



***C. dubia* QA evaluation study**
Stakeholder Committee Meeting

March 28, 2023

Agenda

1. Opening remarks and review of agenda (10 min)
2. Approval of minutes from the last Stakeholder Committee meeting (10 min)
3. Review of progress for Task 2 (30 min)
4. Roundtable workshop (45 min)
5. Timeline for second intercomparison exercise (30 min)
6. Questions from the public (20 min)
7. Schedule and next steps (5 min)

Stakeholder Advisory Committee

- Katie Fong (SWRCB)
- Amelia Whitson (EPA Region IX)
- Veronica Cuevas (RWQCB4)
- Mitch Mysliwec (Larry Walker Assoc/CASA)
- Jian Peng (Orange County Public Works/CASQA)
- Sarah Lopez (Central Coast Water Quality Preservation Inc)
- Josh Westfall (Los Angeles County Sanitation Districts)
- Peter Arth (Enthalpy Laboratories)
- Annelisa Moe (Heal the Bay)
- Steve Boggs (California ELAP)

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Task 2 Training and Education- progress to date

This task is comprised of:

1- Laboratory visits by members of the Science Panel

2- Roundtable workshop with ELAP accredited laboratories and Science Panel

Laboratory Visits

- Goal was to collect more detailed information about the techniques used by a subset of the ELA accredited laboratories
- Laboratories were selected based on size/testing capacity and results of the baseline intercalibration study (ILS)
- Two Panel members conducted the lab visits.
- Name of laboratories visited remained anonymous to other members of the Panel , stakeholders and other laboratories
- This was not an audit. The information solely used by the Science Panel to help narrow the topics of discussion for the Roundtable Workshops.

Laboratory Visits

- Early March: Science Panel met in closed session to
 - Review existing audit and QA checklists
 - Review results of Task 1 historical data and baseline ILS
 - Produce their own list of topics and questions to address
 - Develop a visit schedule
- Mid-March: Two Panel members conducted the lab visits
 - Tour of the facilities
 - Discussion with the lab manager and technicians about their routine operations
 - Focused discussion on topics of interest including water, culture, food, and testing

Laboratory Visits

- Late March: Science Panel met in another closed session to
 - Discuss their findings and observations with the other Panel members
 - Identify the subtopics to be discussed by the laboratories during the roundtables.
 - Finalize the workshop agenda and format

The Panel indicated that the lab visits were “widely successful” with great participation and discussion from the labs.

Roundtable Workshop

- Goal was to determine the lab techniques that should be standardized to reduce intra- and interlaboratory variability in the second ILS
- Two half-day workshops covering four topic areas
 - Culture
 - Food/feeding
 - Testing
 - Documentation and record keeping
- Participants
 - One representative per lab, the lab manager
 - Members of the Science Panel

Roundtable Workshop Topics

Thursday March 23		Friday March 24	
<i>Topic #1 Culture</i>	<i>Topic #2 Feeding</i>	<i>Topic #3 Testing</i>	<i>Topic #4 Documentation</i>
Brood boards	Measurement of algae density and YCT solids concentrations	Test termination triggers	Documenting split broods
Reagents	Holding times	Age of animals	Brood board health as an early warning
Dilution water holding	Feeding procedures	Randomization	Labeling
Water treatment		WQ surrogates	Access to QA records
Air systems		Test volume/containers	Technician training
		Glassware washing	

Roundtable Workshop

- To start each topic, a Panel member gave a ~5 min introduction summarizing their observations and the specific details to discuss
- For every topic, each lab was given 2-3 min uninterrupted to share their experience and identify the lab techniques they would like to standardize
- This was followed by a group discussion to reach consensus on the techniques to standardize and test in the second ILS

List of parameters to test in the second ILS

- Limit age of adults used to start the test to 6-10 days old. Record which brood number is used to start the test.
- Quantify density of food in test chambers. Provide feeding methods.
- Food holding times of YCT ≤ 7 days after thawing and algae ≤ 21 days. Record creation date.
- Evaluate brood boards using common set of health criteria.
- Run test for 8 days AND record when labs would have ended the test.
- Document split broods on bench sheets at the time of observation.
- Document dilution water treatment system and holding time (checklist format).

List of parameters to test in the second ILS

- Renew test boards daily within a + or - 2-hr window from test initiation.
- Evaluate brood board health for 2 weeks prior to testing.
- Require randomization by blocking of known parentage AND randomization of cups on the test board.

Parameters discussed but not standardized

- **Age of animals used to initiate tests**

All labs appear to be following the EPA manual which requires using < 24 hr old neonates produced within an 8-hr window. Not enough evidence to reduce the window or age limit.

- **Surrogate for water quality**

Most labs are not using true surrogate containers. Composites of the 'before renewal' water are produced to have sufficient volume to submerge the probes.

- **Feeding methods**

Feeding small volumes in individual test cups versus in test solution could cause some intra and inter-lab variability. Requiring feeding in cups could complicate operations for labs using the same dilution water for different species.

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Second Intercalibration Study

- Initial plan is to use same study design with implementation of new parameters to assess the following hypothesis

Implementation of standardized parameters does reduce intra- and inter-laboratory variability

- Elements of the study design

Sample Type	Number of Samples	Dilution Series	Number of Rounds
Sample 1- dilution water recipe #1	1	No	3
Sample 2A- dilution water recipe #2	1	No	3
Sample 2B-F- NaCl in dilution water recipe #2 (Perrier)	5	No	3
Sample 3- NaCl solid sample (to be diluted by the labs using their own dilution water)	1	Yes	3

Schedule

Activity	Output	Timeframe
Lab site visits	Inform roundtable topics	Week of March 13 COMPLETED
Two Roundtable Workshops	Create draft list of recommendations for standardization	By end of March COMPLETED
Second intercalibration study design and workplan approval	Testing details for laboratories	By mid-April
Second Intercalibration	Data quantifying improvement for among lab variability	Mid-April thru end of May
Intercalibration data analysis	Refined list of recommendations	By end of July
Draft Guidance Manual	Final product for review	By end of August
Final Guidance Manual and supporting documentation	Final product for Labs and SWRCB	End of September

Next Steps

- Produce draft workplan and QAPP for the 2nd ILS
 - Review and approval by the Stakeholder Committee and the Science Panel
- Produce guidance document for the laboratories to implement the standardized parameters (randomization, brood board health metrics)
- Schedule meeting with participating laboratories
- Next Stakeholder meeting: April 13, 2023 at 1:00 pm

- Recording and minutes available on SCCWRP Cerio project webpage <https://www.sccwrp.org/about/research-areas/additional-research-areas/ceriodaphnia-toxicity-testing-quality-assurance/>