meeting.
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Attachment 2 – Outline of PowerPoint Slides used in Milwaukee Session

Slide 1
WET Expert Committee

Moderators: Michele Potter & Kari Fleming
- Forum on Environmental Accreditation
- Milwaukee, WI
- January 29, 2019

Slide 2
Whole Effluent Toxicity Expert Committee
- Welcome and Introductions
- Michele Potter, NJ DEP
  Kari Fleming, WI DNR
- Meeting time
  - Third Wednesday of each month
  - 1300 hrs ET
  - ~ 1 hr
  - TNI Members are welcome to participate

Slide 3
Committee Members
- Rami Naddy (Chair; Lab) – TRE Environmental Strategies
- Pete De Lisle (Vice Chair; Lab) – Coastal Bioanalysts Inc.
- Ginger Briggs (Lab) – Bio-Analytical Laboratories
- Steve Rewa (Lab) – Environ. Resources Management
- Chris Burbage (Lab) – HRSD
- Chris Pasch (Other) – Alan Plummer Associates Inc.
- Teresa Norberg-King (Other/Affiliate) – U.S. EPA - Duluth
- Elizabeth West (Accreditation Body, AB) – Louisiana DEQ
- Amy Hackman (AB) – Pennsylvania DEP
- Michele Potter (AB) – New Jersey DEP
- Michael Pfeil (AB) – Texas CEQ
- Kari Fleming (AB) - Wisconsin DNR
- VelRey Lozano (Other) – EPA Region 8
- John Overbey (Lab) – American Interplex
- Beth Thompson (Lab) – Shealy Consulting
- Sarah Hughes (Other) – Shell Health
- Program Administrator: Lynn Bradley

Slide 4
Associate Members
- Tom Widera
- Michael Chanov
- Thekkekalathil Chandrasekhar
- Christina Henderson
- Sylvia Bogdan
- Erin Consuegra
- Debmalya Bhattacharyya
Slide 5
Agenda
☐ Accomplishments
  ➢ Webinar available on TNI website Understanding WET Testing
  ➢ 2018 Activities
☐ Activities Underway
  ➢ Revisions to Module 7
  ➢ 2019 Activities
☐ New Business?

Slide 6
WET 2018 Accomplishments
- Revising the Standard – WET Module V1M7
  - Demonstration of Competency requirements for analysts and lab
  - Clarify QC requirements for WET chemistry tests
  - Clarifying Technical Director requirements for WET labs (part of QS module, V1M2)
- WET Proficiency Testing
  - Resolved analyte code usage for WET FoPT tables
  - Requested PTPEC assistance to improve utility of WET PTs
  - Continued conversations with ELAB about improving utility of WET PTs
- Identified person to work with FAC for FSMO Standard revision
- Provided WET Methods for TNI compendium
- SETAC meeting in Sacramento, CA (Nov. 2018) – WETT Session
  - Several committee members present (4 individual talks were presented)

Slide 7
WET 2019 Plans
☐ Revising the Standard Module V1M7
  ➢ DOC for Analyst (some challenges to address)
  ➢ Publish Outline, Receive and Address Comments
  ➢ Possibly Publish Voting Draft
☐ Continue Efforts to Improve Utility of PT Results
  ➢ Work with PTPEC and ELAB/EPA
☐ Continue Interaction with Field Activities Committee to Ensure that WET Testing is Appropriately Addressed in Revised FSMO Standard
Slide 8
Challenges with WET
- Organism is the ‘detector’
- Several different WET / toxicity tests
  - In some instances only difference is the test organism
- Test duration is longer
  - Typically 48-h to 7-d
  - Multiple analysts usually work on same test
- Test specifics typically in NPDES but can also vary based on State and EPA region

Slide 9
The WET methods listed below are codified at 40 CFR 136.3, Table IA

**Acute Toxicity, Freshwater Organisms**
- 2000.0 Fathead Minnow, *Pimephales promelas*, and Bannerfin shiner, *Cyprinella leedsi*
- 2002.0 Daphnia, *Ceriodaphnia dubia*
- 2019.0 Rainbow trout, *Oncorhynchus mykiss*, and Brook trout, *Salvelinus fontinalis*
- 2021.0 *Daphnia pulex* and *Daphnia magna*

**Acute Toxicity, Estuarine/Marine Organisms of the Atlantic Ocean and Gulf of Mexico**
- 2004.0 Sheepshead minnow, *Cyprinodon variegatus*
- 2006.0 Silverside, *Menidia beryllina*, *Menidia menidia*, and *Menidia peninsulae*
- 2007.0 Mysid, *Americamysis bahia*

**Chronic Toxicity, Freshwater Organisms**
- 1000.0 Fathead minnow, *Pimephales promelas*, larval survival and growth
- 1001.0 Fathead minnow, *Pimephales promelas*, larval survival and teratogenicity
- 1002.0 Daphnia, *Ceriodaphnia dubia*, survival and reproduction
- 1003.0 Green alga, *Selenastrum capricornutum*, growth

**Chronic Toxicity, Estuarine/Marine Organisms of the Atlantic Ocean and Gulf of Mexico**
- 1004.0 Sheepshead minnow, *Cyprinodon variegatus*, larval survival and growth
- 1005.0 Sheepshead minnow, *Cyprinodon variegatus*, embryo-larval survival and teratogenicity
- 1006.0 Inland silverside, *Menidia beryllina*, larval survival and growth
- 1007.0 Mysid, *Americamysis bahia*, survival, growth and fecundity
- 1008.0 Sea urchin, *Arbacia punctulata*, fertilization

Slide 10
Other Non-WET Toxicity Tests
Short-term and chronic sediment toxicity tests with invertebrates:
➢ Midge, *Chironomus dilutus*.
  + Survival and growth (10 days).
  + Survival, growth, reproduction, hatchability (20-56 days).
➢ Amphipod, *Hyalella azteca*
  + Survival and growth (10 days).
  + Survival, growth, reproduction (28-42 days).
➢ Amphipod, *Leptocheirus plumulosus*
  + Survival and growth (10 days).
  + Survival, growth and reproduction (28 days).

☐ Others (e.g., plants, earthworm)

Slide 11
DMR-QA for Proficiency Testing

☐ What is the purpose?
  ➢ run it as the NPDES permit (i.e., permit compliance) OR
  ➢ run PTs for data comparability (i.e., laboratory evaluation)

Slide 12
Rationale for PT / DMR-QA Recommendation

☐ The flexibility allowed in 40 CFR 136 or WET Test Manuals (EPA 2002) is not specific enough for proficiency testing
☐ All labs should perform tests using same method, replicates, water type, temperature, renewals, etc.
  ➢ Reduces variability
  ➢ Data more useful & comparable ("apples to apples")
  ➢ Ability to identify labs with deficient techniques
☐ Endpoint standardization – require one reporting value for both acute and chronic
  ➢ LC50 using survival for acute tests
  ➢ IC25 using sublethal endpoints for short-term chronic
  ➢ No negative impact on the PT study power, but not linked to permits
☐ Test parameter summary should be provided with result of Proficiency Testing

Slide 13
WETT CHEMISTRY

☐ What QC procedures should be required of chemistry performed in support of WETT analyses?

Slide 14
WETT Chemistry:

☐ Analytical procedures are required as supporting chemistry for WETT.
☐ These procedures include pH, D.O., temperature, alkalinity, hardness, specific conductance or salinity, TRC, and sometimes ammonia.

Slide 15
Why Revise this Standard:

☐ The Committee agrees that QC is necessary for these supporting procedures; however, not at the level required in Module 4 of the Standard as they are support measures only.
☐ The Committee agrees that some QC guidance is needed to assist auditors in assessing a laboratory’s ability to conduct the supporting chemistry.

Slide 16
**Proposed language**  
The TNI WETT Expert Committee has reached consensus on the following proposed standard language.

**Slide 17**  
**Proposed Standard Language**
- Instruments used for routine measurements of chemical and physical parameters such as pH, DO, temperature, conductivity, salinity, alkalinity and hardness must be calibrated and verified according to the instrument manufacturer's procedures and/or as indicated in the general section on quality assurance of each referenced test method.

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**Proposed Language, cont’d**
- Unless otherwise noted by a mandated method or by regulation, chemical, and physical tests, in toxicity testing are supporting parameters to help aid in the interpretation of toxicity results. As these are support measurements, only the calibration requirements specified in the applicable reference methods apply. Performing matrix spiking, duplicate analysis, and quality control charting of such results is not required during the performance of these tests unless more stringent standards are mandated by a separate State or Federal program.

**Slide 19**  
**Proposed Language, cont’d**
- Documentation of the calibration is required for all support measurements. The preparation of calibration solutions and the identity of the solutions utilized shall also be recorded. The details of initial instrument calibration procedures shall be included the quality system documentation.

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**Proposed Language, cont’d**
- Sufficient raw data records shall be retained to permit reconstruction of the initial instrument calibration (e.g., calibration date, method, instrument, analysis date, analyte name, analysts initial or signature, concentration and response, calibration curve or response factor, or unique equation or coefficient used to reduce instrument responses to concentration).

**Slide 21**  
**Proposed Language, cont’d**
- Sample results shall be quantitated from the initial instrument calibration and may not be quantitated from any continuing instrument calibration verification unless otherwise required by regulation, method, or program. All initial instrument calibrations shall be verified with a standard obtained from a second manufacturer or from a different lot.

**Slide 22**  
**Proposed Language, cont’d**
- Commercially prepared standards shall be traceable to a national standard when commercially available. Criteria for the acceptance of an initial instrument calibration shall be established (e.g. correlation coefficient or relative percent difference). The criteria used shall be appropriate to the calibration technique employed.

**Slide 23**  
**IDOC – CDOC**
- Initial Demonstration of Capability/Competency
- Continuing Demonstration of Capability/Competency
- DOCs / IDOCs well defined for Lab
DOCs / IDOCs for analysts less well defined

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DOC Language in 2009 TNI
- Initial Demonstration of Capability (IDOC).
- Each analyst shall meet the quality control requirements as specified in Section 1.7.1.2.
  - NELAC 2003 Appendix D2 or TNI 2009 V1M7 §1.6 (EL-V1M7-2009).
- Positive and Negative Controls.
  - SRTs and control organism performance.
- Continuing DOC (CDOC).
- Documented procedure describing ongoing DOC.
- Analysts must meet QC requirements of the method, Lab SOP, client specifications, and the standard.
- QC sample data must be reviewed to identify patterns for individuals or groups and make correct actions.

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Proposed Changes to V1M7
- IDOC/DOC:
  + Flexibility in the use of various tools to demonstrate capability (SRT, QC Controls, PTs)
    - Concern that flexibility puts too much responsibility on auditor – how to address?
  + Tests performed as work cells/teams;
    - Less frequently as individual
  + Differentiate between laboratory vs analyst IDOC/DOC.
  + Many phases (e.g., sample prep, water quality measurements, solution renewal, etc.) common to different toxicity tests.
    - Analyst demonstrates competency in test phases, i.e., “demonstration of same technology”

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Module 7
Quality Systems for Toxicity Testing
- Scope of Module 7
  - Not only aquatic toxicity (WET)
  - Sediment (burrowing organisms) and benthic region
  - Drilling fluids and other potentially toxic materials.
  - Soil toxicity
- Revisions to Module 7
  - Demonstration of Competency concepts
  - Reasonable QC for chemistry support measurements

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Questions?
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