Application of NAMs and AOPs to Surface Water Surveillance and Monitoring in the Great Lakes (EPA Region 5) and a Western River (EPA Region 8)

Daniel L. Villeneuve, US EPA, Office of Research and Development, Center for Computational Toxicology and Exposure, Great Lakes Toxicology and Ecology Division

Problem/Need

- Regions, states, tribes, and communities are monitoring an ever-growing list of contaminants in water and other environmental matrices.
- Established water quality standards / guidelines are lacking for many of the chemicals detected.
  - Uncertainty about whether the chemicals detected are likely to be harmful at the concentrations detected
- Need to focus limited resources available for monitoring, research, and/or source reduction on the substances most likely to cause adverse effects.
- Even with extensive contaminant monitoring, undetected compounds and mixtures leave uncertainty about whether assessments based on individual chemicals are sufficiently protective.

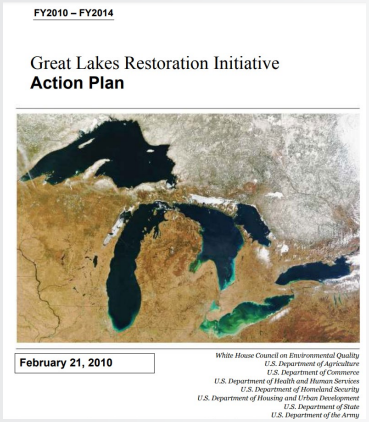
Role for NAMs

- In the absence of traditional animal toxicity data, NAMs can provide a provisional, protective (?), benchmark to support risk-based prioritization

- When traditional animal toxicity data are limited (scope of endpoints or taxa), NAMs can protect against mode of action-based toxicities that may be overlooked in traditional guideline studies or QSARs.

- NAMs can be used to directly test complex mixtures, providing bioactivity data that account for unknowns and cumulative/integrated effects.

EPA Region 5
Great Lakes Restoration Initiative – Emerging Contaminants



Focus Area 1: Toxic Substances and Areas of Concern

Goal 5: The health and integrity of wildlife populations and habitat are protected from adverse chemical and biological effects associated with the presence of toxic substances in the Great Lake Basin.

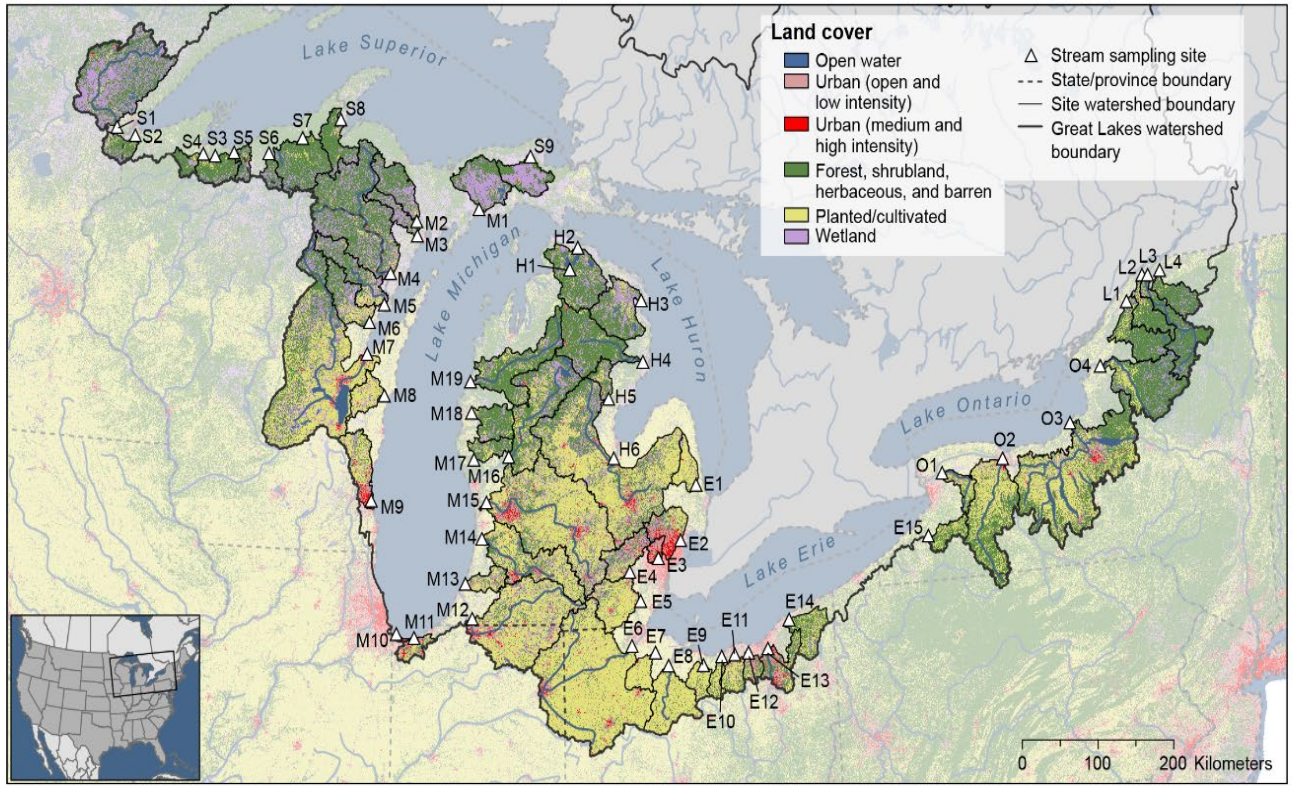
- Identify significant sources and impacts of **new toxics** to the Great Lakes ecosystem ….., in order to devise and implement effective control strategies.

Focus Area 1: Toxic Substances and Areas of Concern

Increase knowledge about contaminants in Great Lakes fish and wildlife

- Identify **emerging contaminants** and assess impacts on Great Lakes fish and wildlife

Chemical monitoring



709 water samples collected 2010-2013

57 Great Lakes tributaries

38 sites sampled 1-2 times

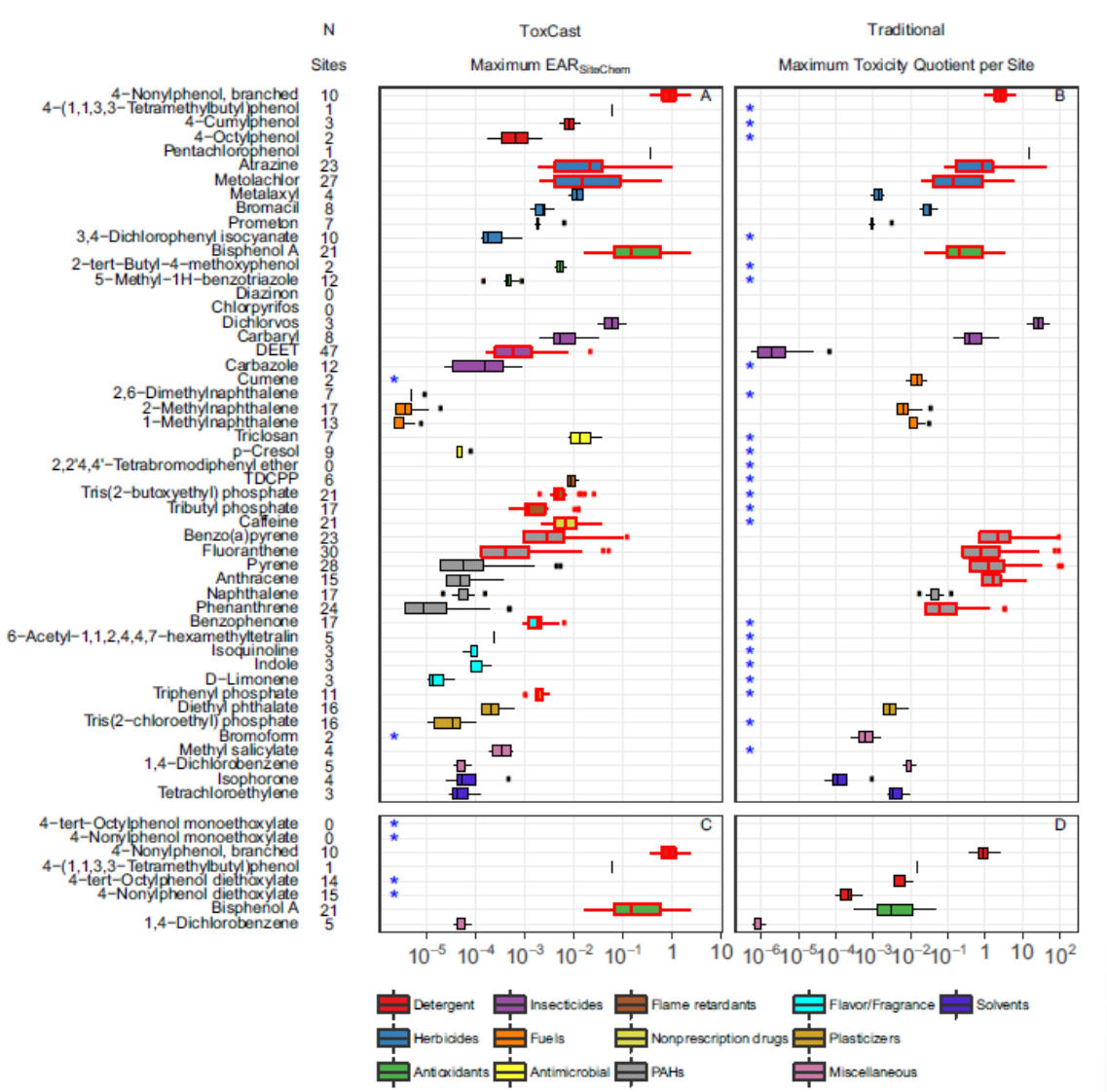
19 sites sampled 7-64 times

Analyzed for 67 organic contaminants

- Water quality benchmarks (27/67 = 40%)
- In vivo toxicity data (34/67 = 51%)
- ToxCast data (54/67 = 81%)

Which chemicals are of concern?
Where are we most likely to see impacts?
What kinds of effects might we expect to see?

Which chemicals?



$$EAR = \frac{Measured\ Concentration\ (\mu M)}{Activity\ Concentration\ at\ Cut{-}off\ (ACC;\ \mu M)}$$

$$TQ = \frac{Measured\ Concentration\ (mg/L)}{In\ vivo\ effect\ concentration\ (\frac{mg}{L})}$$

1. 4 nonylphenol
2. bisphenol A
3. Metolachlor
4. Atrazine
5. DEET
6. Caffeine
7. tris(2 butoxyethyl) phosphate
8. tributyl phosphate
9. triphenyl phosphate
10. benzo(a)pyrene
11. Fluoranthene
12. benzophenone.

Which sites?

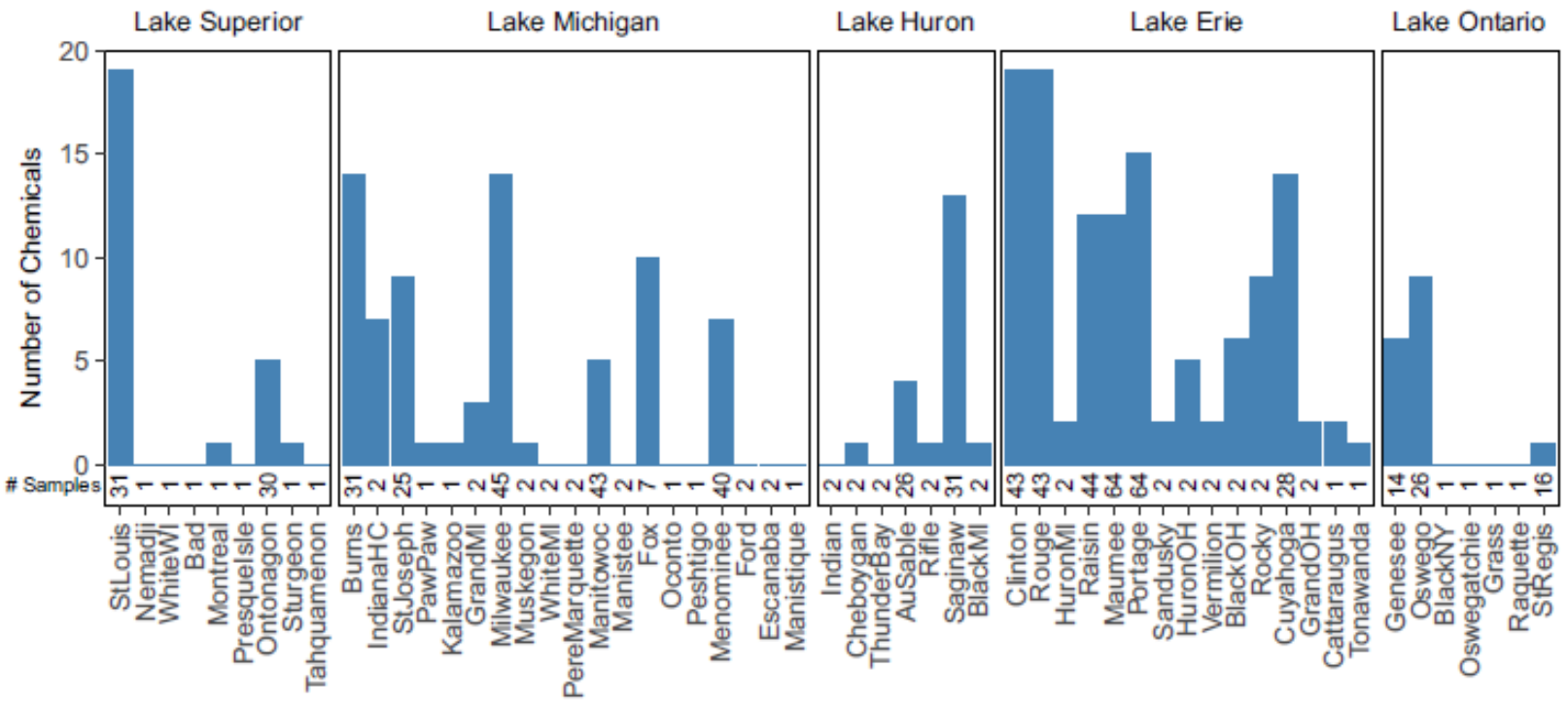
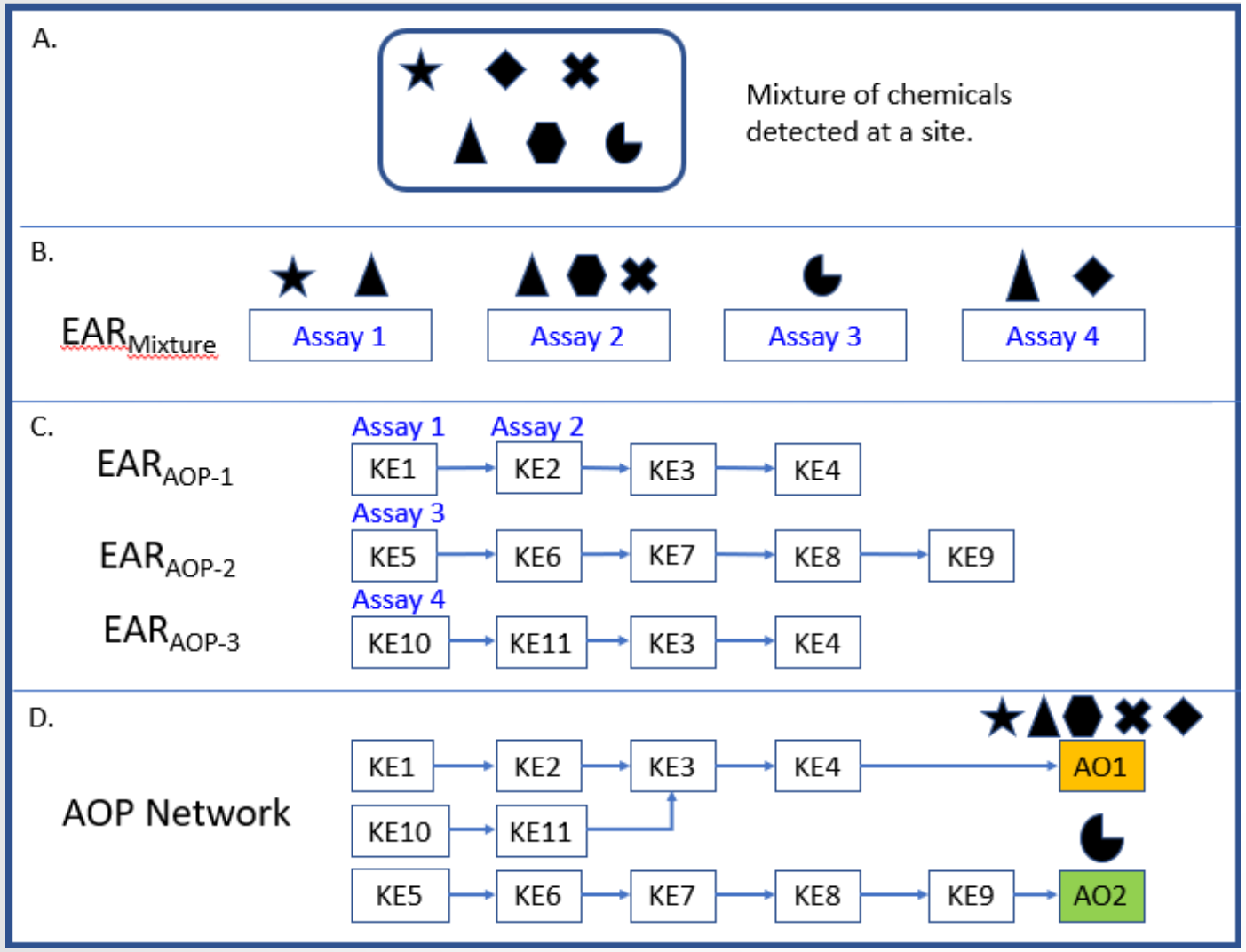


Fig. 3. Number of individual chemicals with at least one sample that resulted in a maximum exposure–activity ratio ($EAR_{SiteChem}$) > 10^{-3} for each site.

Sites link to sources and stakeholders

What effects?



Considers cumulative effects of **detected** chemicals

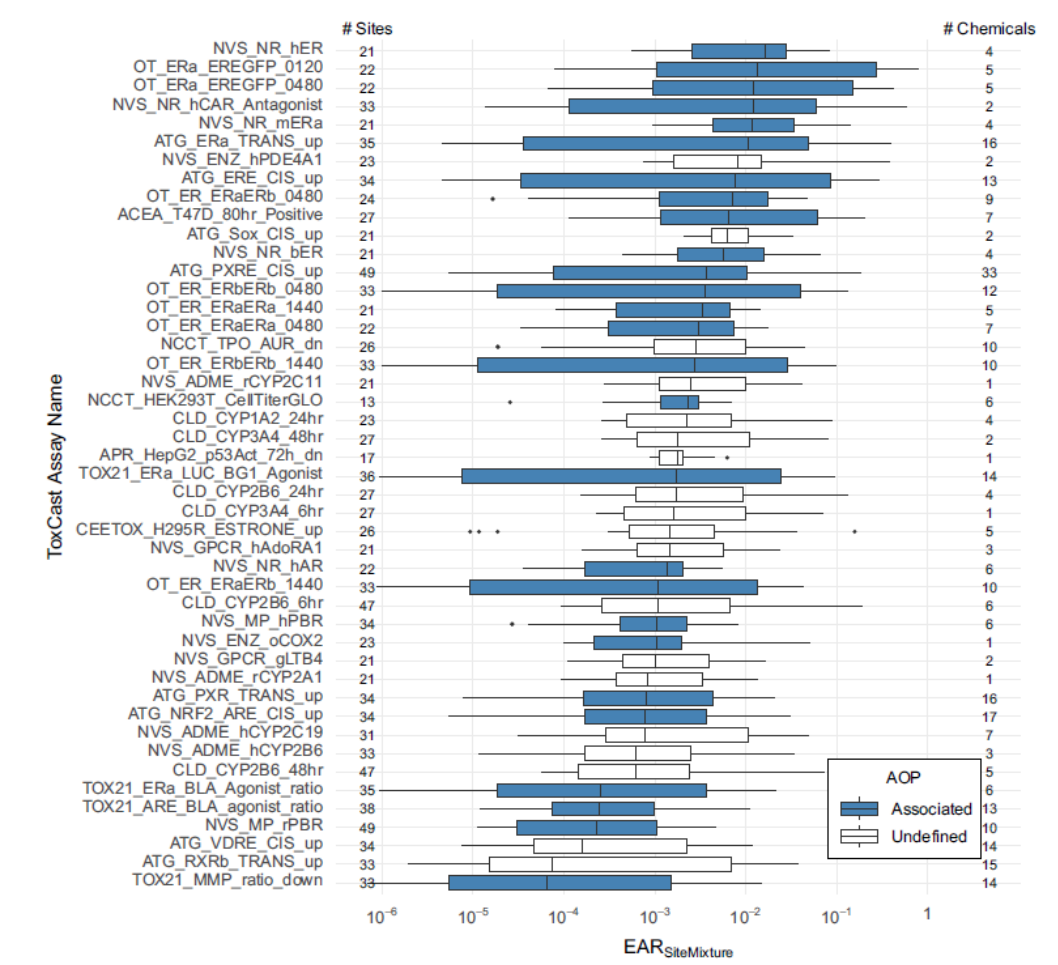
Assume additivity within each ToxCast assay/endpoint

Assay endpoints map to key events
Redundant KEs not double-counted

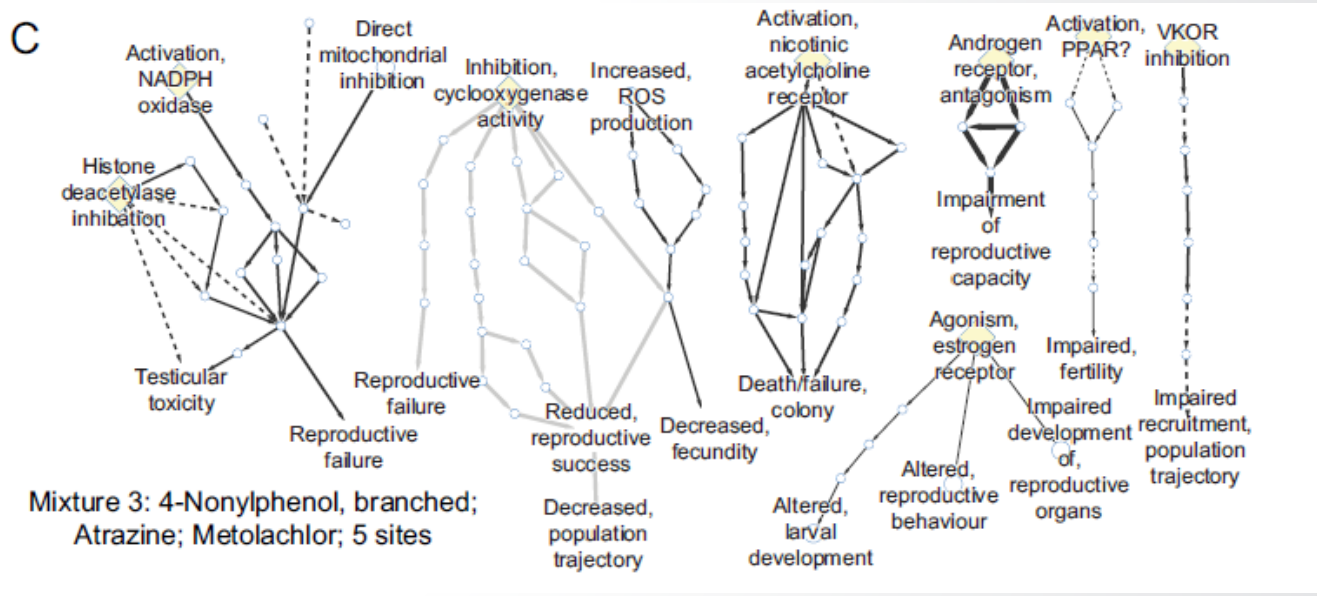
Considers cumulative impacts of multiple pathway perturbations on potential adverse outcomes.

What Effects?

Assay endpoints associated with higher EARs



Associated AOPs / AOP networks



Mixture 3: 4-Nonylphenol, branched; Atrazine; Metolachlor; 5 sites

GLRI-CECs, On-going research

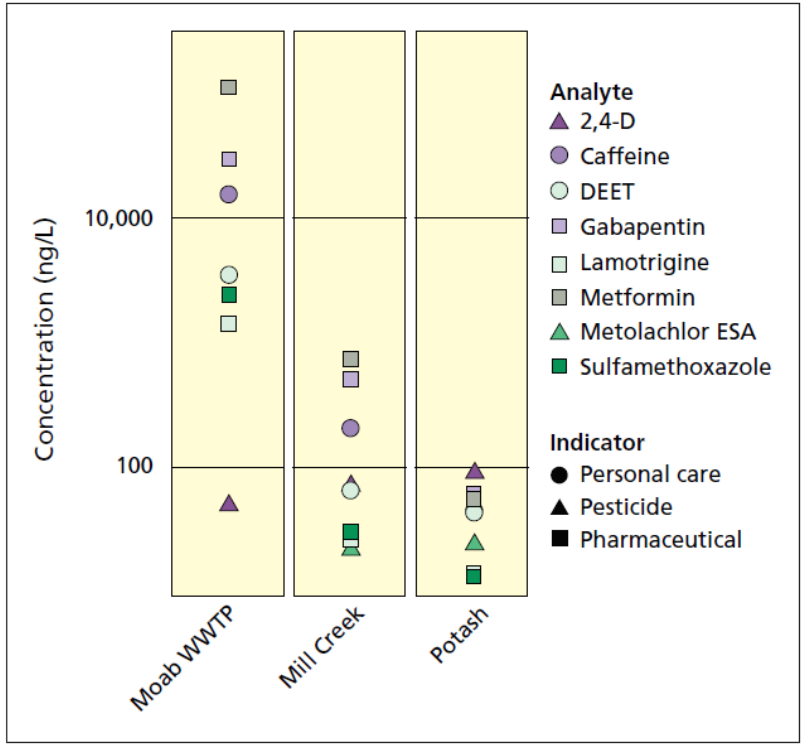
- NAMs-based prioritization being applied to other data sets
  - Fill gaps when water quality benchmarks and in vivo toxicity data are lacking or limited
  - Additional GLRI data sets
  - Other USGS monitoring studies (including drinking water)

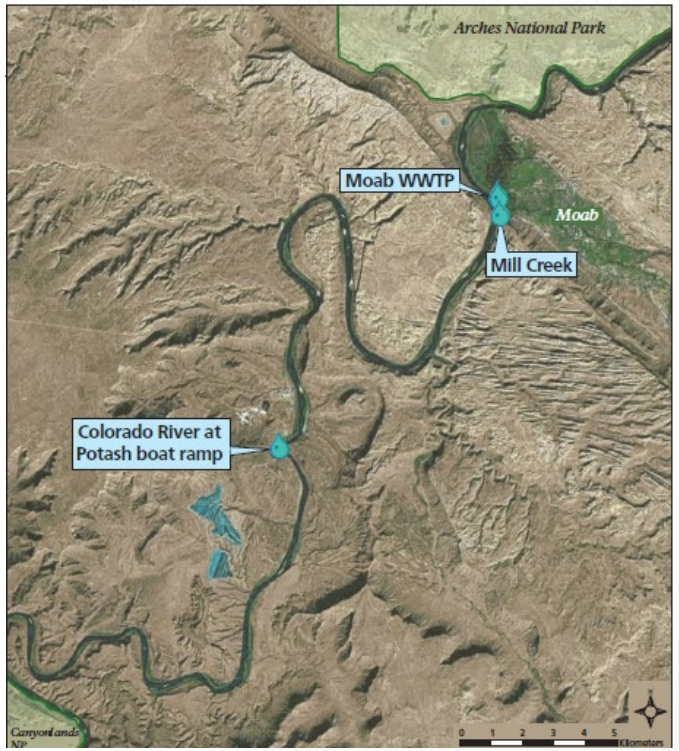
- Risk-based prioritization (incorporating NAMs) is now being applied to over 800 organic contaminants detected over 10 years of CEC monitoring
  - Includes water, sediment, passive samplers, mussels, fish
  - Help inform nomination of potential chemicals of mutual concern as defined through Annex 3 of binational Great Lakes Water Quality Agreement.

EPA Region 8
Waste-water treatment upgrade, Moab, UT

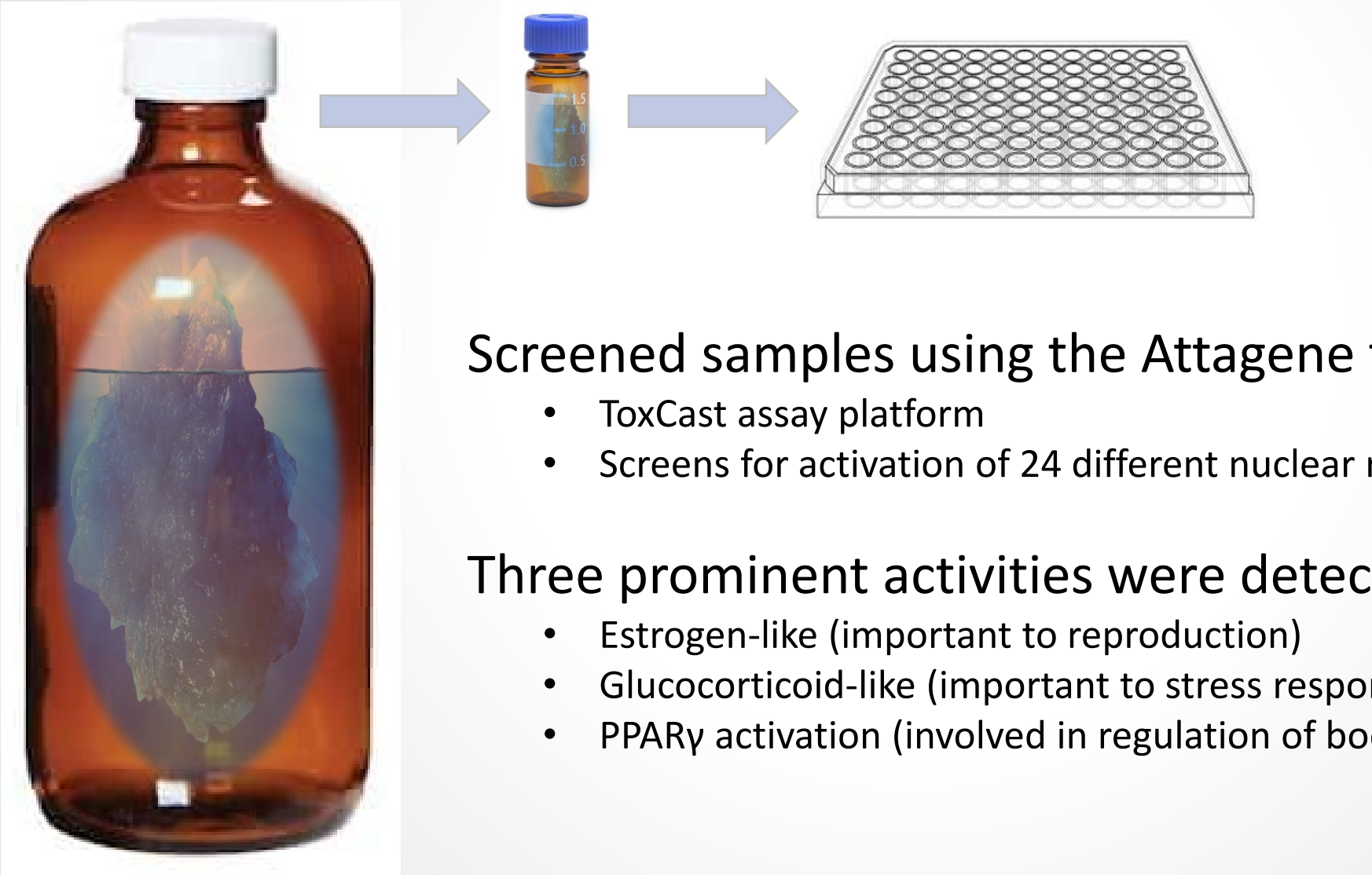
- 2013 National Park Service and USGS measured contaminants along Colorado River between Arches NP and Canyonlands NP

- Variety of pharmaceuticals, pesticides and personal care products detected
  - Greatest concentrations at the Moab WWTP discharge
  - Detectable concentrations extended > 15 km downstream





What about chemicals that weren't monitored



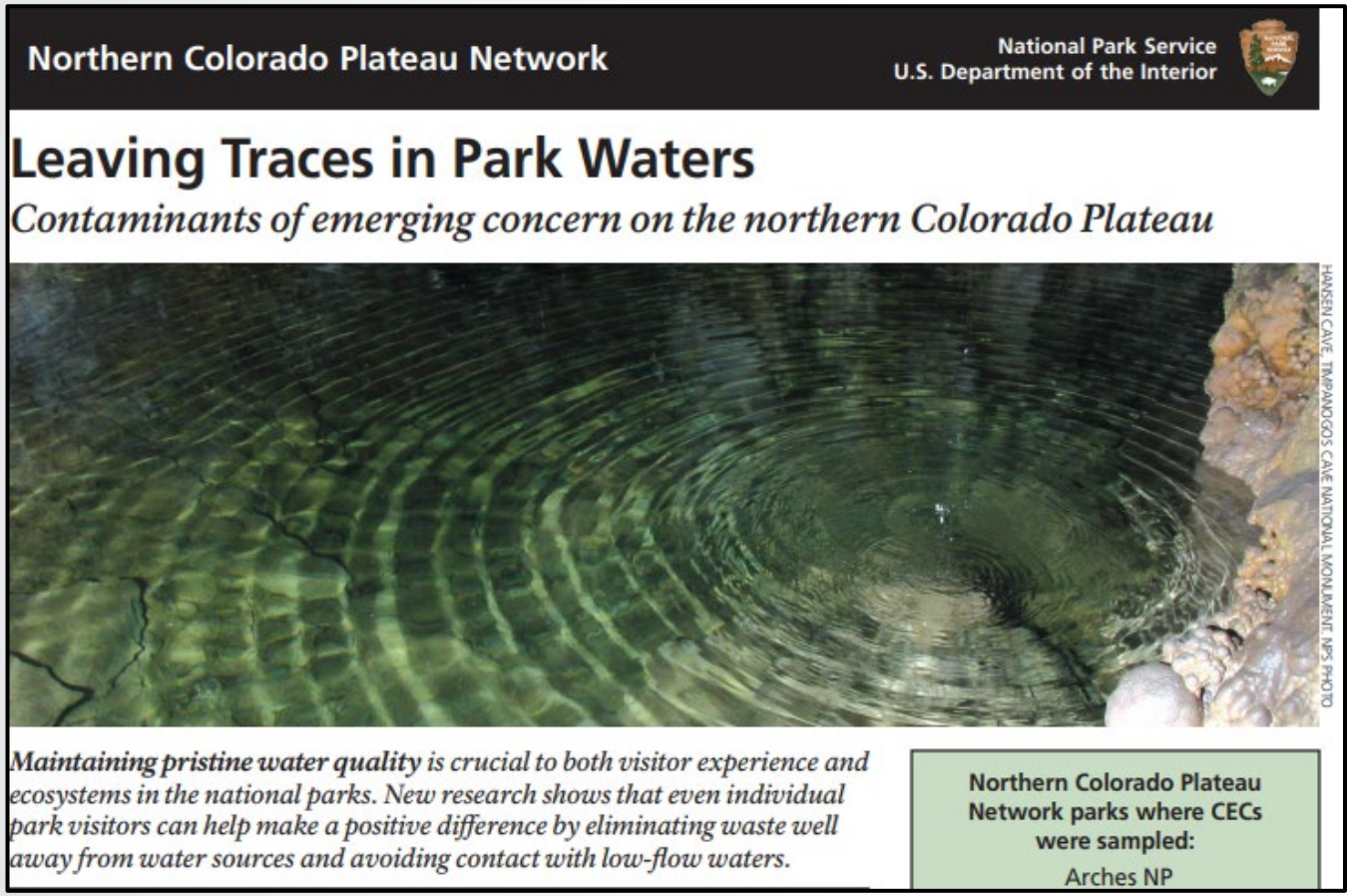
Screened samples using the Attagene trans-Factorial assay

- ToxCast assay platform
- Screens for activation of 24 different nuclear receptors

Three prominent activities were detected

- Estrogen-like (important to reproduction)
- Glucocorticoid-like (important to stress response)
- PPARγ activation (involved in regulation of body fats)

EPA Region 8
Waste-water treatment upgrade, Moab, UT



Moab UT

- 5000 year-round residents
- >1 million visitors per year

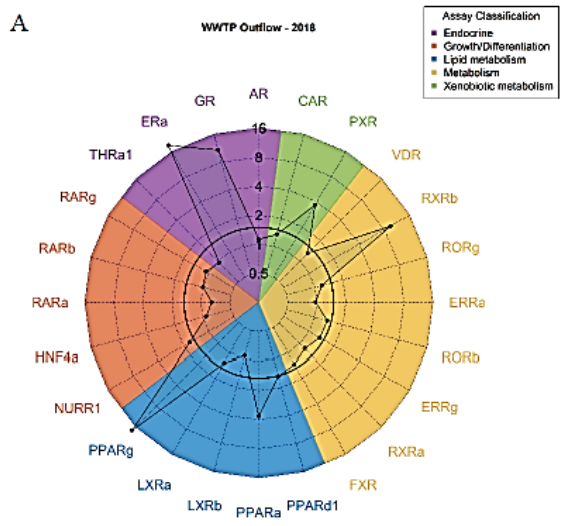
Moab WWTP

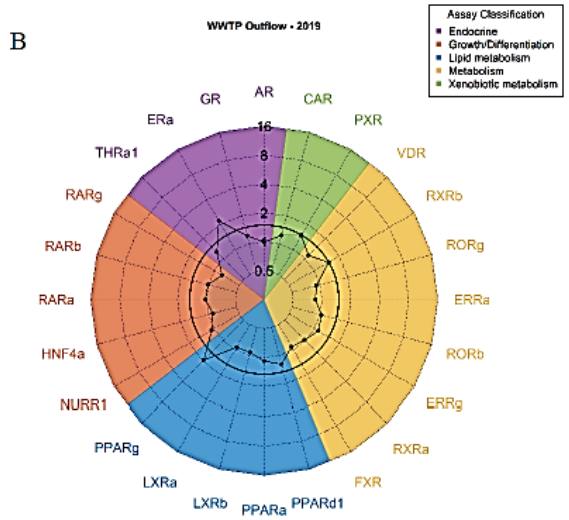
- Originally built in the 1950s
- Upgraded 1996 (trickling filter, chlorine disinfection)
  - Ammonia and nutrient violations with increasing tourism pressure and age
- 2018 new WWTP (activated sludge, UV disinfection)

- Parks and tourism are important to the local economy

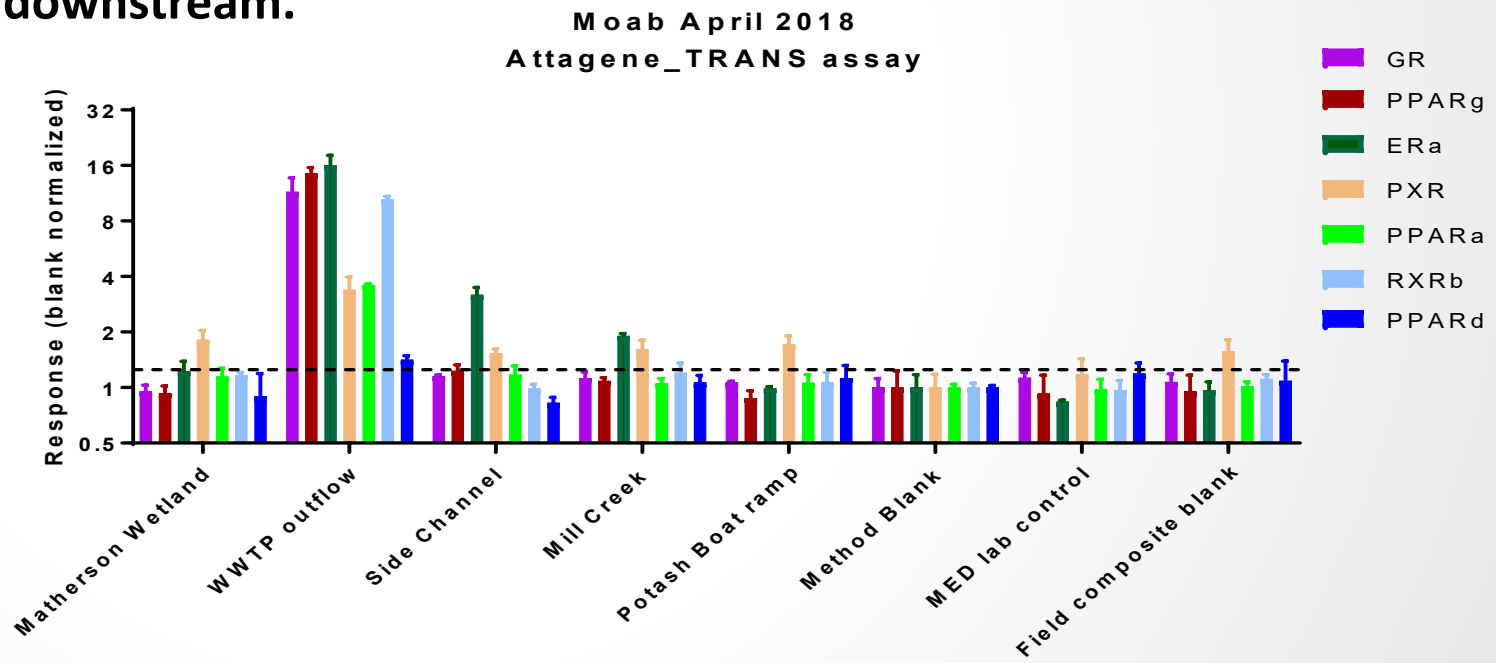
Would the treatment upgrade reduce the loading of bioactive CECs to the Colorado River?

Bioactivity Screening with Attagene



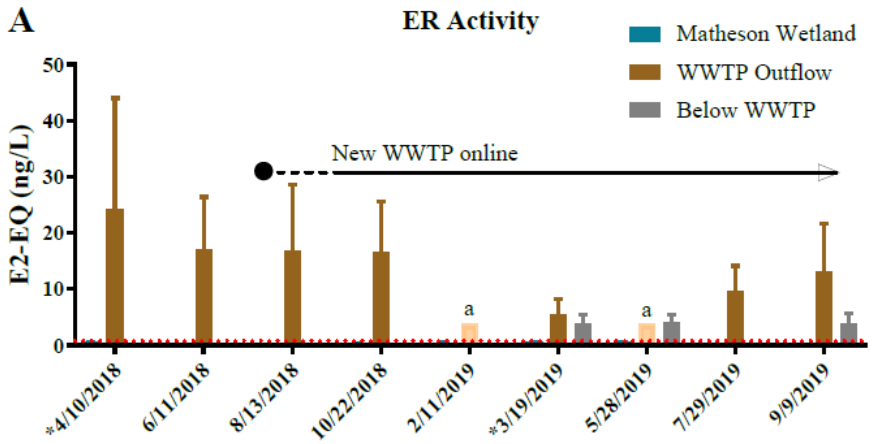


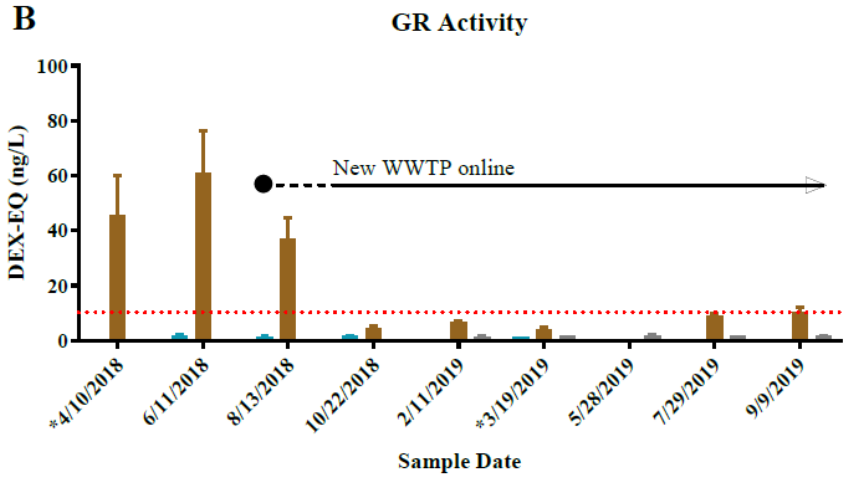
- Six sites, once per year
- Biological activities observed (ER, GR, PPARg) were consistent with pilot years.
- Activity was greatest at the WWTP outflow, diminished rapidly downstream.



- Activity in 2019 was much lower than in 2018

Targeted Bioassays





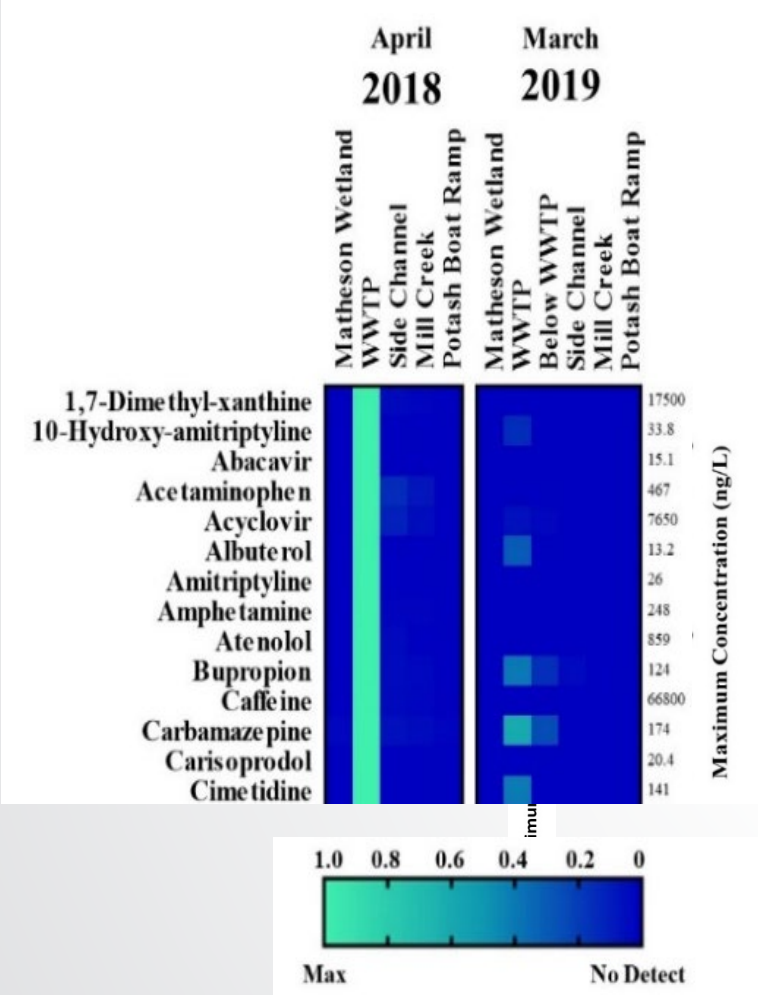
12 sites, bi-monthly, spring to fall over two years

- ER activity declined shortly after WWTP replacement
  - A little lag
  - Possibly trending back up in summer
  - Much lower immediately downstream

- PPARγ activity not detected in targeted assay
  - Slightly less sensitive

- GR activity declined immediately after WWTP replacement
  - Only detected at WWTP outflow

Chemical Monitoring



Only partial heat map shown

- **2018**
  - 62 (out of 131) chemicals detected at outflow
- **2019**
  - 36 (out of 131) chemicals detected at outflow
  - Generally lower concentrations than 2018
- Consistent with bioassay results
- Detections and concentrations quickly decrease away from WWTP
- Guanylurea increased in 2019
  - WWTP transformation product of metformin
  - Metformin below detection limits
  - Recent studies in our lab suggest very low toxicity to aquatic organisms

Good news!

Community investments in upgraded WWTP infrastructure appear to have had a positive effect on the loading of biologically active contaminants to the Colorado River.

- In vitro bioactivities (ER, GR, and PPARγ) reduced and rapidly decline downstream
- Fewer contaminants and lower concentrations
- Caged-fish survival drastically improved

- **Additional contaminant and bioactivity monitoring, if desired, can be focused in close proximity to the WWTP outflow**
  - Some on-going sample collection in 2020-2021 monitor trends in ER- and GR- activity

Conclusions

- Practical applications of NAMs and NAMs data in chemical safety assessment is not limited to prospective assessments of individual chemicals.
- NAMs data can help inform risk-based screening based on environmental monitoring, particularly where traditional toxicity benchmarks are lacking.
- NAMs can be applied to evaluate complex mixtures with both known and unknown compositions.
- NAMs applications can aid in environmental decision-making

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Great Lakes Restoration Initiative, Action Plan II. https://www.glri.us/sites/default/files/glri-action-plan-2.pdf