

**Minutes of  
THE NELAC INSTITUTE'S  
Whole Effluent Toxicity Testing (WET)  
Subcommittee for Acceptance criteria  
January 12, 2011**

---

The meeting was called the order at 1:00pm EST by RaeAnn Haynes. Members present are listed in Attachment B.

The first order of business was the review and acceptance of the minutes from December. The group voted to accept the minutes a typo correction and the addition of WET to the Mission/Goal statement.

Jeff Lowry addressed the statement by some of the laboratory representatives which stated that "5% of the laboratories will fail a PT study". The acceptance criteria is based on the study mean and the acceptance is +/- 2 standard deviations from the study mean. This means that there is always a 5% chance that any laboratory may fail the PT study, however, it cannot be inferred that in every study 5% of the laboratories participating in the study are expected to fail. All laboratories in a study may pass.

The group is satisfied with the Goal statement proposed last meeting:

To purpose an alternative WET FoPT acceptance criteria Table that is based on good biological science.

RaeAnn Haynes put together all the comments received from the group proposing changes to the WET FoPT Table. (See Attachment C). The group started at the top of the comments and got about half way through the list. The responses to the comments are described in the Attachment C. Some general suggestions are that NOEC tests are hypothetical estimates and therefore may not be appropriate measures for PT studies. All NOEC tests include a IC25 which is an endpoint test and as such may be a better measure of the laboratory's ability to acceptably report NOEC. This would be a major change to the current Table and if the group determines this would improve the PT program for WET laboratories we would have to get the State DMRQA coordinators informed and solicit their input as well as the NELAC AB's before going forward with such a change.

Jeff Lowry agreed to send RaeAnn the format in which to request data from the PT Providers so that request can be emailed out to the 3 PTP's.

The Chair, RaeAnn Haynes, initialed a discussion about decision making as the group moves forward. The group agreed that a consensus model where all members of the committee at least can agree to move forward with all proposals is ideal. However, the group also thought that a 2/3 vote will be enough to recommend any changes to TNI.

The next teleconference is planned for February 9, 2011.

**Attachment A**

**ACTION ITEMS  
TNI PROFICIENCY TESTING COMMITTEE**

<b>Item</b>	<b>Action Item</b>	<b>Assigned To</b>	<b>Due Date</b>	<b>Date Complete</b>
1	Keep the PT Board in formed	RaeAnn Haynes	On-going	
2	Send original comments to this subcommittee for evaluation	Stacie Metzler	1/12/2011	Not Complete
3	Request data from PT Providers for evaluation	RaeAnn Haynes	1/12/2011	Not Complete
4	A summary for non-biologists about the stream-lined WET test proposed by EPA in 2002.	Teresa Norberg-King	2/9/2011	
5				

**Attachment B**

**TNI WET FOPT SUBCOMMITTEE MEMBERS 2010-2011**

<b>Member</b>	<b>Affiliation</b>	<b>Email</b>	<b>Phone</b>	<b>Category</b>	<b>Present on the Call Y/N</b>
Raeann Haynes (Chair)	Oregon DEQ	<a href="mailto:Haynes.raeann@deq.state.or.us">Haynes.raeann@deq.state.or.us</a>	503-693-5757	AB	Y
Stacie Metzler	Hampton Roads Sanitation District	<a href="mailto:smetzler@hrsd.com">smetzler@hrsd.com</a>	757-460-4217	Lab	Y
Ginger Briggs	Bio-Analytical Laboratories	<a href="mailto:bioanalytical@wildblue.net">bioanalytical@wildblue.net</a>	318-745-2772	Lab	Y
Pete De Lisle	Coastal Bioanalysts Inc	<a href="mailto:pfd@coastalbio.com">pfd@coastalbio.com</a>	804-694-8285	Lab	Y
Robert Kelley	ETT Environmental Inc	<a href="mailto:bobkelley@ettenvironmental.com">bobkelley@ettenvironmental.com</a>	864-877-6342	Lab	Y
Jeff Lowry	Environmental Resources Assoc.	<a href="mailto:jlowry@eraqc.com">jlowry@eraqc.com</a>	303-431-8454	Other (PTP)	Y
Jamie Mitchell	Hampton Roads Sanitation District	<a href="mailto:jmitchell@hrsd.com">jmitchell@hrsd.com</a>	757-460-4220	Lab	Y
Rami Naddy	AE Com	<a href="mailto:rami.naddy@aecom.com">rami.naddy@aecom.com</a>		Lab	Y
Bob O'Brien	RT Corporation	<a href="mailto:bobrien@rt_corp.com">bobrien@rt_corp.com</a>	307-742-5452	Other (PTP)	Y
Faust Parker	PBS&J Environmental Toxicology Lab	<a href="mailto:frparker@pbsj.com">frparker@pbsj.com</a>	713-977-1500	Lab	Y
Steven Rewa	Environmental Resources Management	<a href="mailto:steven.rewa@erm.com">steven.rewa@erm.com</a>	616-738-7324	Lab	N
Chuck Wibby	Wibby Associates	<a href="mailto:cwibby@wibby.com">cwibby@wibby.com</a>	303-940-0033	Other (PTP)	Y
Patrick Yellin	USEPA	<a href="mailto:yellin.patrick@epa.gov">yellin.patrick@epa.gov</a>	202-564-2970	Other (EPA)	Y
Teresa Norberg-King	USEPA	<a href="mailto:Norberg-King.Teresa@epamail.epa.gov">Norberg-King.Teresa@epamail.epa.gov</a>		Other (EPA)	Y

## Attachment C

### Comment

The bulk of historic data is based on pre-NELAC labs: is it reliable?

Restrict endpoints to IC25 (reproduction or growth) and EC50 (survival) and do away with NOEC endpoints.

Have each field (test type / species) have two criteria - the IC25/EC50 for "accuracy" and the PMSD for "precision". Both would be reported and a lab would have to fail both for the result to be considered a failure.

The limits for the IC25 and EC50 should be the 99% confidence interval and the limits for the PMSD would be the upper limit only as defined in the EPA Method Variability document for toxicity. I don't think the lower limit should apply in as much as it is not a bad thing if the lab has very precise results.

That the WET FOPT table be improvised and easier to understand

That the WET PT studies be modified to better reflect a laboratory's performance, either by modifying the endpoint (i.e. required reporting of IC25 instead of NOEC) or making AB bodies understand that this is a monitoring tool and not to put so much emphasis on it.

NOEC endpoints should not be used and only point estimates (i.e. IC25 of LC50) should be used.

The TNI criteria need to work with the DMRQA criteria. (But I don't see why they have to be same).

### Response

What's the background for the pre-NELAC? Used EPA Tables to generate current NELAC Table. Methods were changed in 2002. Or methods were streamlined but not so much to affect accuracy and precision?

Cannot get rid NOEC for the permits, point estimates are necessary. PT study needs a point estimate. However, all NOEC tests include an IC25. Seems appropriate to use an endpoint test rather than a point estimate hypothesis result.

PMSD no data for PTP's, has established limits. Limits not hard. Can be used to repeat tests. May not reflect lab performance. Samples different than controls. No toxicity then no limits for PMSD. If we don't require NOEC then the PMSD is not necessary. 2 SD vs 3 SD? Answer by looking at the data? May depend on the test and study size.

The NOEC test uses the IC25 and the two results are normalized by the control. Do we really need two results for the same test?

The IC25 is an endpoint. The NOEC is more like a DL (detection limit).

### Attachment C

Use of 95% probability limits for TNI WET limits is probably not a good idea. TNI chemistry wastewater limits are based on 99% probability values.

These really need to be talked about in terms of standard deviations. EPA has stated +/- 2 std dev is appropriate for WET however, the data evaluation will show which is most reliable.

There are multiple variables in WET testing to begin with (live organisms, which have their own set of issues like age, condition, sex, etc.; multiple technicians handling test and organisms every day; variations in test water between labs (both salt and fresh); different test vessel configurations and on and on. With PT we are adding the most significant variable of all, which is the preparation of the "neat" PT sample for use in the test. A relatively small error in preparing the simulated effluent probably explains most of the differences between lab results; in the Chronic test series there is a daily preparation of the PT sample, almost always by different lab techs (even when you try to have the same person prepare the sample). This step in the process is not part of the normal WET test protocol.

Losing Accreditation for one species could have major ramifications for all tests (either salt or fresh water). In Texas, permittees run either chronic or 48 hour acute tests, in addition to a 0 % & 100% acute. If a lab loses accreditation for any one method in these groups they will lose all clients that have that requirement. If you are a freshwater only lab you are out of business. Even if you run both fresh and salt your revenues could be cut in half with the same result. This is not appropriate for such a variable screening tool.

The reality is, that with the exception of the *C. dubia* tests, there just isn't any significant difference in the various methods. If you can run one method you can pretty much run them all. So assessing on the OVERALL performance of the lab on PT's (even across methods) makes a lot of sense. And we have to keep in mind that we are, in some, cases grading a method, within a method, that has the same potential for error and variability.

### Attachment C

The dilution series used (.5) is inappropriate. A .75 series based on where the end point is projected would provide better data. In the old days, as I recall, we ran a range finder and then determined our own dilution series on the acute tests. An appropriate response curve would make much more sense for PT testing.

The end points being used are dumb! LC 50s are slightly better than point estimates. But if you do a literature search on the toxicity (LC 50's) of various toxicants you will find much variability and in some cases extreme variability. WET is a screening tool for complex effluents. We are only looking for a toxic response at some critical dilution. The agencies understand this variability and allow for additional confirmation showing that there is a REAL issue before holding the permittees feet to the fire.

Both IC25s and NOEC (i.e., hypothesis test) endpoints should be required for ALL short-term chronic WET studies for sublethal data (possibly for survival data as well). This should help evaluate response data and gain a better understanding of the variation of the given endpoint from all the testing labs. Currently, all labs may not be reporting both endpoints.

When reporting sublethal hypothesis test endpoints (i.e., NOEC, LOEC) require the reporting of the percent minimum significant difference (PMSD). This is a requirement for WET testing results and should be required for PT studies. This should help interpret / explain results among various laboratories and explain out-of-range results (as I mentioned on the previous call). It will also help evaluate the PMSD range among WET laboratories and ensure that invalid studies are not being reported.

### Attachment C

Suggestion (based on our discussion): For hypothesis test endpoints include results at 95% and 99% statistical significance (i.e.,  $\alpha = 0.05$  &  $0.01$ ). Don't think we can exclude results at 95% significance because most WET dischargers with NOEC requirements have same limitation (i.e., they can't use 99% statistical significance for reporting purposes).

Suggestion: Provide a dataset to all labs to statistically analyze and report endpoints. This may help provide some (initial) feedback on variance associated with data interpretation. Data set should result in anomalous statistical results that would require interpretation of the NOEC endpoint (e.g., USEPA 2000).