C. dubia QA evaluation study

Stakeholder Committee Meeting

Wednesday April 6, 2022

Agenda

1. Opening Remarks and Review of the Agenda (10 min)

2. Minutes of Stakeholder Committee Meeting #5 (5 min)

3. Proposed Testing Plan (45 min)

4. Schedule and Next Steps (5 min)

5. Public Comments (15 min)

Proposed Testing Plan

Background

- Previous studies have investigated possible sources of intra and inter-lab variability in the *C. dubia* chronic reproduction toxicity test

- This study aims to build on previous efforts, working with all CA-accredited labs, to provide lab technique guidance to improve both:
  - Consistency of individual lab performance
  - Comparability in *C. dubia* test results among laboratories

Study Questions

1) What are the *C. dubia* test laboratory techniques used by ELAP accredited laboratories in the state of California?

1) How does variability in *C. dubia* control reproduction and/or reference toxicant compare within and among laboratories? How do lab techniques influence the observed variability?

1) Does standardizing select test laboratory techniques reduce intra- and inter-laboratory variability in control reproduction and/or reference toxicant responses?

Approach

Completed

1) What are the *C. dubia* test laboratory techniques used by ELAP accredited laboratories in the state of California?

- **Develop a comprehensive database documenting historical data and lab techniques**

Completed

2) How does variability in *C. dubia* control reproduction and/or reference toxicant compare within and among laboratories? How do lab techniques influence the observed variability?

- **Conduct in depth data analyses to identify lab techniques contributing to intra- and inter-lab variability**

3) Does standardizing select test laboratory techniques reduce intra- and inter-laboratory variability in control reproduction and/or reference toxicant responses?

- **Perform intercomparison laboratory exercises using existing and optimized lab procedures**

Conclusions From Our Last Meeting

- Analyses of historical data and lab methods are inconclusive
  - Lab practices vary greatly among labs
  - No consistent or predominant lab technique identified from statistical analyses

- But some test parameters emerged as possible sources of variability
  - Age window at test initiation, test duration, water recipe, food recipe

- Many of you would like to see some lab testing to demonstrate that multiple labs testing split samples can get comparable results
  - SCCWRP was tasked to draft a study plan for review and feedback by the stakeholders

Lab Testing: Questions and Testable Hypotheses

Q1: Does standardizing lab practices improve **consistency and comparability in** C. dubia toxicity test results?

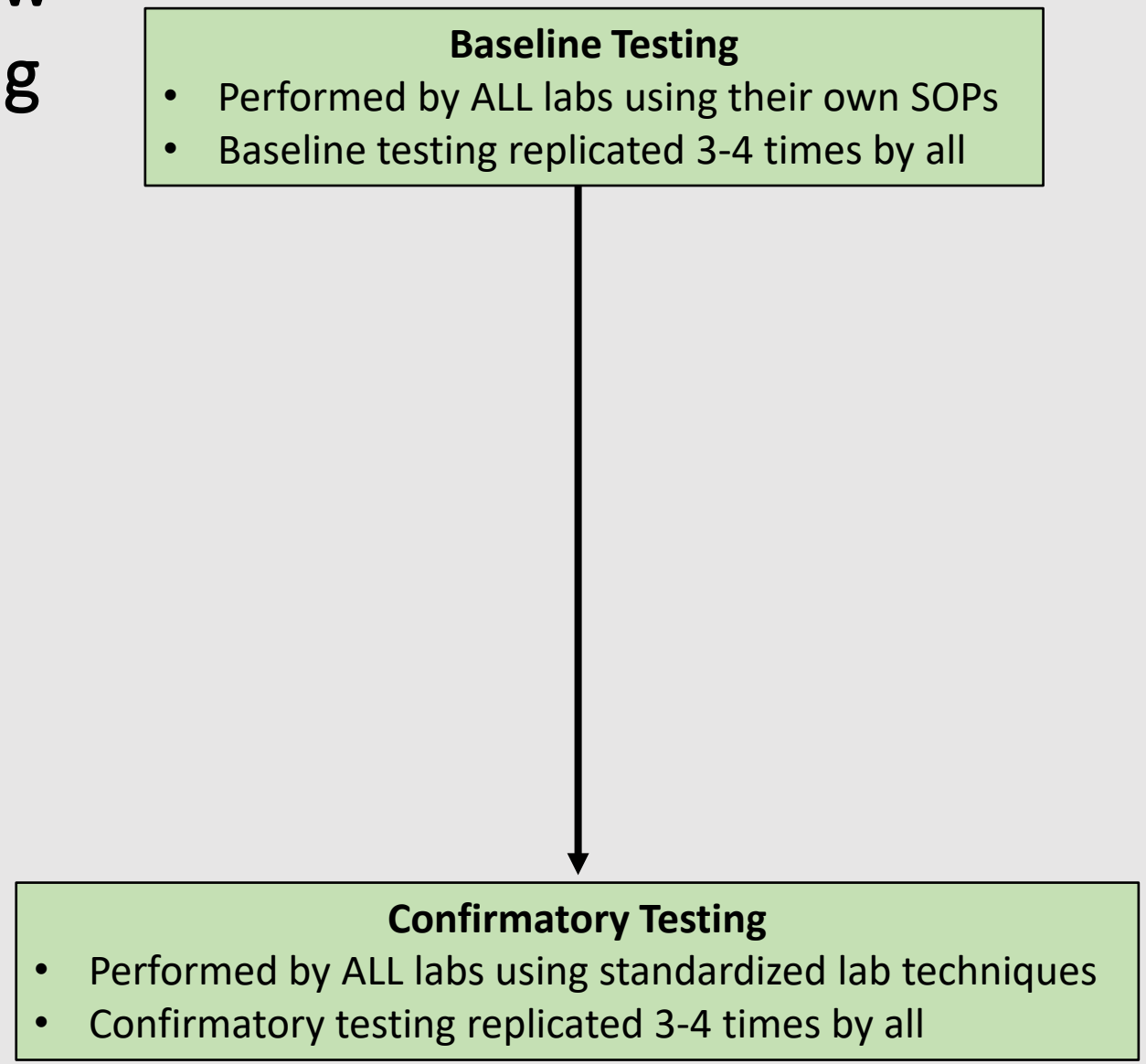
$H0^1$: Standardizing lab practices does not reduce **intra- and inter**-variability in "control" **test samples** (# neonates/ female, mean, SD, **CV**) and ref. toxicant responses (EC50, mean, SD, **CV**)

Q2: Which lab practice should be standardized to reduce intra- and inter-lab variability?

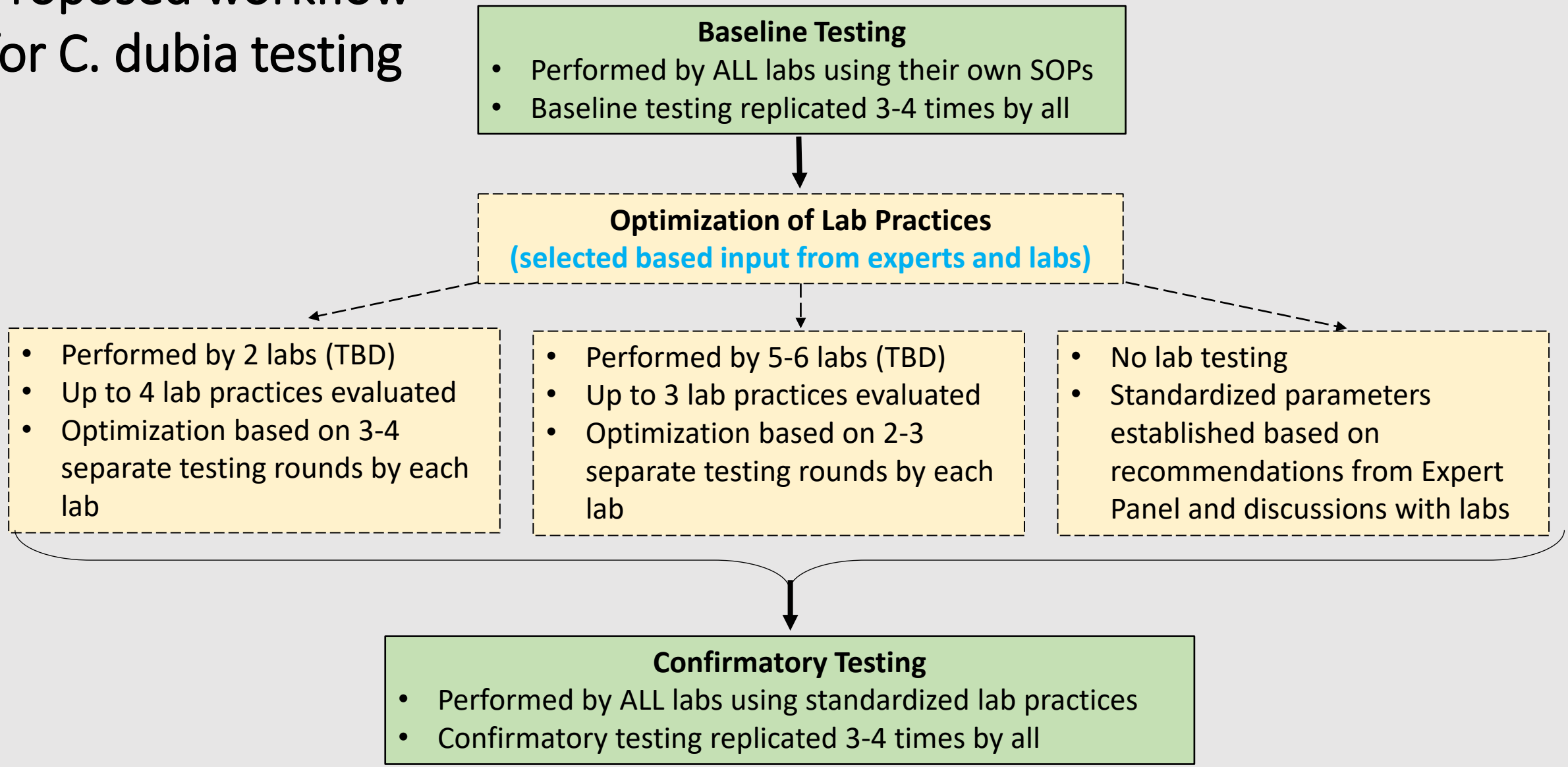
$H0^2$: "*Lab practice x*" does not have an impact on variability in "control" test sample reproduction (mean, SD, CV) and ref tox response (EC50, mean, SD, CV)

Testing 4 lab practices: age of neonates at test initiation, water recipe, YCT food recipe, test duration

Proposed workflow for C. dubia testing



Proposed workflow for C. dubia testing



Testable Hypothesis 1

H0: Standardizing lab practices does not reduce variability in “control” test samples (# neonates/ female, mean, SD CV) and ref. toxicant responses (EC50, mean, SD, CV) among laboratories

Proposed Study Design to Test H0[1]

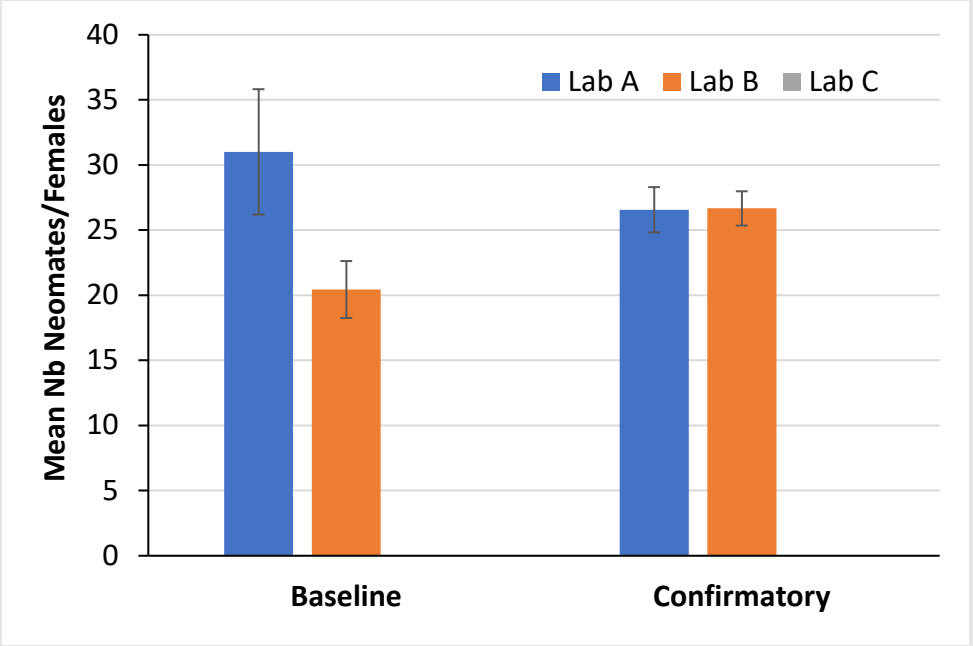
Two rounds of testing by <u>all laboratories</u>

- Baseline testing using current SOPs
- Confirmatory testing using standardized lab practices

Each round will be performed multiple times (e.g., 3 or 4 times) to calculate CV

- Blind sample analyses (incl. dilution waters and ref tox)
- 8-day tests with daily neonate production
- Additional data collection (e.g., specific age window, brood board health metrics tbd, etc..)

Example of key graphic (not REAL data)



Testable Hypothesis 2

H0: “Lab practice x” does not have an impact on variability in “control” test reproduction (mean, SD, CV) and reference toxicant response (EC50, mean, SD, CV)

Proposed Study Design to Test H0[2]

Multiple rounds of testing by select labs (selection criteria TBD).

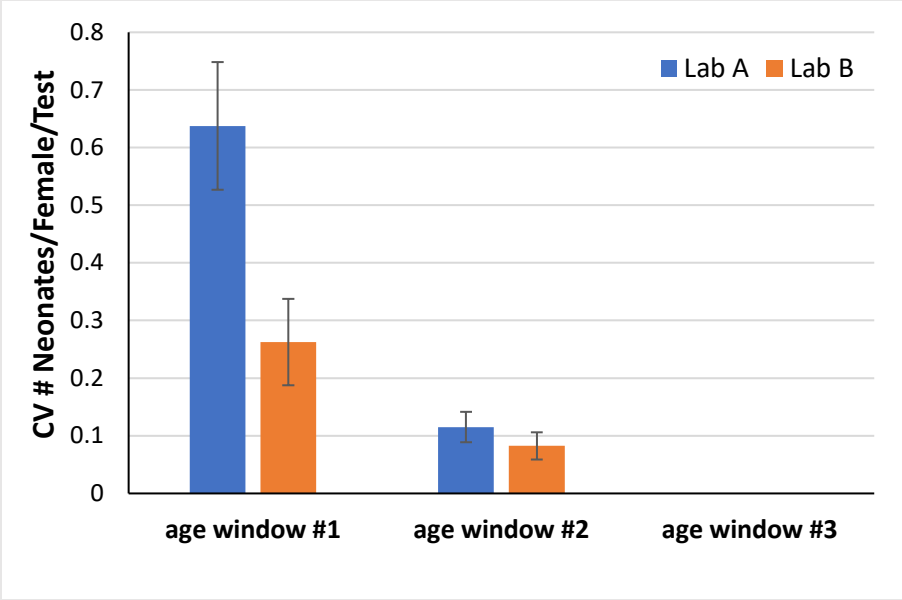
Lab techniques evaluated

- 3 different age windows
- 3 different water recipes
- 3 different YCT food recipes

These will be 8-day tests with dilution water and reference toxicant

Tests will be replicated 3-4 times to calculate CV

Example of key graphic (not REAL data)



Agenda

1. Opening Remarks and Review of the Agenda (10 min)

2. Minutes of Stakeholder Committee Meeting #5 (5 min)

3. Proposed Testing Plan (45 min)

4. Schedule and Next Steps (5 min)

5. Public Comments (15 min)

Next Steps and Schedule

This week: SCCWRP will revise proposed study design based on stakeholders' feedback

Next **Tue** 4/12/22: SCCWRP will present the proposed testing plan and options to the Expert Science Panel. The Panel will make their recommendations on how to move forward

End of April: SCCWRP will draft a written lab testing plan for review **by SAC and ESP**

~~Early~~ May: Upon approval of the plan by the ESP, SCCWRP will initiate testing coordination and logistics with the participating labs