

Building a centralized database of watermilfoil strain geographic distribution and herbicide response for use in management decision-making



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Introduction, Background, & Problem

- Genetic variation can influence management outcomes
- Our primary goal is to use genetic information to predict herbicide response, and thus improve management outcomes.
- Our research includes both short and long term approaches to relate control efficacy to genetic variation.



Objectives

1. Build a bioinformatics pipeline to integrate new genetic strain data into a central database capable of real-time updates (similar to COVID variant tracking).
2. Explore several options to interface the centralized database with a stakeholder-facing dashboard that can be used to inform herbicide management options and permitting.
3. Add to our existing database of strain herbicide responses by characterizing several Minnesota strains.
4. Use simulations to identify the best methods to estimate in-field response to herbicides for different strains, which will in turn help identify strains for herbicide resistance testing.

Objective 1: Build a bioinformatics pipeline to integrate new genetic strain data into a central database capable of real-time updates.

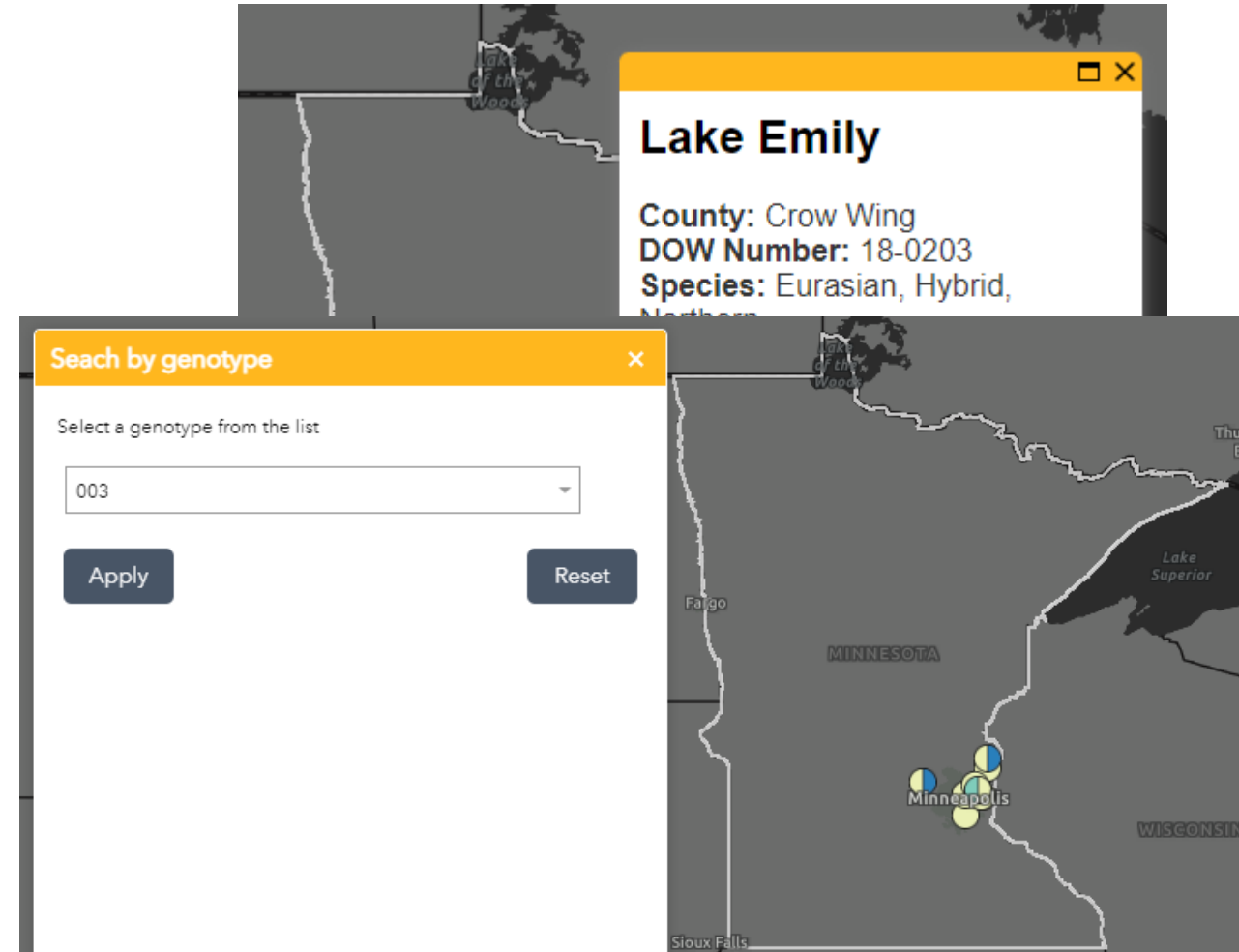
- Iowa – 4 lakes (17 samples)
- Illinois – 5 lakes (81 samples)
- Indiana – 18 lakes (180 samples)
- Michigan – 39 lakes (400 samples)*
- Minnesota – 91 lakes (5,000+ samples)
- Nebraska – 10 lakes (185 samples)
- New York – 14 lakes (215 samples)
- Ohio – 26 lakes (453 samples)
- Pennsylvania – 6 lakes (102 samples)
- Wisconsin – 97 lakes (1,097 samples)

Objective 1: Build a bioinformatics pipeline to integrate new genetic strain data into a central database capable of real-time updates.

- Developed a naming scheme for all strains
- Current Workflow
 - Sample receipt and processing - ~May – November
 - Scoring and analysis - ~December
- Future Directions
 - New DNA fingerprinting method
 - Automated analysis of fingerprints, and update of database

Objective 2: Interface database with a stakeholder-facing dashboard that can be used to inform herbicide management options and permitting.

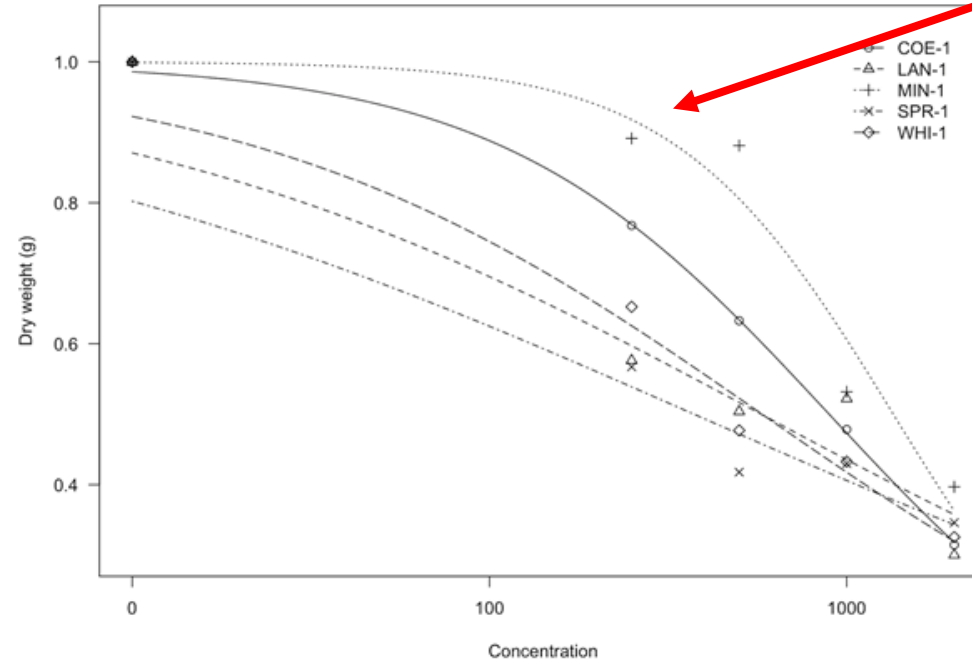
- A prototype database of genetic information, with qualitative flags for herbicide response
 - **MAISRC milfoil app:**
<https://umn.maps.arcgis.com/apps/webappviewer/index.html?id=e59de1333bf64e02997b112881361e4c>
- Have consolidated all previous microsatellite data with more recent data into a centralized database
- No herbicide flags, yet, but can identify some resistant and susceptible strains, and I relay this information to stakeholders



Objective 3: Add to our existing database of strain herbicide responses by characterizing several Minnesota strains.

- Identified a 2,4-D resistant genotype

- Submitted a manuscript to *Evolutionary Applications*



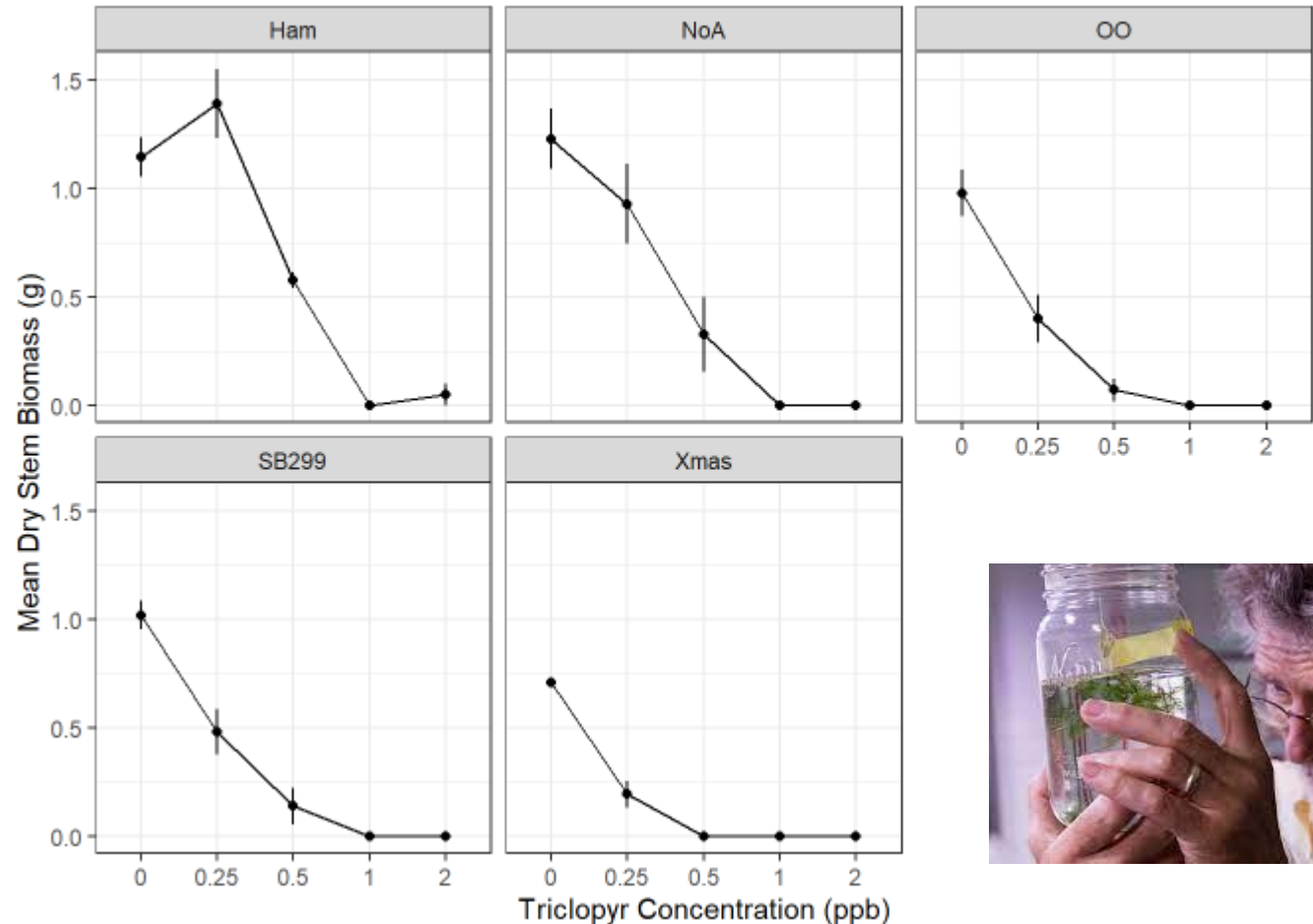
**North Arm strain
(Lake Minnetonka)**



Objective 3: Add to our existing database of strain herbicide responses by characterizing several Minnesota strains.

Triclopyr

- North Arm strain does not look like it is as resistant to triclopyr as 2,4-D
- Ham Lake strain appears resistant to lower concentrations of triclopyr
- 2,4-D and triclopyr response not necessarily the same

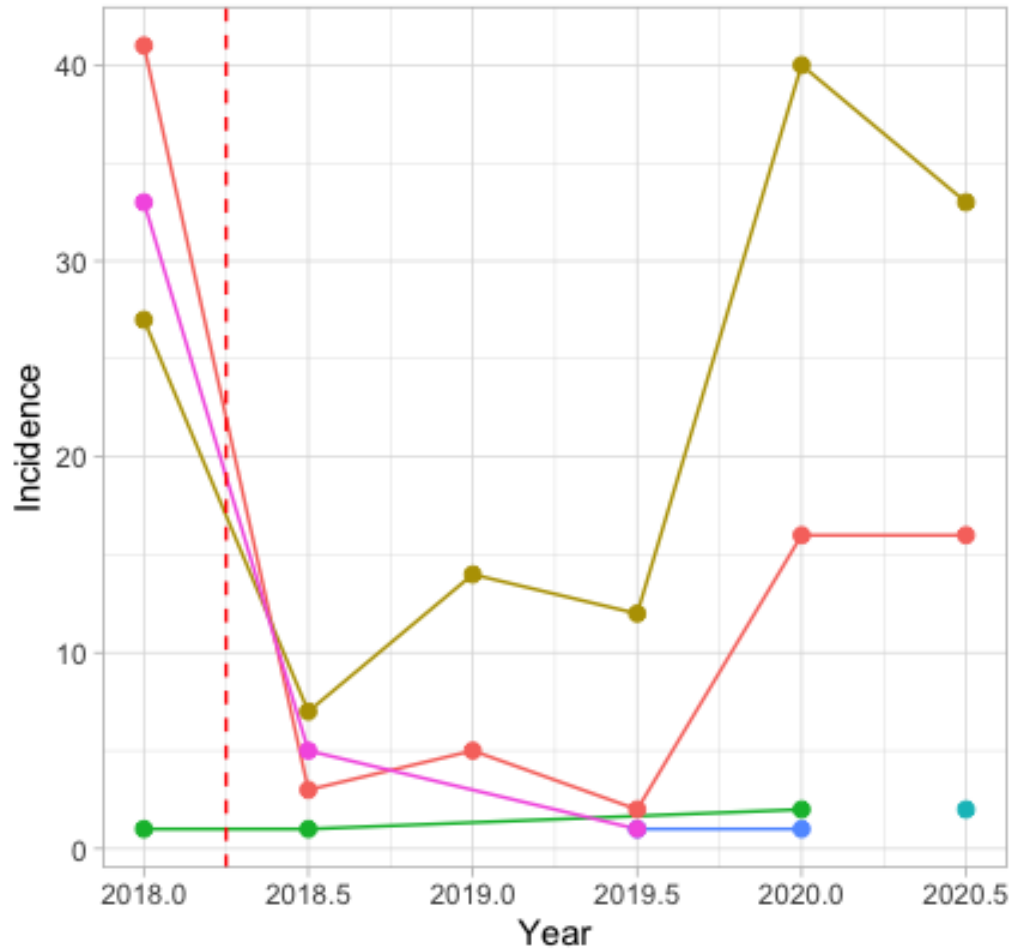


Objective 4: Simulations to estimate in-field response to herbicides for different strains.

- How do we prioritize strains for herbicide characterization?
 - Common/widespread
 - Credible manager account of resistance
 - **Quantitative field evidence – disproportionately low efficacy relative to other strains present**
- Typical aquatic vegetation surveys may not provide adequate statistical power
- Simulations to examine power under different sampling designs and factors, and inform strain tracking design and sampling



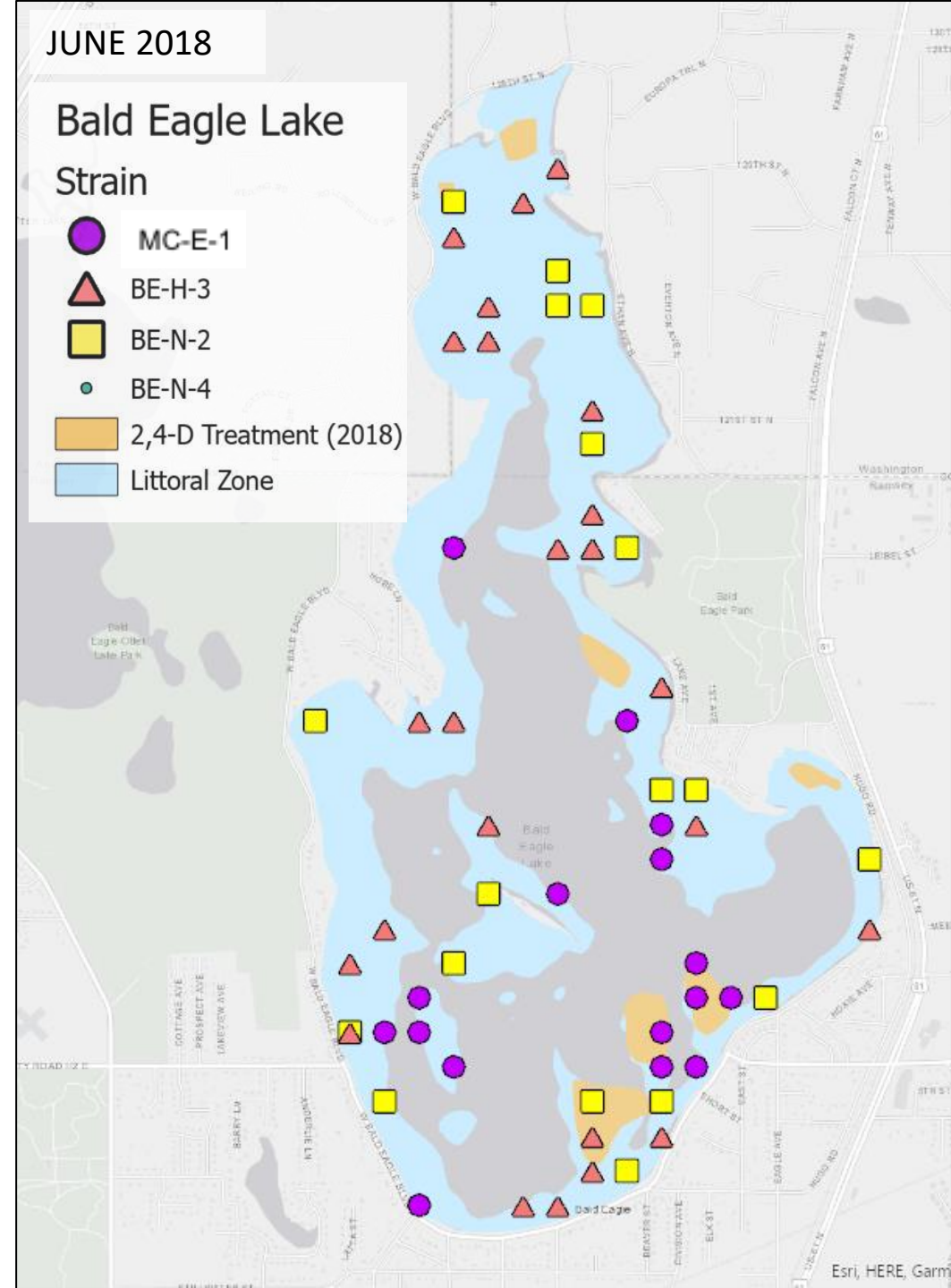
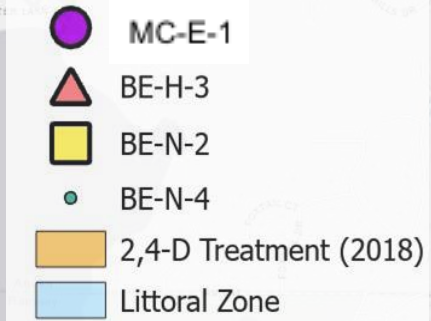
Bald Eagle Lake



JUNE 2018

Bald Eagle Lake

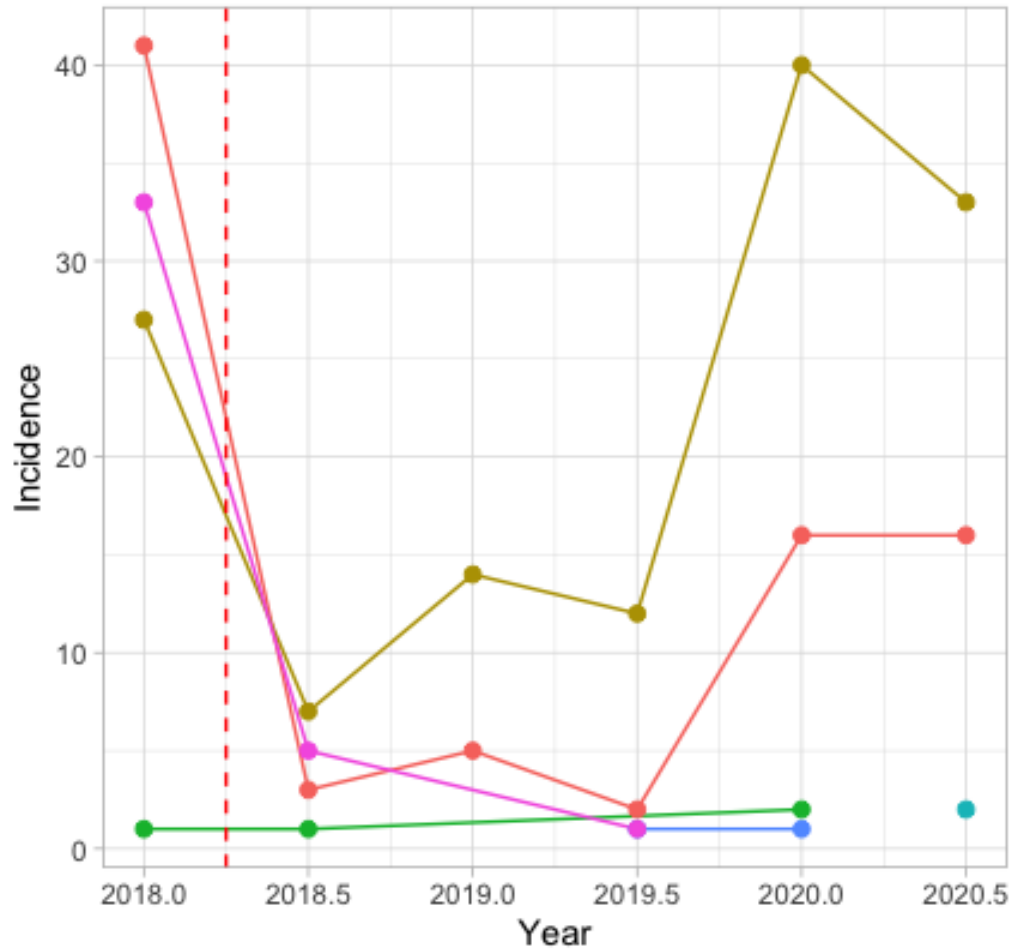
Strain



Simulation Based Chi Squared

Lake	2018-2019	2019-2020
Bald Eagle	<.001	1

Bald Eagle Lake

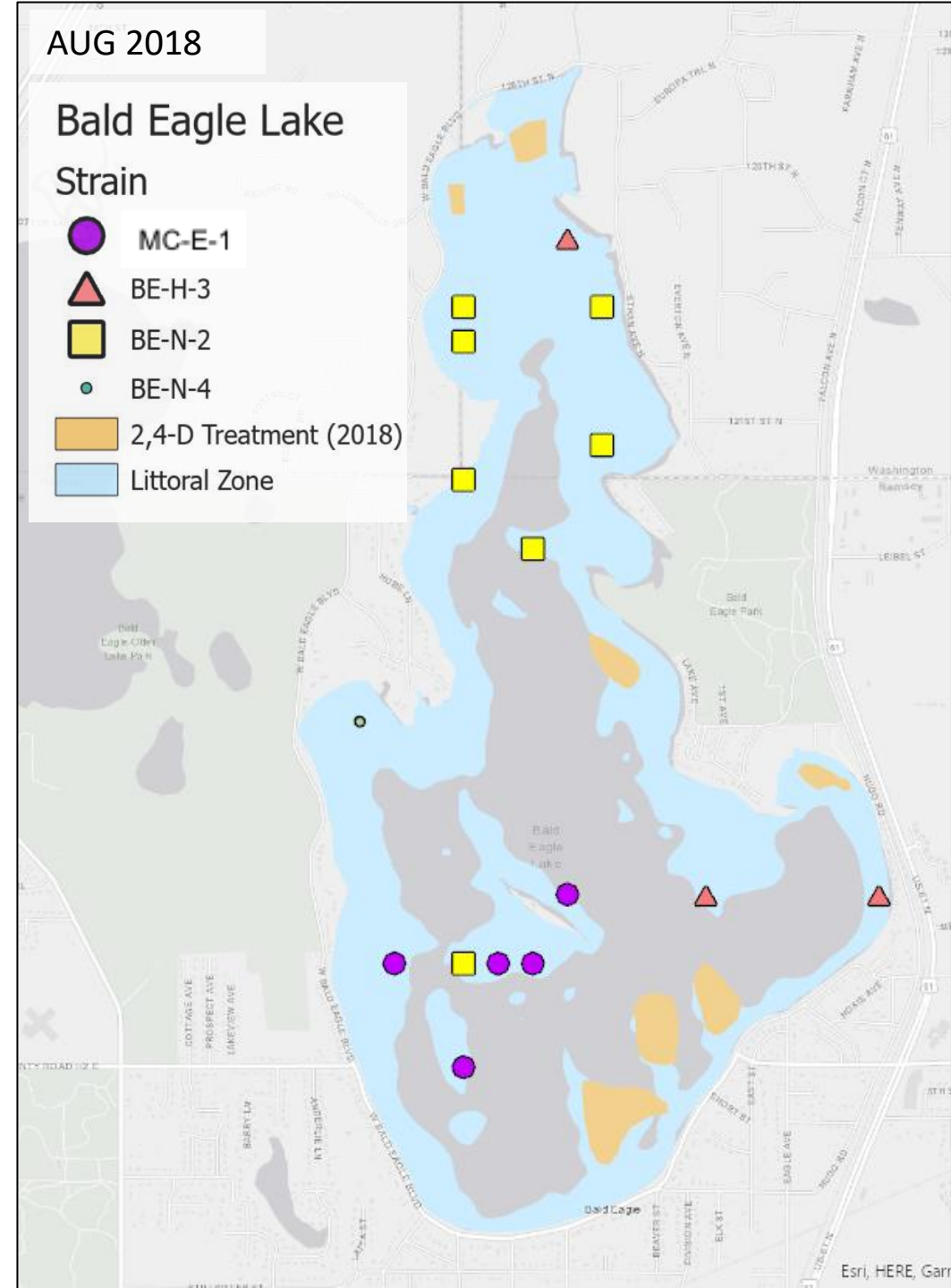


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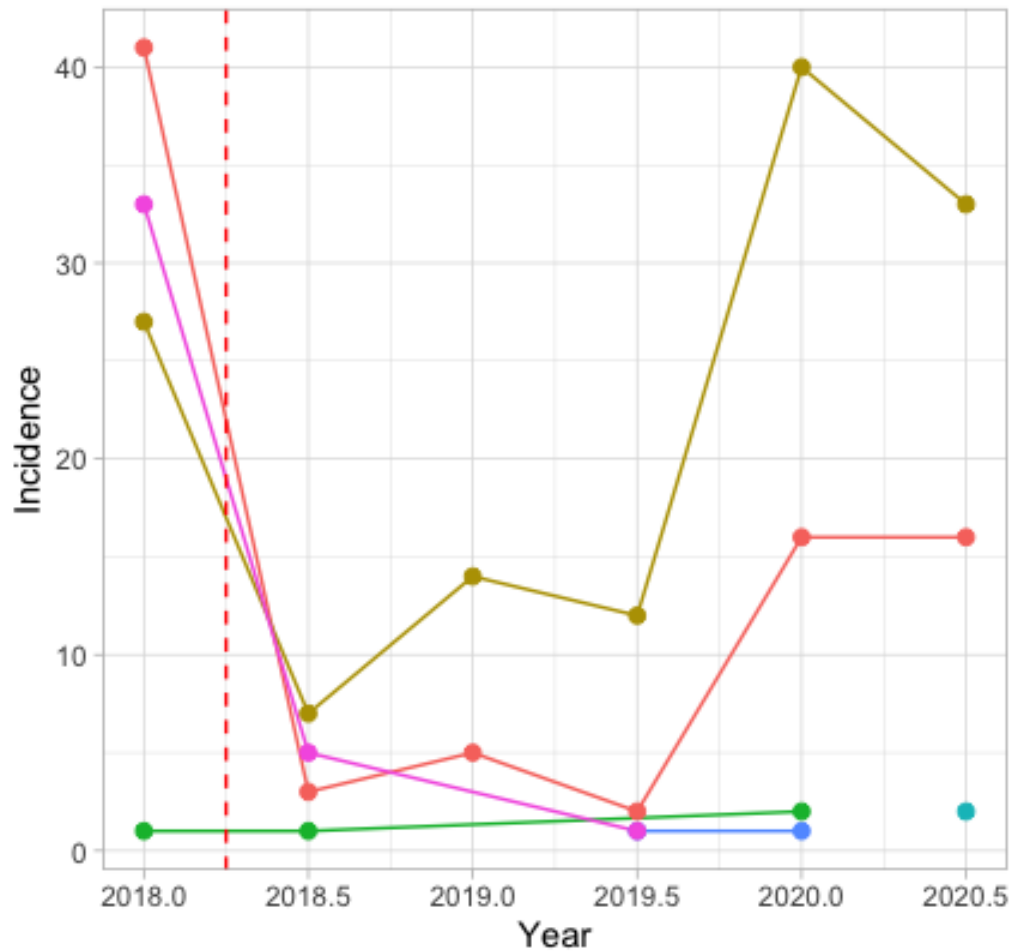
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AUG 2018

Bald Eagle Lake Strain



Bald Eagle Lake

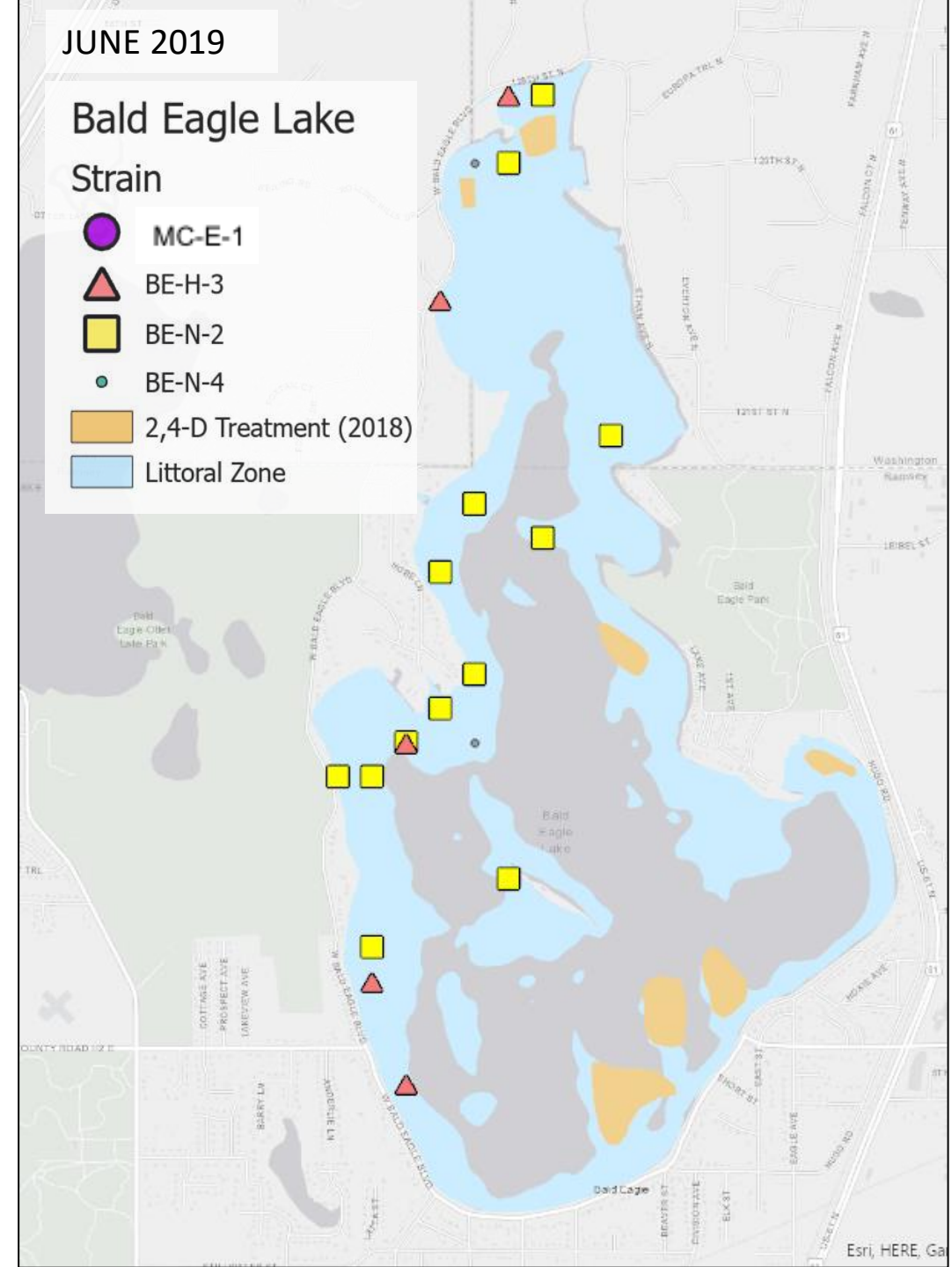


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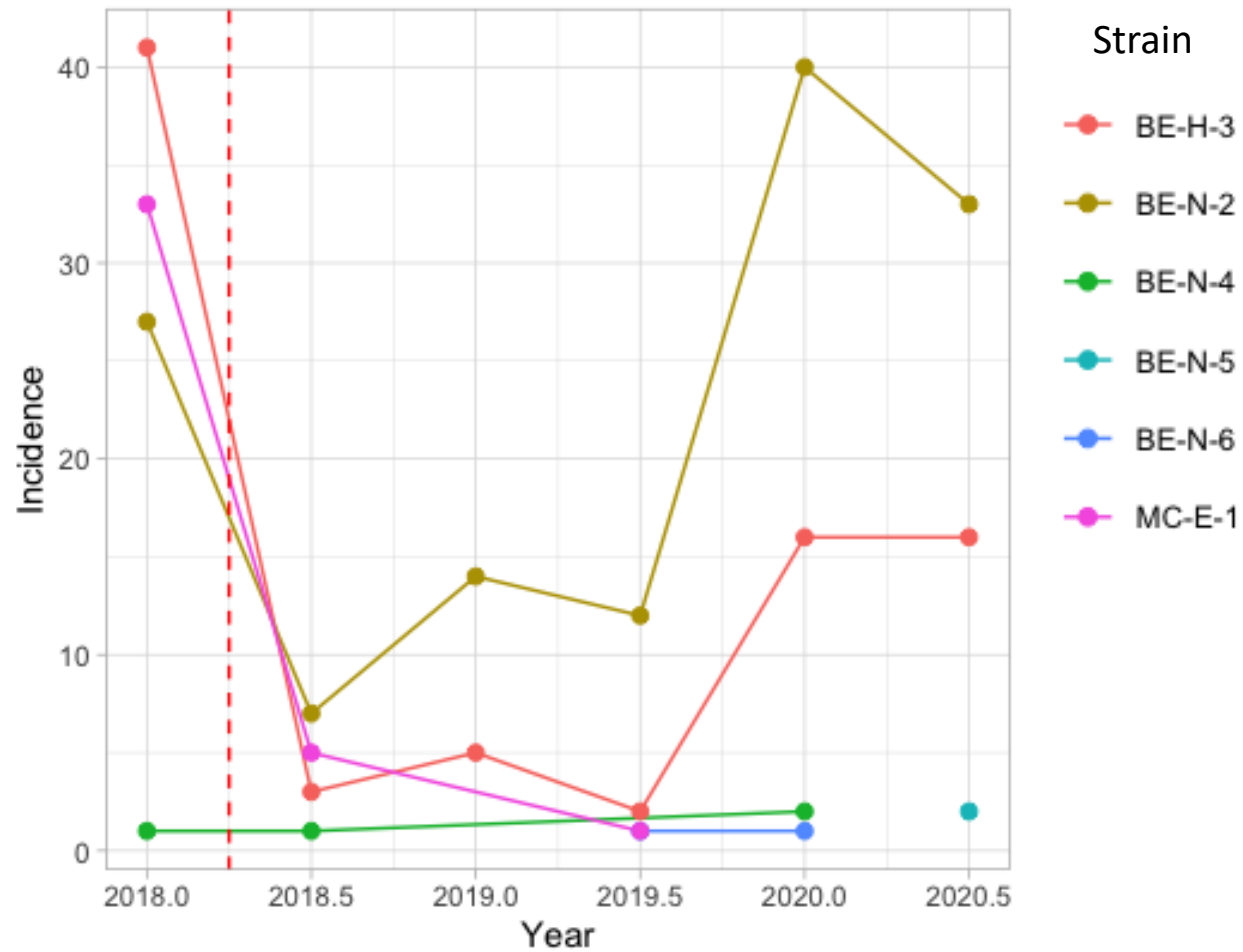
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JUNE 2019

Bald Eagle Lake Strain



Bald Eagle Lake



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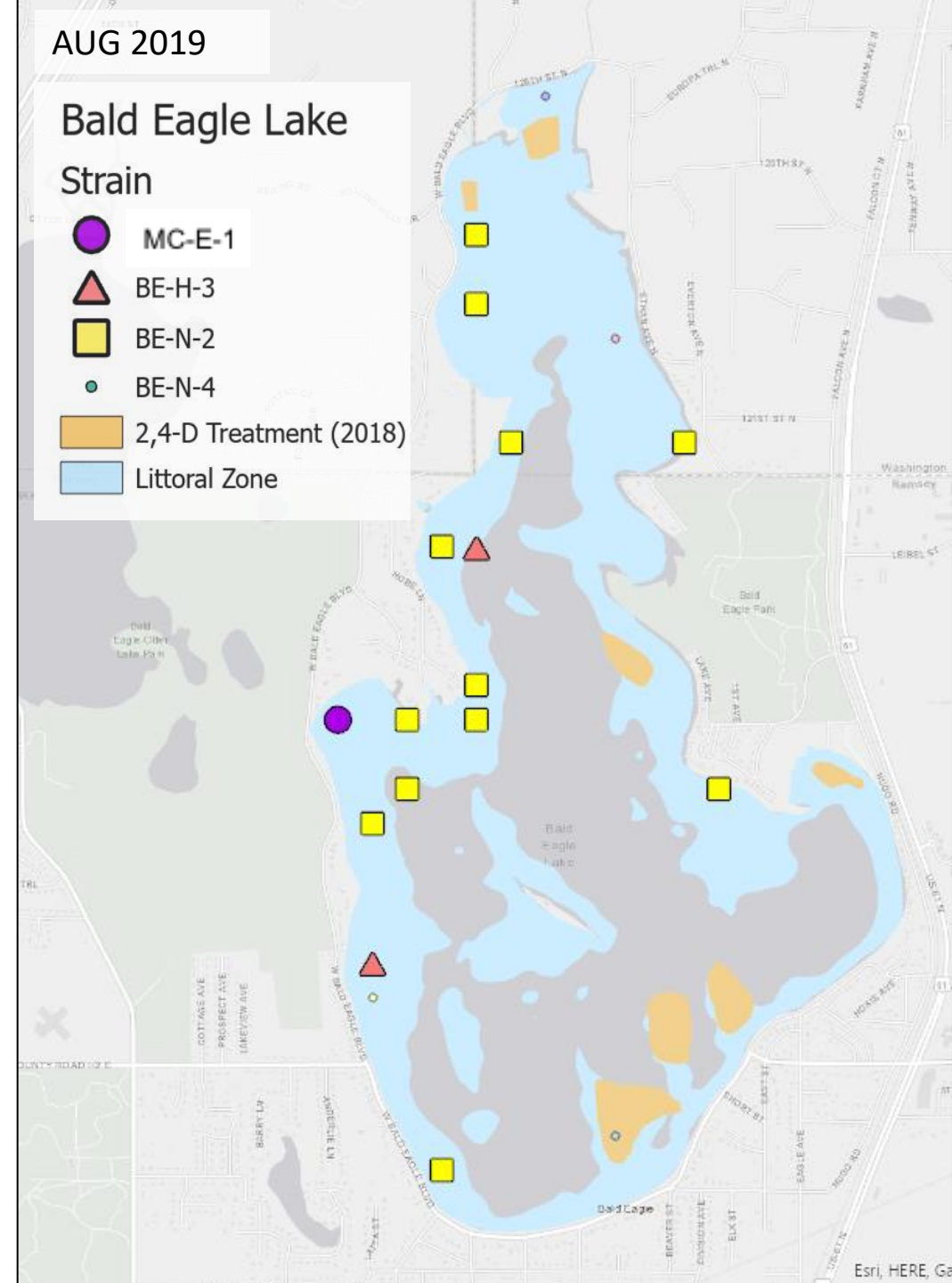
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AUG 2019

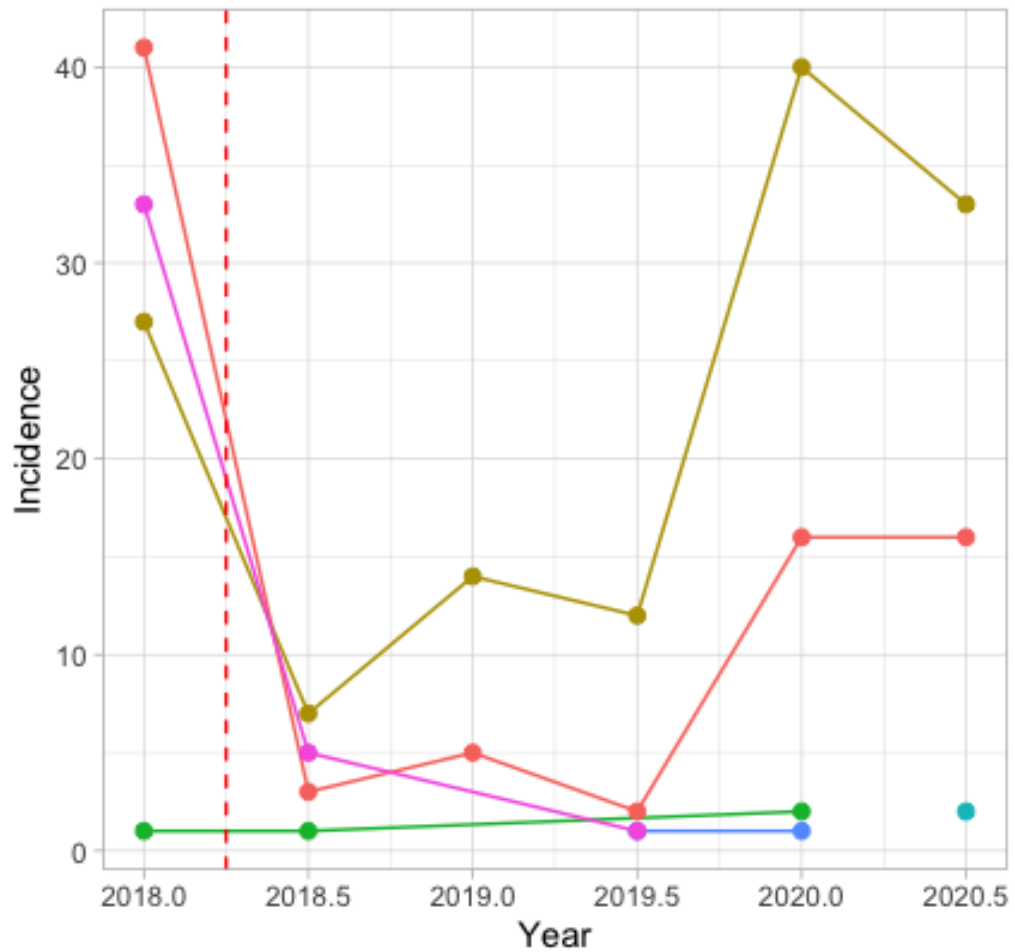
Bald Eagle Lake

Strain

- MC-E-1
- ▲ BE-H-3
- BE-N-2
- BE-N-4
- 2,4-D Treatment (2018)
- Littoral Zone



Bald Eagle Lake



- Strain
- BE-H-3
 - BE-N-2
 - BE-N-4
 - BE-N-5
 - BE-N-6
 - MC-E-1

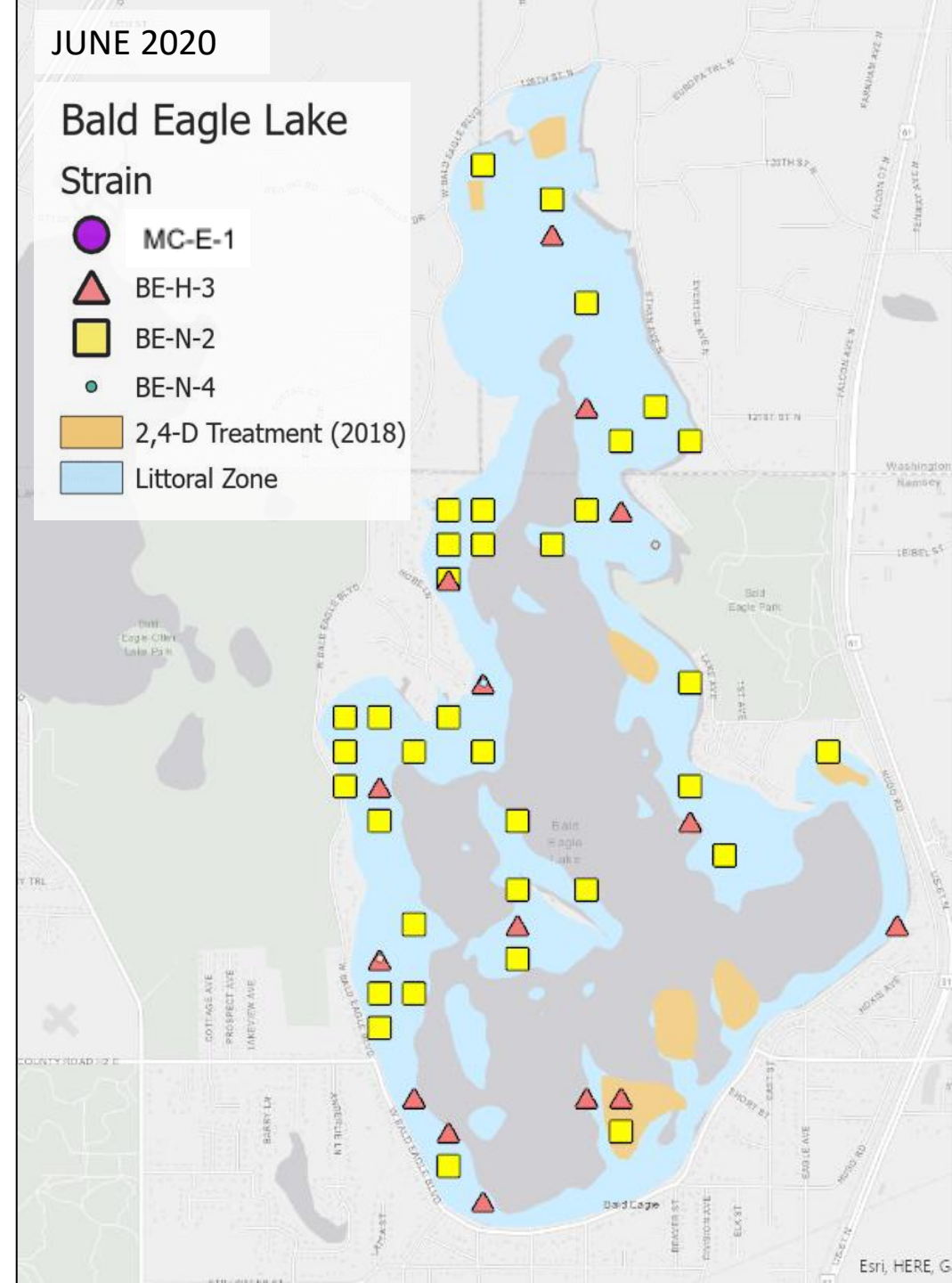
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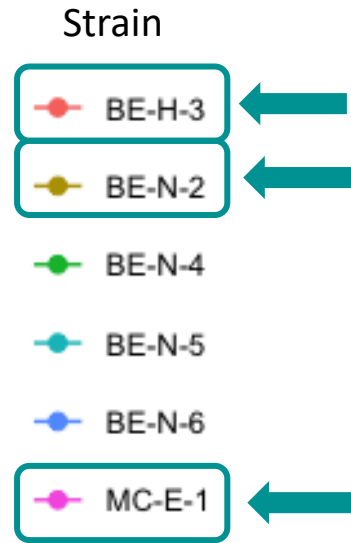
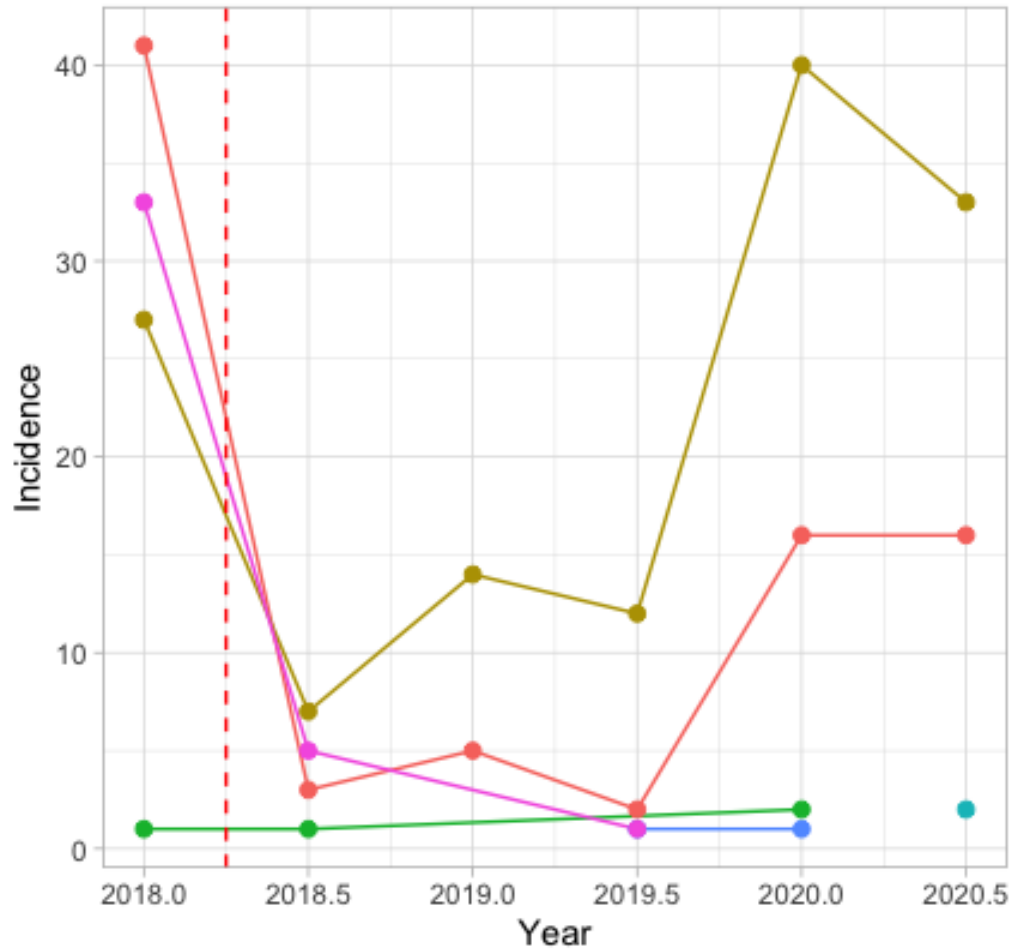
JUNE 2020

Bald Eagle Lake Strain

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Bald Eagle Lake



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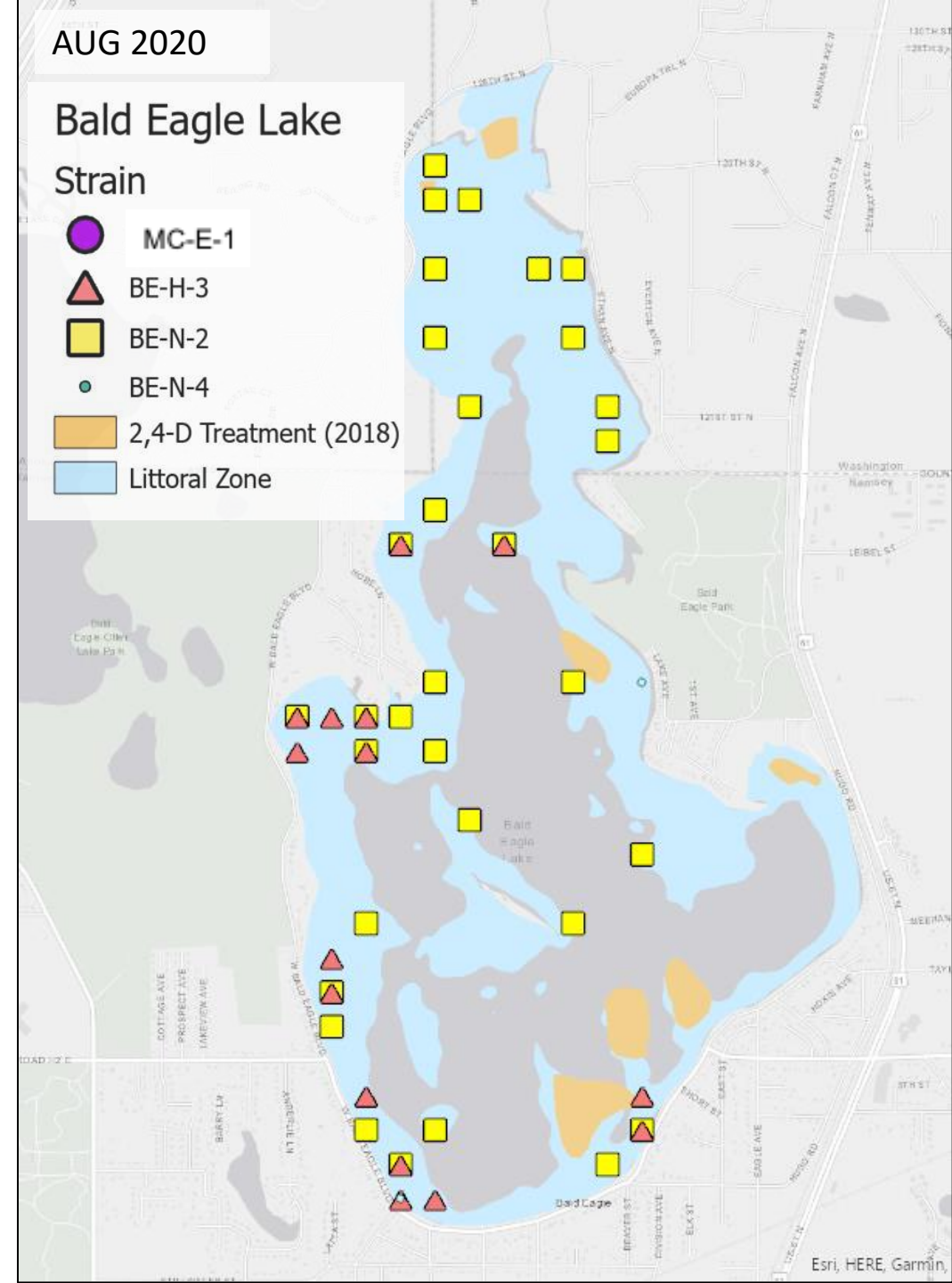
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2,4-D Treatment (2018)

Littoral Zone



Objective 4: Simulations to estimate in-field response to herbicides for different strains.

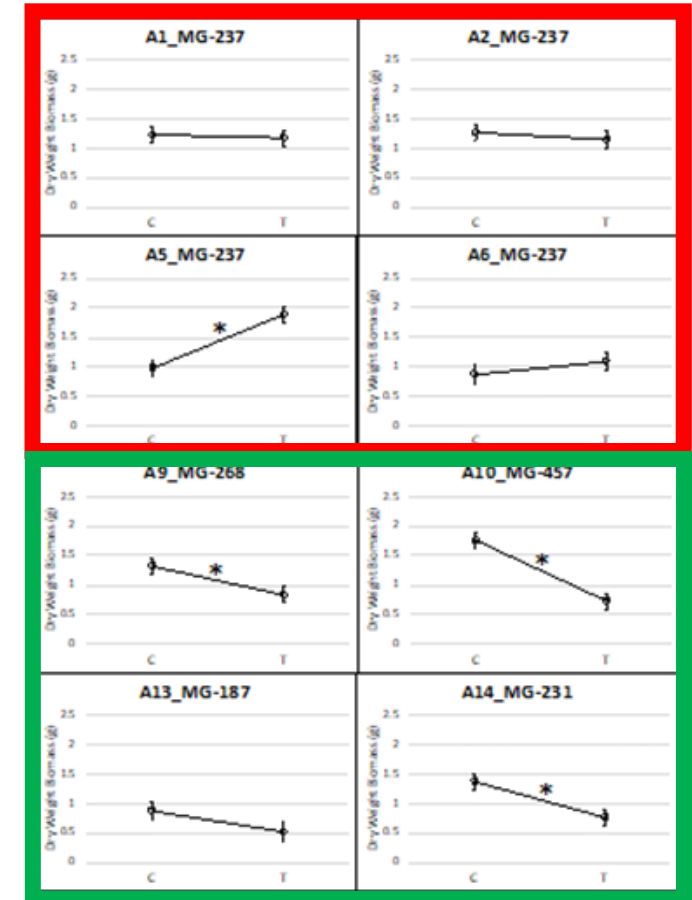
Key Factors

- “Spot” treatments vs whole-lake treatments
 - Sampling inside versus outside of treatment areas
 - Dilution and dissipation effects
- Frequencies of strains
 - 10/90 vs 50/50
 - Few vs many
- Spatial distributions (autocorrelation)
 - Random vs clumped
- Vegetation survey design
 - Point-intercept
 - Meandering shoreline
 - Transect



Why we need to identify genes for herbicide resistance versus susceptibility

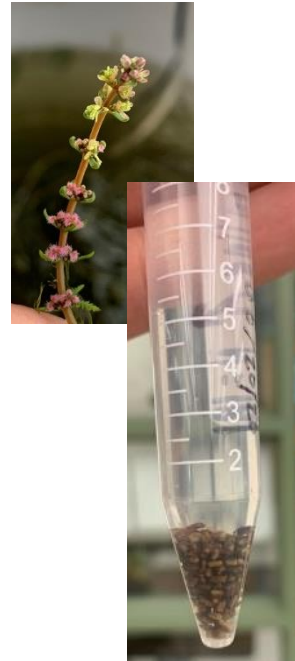
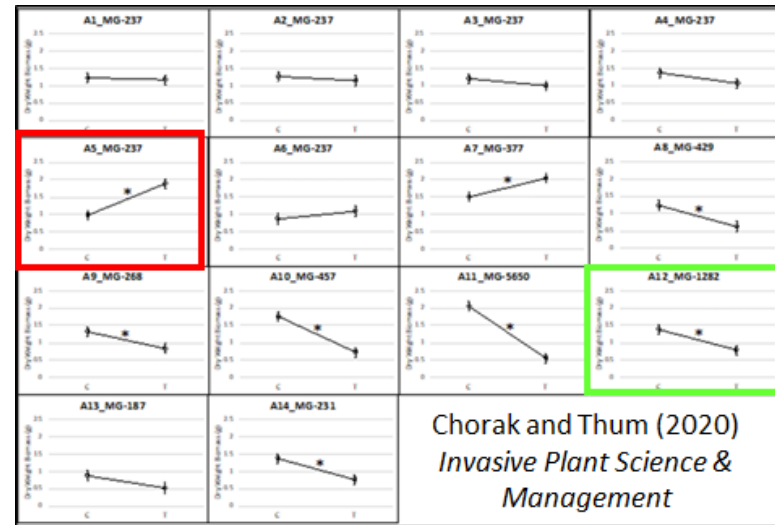
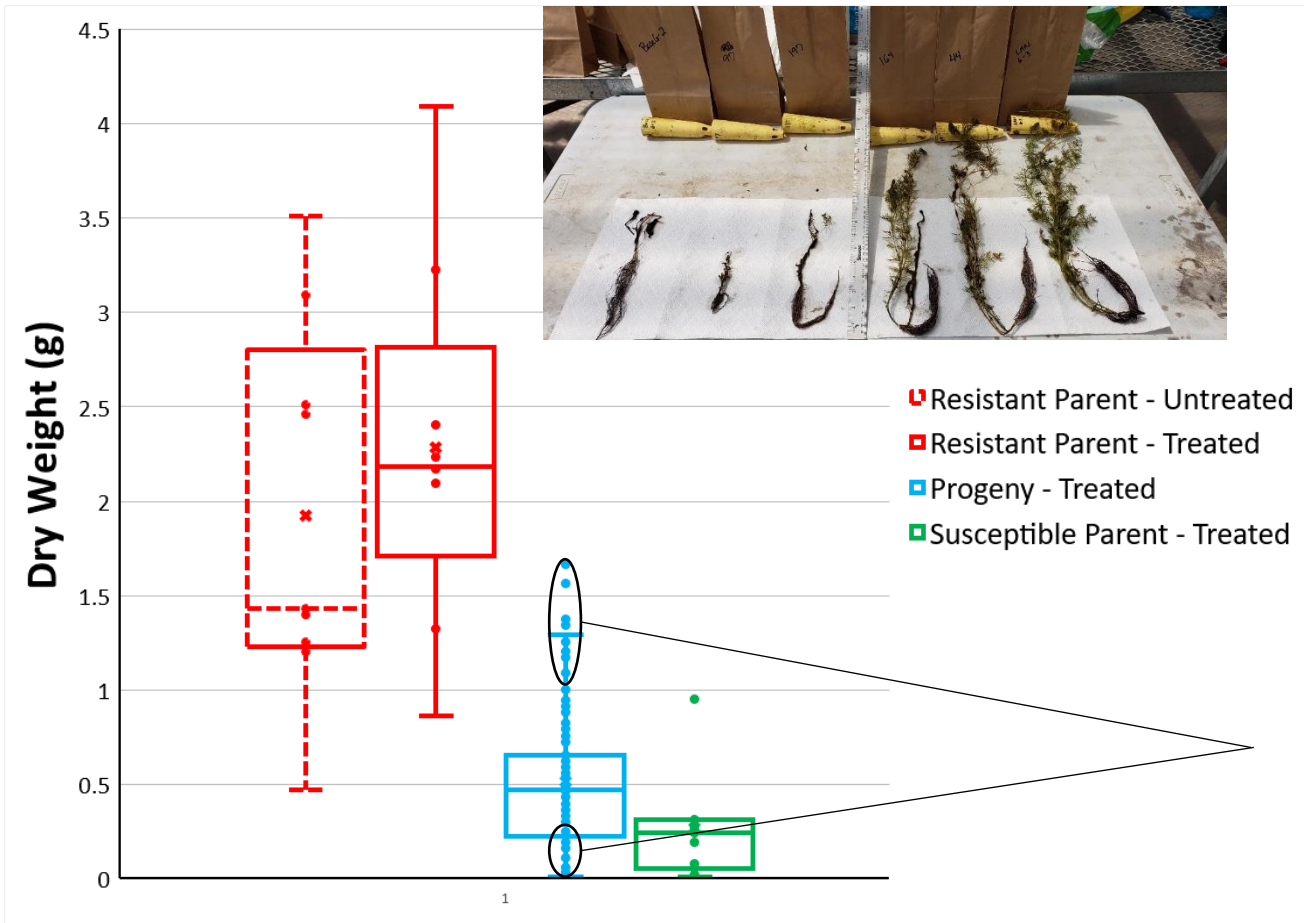
- Using 'neutral' molecular markers, we can distinguish strains that have different herbicide responses.
- However, because watermilfoil can reproduce sexually, new strains are produced all of the time.
- Resistant strains can pass on their resistance genes to new strains, which would not be detected unless and until they were identified in the field, and tested in the lab.
- Therefore, we need to identify the specific gene(s) for resistance, so that molecular tests can identify resistance in new strains.



Chorak and Thum (2020)
Invasive Plant Science & Management

Genetic mapping population 1 (fluridone resistance)

- Cross between fluridone resistant and fluridone susceptible genotype
- Segregation for resistance/susceptibility in progeny

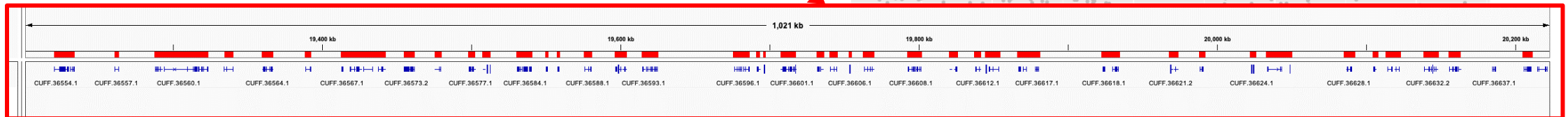
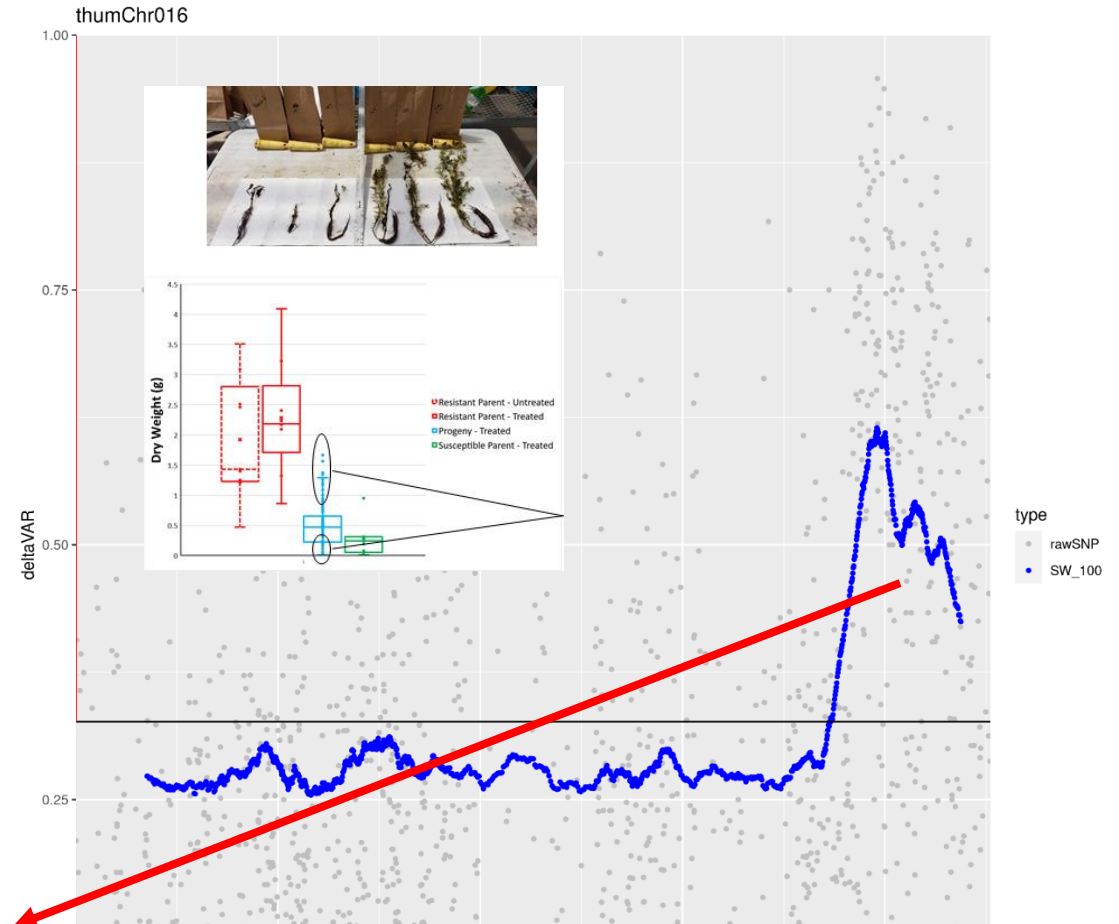


Whole-genome resequencing of ~25 best + ~25 worst growers to look for genes associated with herbicide response.



Genetic mapping population 1 (fluridone resistance)

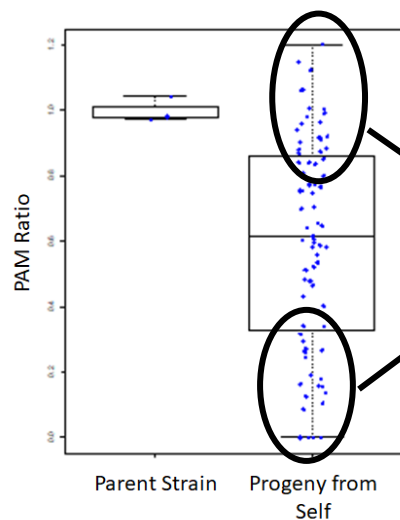
- **Large-effect QTL**
 - ~1MB on “chromosome 16”
- **Not phytoene desaturase (PDS)**
 - Located on “chromosomes 4-6”
- Evaluating candidate genes that occur within the QTL



Additional genetic mapping populations

Second Fluridone Resistance

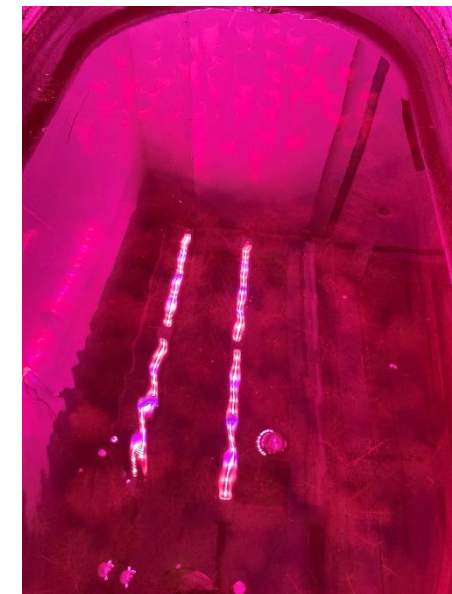
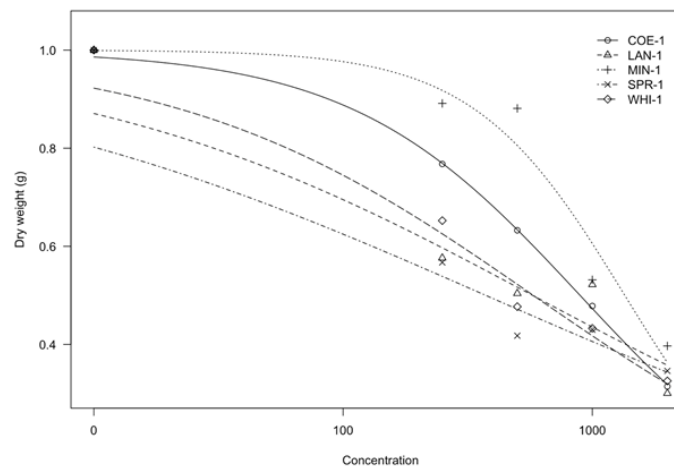
- Mapping population of a second fluridone resistant strain
- **Do the two strains have the same or different resistance mechanisms and genes?**



Whole-genome resequencing of resistant and susceptible bulks.

2,4-D Resistance

- Can we identify any major-effect QTL for 2,4-D resistance?



Acknowledgements



LOTS of people who collected and sent samples, and identified interesting strains to work on



Genetic mapping of fluridone resistance

= 10 MB
(10,000,000
base pairs)

