

**Whole Effluent Toxicity Testing Expert Committee Meeting Summary
December 16, 2020 1:00 pm Eastern**

1. Welcome and Announcements

Rami welcomed everyone to the meeting. Attendance is recorded in Attachment 1, below. The newest associate member, Ila Meyer-Fritzsche, was invited to introduce herself. There were no changes to the agenda, which is in Attachment 2, below. Sarah moved and Pete seconded that the minutes of November 18 be approved, and the vote was unanimously in favor.

2. Membership Election

With a majority of the original committee members completing their second 3-year term in January, applications for both AB and lab stakeholders have been actively solicited over the past several months from associate members of the committee. All applications for membership were distributed to all committee members for consideration.

There are no current member AB stakeholders eligible for another term, but one “other” stakeholder is still in the first term. The affiliate member requires re-election annually. For lab stakeholders, three members will carry over – Rami with an approved third term to continue as Chair while the standard is being revised plus both John and Mark requesting to continue with a second 3-year term.

With three continuing lab stakeholders, only four of the six applicants could be brought in as voting members, so that a run-off election was deemed necessary. That run-off election was held by email during the week prior to the committee meeting, among T.M. Chandrasekhar (7), Stephen Clark (11), Natalie Love (9), Katie Payne (12), Bruce Weckworth (4) and Tom Widera (7); the number of votes for each is indicated in parenthesis. This led to a tie for the fourth person, between Tom and Chandra, so one of them was requested to withdraw and Tom did so by graciously deferring to Chandra’s longer and more active participation.

With the lab candidates established, all five of the AB candidates could be approved as could the affiliate member. It seemed advisable to affirm Rami’s continuation as Chair since there could be ambiguity about whether his third term election request and approval included that role. Thus, only Sarah continues membership without being on the following slate that was presented for the election.

| WET Expert Committee (15 member limit) | Lab Stakeholders | AB Stakeholders | “Other” Stakeholders (Sarah Hughes continuing first term) |
|--|--|--|--|
| New Applicants with application number | Stephen Clark Natalie Love Katie Payne T.M. Chandrasekhar | Dwayne Burkholder David Caldwell Ila Meyer-Fritzsche Rosana McConkey Caitie Van Sciver | Teresa Norberg-King, continuing Affiliate member |
| Members renewing for a second 3-year term | Mark O’Neil John Overbey | | |
| Chair of Committee | Rami Naddy | | |

This slate of members was offered to the full committee for approval. Sarah moved that it be approved and Kari seconded the motion. All present voted in favor of the motion, with Rami abstaining (9 votes in favor of the slate as presented).

Congratulations to the new members! Your terms take effect following the conference in January.

And many thanks to the eleven members who have completed the maximum number of years allowed on a committee! You have all worked hard to bring the WET Committee into one of the largest and most enthusiastic committees in all of TNI!

3. Updates

PT Instructions for PT Providers – Rami met with PTPEC and presented the committee's recommendations for improvements in the WET PT process. There will be a joint meeting (PTP Executive Committee, PT Expert Committee and WET Expert Committee) during the Winter Conference to discuss WET PT concerns [Monday afternoon, January 25, 2 pm Eastern] to discuss whether and how improvements can be made. For standardizing test parameters, there is no mechanism by which PT Providers (PTPs) can require this, and the options appear to be either writing it into V1M1 (the PT module) or V1M7 (WET module); the DMR-QA letter now contains much of the information about standardization but requirements in the standard would not be required of non-accredited labs. The original recommendations along with PTPEC's response are included in Attachment 3, below.

Assessor Training Errata Sheet – Marlene is supposed to be reviewing the errata sheet and providing edits by mid-January. One stumbling block is how to ensure that non-accredited measurements for water quality are properly identified as being "not accredited" in reports. Participants seemed to agree that an additional clause will be needed to the chemistry support measurements language requiring a disclaimer to this effect.

Draft Outline for Data Interpretation Training – Katie, Rami, Stephen and John volunteered to work with Natalie and Teresa to develop this webinar. Target date is in late summer.

Method Codes for WET Analyses (Michele, Ginger and maybe Teresa) – re-start this review in 2021

LAMS Clean-up for WET Methods (Rami, Michele and Elizabeth) – still on hold

4. Continued Discussion of Analyst DOC Write-up

Rami circulated a final draft of this concept paper describing individual analyst initial and ongoing Demonstrations of Competency along with two attachments intended to define more details of the minimum requirements that will be incorporated into the WET module of the TNI Standard. The drafts are included as Attachments 4, 5 and 6 below.

Discussion points were that the analyst ODOC would be adequately addressed with one SRT from each of the seven listed in Attachment 1 and that the lab DOCs are not addressed in this document, as they will be in a separate section of the standard. Also, the lab DOC requirements are well covered in the method manuals. A few minor edits may still be made but the concept itself was agreeable.

Mark moved and Pete seconded that the concept paper be approved; the vote was unanimously in favor of approval. Rami will present this to the NELAP AC at its January 4 meeting.

5. Method Validation

Time was expired. This topic will definitely be a priority for the January 20 WET meeting

6. New Business

There was no new business.

Pete moved and Mark seconded that the meeting be adjourned.

7. Next Meeting

The next teleconference meeting will be on Wednesday, January 20, 2020, at 1 pm Eastern. An agenda and any needed documents will be sent in advance.

For individuals registered for the winter conference, the joint committee meeting will be on Monday afternoon, January 25, at 2 pm Eastern.

Attachment 1

WET Expert Committee Membership

| Member | Affiliation | Email | Category | Term Expiration | Present |
|----------------------------|-------------------------------------|--|-------------------|------------------------|----------------|
| Ginger Briggs | Bio-Analytical Laboratories | bal@bioanalyticallabs.com | Lab | Jan. 2021 (2) | No |
| Chris Burbage | Hampton Roads Sanitation District | cburbage@hrsd.com | Lab | Jan. 2021 (2) | Yes |
| Kari Fleming | WI DNR | kari.fleming@wisconsin.gov | AB | Jan. 2021 (2) | Yes |
| Amy Hackman | PA Dept. Environ. Prot. | ahackman@pa.gov | AB | Jan. 2021 (2) | Yes |
| Sarah Hughes | Shell Oil Co. | s.hughes@shell.com | Other | Jan. 2022 (1) | Yes |
| Pete De Lisle (Vice Chair) | Coastal Bioanalysts Inc. | pfd@coastalbio.com | Lab | Jan. 2021 (2) | Yes |
| Rami Naddy (Chair) | TRE Env. Strat. LLC | naddyrb.tre@gmail.com | Lab | Jan. 2021 (2) | Yes |
| Teresa Norberg-King | USEPA | norberg-king.teresa@epa.gov | Other (Affiliate) | Jan. 2021 (2) | Yes |
| Mark O'Neil | Environmental Enterprises USA, Inc. | moneil@eeusa.com | Lab | Jan. 2021 (1) | Yes |
| John Overbey | American Interplex Corp. | joverbey@americaninterplex.com | Lab | Jan. 2021 (1) | Yes |
| Chris Pasch | Alan Plummer Associates, Inc. | cpasch@apaienv.com | Other | Jan. 2021 (2) | No |
| Michael Pfeil | Texas Comm. Environ. Quality | Michael.pfeil@tceq.texas.gov | AB | Jan. 2021 (2) | Yes |
| Michele Potter | NJ Dept. of Environ Protect. | Michele.Potter@dep.nj.gov | AB | Jan. 2021 (2) | Yes |
| Steven Rewa | Env. Resources Management | steven.rewa@erm.com | Lab | Jan. 2021 (2) | No |
| Elizabeth West | LA DEQ LELAP | elizabeth.west@la.gov | AB | Jan. 2021 (2) | No |
| Associate Members | | | | | |
| Sylvia Bogdan | EPA R6 | Bogdan.sylvia@epa.gov | Other (Assoc.) | | No |
| Steve Boggs | CA ELAP | steve.boggs@waterboards.ca.gov | Other (Assoc.) | | No |
| Dwayne Burkholder | PA DEP | dburkholde@pa.gov | AB (assoc.) | | No |
| David Caldwell | OK DEQ | David.caldwell@deq.ok.gov | AB (assoc.) | | Yes |

| | | | | | |
|--|--------------------------------------|--|-----------------------|--|-----|
| Antoine Chamsi | East Bay Municipal Utility Dist. | antoine.chamsi@ebmud.com | Lab (Assoc.) | | Yes |
| Thekkekalathil "Chandra" Chandrasekhar | FL DEP | Thekkekalathil.Chandrasekhar@dep.state.fl.us | Lab (Assoc.) | | No |
| Michael Chanov | EA Eng., Sci. &Tech. | mchanov@eaest.com | Lab (Assoc.) | | Yes |
| Stephen Clark | Pacific EcoRisk | slclark@pacificecorisk.com | Lab (Assoc.) | | Yes |
| Erin Consuegra | ERA LAB | econsuegra@eralab.com | Lab (Assoc.) | | No |
| Kevin Dischler | Element Materials Technology | Kevin.dischler@element.com | Lab (Assoc.) | | No |
| Monica Eues | CK Associates | Monica.eues@c-ka.com | Lab (Assoc.) | | No |
| Nicole Fortin | Honolulu City Lab | nfortin@honolulu.gov | Lab (Assoc.) | | No |
| Christina Henderson | Bio-Aquatic Testing, Inc. | chenderson@bio-aquatic.com | Lab (Assoc.) | | No |
| David Johnston | Valero Refining Co - Benecia | david.johnston@valero.com | Lab (Assoc.) | | No |
| Paul Junio | Northern Lake Service, Inc. | paulj@nslslab.com | Lab (Assoc.) | | Yes |
| Natalie Love | GEI Consultants | nlove@geiconsultants.com | Lab (Assoc.) | | Yes |
| VelRey Lozano | USEPA Reg. 8 | Lozano.VelRey@epa.gov | Other (Assoc.) | | Yes |
| Rosana McConkey | WA Dept of Ecology | rosa461@ECY.WA.GOV | Non-NELAP AB (Assoc.) | | Yes |
| Marlene Moore | Advanced Systems | mmoore@advancedsys.com | Other (assoc.) | | No |
| Ila Meyer-Fritzsche | VA DCLS | ila.meyer-fritzsche@dgs.virginia.gov | AB (assoc.) | | Yes |
| Linda Nemeth | | lkn1304@gmail.com | Other (assoc.) | | No |
| Katie Payne | Enthalpy Analytical | katie.payne@enthalpy.com | Lab (Assoc.) | | Yes |
| Christina Pottios | Los Angeles Cty Sanitation Districts | cpottios@lacsdsd.org | Lab (Assoc.) | | No |
| Greg Savitske | US EPA OECA | Savitske.gregory@epa.gov | Other (Assoc.) | | No |
| Justin Scott | Cove Sciences | justin@covesciences.com | Lab (Assoc.) | | Yes |
| Caitie Van Sciver | NJ DEP | Caitie.VanSciver@dep.nj.gov | AB (assoc.) | | No |

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|--|-------------------------|--|-------------------|--|-----|
| Lem Walker | USEPA OW/OST | Walker.lemuel@epa.gov | Other (Assoc.) | | No |
| Craig Watts | Hydrosphere Research | cwatts@hydrosphere.net | Lab (Assoc.) | | Yes |
| Bruce Weckworth | HRSD | Bruce.weckworth@hrsd.com | Lab (Assoc.) | | No |
| Tom Widera | Pace Labs | Thomas.Widera@pacelabs.com | Lab (Assoc.) | | yes |
| Program Administrator: Lynn Bradley, lynn.bradley@nelac-institute.org | | | | | |

Attachment 2

WET Expert Committee Meeting Agenda, December 16, 2020

- Welcome and Roll Call
- Approval of Agenda
- Approval of Minutes (November minutes attached)
- Election -- Slate of New Members, Vote expected (slate will be sent by Tuesday, December 15)
- Brief Updates
 - PT Instructions for PT Providers – (from Board agenda: “The Committee met with Rami from the WET Expert Committee to understand their PT concerns. There will be a joint meeting (PTPEC, PT Expert and WET Expert) during the Winter Conference to discuss WET PT concerns [Monday afternoon, January 25, 2 pm Eastern]. The discussion this month centered around whether language in the Standard could change, would additions to FoPT tables be appropriate, is there a way to request PT Providers to include information in their instructions, are changes needed to the WET Standard, etc.”)
 - Assessor Training Errata Sheet – Marlene and Ilona meeting December 14 to discuss
 - Draft Outline for Data Interpretation Training (Teresa and Natalie) – ready to proceed, need volunteers to work with Natalie and Teresa
 - Method Codes for WET Analyses (Michele, Ginger and maybe Teresa) – re-start this review in 2021
 - LAMS Clean-up for WET Methods (Rami, Michele and Elizabeth) – still on hold
- Analyst DOC Paper and Attachments – possible vote to approve for distribution to NELAP AC (most recent versions attached)
- V1M7 Method Validation, time permitting (draft V1M7 attached)
- Affirm January 20 Meeting Date
- New Business, if any
- Adjourn

Attachment 3

DRAFT: Suggested Proficiency Testing (PT) Instructions for PT Providers

These are suggested steps to standardize PT instructions for Whole Effluent Toxicity DMR-QA/PT testing to assure and increase the comparability and usefulness of the data generated the studies.

Suggested steps include: These can be included in the FoPT table for requirements.

1. Standardize the required number of replicates per test.
2. Standardize the required number of organisms per replicate.
3. Standardize and reduce the age range of test organisms used in the following tests:
 - a. DMR-QA Test code 13 and 14 (EPA Method 2000): Pimephales acute tests reduce age range from 1 – 14 days down to 1 – 5 days with a 24 hr range in age.
 - b. DMR-QA Test code 46 (EPA Method 2004): Cyprinodon acute test reduce age range from 1 – 14 days down to 1 – 5 (or other such consensus range) days with a 24 hr range in age.

The following additional suggested steps may be best placed into the TNI standard as requirements for the labs to implement.

1. Require labs to affirm that DMR-QA/PT tests were conducted according to the specified test conditions listed in the PT instructions.
2. Require labs to document if any deviations from required test conditions occurred and whether a deviation invalidated the test or not. Some deviations from test conditions would invalidate a test such as incorrect number of replicates used, incorrect number of test organisms per replicate, incorrect test organism age, etc. would not.
3. Require labs to document each test's test acceptability criteria data, for example:
 - a. For the negative laboratory performance control in acute tests, document the % survival.
 - b. For the negative laboratory performance control in chronic tests, document the % survival and the mean weight per surviving test organism or the mean 3rd-brood reproduction per surviving *C. dubia*.
4. Require labs to document the sublethal PMSD evaluation for tests where PMSD bounds are established in the EPA test method and when a chronic NOEC test endpoint was reported.
 - a. If a test's PMSD is less than or equal to the lower PMSD bound for the test method reported, then the lab must document that the relative % difference from the control of each test concentration tested and that the % relative difference reported for the NOEC is greater than the lower PMSD bound.
 - b. If a test's PMSD is above the maximum PMSD bound for the test method then the NOEC shall not be reported.
5. Require labs to document the evaluation of interrupted dose-response curves for tests where an interrupted dose-response occurs and an NOEC test endpoint is reported. The lab shall document the statistical significance or non-significance of every test concentration subsequently to the PMSD evaluation in #4 above
 - a. Lab shall evaluation dose-response curves per EPA 821-B-00-004 Method Guidance and Recommendations for Whole Effluent (WET) Testing (40 CFR Part 136).
6. Require labs to document the source of test organisms used in a DMR-QA/PT test.

SMK Response (NOTE, this is Shawn Kassner, Chair of PTPEC)

I have read 2016 TNI Vol 1 Mod, Vol 1 Mod 7 and the proposed changes to the standard and the FoPT tables. A couple of things to note, the FoPT tables are not just for PTPs anymore. The 2016 Vol 1 Mod 1

references that laboratories shall use the tables for the purposes of reporting data multiple times. So, the 2016 TNI standard directs laboratories to the FoPT tables currently.

Historically, the ABs and the PTPAs (A2LA and ANAB) have frowned upon PTPs providing laboratories more guidance than they thought went beyond instructions; such as helpful hints etc. The items 1 – 3 as listed below are not helpful hints to perform the method, they are an attempt to standardize the test conditions for the sake of statistical evaluation. The WETT expert committee can work with the PTPEC to evaluate whether these should be added to the table. We will then seek the input from the AC, the understanding must be that these criteria are needed to develop study-based statistics that allow for comparability and appropriate evaluations for the WETT labs. I have specific questions surrounding these for statistical impact but will save those for the WETT committee. If these are added to the FOPT table TNI and the PTPs will need to direct people to the table for their review. The Vol 3 specifically allows for the table to supersede it for acceptance criteria determination in section 5.9.2.2 “Analyte- or study-specific evaluation criteria defined in the TNI FoPT Tables shall supersede the criteria in this Section.” This was done purposely to allow for more rapid changes to the acceptance criteria.

1. Standardize the required number of replicates per test.
2. Standardize the required number of organisms per replicate.
3. Standardize and reduce the age range of test organisms used in the following tests:
 - a. DMR-QA Test code 13 and 14 (EPA Method 2000): Pimephales acute tests reduce age range from 1 – 14 days down to 1 – 5 days with a 24 hr range in age.
 - b. DMR-QA Test code 46 (EPA Method 2004): Cyprinodon acute test reduce age range from 1 – 14 days down to 1 – 5 (or other such consensus range) days with a 24 hr range in age.

The remaining standard changes are covered in Vol 1 Mod 1 and Mod 7 to some extent, but let's review. My initial comment is that if the labs are required to document this information for PTs, who is going to evaluate it, deem it acceptable or not, and to what criteria? So there are a few questions surrounding these additions. My presumption is that the ABs would review these as part of their normal assessment.

Item# 1 is in 2016 TNI Vol 1 Mod 1 section 4.2.1 and can be removed as redundant.

Item #2 The requirement to document deviations from a method is required in TNI 2016 Vol 1 Mod 2 section 5.4.1. “Deviation from test and calibration methods shall occur only if the deviation has been documented, technically justified, authorized, and accepted by the customer. “So this is also a redundant clause to the current standard. The other issue I have with this item is who is going to be the arbiter of what method deviations are technically acceptable or not to the test results for the PTs. This must fall to the ABs as the PTPs do not have the technical expertise to evaluate these deviations nor is this their role.

Items# 3 – 6 are somewhat addressed in Mod 7 but nowhere near as much as here. I do believe that these are great QC tacking and practices that labs should be performing all the time. And perhaps these should be amended into Mod 7!? Again, I am going to ask what is the purpose, evaluation criteria, and who is reviewing the data? It also appears to be a great place to start a corrective action for a PT failure. These in general are potentially great improvements to the WETT program and standard, I am not sure that Vol 1 Mod 1 is the place for these and just be used for PT.

The next steps should be for Kirstin and I to validate what our committees think if these changes and return that information to this group. Everyone took the time to ask good questions, provide good answers and it is important for Kirstin and I to review these between us and then with our individual committees.

Regardless, I would also like Rami and the WET Expert committee to evaluate whether the items 3 – 6 should be adopted as the normal QC practices for Mod 7 or for corrective action investigation for failed PTs.

Attachment 4

Analyst Demonstration of Capability (DOC) WETT

V1M7 1.6.2.2

Demonstration of capabilities for analysts (both initial and continuous) is a challenging topic for toxicity tests (including WETT) because of some of the inherent aspects of toxicity tests and how they are performed. Some of these are listed below.

- Test durations for WET / toxicity methods are typically anywhere from 24 hours to 7 days (for typical WET studies; other toxicity tests can go even longer).
- Laboratory staff work as a team when performing tests, and therefore multiple analysts may work on a single test (i.e., one analyst does not typically conduct an entire test from start to finish but may work on it a few times while it is up).
- Many of the steps in toxicity tests are very similar / are almost exactly the same from test to test / day to day, with the main difference being the use of a different organism (i.e., detector) which can allow for cross-training as provided in Attachment 1.
- Some methods (e.g., sediment tests) allow for using a shorter standard reference toxicity (SRT) test (e.g., 96-hours) using water only exposures compared to the test method itself (e.g., 10-day sediment toxicity tests).
- Auditors typically have expertise in chemical but less so in biological test methods
- Differences in requirements for WET methods across the country by different accrediting/regulatory bodies.

Because of these challenges that are inherent to toxicity testing, the WET Expert Committee is providing what we view as minimum requirements for the analyst DOCs from individual WET / toxicity testing laboratories.

There are certain general concepts that will apply to any WET / toxicity testing laboratory training approach for analyst training:

- The laboratory must have a detailed written approach for analyst training including initial and continuing DOCs. This approach must be well documented and must make it understandable for anyone that has to document the analyst training. The laboratory can include how it handles cross-training between methods of similar technology.
- An individual who performs any activity involved with preparation and/or analysis of samples must have constant, close supervision as defined in the laboratory's training procedure until a satisfactory initial analyst DOC is completed.
- Where the analyst performs the toxicity test from start to finish, that analyst must perform and document all major tasks of the test method they perform
 - The WET Expert Committee has provided a list of tasks that may be included for analyst training (Attachment #2)
 - The WET Expert Committee has provided a table illustrating a list of tests that can be used in substitution of other tests (Attachment #1). For instance, training on a fathead minnow chronic WET test should cover training on a fathead minnow acute WET test (a mysid chronic for a mysid acute, etc.) because of the similar / same tasks conducted in each (flow-through test or other specialty tests would have to be documented separately).
- Where the analyst does not perform the entire toxicity test, task-based performance must be demonstrated and documented for each step in the test they perform
- See cross-training discussion for secondary tests.
- Training on acute tests cannot substitute for training on chronic tests

- although similar steps / tasks within each test may be used for dual task-based (cross) training purposes e.g., prep of test solutions, sample renewal, etc., i.e., similar technology items.
- Cross-training of methods are allowable as long as the secondary method has the same tasks as the primary method (e.g., fathead chronic test training covering training for acute fathead tests with similar technology (static or static-renewal)).
- While work on SRTs is the preferred approach when documenting analyst's DOCs, some analyst DOCs can be documented prior to working on SRTs.
 - Most analyst training consists of training on cultures, demo tests, job shadowing, tests with constant supervision, etc. in which some aspects of tests may be documented quantitatively (e.g., counting of the number of *C. dubia* neonates in a given container). Therefore, laboratories may use these non-SRT situations to serve and document analyst training, especially for analyst iDOCs.
 - NOTE: This highlighted section is not written in regulatory / enforceable language b/c it's intended to be a note in V1M7.
- Each analyst must be involved in the performance of at least 1 acceptable SRT for each primary test method they have competency or for the specific tasks.
 - SRTs are likely to be performed as a team unless the analyst performs the entire test
- iDOCs for sediment toxicity tests (or similar tests where the SRT does not have a similar test duration) the analyst training program must include acceptable performance on one SRT and assessment of laboratory controls, or simulated controls, as appropriate (e.g., $\geq 90\%$ recovery of organisms after at least 1-h in sediment tests, measurement of weights or lengths, etc., that produce acceptable control performance criteria).

Attachment 5

Attachment #1: Demonstration of Capability – Toxicity Testing Substitution List of Common WET tests

| Primary Methods listed below (more common methods) can substitute for secondary methods to the right because they include the same analyst skillset / similar technology, i.e., satisfies DOC for corresponding methods | 1000.0 Chronic Fathead | 1002.0 Chronic <i>Ceriodaphnia</i> | 1003.0 Chronic Algae | 1004.0 Chronic Sheepshead | 1007.0 Chronic Mysid | 2000.0 Acute Fathead | 2002.0 Acute <i>Ceriodaphnia</i> | 2004.0 Acute Sheepshead | 2007.0 Acute Mysid | 2019.0 Acute Trout | 2021.0 Acute <i>D. pulex / magna</i> |
|---|------------------------|------------------------------------|----------------------|---------------------------|----------------------|----------------------|----------------------------------|-------------------------|--------------------|--------------------|--------------------------------------|
| 1000.0 Chronic Fathead | X | | | | | X | | | | | |
| 1002.0 Chronic <i>Ceriodaphnia</i> | | X | | | | | | | | | |
| 1003.0 Chronic Algae | | | X | | | | | | | | |
| 1004.0 Chronic Sheepshead | | | | X | | | | X | | | |
| 1007.0 Chronic Mysid | | | | | X | | | | X | | |
| 2000.1 Acute Fathead | | | | | | X | | | | | |
| 2002.0 Acute <i>Ceriodaphnia</i> | | | | | | | X | | | | X |
| 2004.0 Acute Sheepshead | | | | | | | | X | | | |
| 2019.0 Acute Trout | | | | | | | | | | X | |
| 2021.0 Acute <i>D. pulex / magna</i> | | | | | | | X | | | | X |

Note: For a freshwater method to satisfy a saltwater method the analyst must also work on at least one saltwater DOC (besides the initial freshwater DOC). Other less frequent test species (topsmelt, silversides, shiner, etc.) may also be substituted with a more common test method, as appropriate (i.e., similar method, species, etc.). Other experimental design differences (static vs static-renewal vs flow-through) should be taken into consideration when training. Additional training may also include: reading SOPs, nuances of tests, publications, etc.

Attachment 6

Attachment #2: Steps for Individual DOC for Revised WET Module

Sample handling

- Proper temp upon receipt
- Holding time criterion met
- Support chemistry measurements
 - Calibration and use of meters (as appropriate)
 - pH, DO, conductivity, alkalinity, total residual chlorine, hardness, and/or salinity measurements

Initiation of test

- acclimation
- randomization
- collection of organisms
- age of organisms
- handling of organisms
- organism acceptability/selection
- prep of test dilutions
- test temperature
- food prep and addition
- dilution water prep and use

Renewal of test dilutions (Maintenance phase)

- temperature
- counting organisms
- organism observations
- feeding
- transfer of organisms
- food prep and addition
- prep of test dilutions

Ending of test

- transfer and counting organisms
- observations of organisms
- drying and weighing (as appropriate)
- balance calibration and use
- data gathering (i.e., weights, neonate production, survival data, etc.)
- QC data / bench sheets
- test acceptability criteria

Statistical analyses of data

- Crunch data (survival data, reproduction data, weight data)
- Determine appropriate endpoints for method (e.g., LC50s, IC25s, NOEC, NOAEC, etc.)
- Confirm that study meets test acceptability criteria
- Reporting

Footnote: all the requirements in Module 2 apply to this section (i.e., reading of appropriate SOPs, test methods, and any other organism / test specific information)