



COUNTY SANITATION DISTRICTS OF LOS ANGELES COUNTY

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March 2, 2018

Keith A. Maruya
Principal Scientist
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Dear Mr. Maruya:

Comments on Draft Final Report "Monitoring Strategies for Constituents of Emerging Concern (CECs) in Recycled Water"

The County Sanitation Districts of Los Angeles County (Sanitation Districts) appreciate the opportunity to provide comments on the draft final report entitled "Monitoring Strategies for Constituents of Emerging Concern (CECs) in Recycled Water" (Report). This Report contains the draft final recommendations of the Science Advisory Panel for CECs in Recycled Water (Panel). As background, the Sanitation Districts consist of 23 independent special districts serving approximately 5.7 million people in Los Angeles County, covering an area of 820 square miles and encompassing 78 cities and unincorporated territory within the county. The Sanitation Districts own and operate 11 wastewater treatment plants. One of these plants is a secondary treatment plant with a deep ocean outfall; eight are water reclamation plants with existing discharges to effluent-dominated surface water bodies; and two do not have surface water discharges.

Overall, the Sanitation Districts support the approach taken by the Panel, and support the Panel's recommendation that "the State Water Board continue to rely on a transparent, science-based framework to guide prioritization of which CECs should be included in recycled water monitoring programs both now and in the future as additional data become available." We also support the Panel's recommendation that "the State Water Board consider the results of more definitive research showing an actual relationship of antibiotic resistance to reuse water before changing its current policy" (Section 9.3, page 97).

However, while we are supportive of the overall approach adopted by the Panel, as well as most aspects of the Report, we do have some specific concerns. These mainly regard the recommendation to require monitoring using bioanalytical methods, along with the recommendation to require specific actions to be taken based on the results of these methods.

Areas of Concern

Use of Bioanalytical Methods

In Section 7, the Panel recommends using bioanalytical methods as an initial screen for CECs, which would then lead to further required management actions. In particular, the Panel has recommended use of two receptor/bioassays that both have established adverse outcome pathways: the estrogen receptor (ER) and aryl hydrocarbon receptor (AhR) assays. The Sanitation Districts believe that the bioanalytical tools and our ability to interpret results from these tools have not reached a point where they should be used on a required basis. In order for the tools to be required, there first needs to be standardized testing protocols specified, and the specific protocols listed need to be available at commercial laboratories. Additionally, triggers need to be established for each standardized protocol.

The Panel acknowledges these issues (p. 65), stating that, “The recent Direct Potable Reuse (DPR) Expert Panel; however, was more critical of bioanalytical tools, raising concerns regarding the lack of standardization, interpretation of results, and regulatory applications, which ultimately may limit their use (Olivieri et al., 2016).” The DPR panel was critical for good reason, and it is not at all clear that the state of the science has advanced enough since the DPR panel met to justify with moving forward with bioassays as a regulatory tool at this time.

At an absolute minimum, the Panel should recommend that bioanalytical monitoring not be required until the “bioanalytical advisory group” discussed in Section 7.5.5 has been formed and has provided guidance on the required sampling, extraction, measurement, data reporting, interpretation, and response. More specific comments on use of bioanalytical methods are provided below.

Standardization and Validation

The Panel provides examples of "standardized" methods from USEPA, OECD, the European Union, etc. (Section 7.2, page 66), however the Panel does not recommend a specific ER or AhR bioassay out of the variety that are available. Identification of a single standardized, validated method that includes both an extraction procedure as well as the bioassay needs to be made before a requirement is established for utilities to perform routine monitoring using these bioassays. Additionally, none of the assays have been validated at a federal level, which suggests that it may not be appropriate for them to be used as a required method for assessing recycled water quality at this time. Moving forward, the panel should recommend a specific ER or AhR bioassay or bioassays out of the variety that are available, so that efforts can be focused on standardizing and validating these particular assays.

Furthermore, the Panel should specify who would have oversight over the laboratories that would conduct these bioassays to ensure the labs are qualified, that appropriate QA/QC is performed, and that data quality management systems are in place. The state of California (ELAP) does not currently certify labs for bioassays and it may be many years before they do.

Availability of Laboratories

The Panel should be aware that many of the ER assays require extensive lab infrastructure and expertise that goes along with a mammalian cell culture operation, which most small utilities and many large utility labs do not have. The only methods that most of the larger utilities could potentially use would be the "off-the shelf" kits such as the GeneBLAzer-ER α assay.

The Panel does refer to three commercial labs that could "currently provide services, including bioanalytical screening of organic extracts of water samples" (Section 7.2, page 67). However, the Panel does not specify whether these three labs have the capacity to handle samples from all of the DPR projects in California that would be required to use these bioassays.

Quality Assurance/Quality Control (QA/QC)

The Panel provides good guidance regarding QA/QC for sample extraction and bioassays but should make more specific recommendations. The Panel refers to an inter-laboratory agreement and literature reports that compare results from different labs using similar methods (Section 7.3.2, pages 69-70; Section 7.5.2, page 74); however, more rigorous inter-laboratory calibration studies with well-defined standard operating procedures and QA/QC parameters would provide the necessary guidance to address sample extraction and QA/QC procedures.

Bioassay Equivalency

There are many bioassays that detect estrogenicity or that detect upregulation of one or more estrogen receptors. The Sanitation Districts' lab has some experience with three different estrogenicity bioassays. The two that we have the most experience with, GeneBLAzer and T47D-KBLuc, have been shown to give comparable results. The most difficulty we have had with these methods has been with the sample extraction step rather than the bioassay step. The Panel indicates that there are available EPA methods with standardized extraction procedures that could be used, but it is not clear if these extraction procedures are all equally well suited for use with the ER and AhR bioassays in terms of compatibility of the extract solvents and the cells used in the bioassays. The Panel should assess combinations of extraction procedures and bioassays, and recommend the appropriate combinations.

The Panel should also define equivalency between the different methods for each bioassay type, depending on a desired level of accuracy and variability. A recent literature article¹ compared five different estrogen transactivation assays (YES, ER α -CALUX, MELN, T47D-KBLuc, and GeneBLAzer-ER α) and indicated a coefficient of variation of 32% between all five assays across all tests performed. It does not make sense to recommend a specific receptor type to be used for monitoring knowing that the end users may get significantly different results because they used different extraction procedures and different assays, even though they targeted the same receptor (i.e., the ER or the AhR). The Panel should provide recommendations regarding acceptable levels of variation between the different bioassays available for each receptor type. Any recommendations should be based on analytical performance or results from a rigorous intercalibration study.

Establishment of Bioassay Triggers

The Panel should identify the standardized trigger level for each particular bioassay protocol that it recommends for use. In the Report, the Panel directs dischargers to use the methods outlined in Textbox 7.1 to develop their own triggers. This could result in inconsistent triggers and thus inconsistent interpretation of results around the state. Additionally, while Textbox 7.1 provides an indication of the basis for an ER bioassay based on its response to EE2 (3.5 ng/L EE2 equivalent), for the AhR bioassay the Panel does not make a suggestion as to the specific compound to be used to set a trigger level, nor does it make a recommendation for the specific concentration of that compound at which the trigger would be set. Any trigger chosen must be based on sound toxicology, wherein the trigger value is clearly linked to well established adverse biological effects.

¹ Kunz, P.Y., et al. (2017) Water Research. 110: 378-388.

Response to Bioassay Triggers

The Panel recommends actions to follow based on bioassay results (Section 7.5.3, page 76). If the bioassay gives a response greater than the trigger level, the recycled water producer is required to resample within 72 hours. If the result is confirmed, the recycled water producer would be required to increase the frequency of their sampling and notify the appropriate regulatory authorities. This degree of required response is unwarranted at this time, given the state of knowledge and experience related to bioassays. Additionally, the bioassay results do not represent an immediate threat to public health protection so the 72 hour requirement is inappropriate. At this time, the required initial response to an elevated bioassay response should be simply confirmatory sampling. There could also be a recommendation that agencies implement additional chemical and/or bioassay monitoring on a voluntary basis, and consider taking the steps listed in Step 3 of Section 7.5.3, again on a voluntary basis.

Cost Estimates

Cost estimates for the estrogen receptor (ER) and aryl hydrocarbon receptor (AhR) assays should be provided so that stakeholders can evaluate the cost-benefit of such analyses. Utilities could use information on how many illnesses/adverse outcomes could be expected for waters in each of the BEQ/AL tiers (section 7.5.3, page 76) to determine when these assays are appropriate and the most beneficial way to use these assays.

CEC Data Used for the Report

In the preparation of the Report, the Panel spent considerable resources to collect and analyze extensive data on the presence of CECs in recycled water. These data can serve as a valuable resource for other entities going forward. However, the only summary of the data is in Figure 4.1, and does not include all of the compounds analyzed (e.g., 1,4-dioxane). It would be extremely helpful if the final report could contain a more comprehensive summary of the data, as included in Appendix K of the 2010 Final Report on Monitoring Strategies for Chemicals of Emerging Concern in Recycled Water. At a minimum, it would be useful to have a table that includes the 90th percentiles of the data that were used to compare results to MTLs. This would allow the Panel's work to be referenced and used by a wider audience.

Panel Guidance and Review for Future Changes

The Panel recommends that the State Water Board conduct a number of tasks, including updating the list of priority CECs and updating guidance on selecting viable surrogate parameters and performance indicator CECs. While we have no objection to State Water Board staff conducting these tasks, we strongly recommend that no changes be made to CEC monitoring requirements until the Panel has had a chance to review and provide guidance on them.

Recommendations for Submission of CEC Data

The Panel recommends that the State Water Board develop protocols for submission of CEC data in an electronic, machine-readable format (Section 2.3, page 12; Section 5.2, page 42). Publicly owned treatment works (POTWs) already conduct extensive reporting of their data, much of which is already done in an electronic, machine-readable format through programs such as the California Integrated Water Quality System (CIWQS). All efforts should be made to make use of existing data submission routes, so that duplicative reporting efforts are not put in place. Duplicative reporting draws upon the resources of both the regulated and unregulated community, without providing any improvements in water quality or public health.

Thank you for the opportunity to provide comments on the Report. If you have any questions or require additional information, please contact Ann Heil at (562) 908-4288, extension 2801.

Very truly yours,



Ann Heil
Division Engineer
Reuse and Compliance Section

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Appendix A. Minor Comments and Clarifications

- 1) Page 14 (end of second paragraph): outdated estimate of the potential for future water recycling in California. This estimate was made in 2010, well before the recent drought and the resulting unprecedented efforts to conserve water. Since that time, wastewater flows have dropped tremendously and this in turn has affected the amount of recycled water that is available for reuse. This estimate should be updated or deleted.
- 2) Footnote 11 on page 15: There is no source of information for the estimate of the percentage of edible food crops that are irrigated with recycled water and the extrapolation of how this may change in the future. The source should be included in the footnote.
- 3) Page 56: Method SM6810 “is claimed to achieve detection limits ranging from 1 to 2,000 ng/L.” Actual detection levels are typically lower than this range.
- 4) Page 57: Additional methods for non-regulated substances should include EPA method 1625 for NDMA, in addition to EPA method 521.
- 5) Global: The term “recycled water” should be used instead of “reclaimed water” throughout the Report.