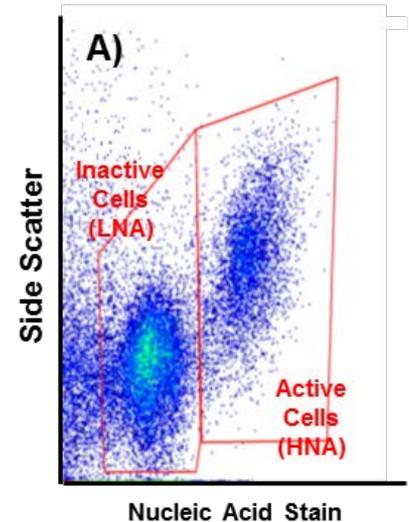
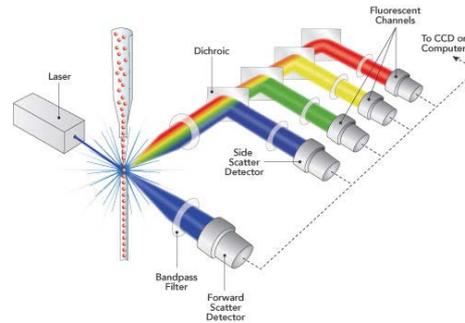


Flow Cytometry: A novel, rapid, screening and research tool for methylmercury production activity in aquatic ecosystems?



Collin Eagles-Smith¹, James Willacker¹, David Krabbenhoft²

¹US Geological Survey, Corvallis, OR

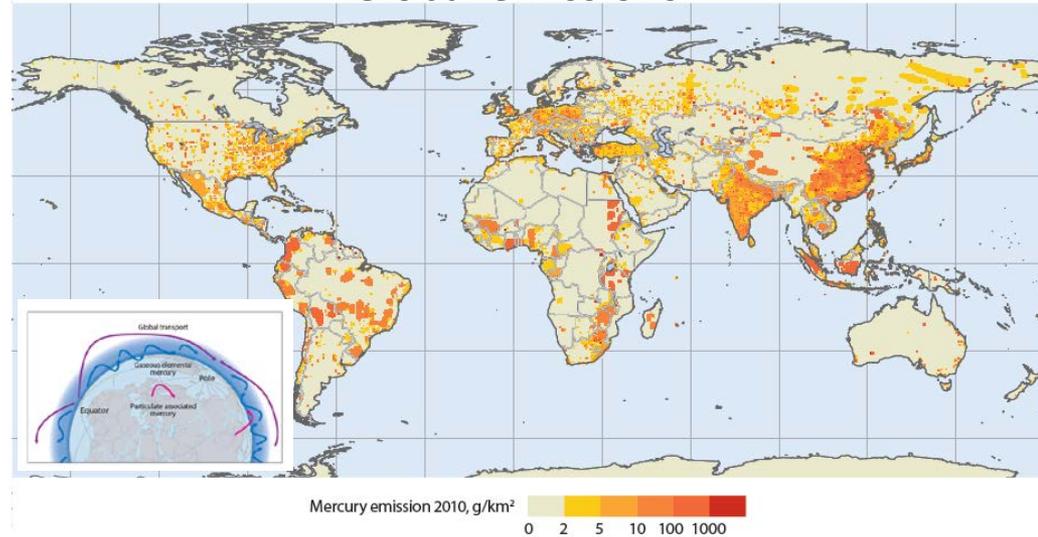
²US Geological Survey, Middleton, WI

Mercury

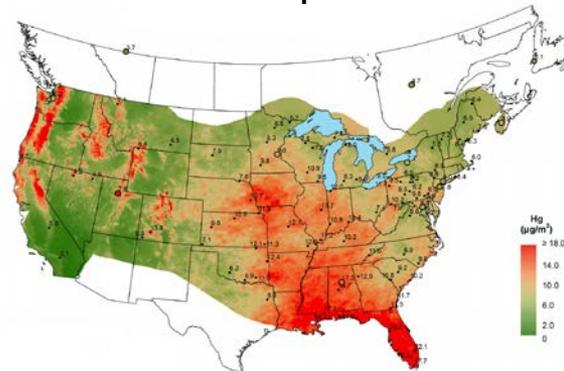
A contaminant of global consequence

- Responsible for 80% of all fish consumption advisories
- Global burden of disease estimate 1.2-2.4 million years living with disability
 - Higher than hepatitis or Parkinson's
- Only element with its own international treaty

Global emissions



National deposition

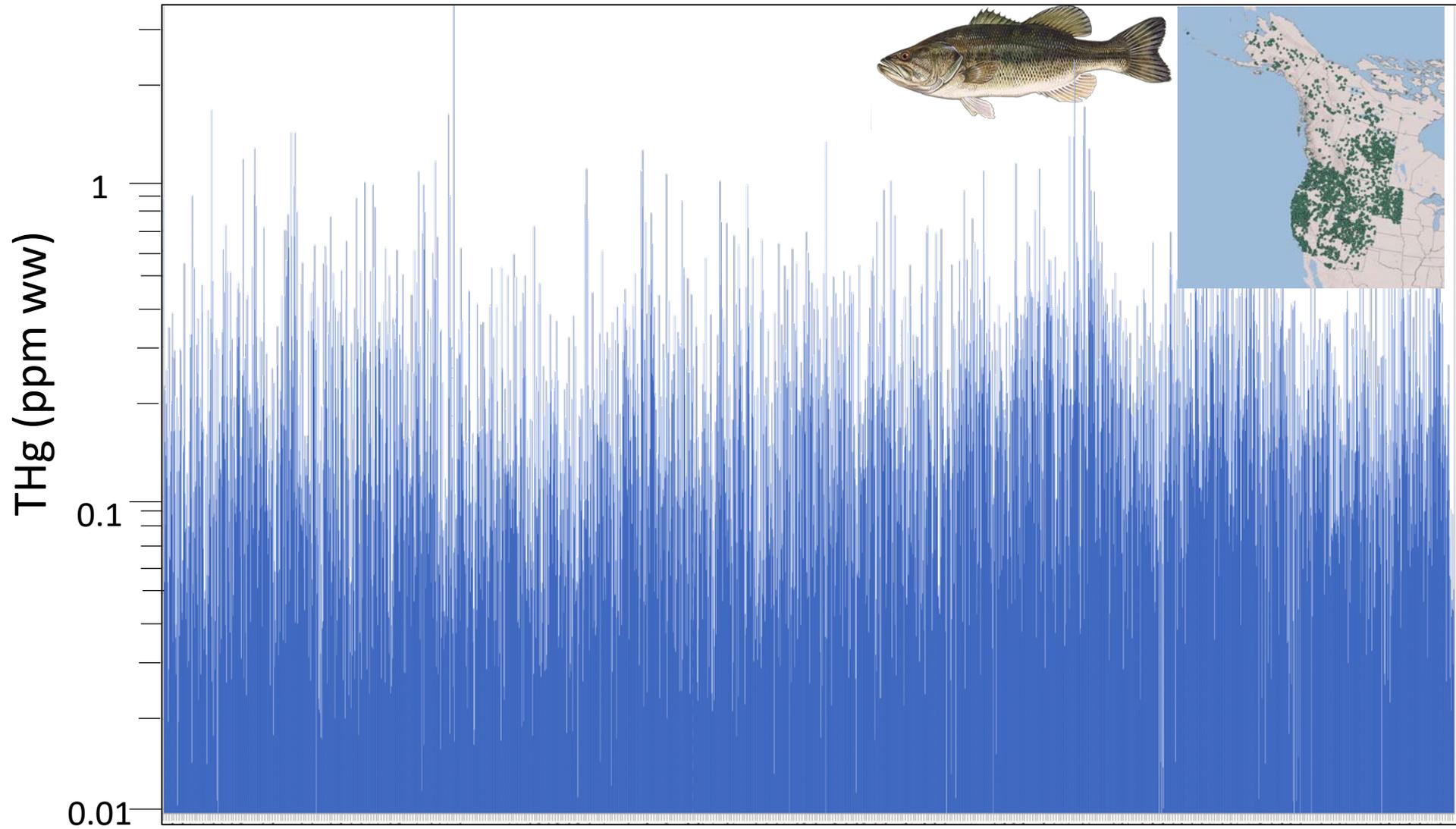


Consumption advisories

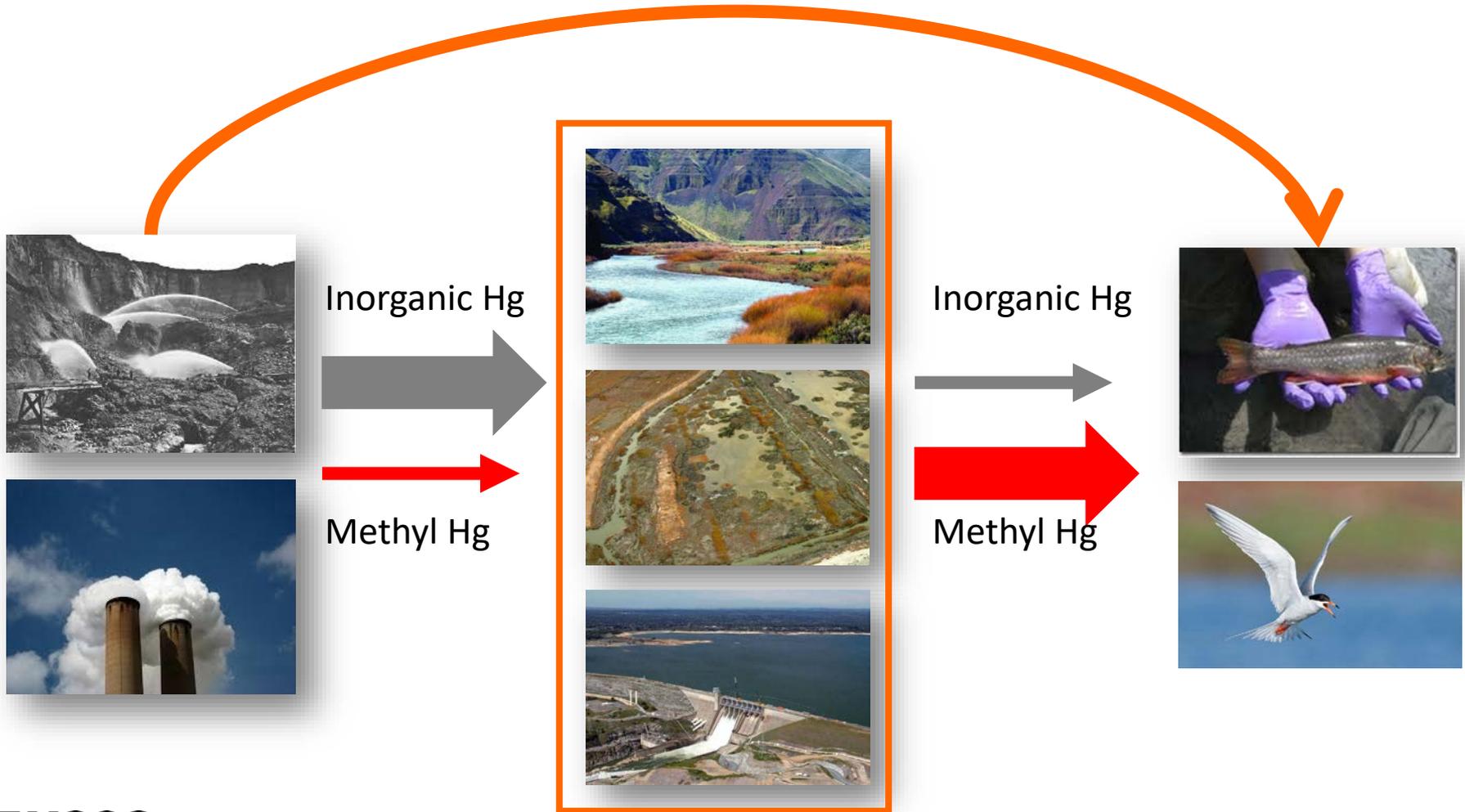


Variability of mercury in ecosystems

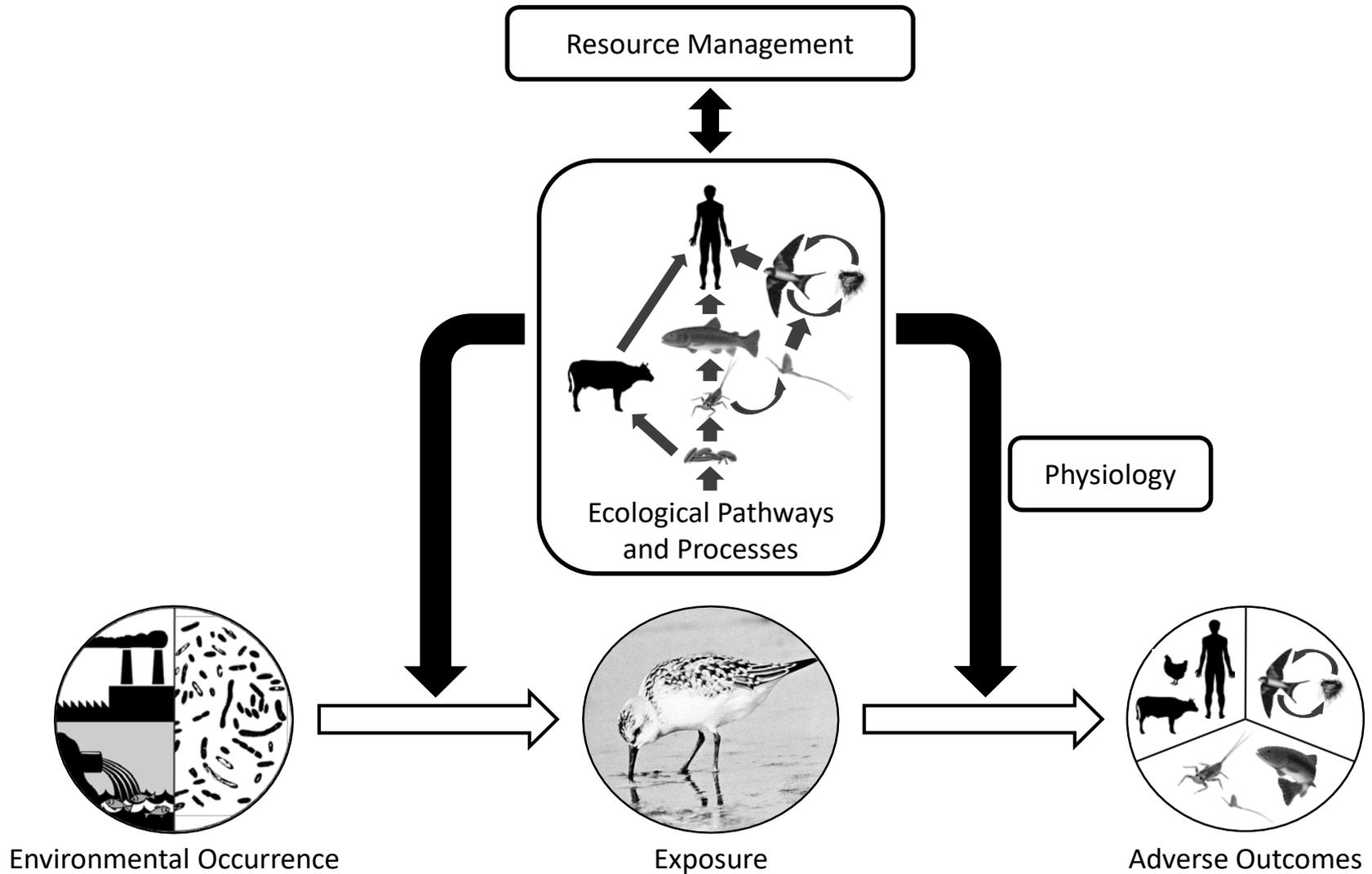
N=4262 locations



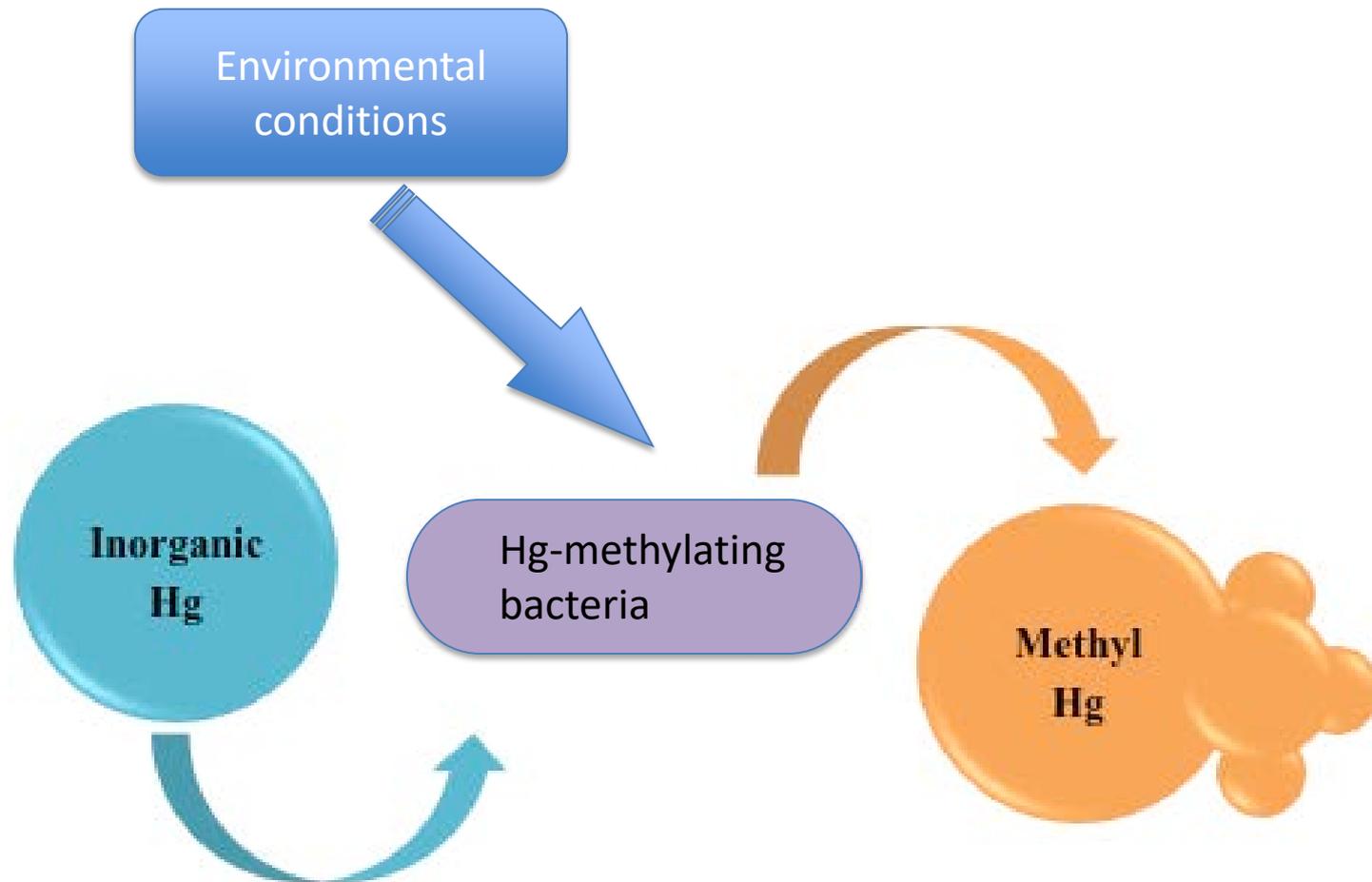
Pathways of inorganic Hg to bioaccumulation



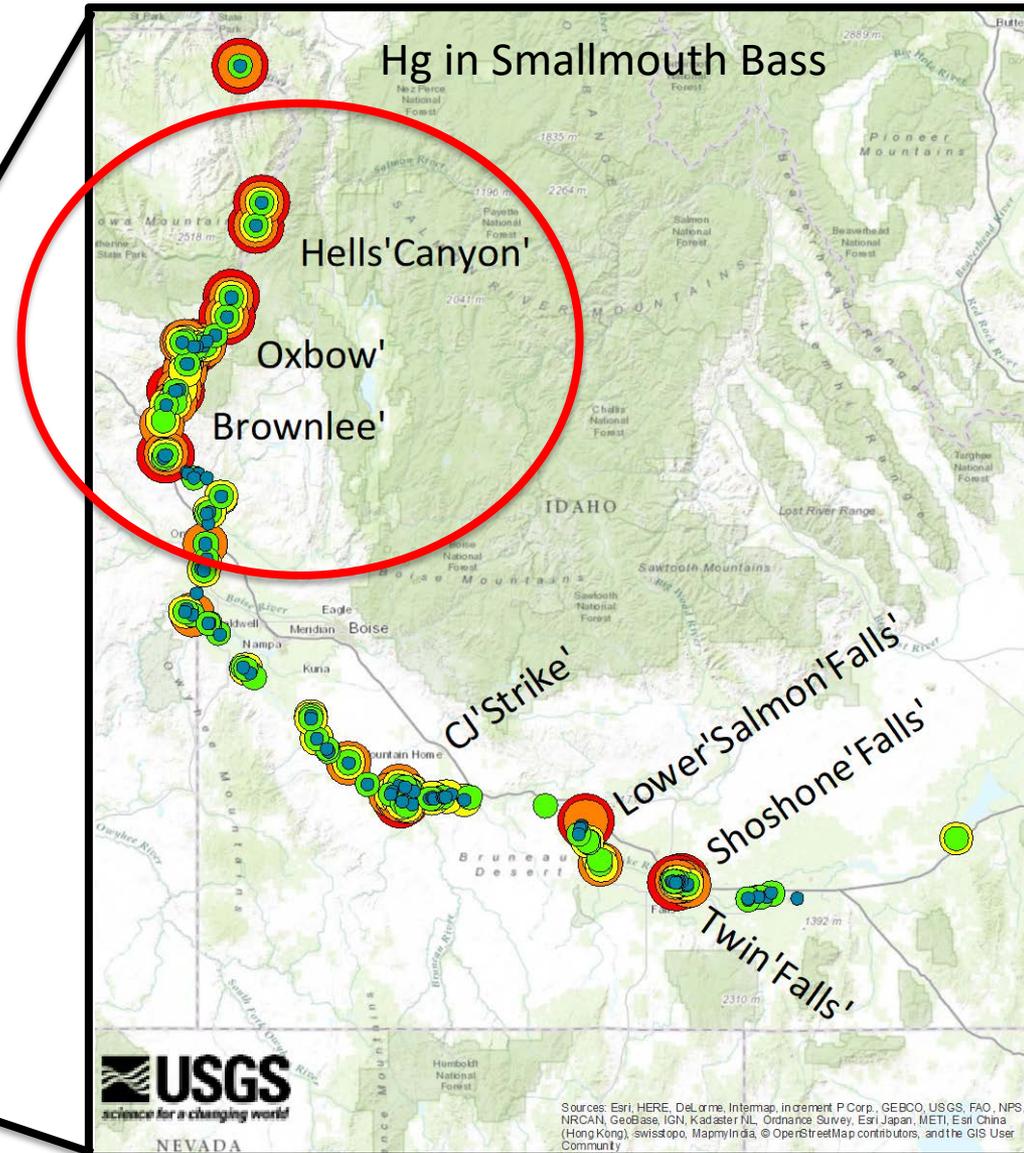
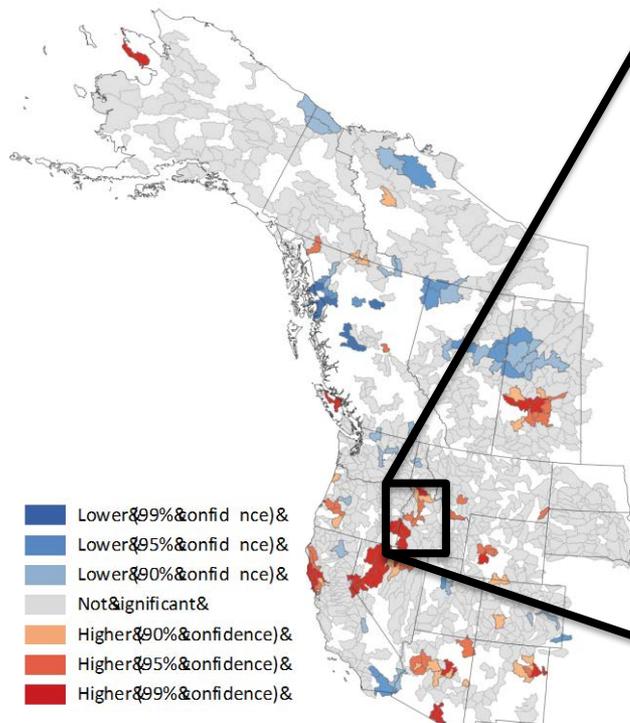
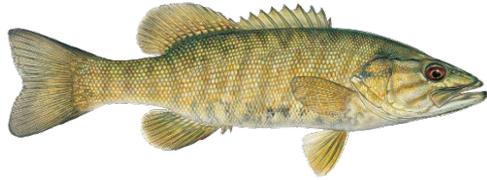
Ecological Pathways Conceptual Model

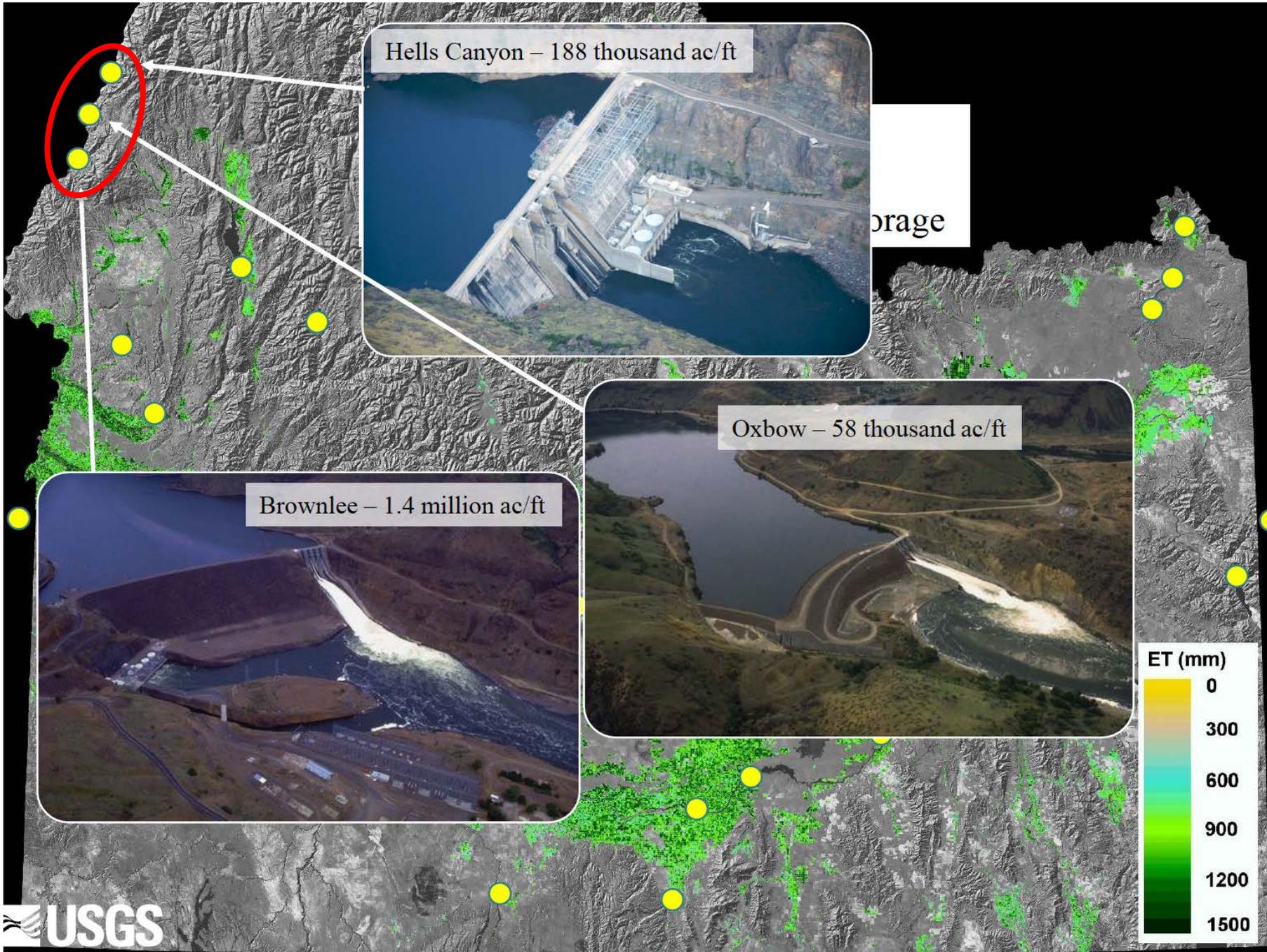


Methylmercury production – a microbial process

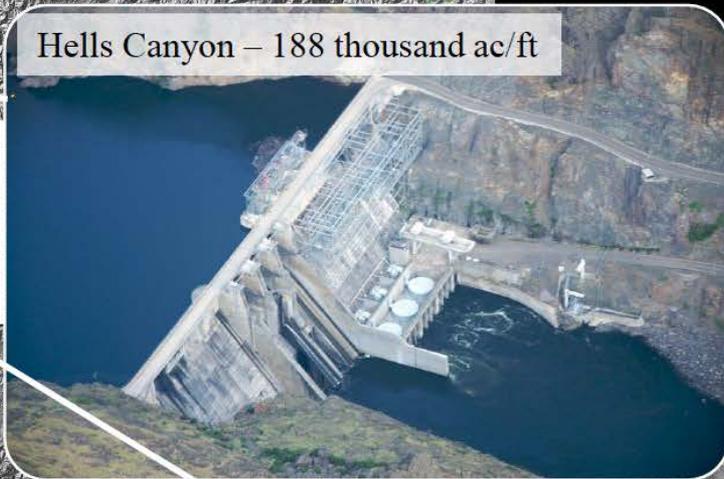


Hg in the Great Basin and Snake River Drainage



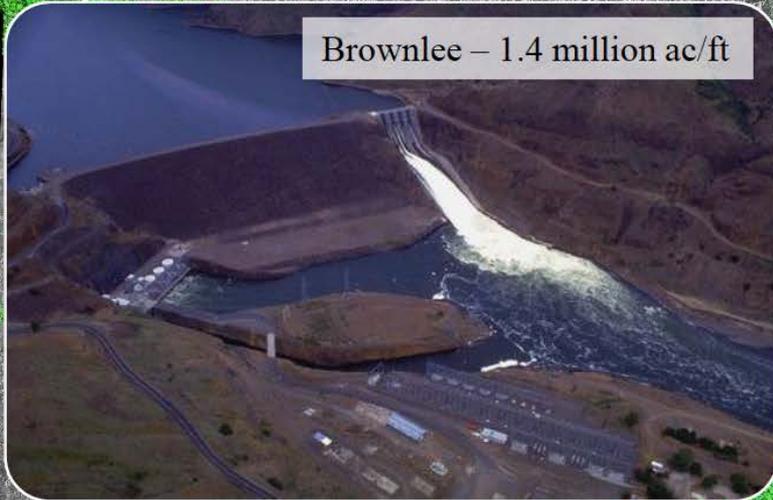


Hells Canyon – 188 thousand ac/ft



Storage

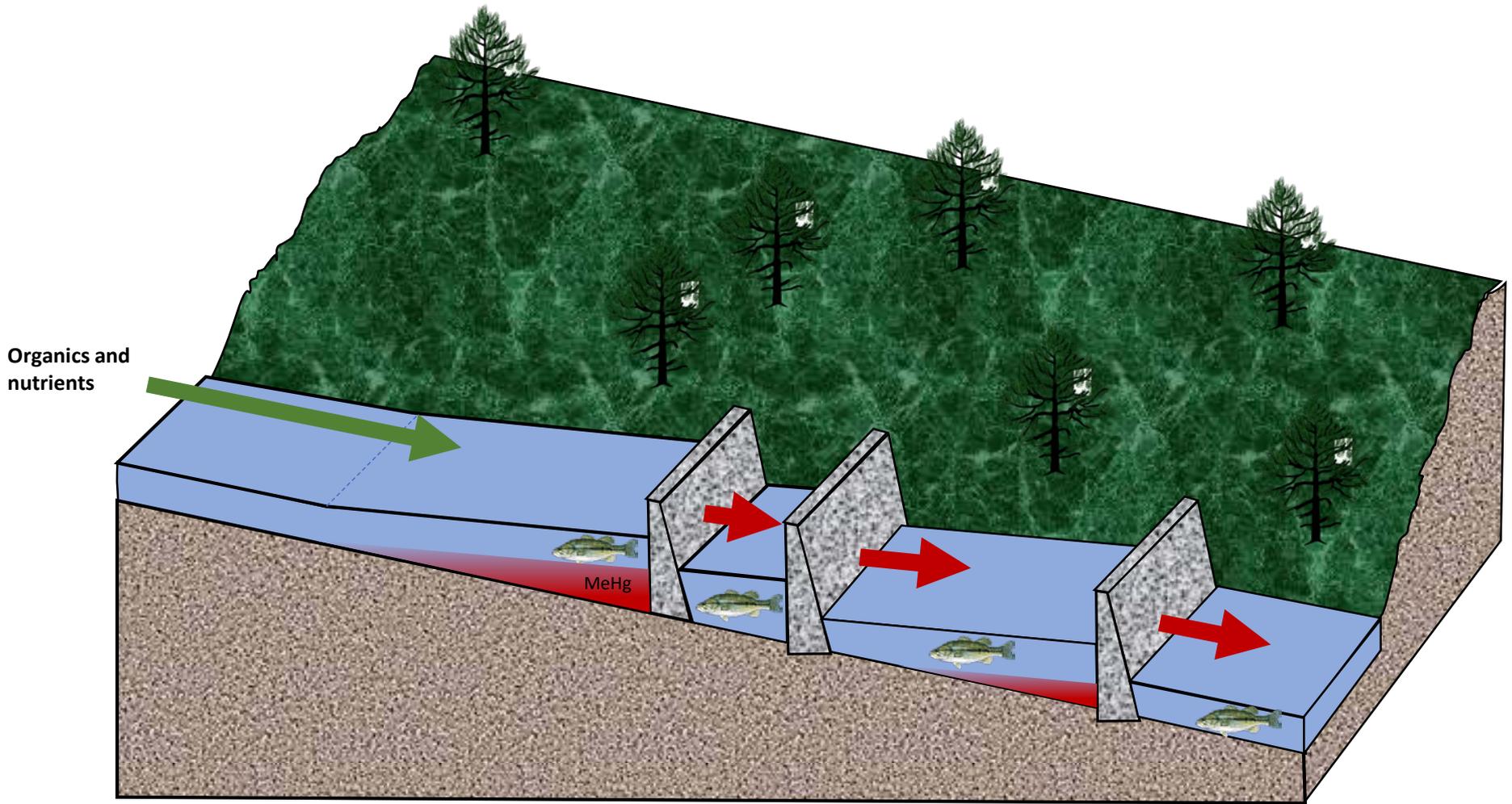
Brownlee – 1.4 million ac/ft



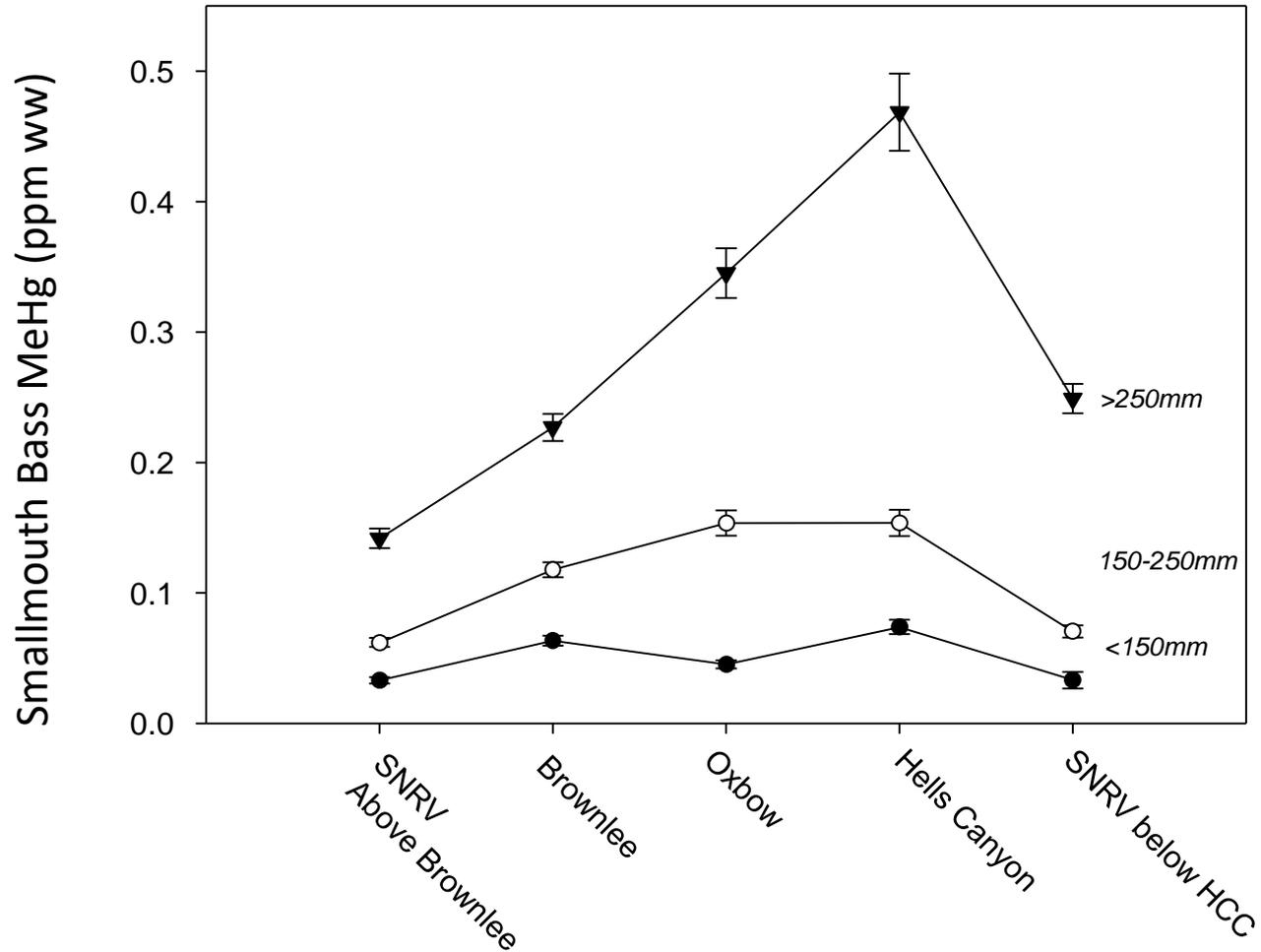
Oxbow – 58 thousand ac/ft



Mercury bioaccumulation through a three-reservoir complex



Fish mercury concentrations increase through the complex



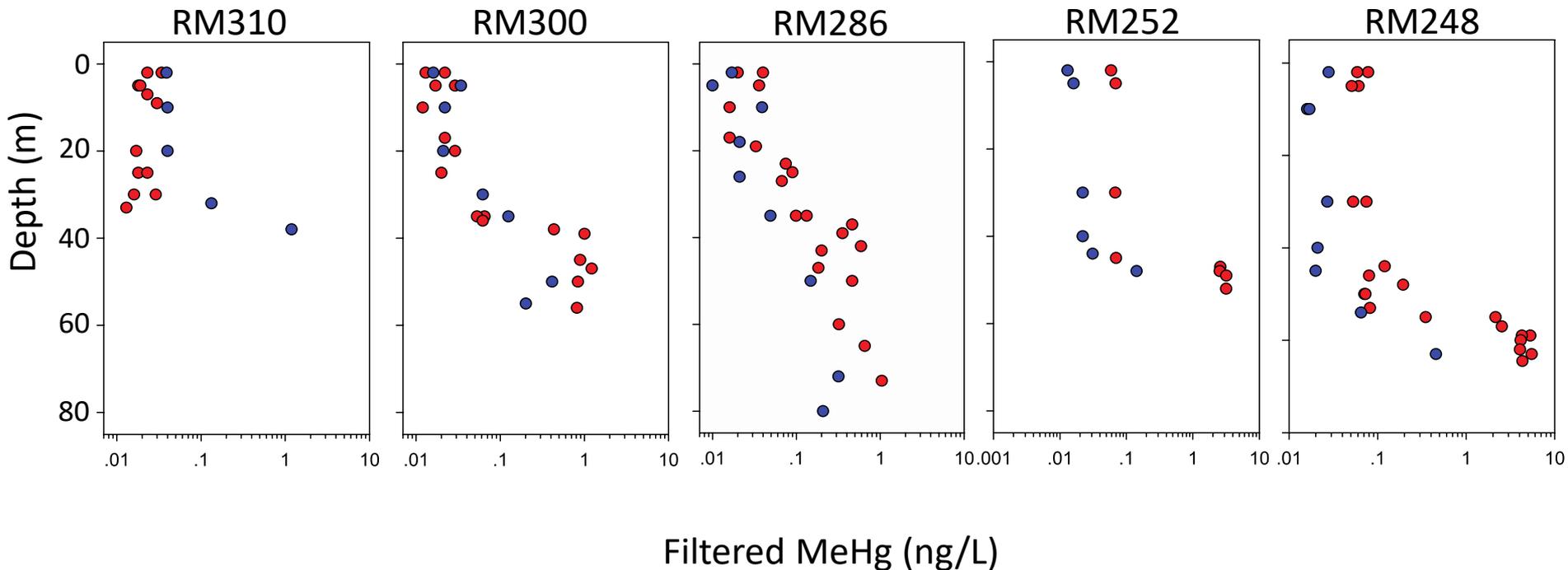
Aqueous MeHg concentrations through the complex

● Spring ● Fall

Up-reservoir

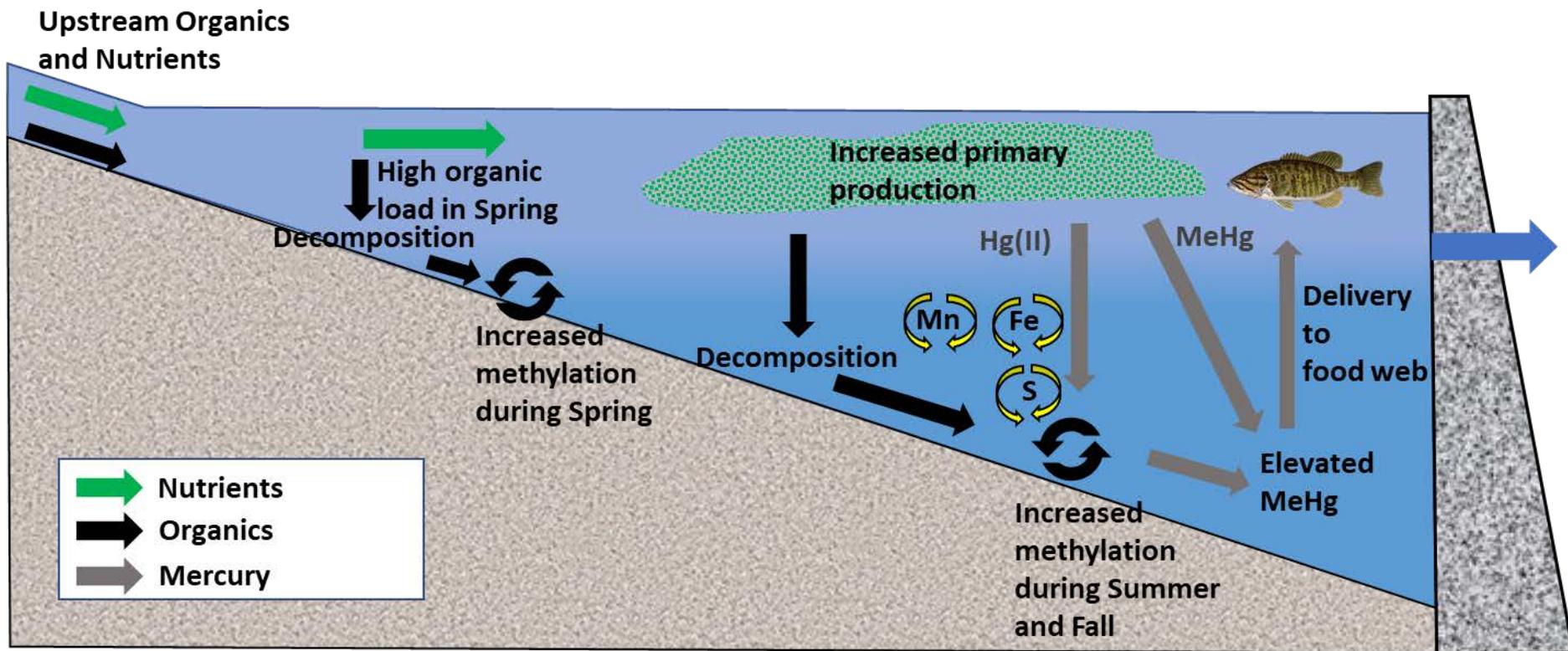


Down-reservoir

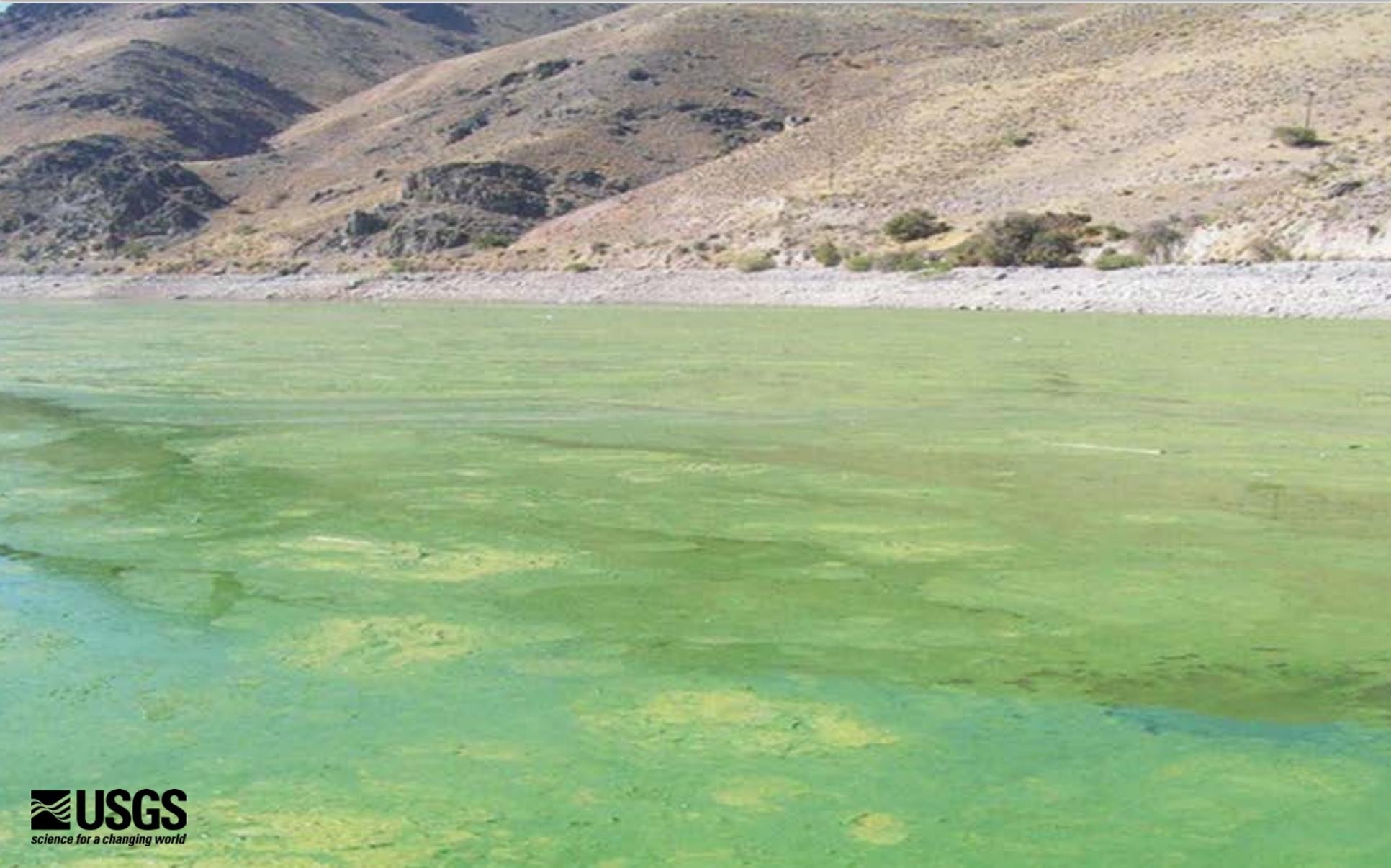


Variation greater within site (by depth) than across sites

Mercury cycling model for the complex

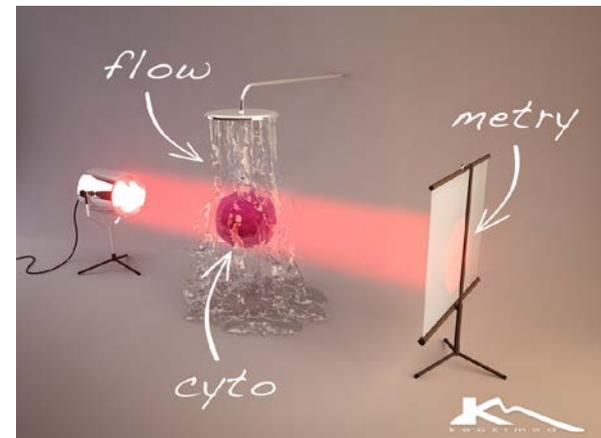


Primary productivity as a driver of MeHg production



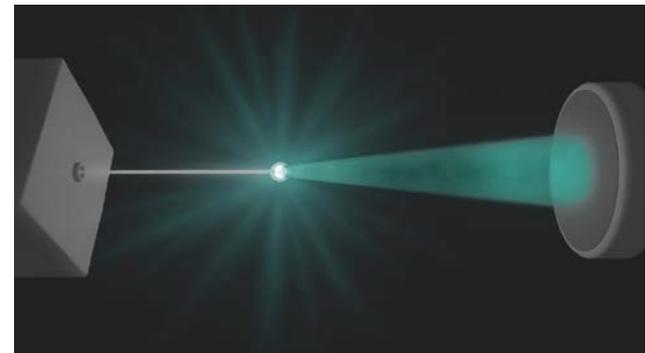
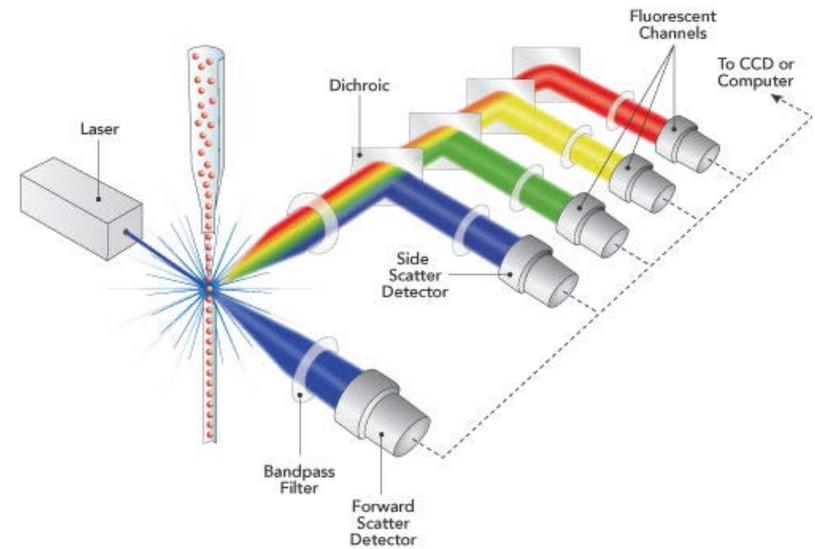
Flow Cytometry

- Instrumented method for quantifying size and type of cells in a fluid
- Originally developed for medical uses (blood cells, etc.)
- Adopted by oceanographers and limnologists for algal cells
- Employed by microbiologists



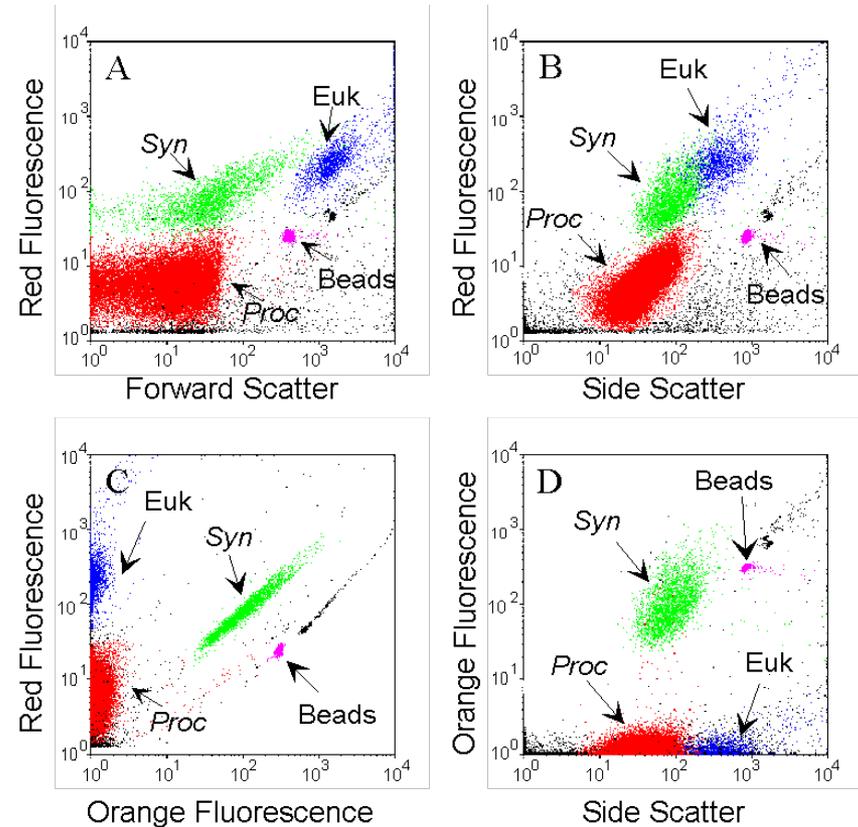
Flow Cytometry

- Forward scatter estimates cell size
- Side scatter and fluorescence indicative of cell structure and granularity
- Measures light absorption and excitation



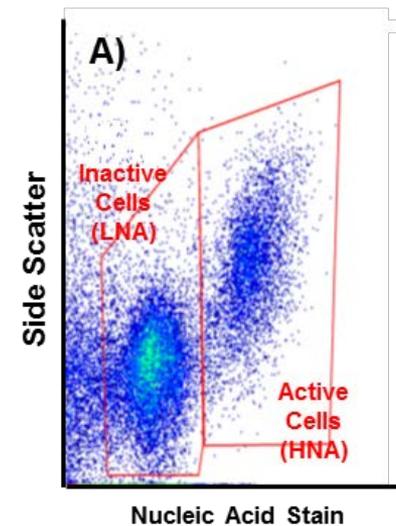
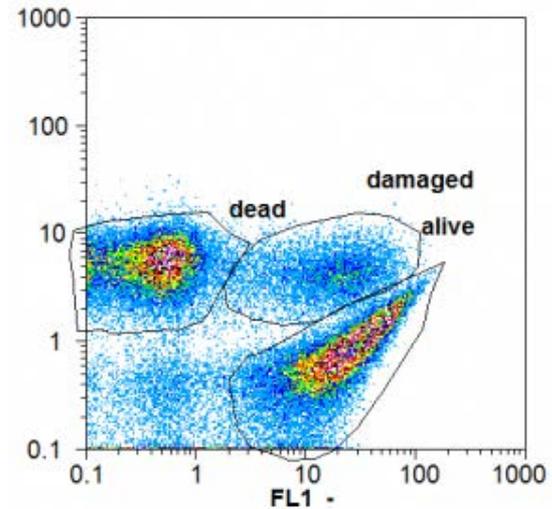
Flow Cytometry

- Biplots of scatter and fluorescence allow for differentiation of different cell types



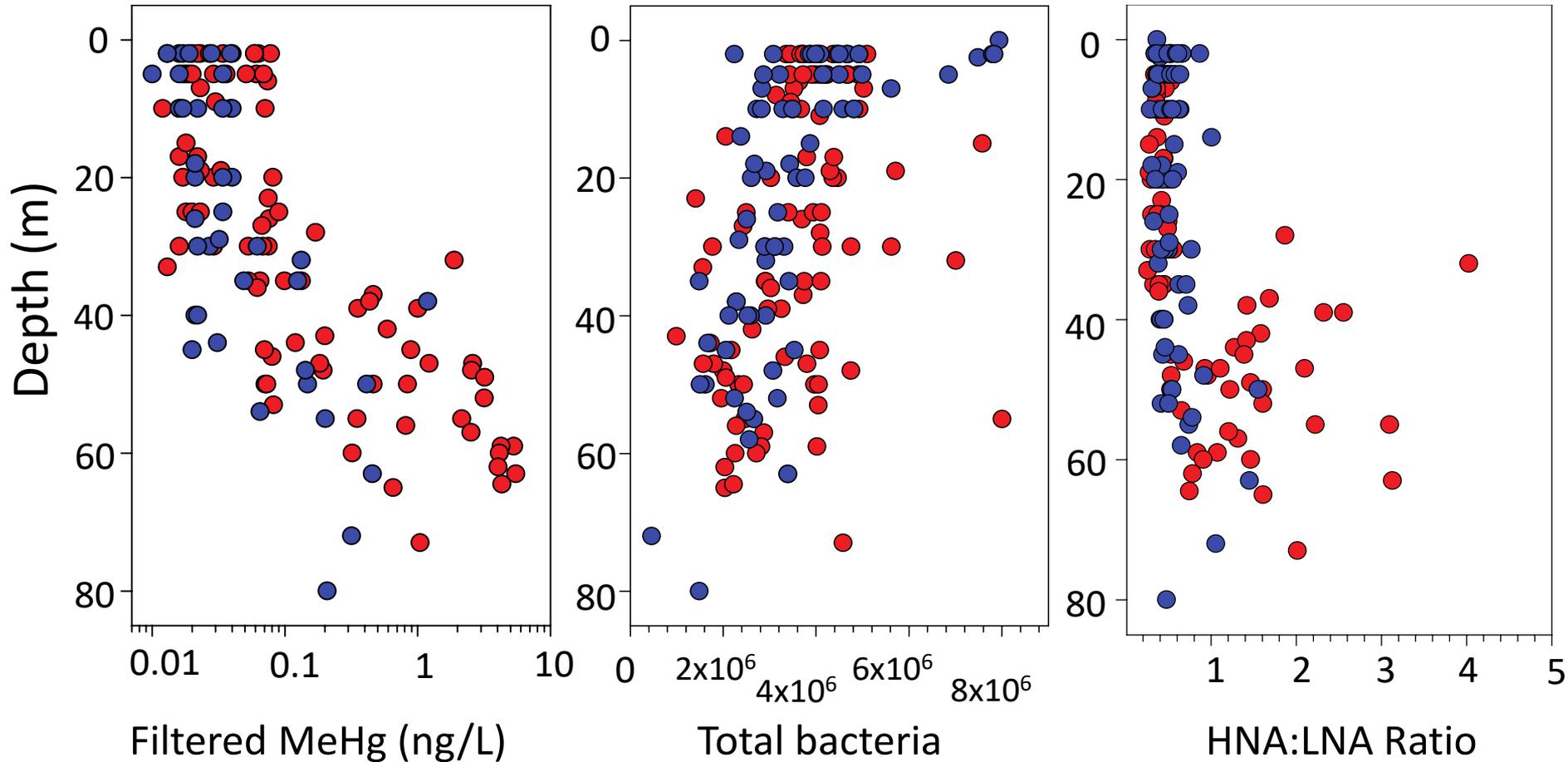
Flow Cytometry

- Bacteria determined by size and cell stains
- Coarse differentiation
- Nucleic acid stains differentiate two primary types
- HNA = High nucleic acid content (active cells)
- LNA = Low nucleic acid content (inactive cells)

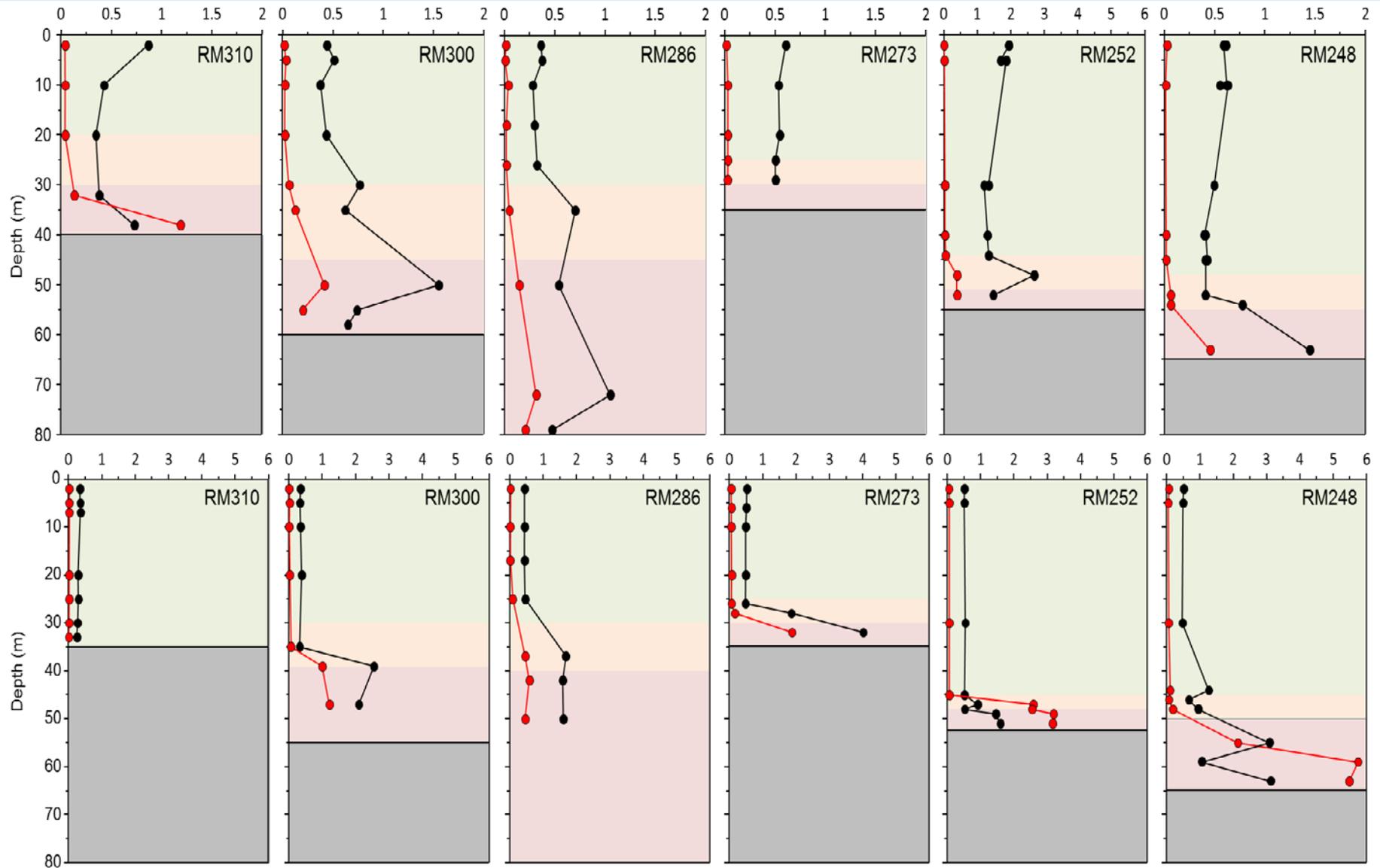


MeHg concentrations and microbial activity through the water column

● Spring ● Fall



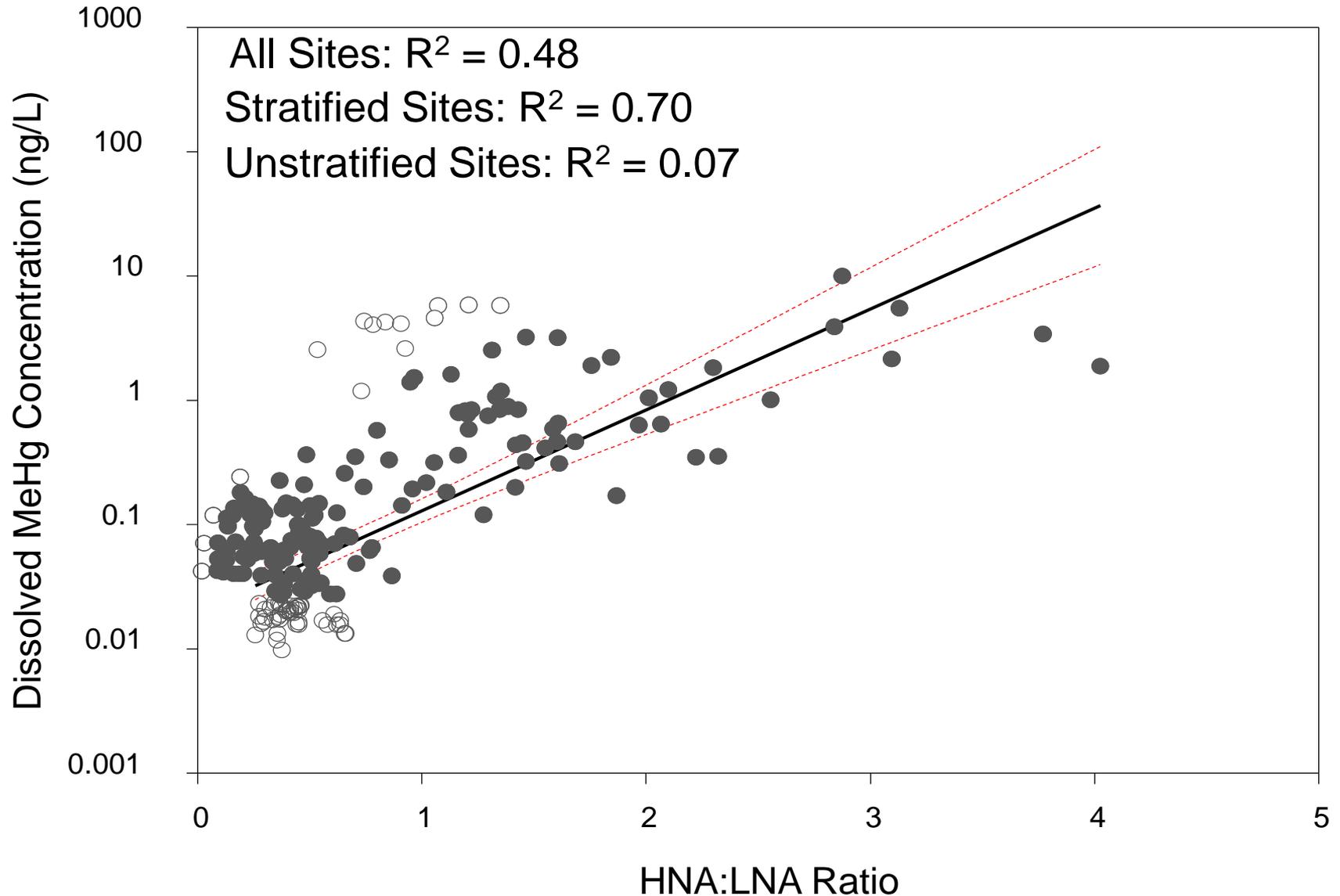
HNA:LNA ratio & Dissolved MeHg (ng/L)



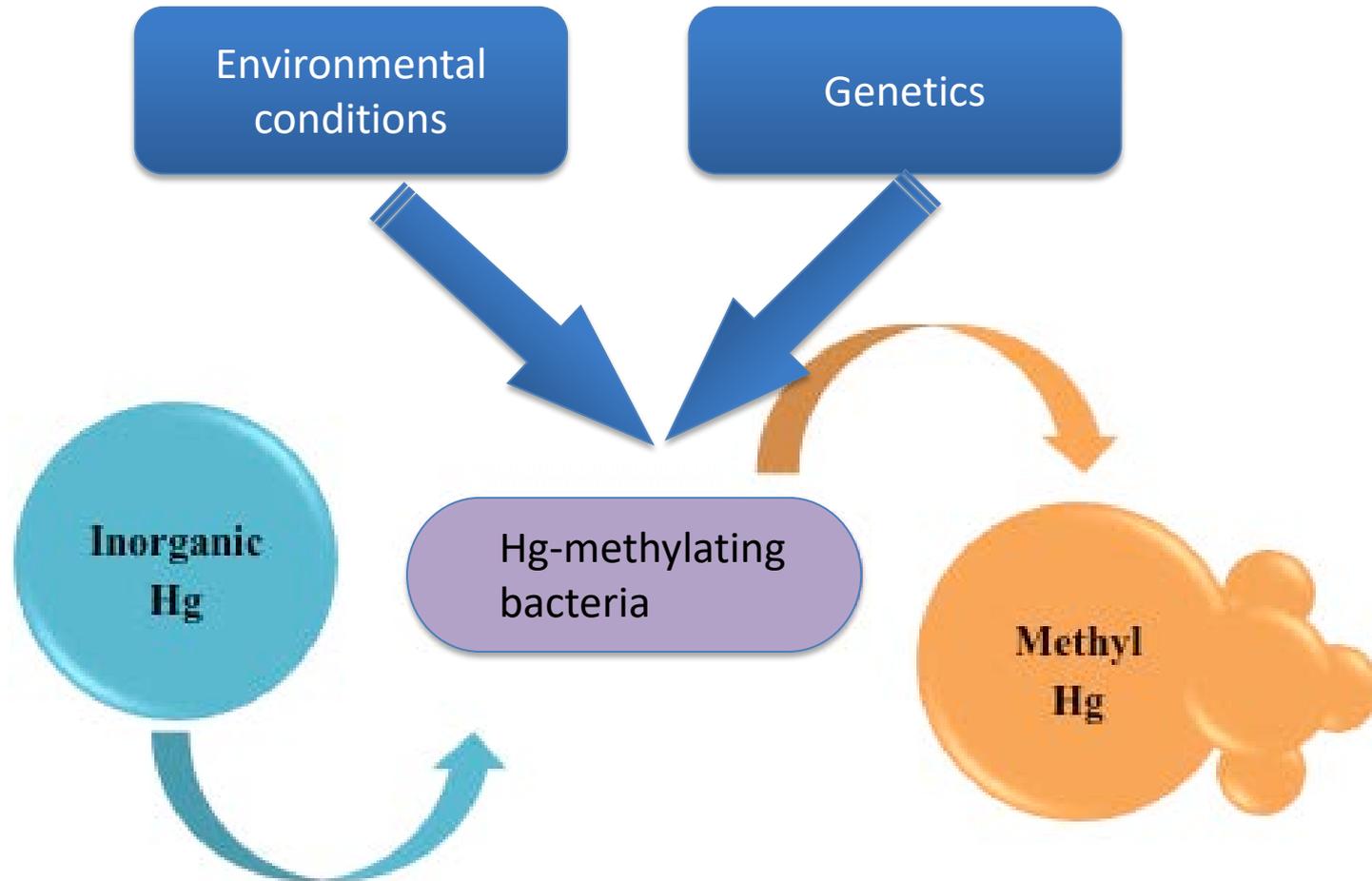
June 2015

October 2015

HNA:LNA ratio correlated with aqueous MeHg

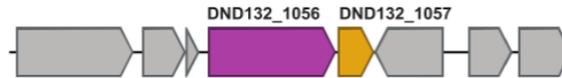


Methylmercury production – a microbial process



Discovery of hgcAB gene cluster (2013)

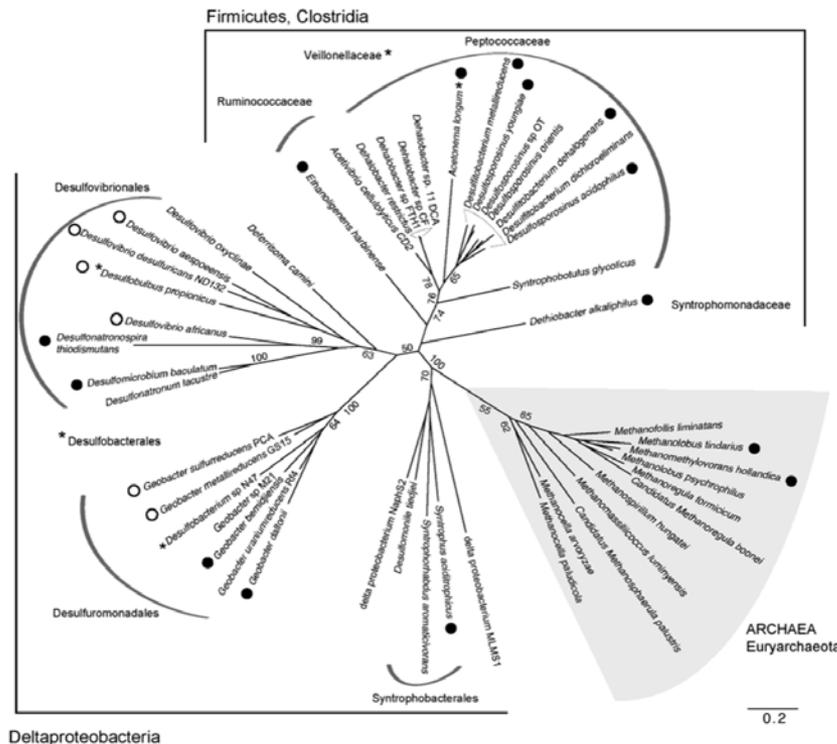
D. desulfuricans ND132



G. sulfurreducens PCA

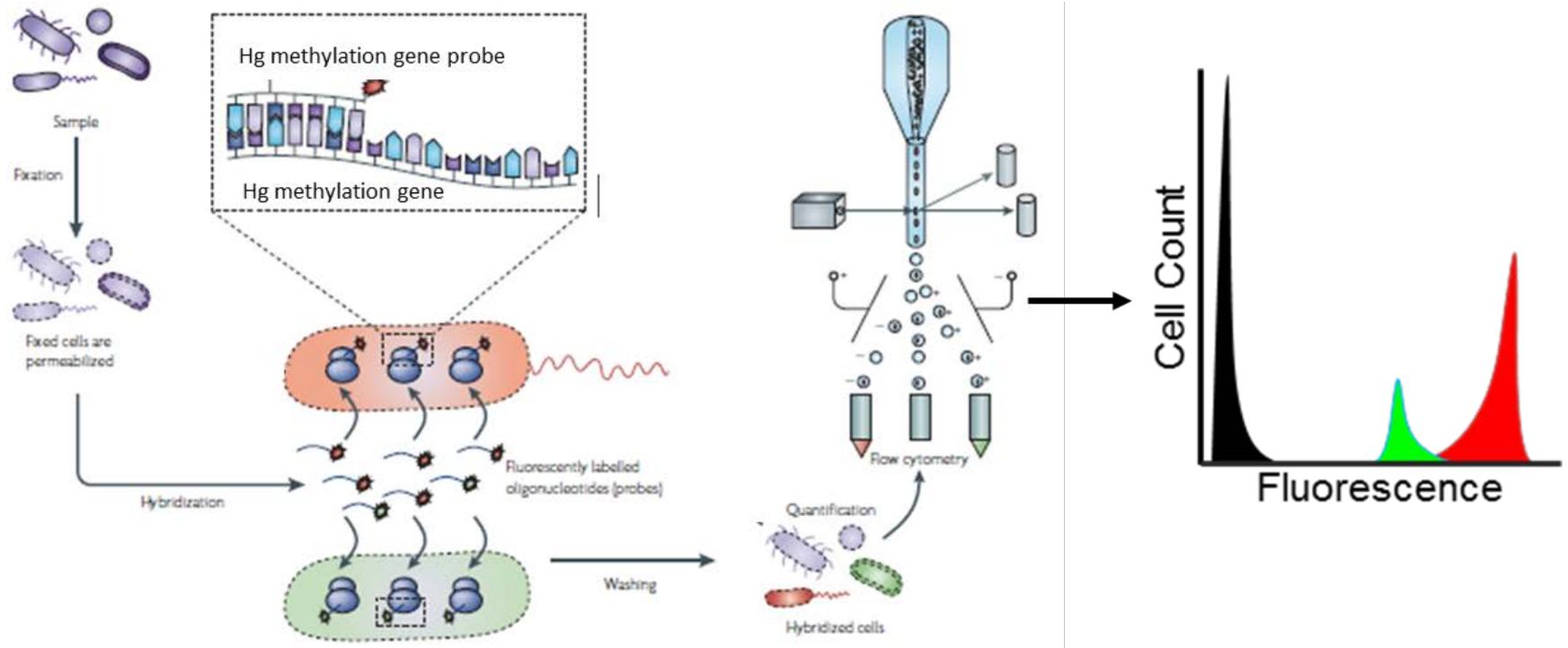


Modified from Parks et al, 2013

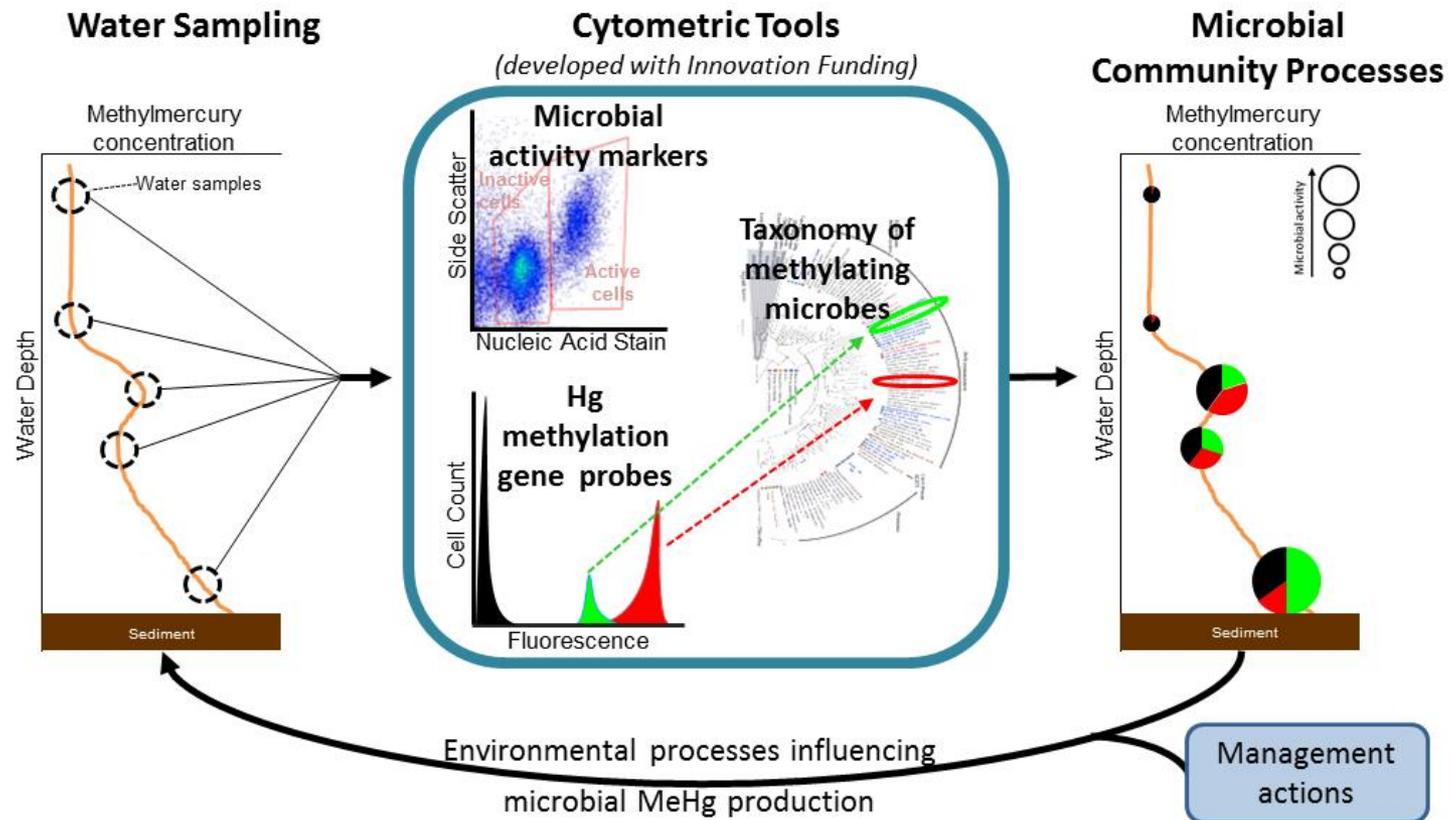


- Insights into molecular mechanisms
- Probing of existing cultures and genetic databases
- Environmental marker

Potential applications of flow cytometry in MeHg monitoring and research

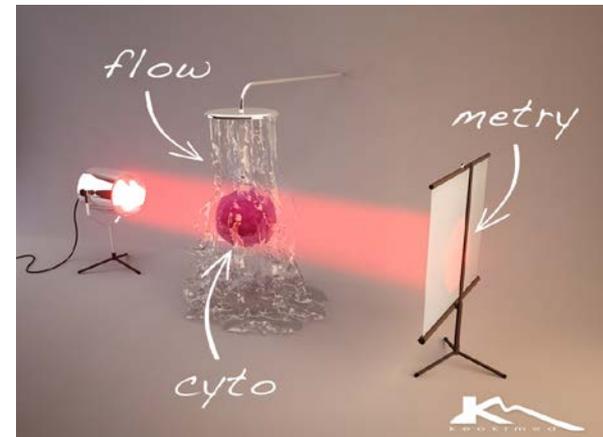


Incorporation of cytometric tools



Summary

- Flow cytometry provides a rapid, low-cost estimate of microbial community composition and activity
- Ratio of “active” to “inactive” bacteria profile closely matched aqueous MeHg profile
- Application to other systems?
- Development of molecular/genetic markers?



Acknowledgments

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Idaho Dept Env Quality
US Geological Survey

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Randolph, Chelsea Wisotzky, Kali Doten, Erica Johnson, Caitlin
Rumrill, Alex Woolen

