U.S. Environmental Projection Agency, Office of Research and Development

SAFE AND SUSTAINABLE WATER RESOURCES RESEARCH PROGRAM

USEPA Research Update:

2014 Bloom Season Monitoring - Brief Recap

Permanganate Oxidation – Preliminary Results

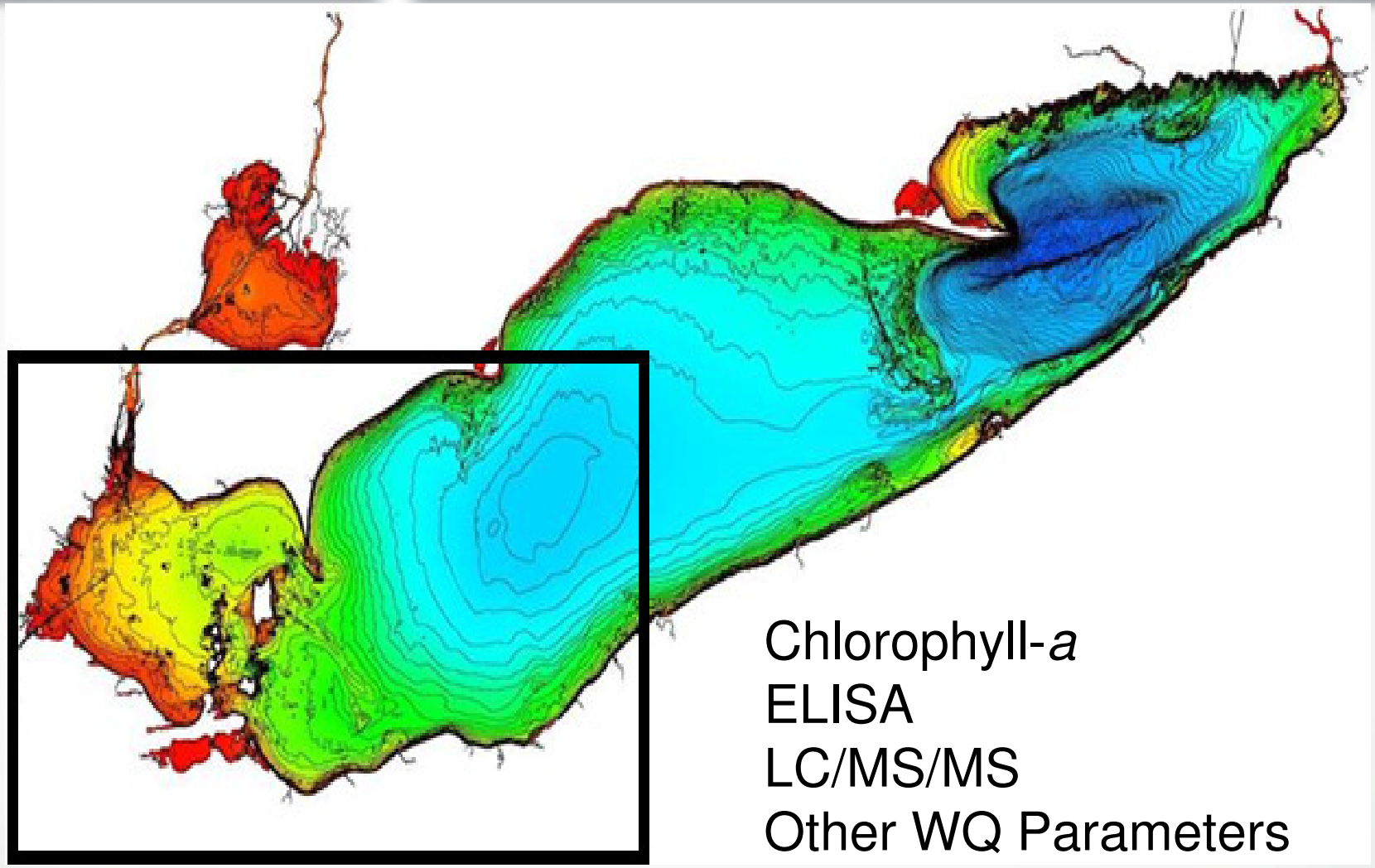
2015 Bloom Season Monitoring – Preliminary Plans

Nicholas Dugan, Toby Sanan, Samantha Smith, Randall Marsh, Heath Mash, Darren Lytle

Ohio EPA Regional Harmful Algal Bloom Meeting, Avon Lake OH, March 24, 2015

circle of blue

2014 DWTP Study on Lake Erie



Chlorophyll-*a*
ELISA
LC/MS/MS
Other WQ Parameters

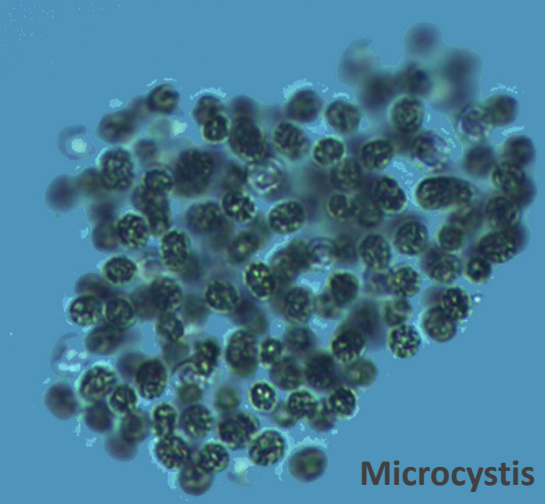
Occurrence and effects of treatment on cyanobacteria/microcystins

Treatment for Cyanobacterial Toxins

Toxin within the cell and those that are dissolved require different treatment processes

Intracellular (toxin inside cells)

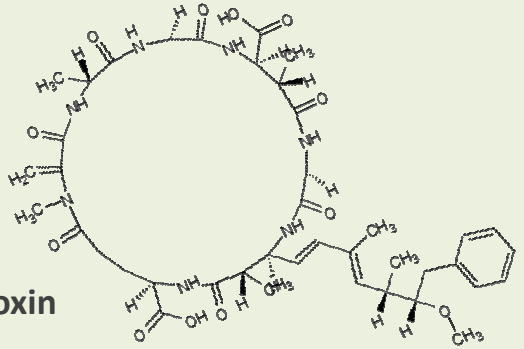
- Solids removal processes effective prior to toxin release



Microcystis (cells)

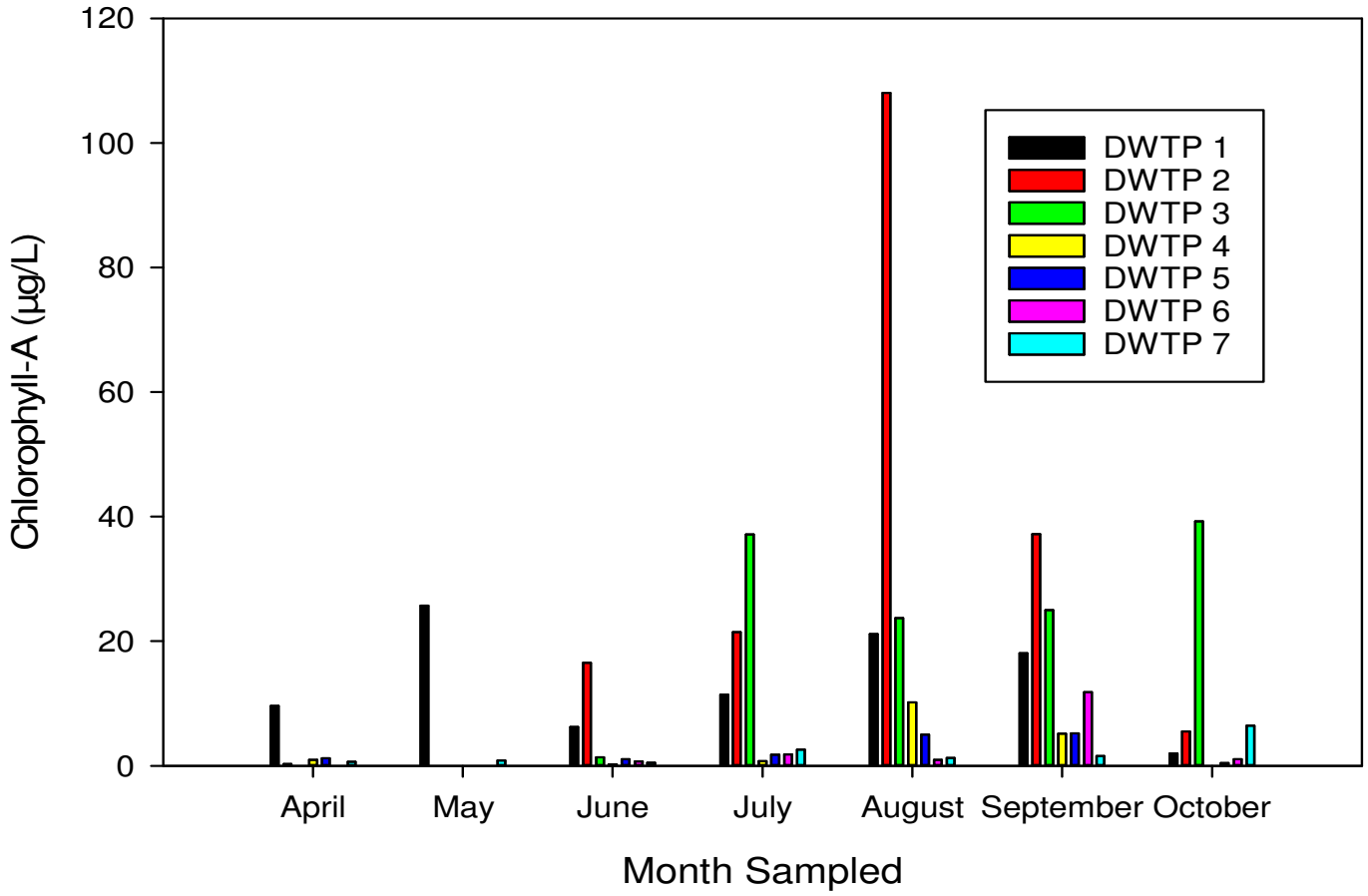
Extracellular (toxin released from cell)

- Solids removal processes ineffective
- Typical disinfectants may not be effective enough (e.g., permanganate, chlorine)
- More effective treatments are expensive (e.g., GAC)



Microcystin Toxin

Chlorophyll-A Levels in 2014

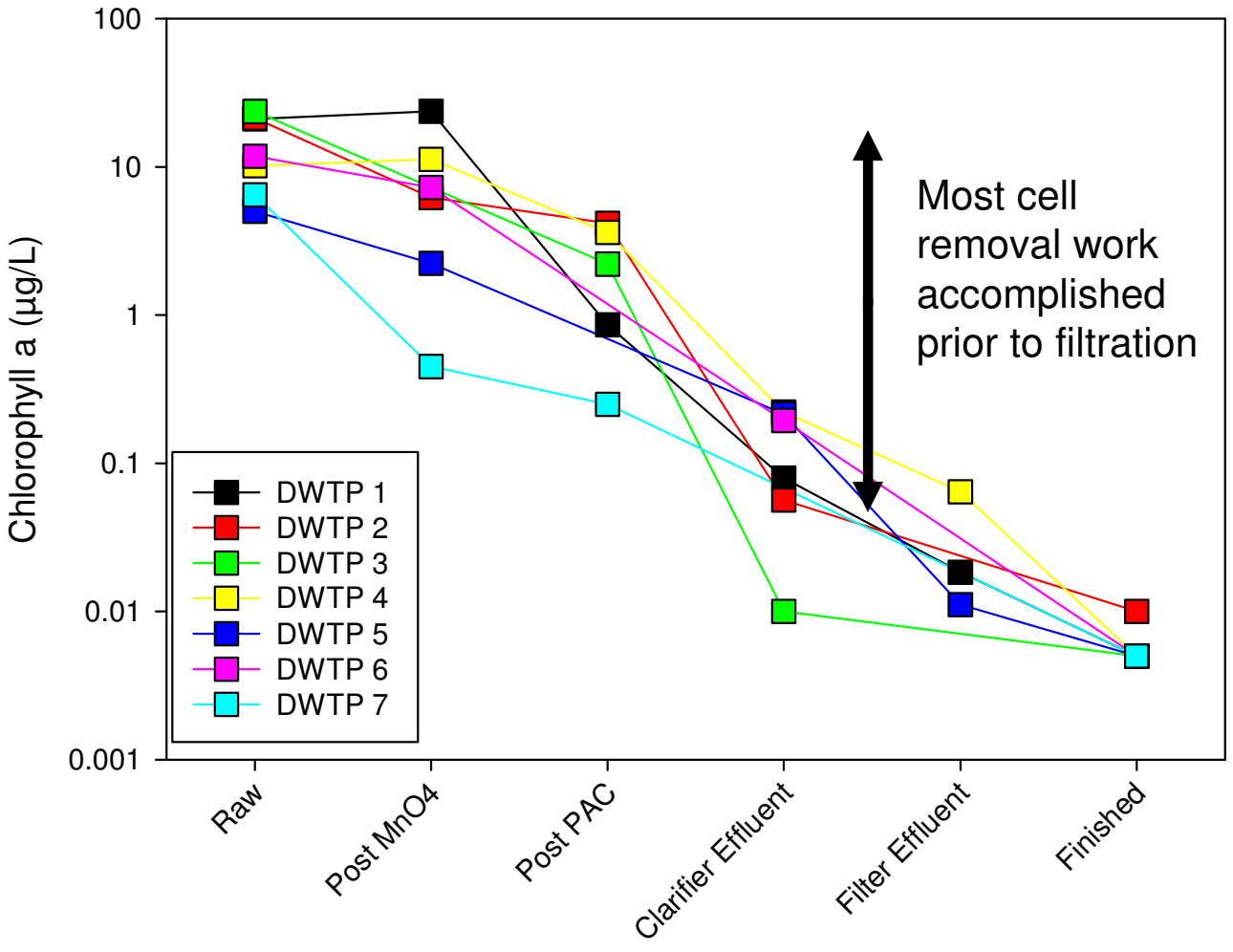


Chlorophyll-*a* peaks in August-September, consistent with HAB maxima
Large fluctuations show need for an increased sampling frequency

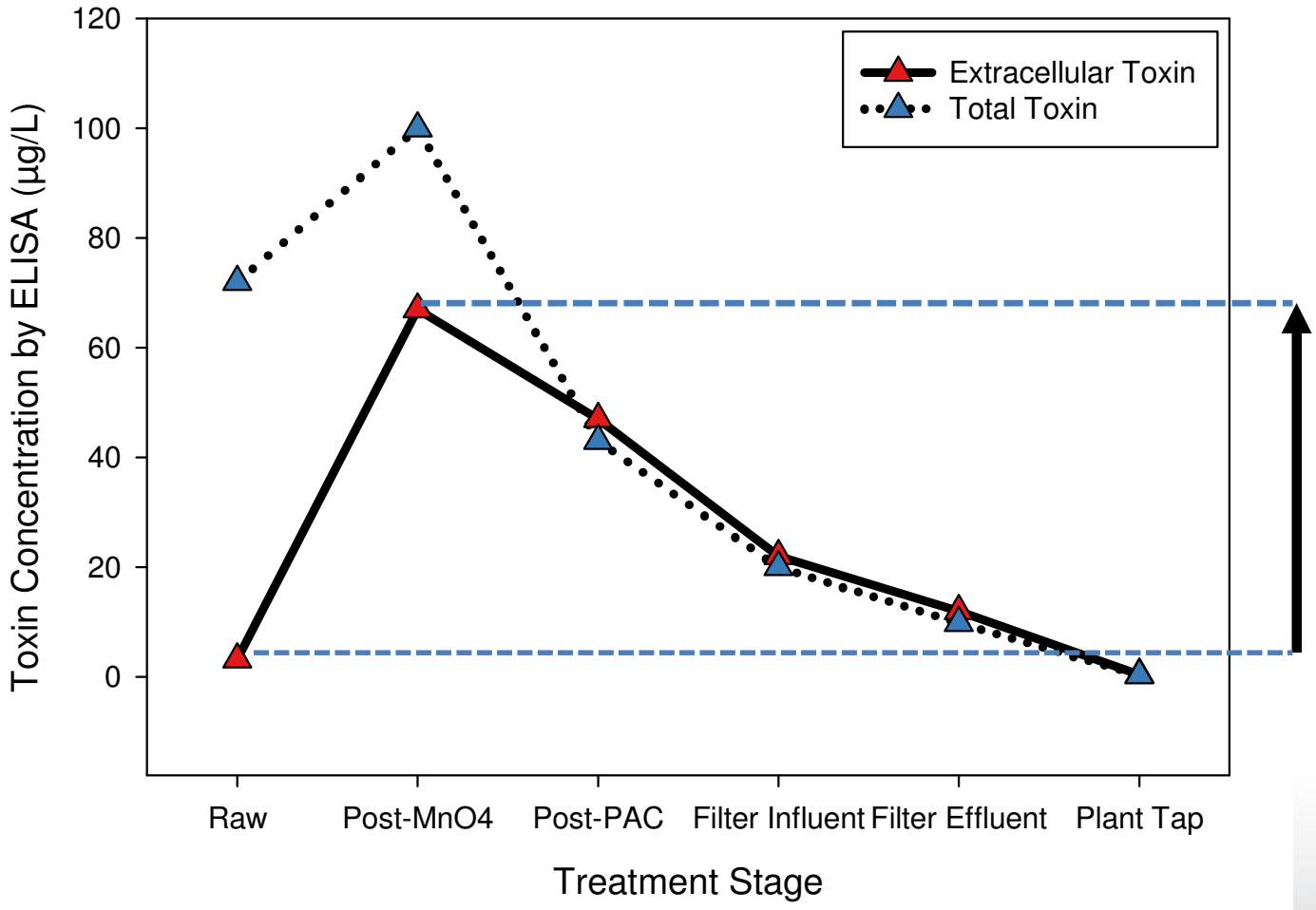
DWTP/ Month	Chlorophyll-a, µg/L	ELISA, Extracellular, µg/L	LC/MS/MS, Extracellular, µg/L	ELISA, Total, µg/L	LC/MS/MS, Total, µg/L
DWTP 1, August	21	0.4	< DL	> 5.0	1.4
DWTP 2, August	108	3.2	< DL	72	8.4
DWTP 3, August	24	< DL	< DL	2.4	0.6
DWTP 4, August	10	0.5	< DL	2	0.5
DWTP 1, September	18	0.8	< DL	3.9	0.5
DWTP 2, September	37	0.7	< DL	1.4	0.5
DWTP 3, September	25	0.7	< DL	3.7	0.3

Seven sampling events with influent waters over 1 µg/L by ELISA
Only in August/September
No finished water detections above 1 µg/L by ELISA or LC/MS/MS

Chlorophyll-a measurements show removal by clarification

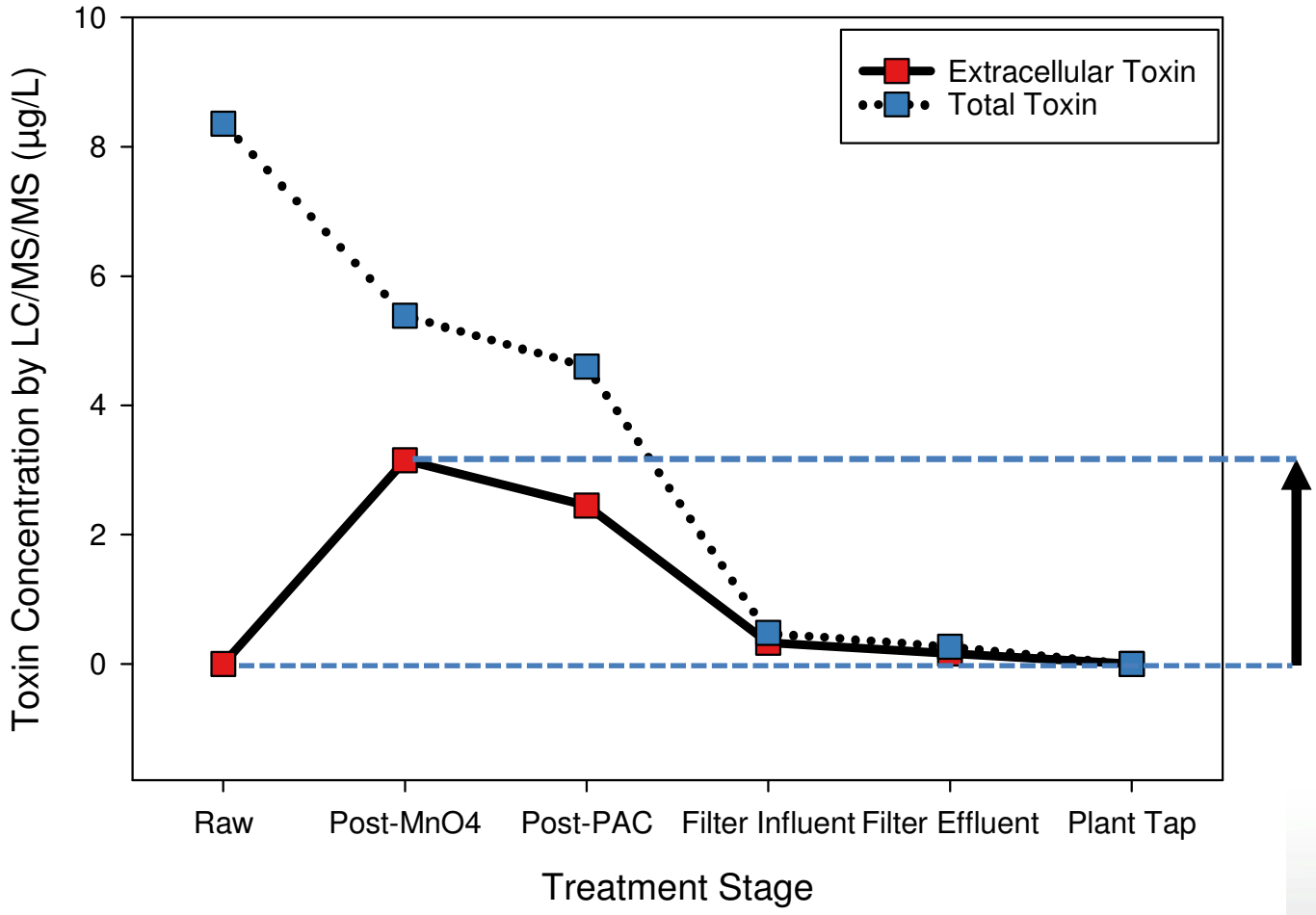


Toxin Propagation through DWTP as measured by ELISA



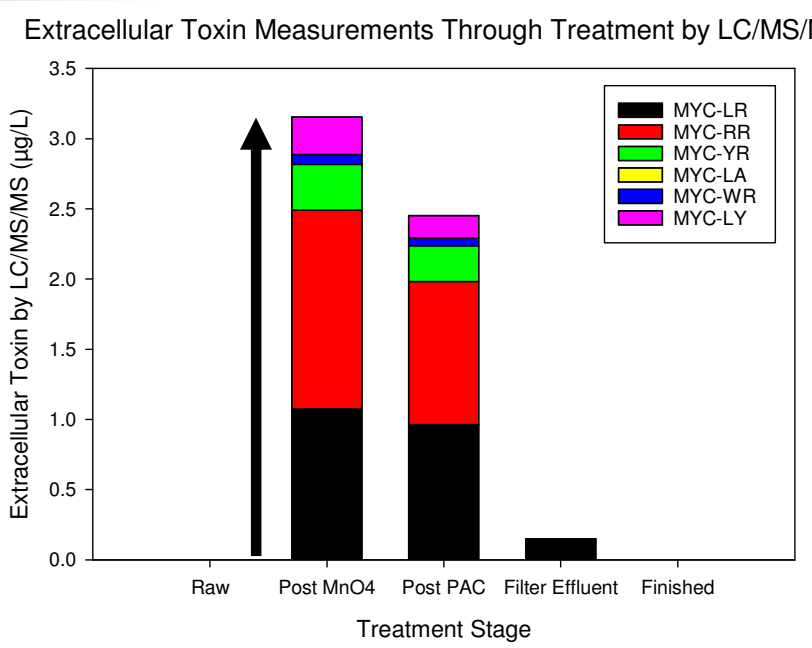
Intracellular toxin release following MnO_4 addition

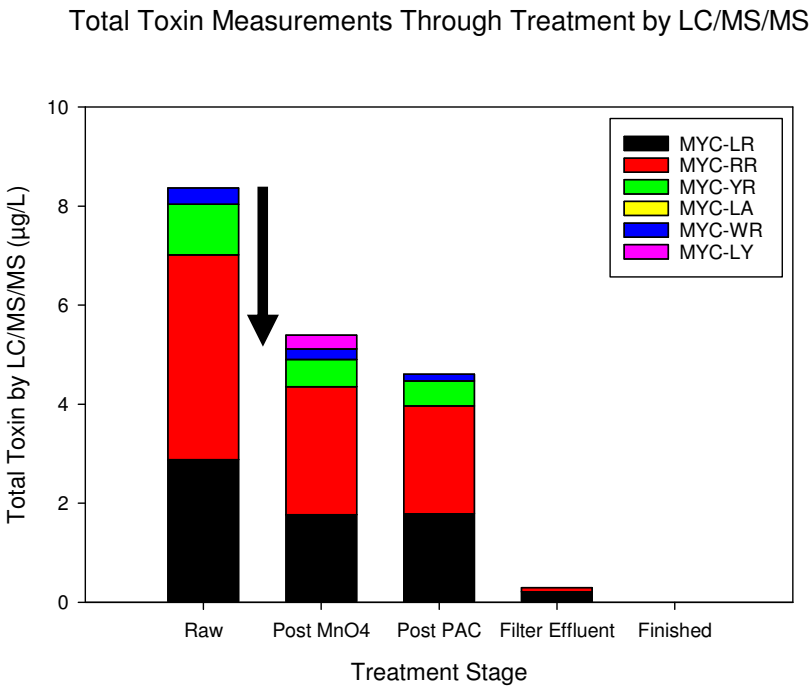
Toxin Propagation through DWTP as measured by LC/MS/MS



Intracellular toxin release following MnO_4 addition

Toxin Speciation and Propagation as Measured by LC/MS/MS





Significant diversity in toxin speciation

More research needed to investigate effects of $KMnO_4$ and PAC on toxin release/removal

Jar Test Methodology

- Intact, toxin-producing cyanobacterial cells, grown in culture
- Cells suspended in de-chlorinated tap water
- Jar tests performed at pH 7, 9
- Potassium permanganate doses of 1, 2.5 and 5 mg/L
- Tests performed in 2 L beakers on a standard jar test apparatus
- Samples collected prior to oxidant addition as well as at t = 15, 30 and 90 minutes

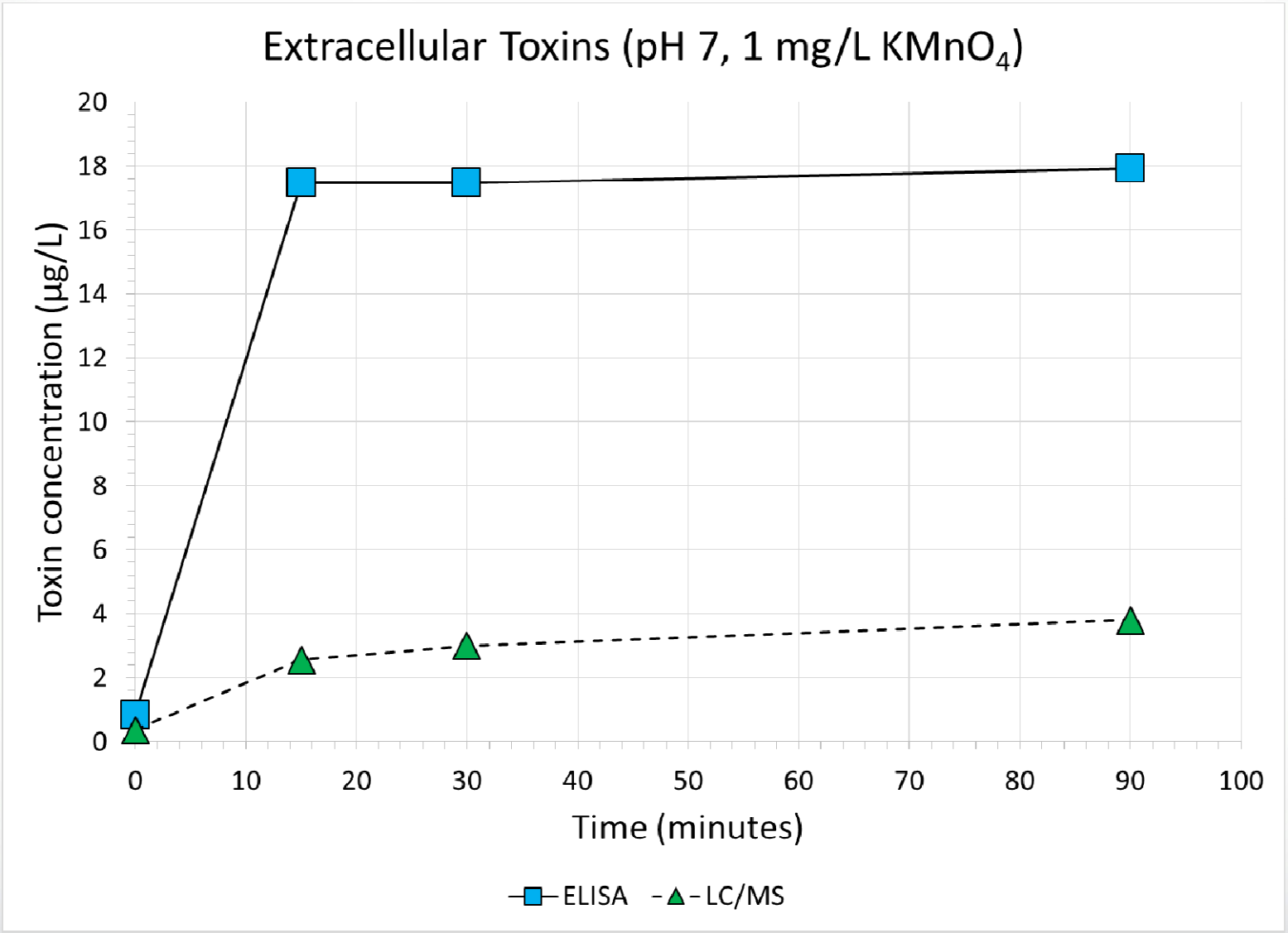
Jar Test Methodology

- Total toxins:
  - Three (3) freeze/thaw cycles @ -20° C
  - Centrifugation
  - Sample collected from the supernatant
- Extracellular toxins:
  - Filter through glass fiber filter

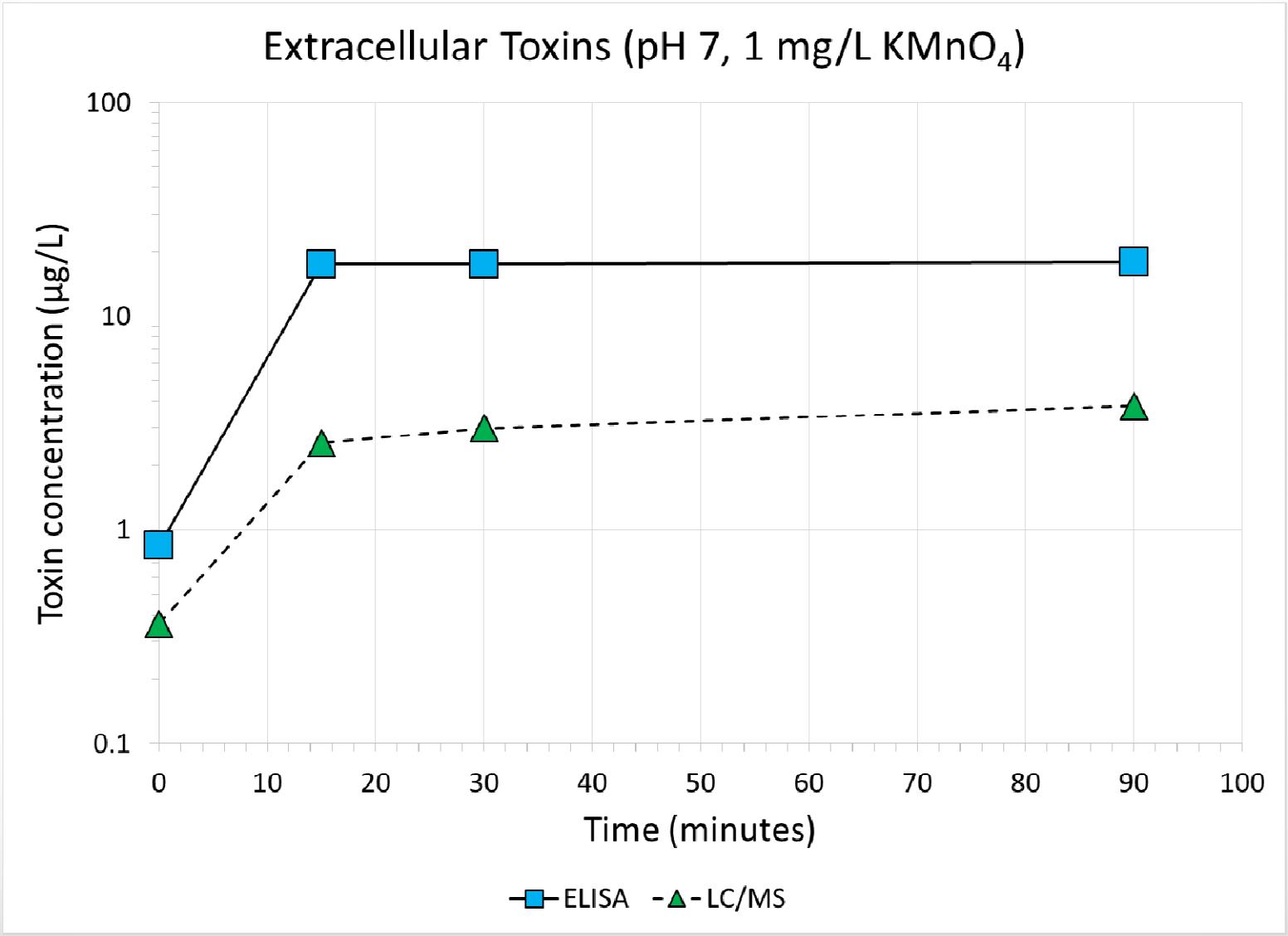
Jar Test Methodology

- Extracellular and total toxin concentrations measured by ELISA and LC/MS
- ELISA measures all microcystin congeners + nodularin
- USEPA/ORD's current LC/MS method measures MC-LR, -RR, -YR, -WR, -LA, -LF, -LY, -LW and nodularin
- Cell concentrations measured by microscope:
  - Membrane integrity measured by application of cell permeant fluorescent stain – stain only penetrates membranes that are compromised in some way

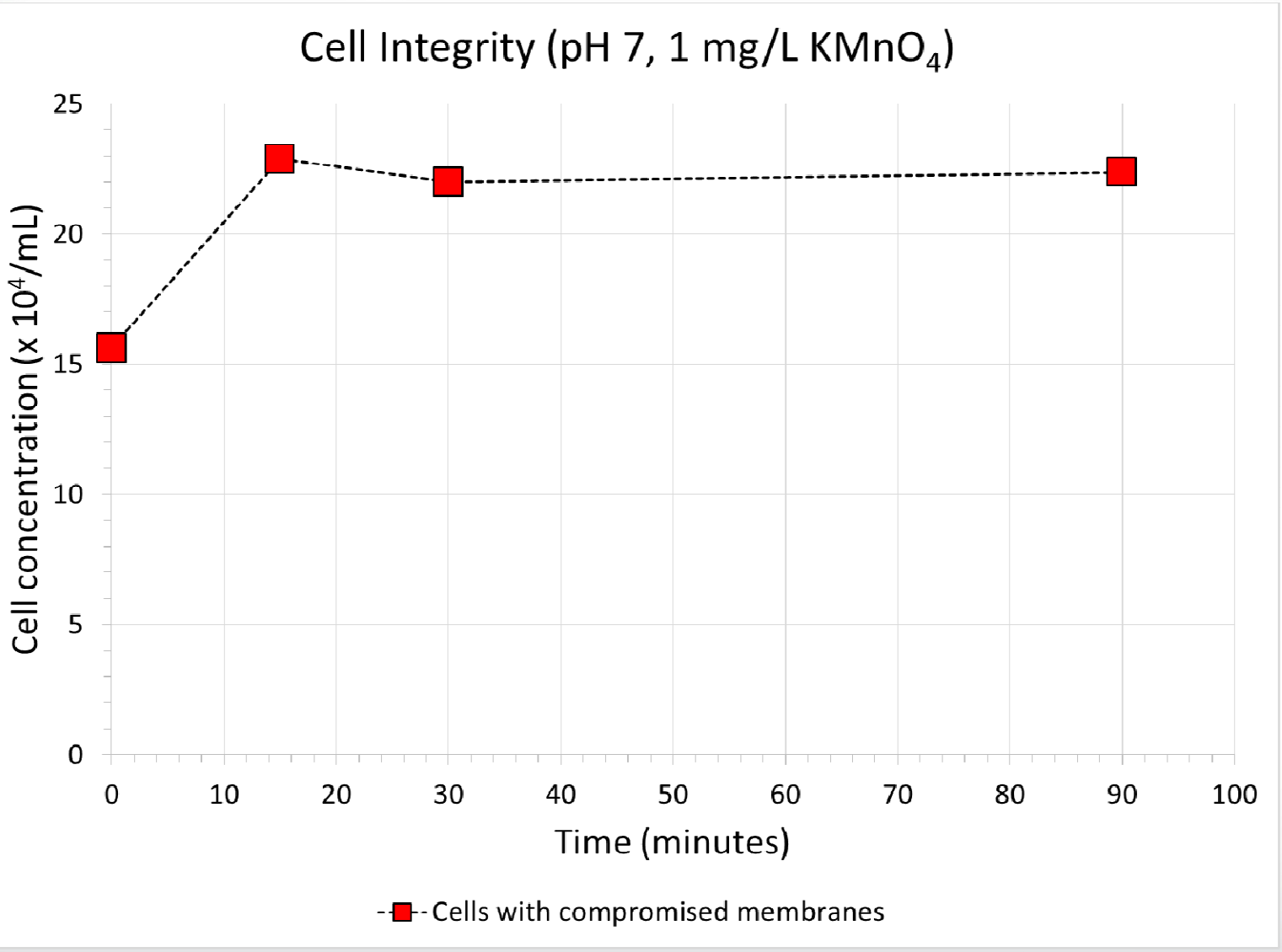
Jar Test Results



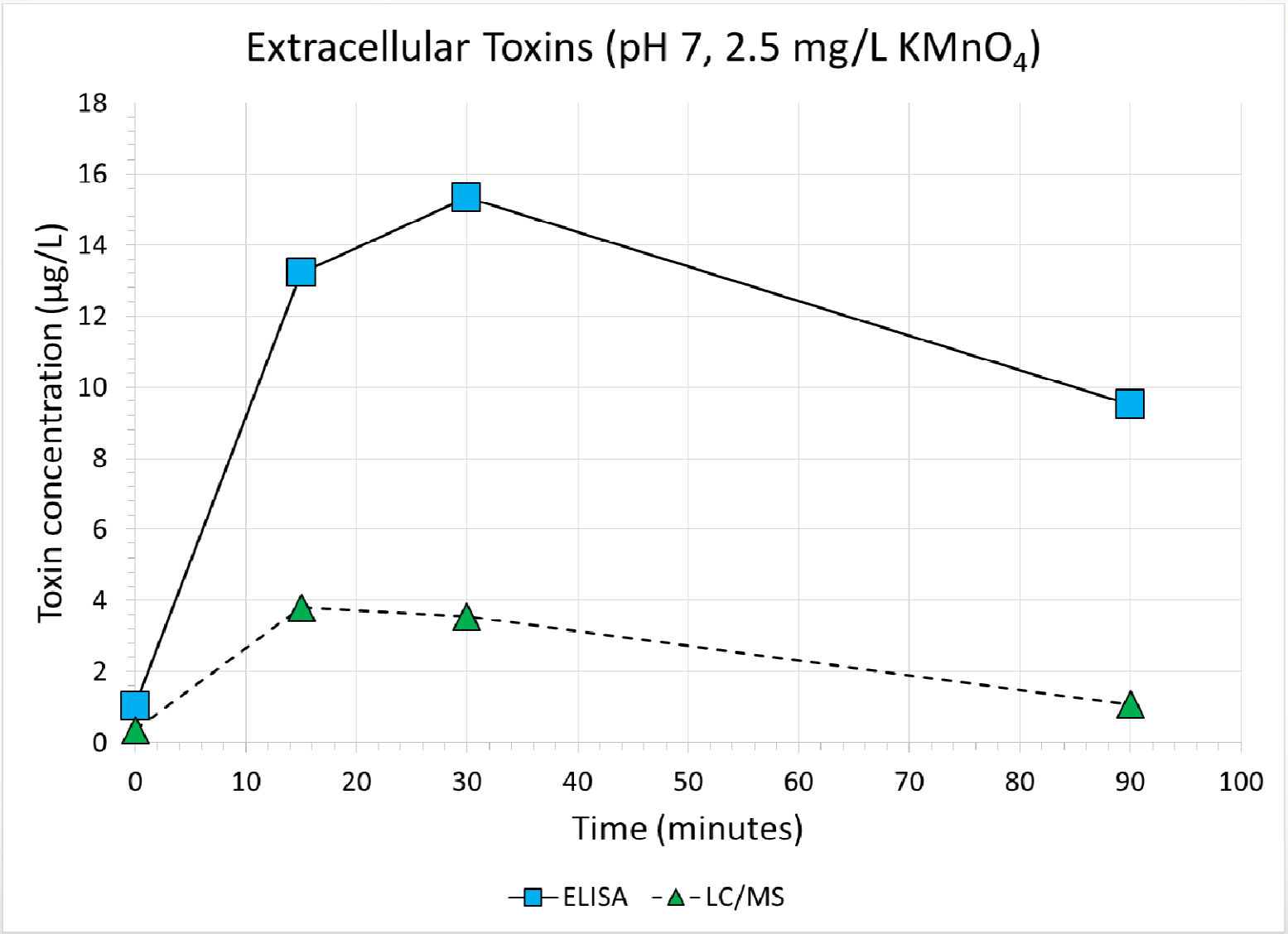
Jar Test Results



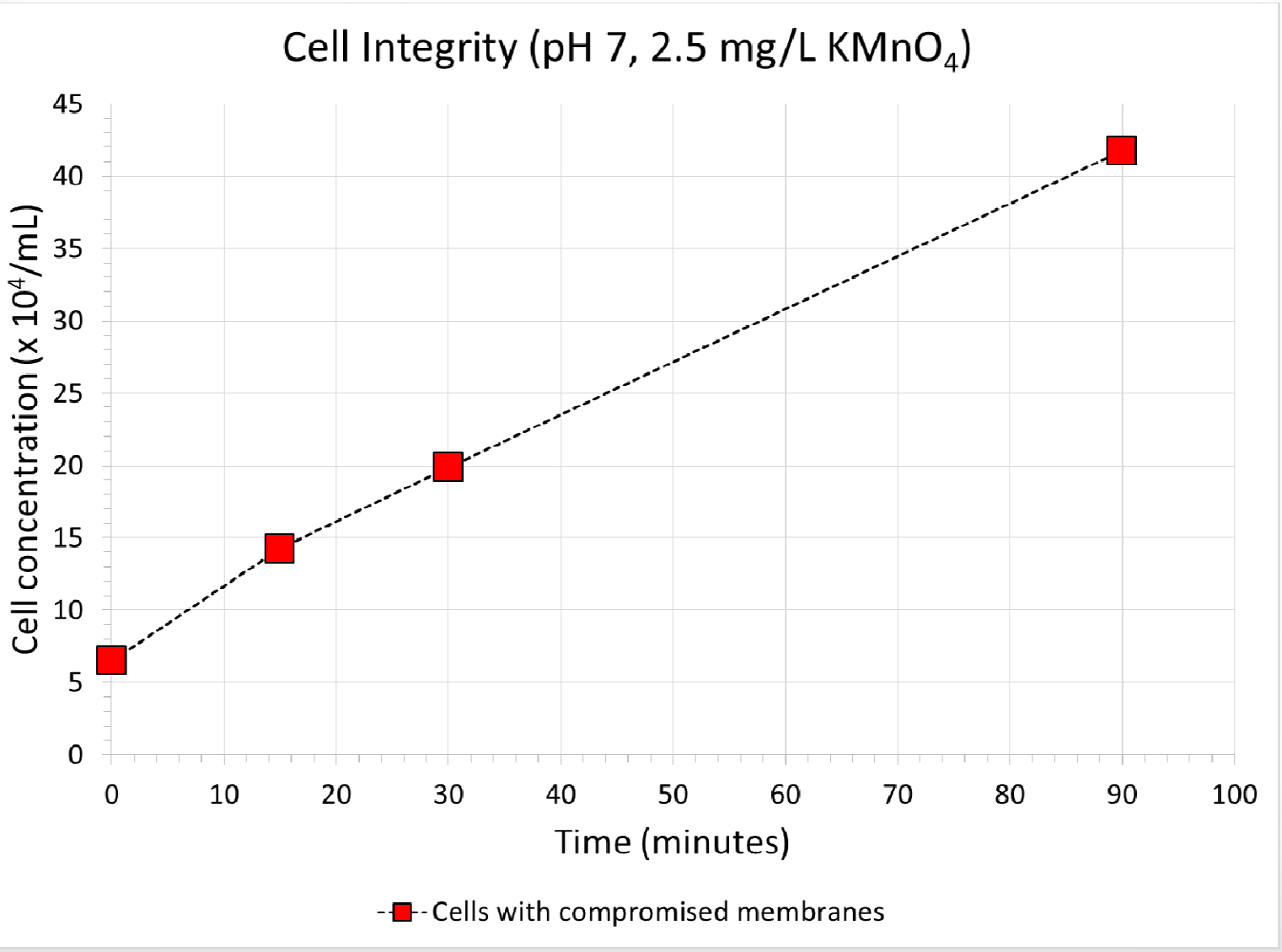
Jar Test Results



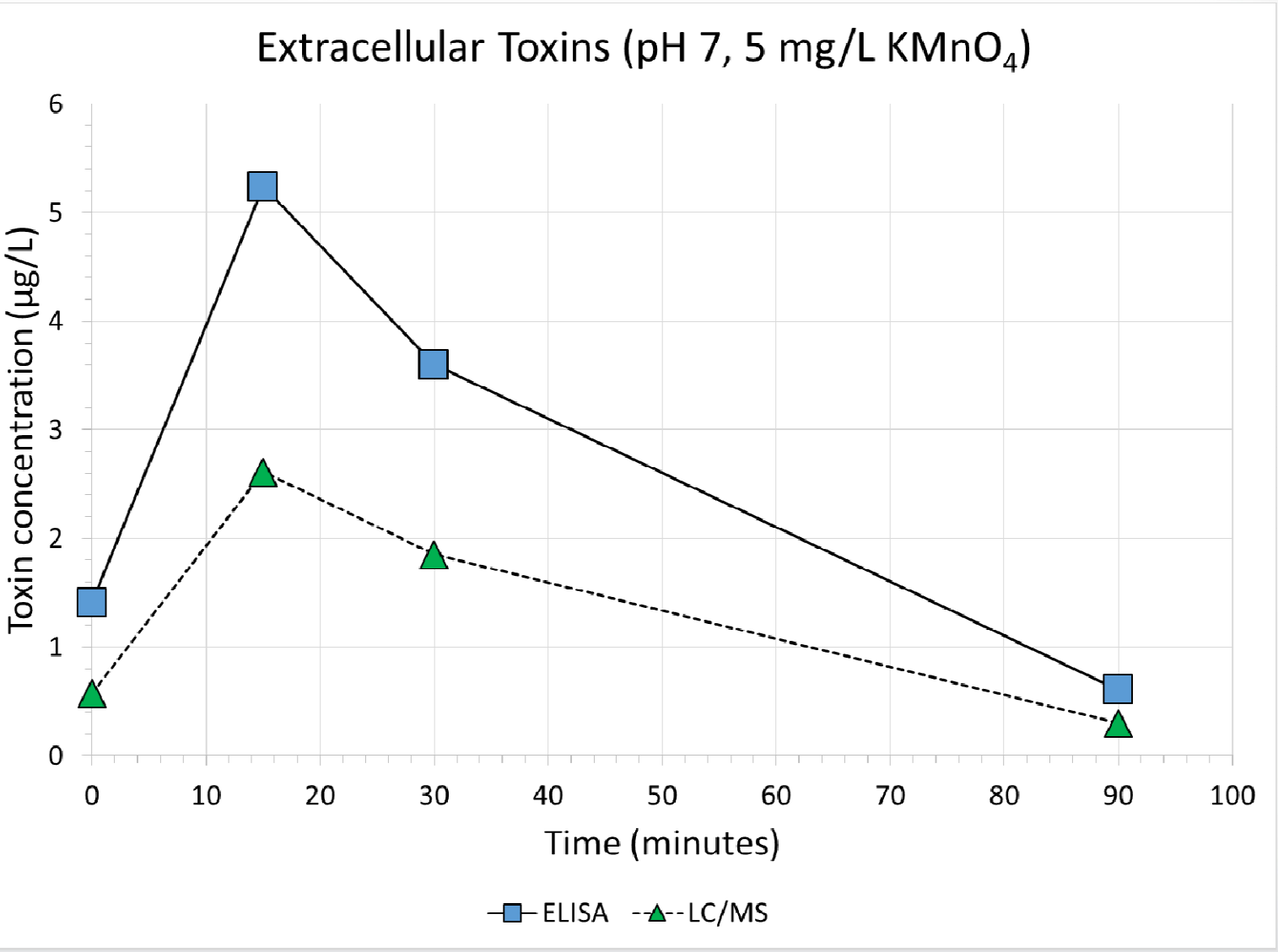
Jar Test Results



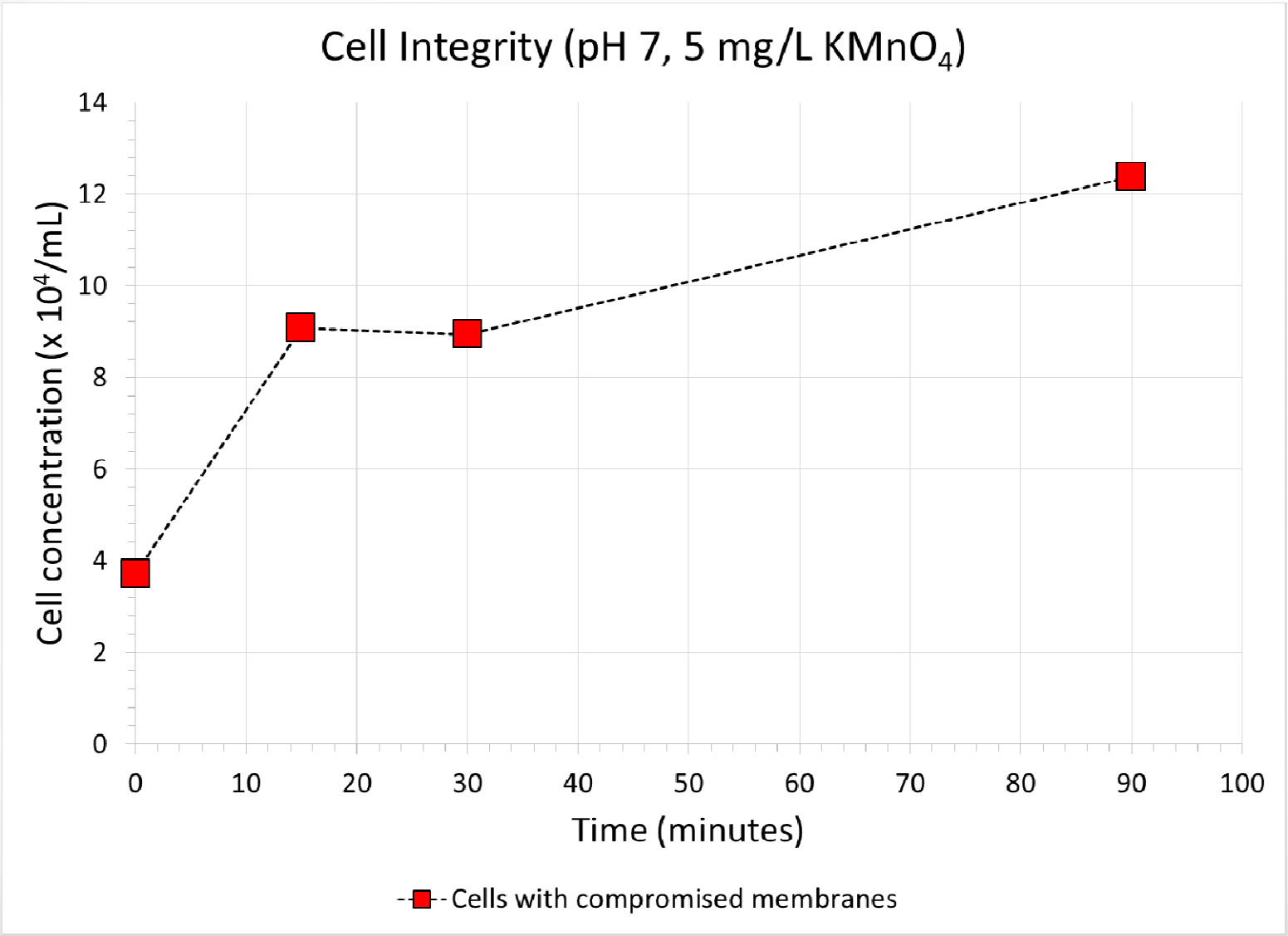
Jar Test Results



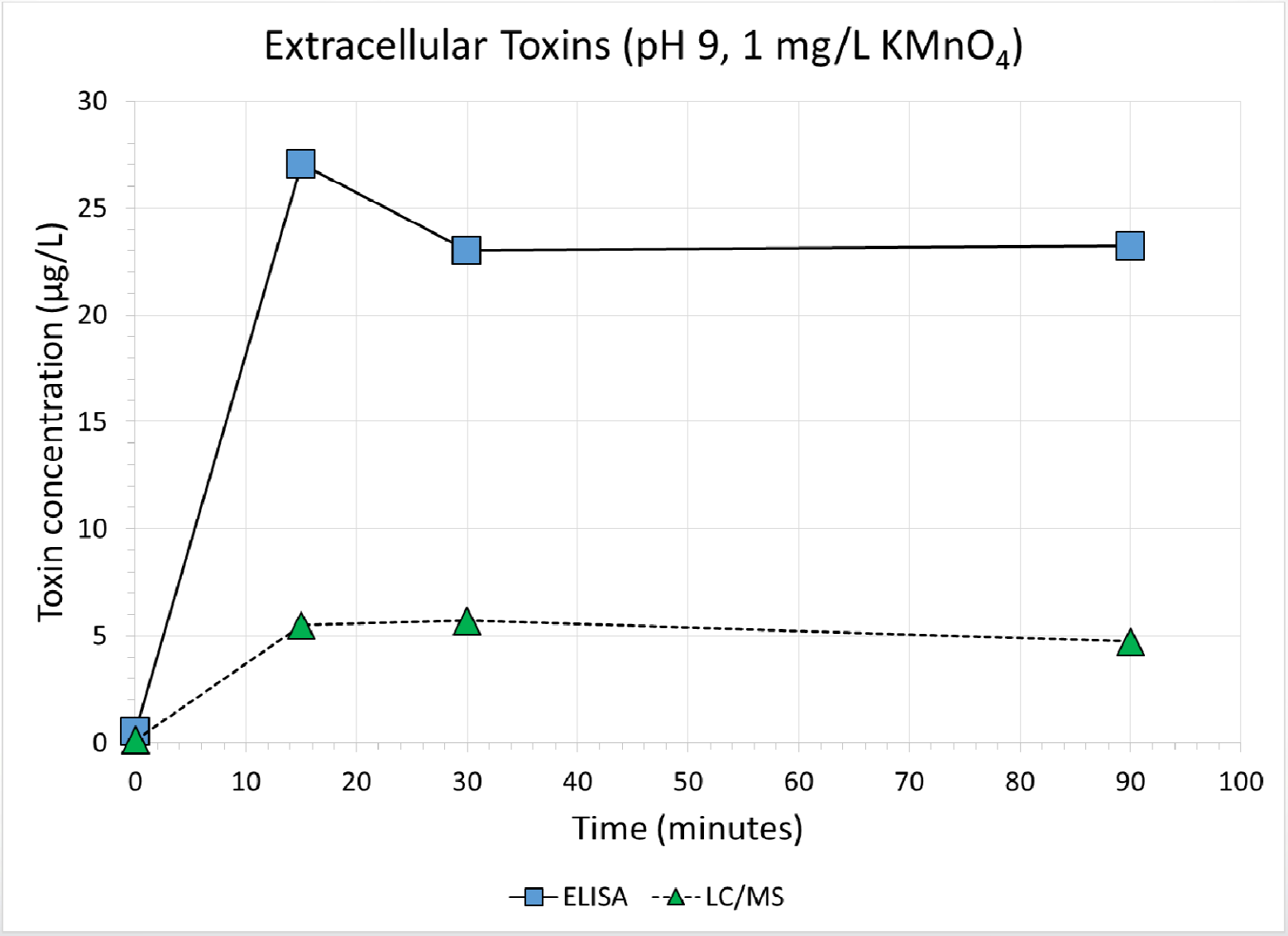
Jar Test Results



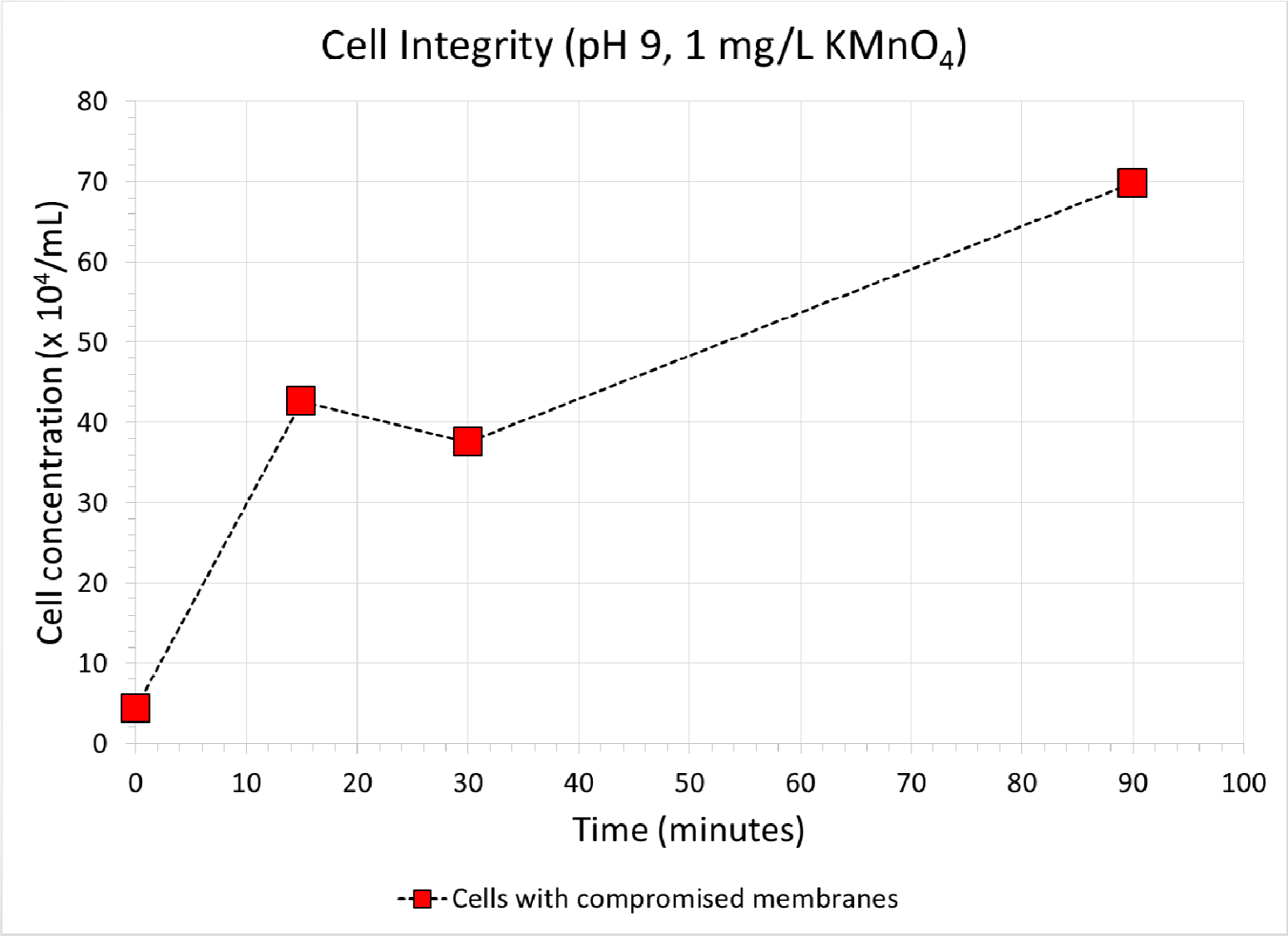
Jar Test Results



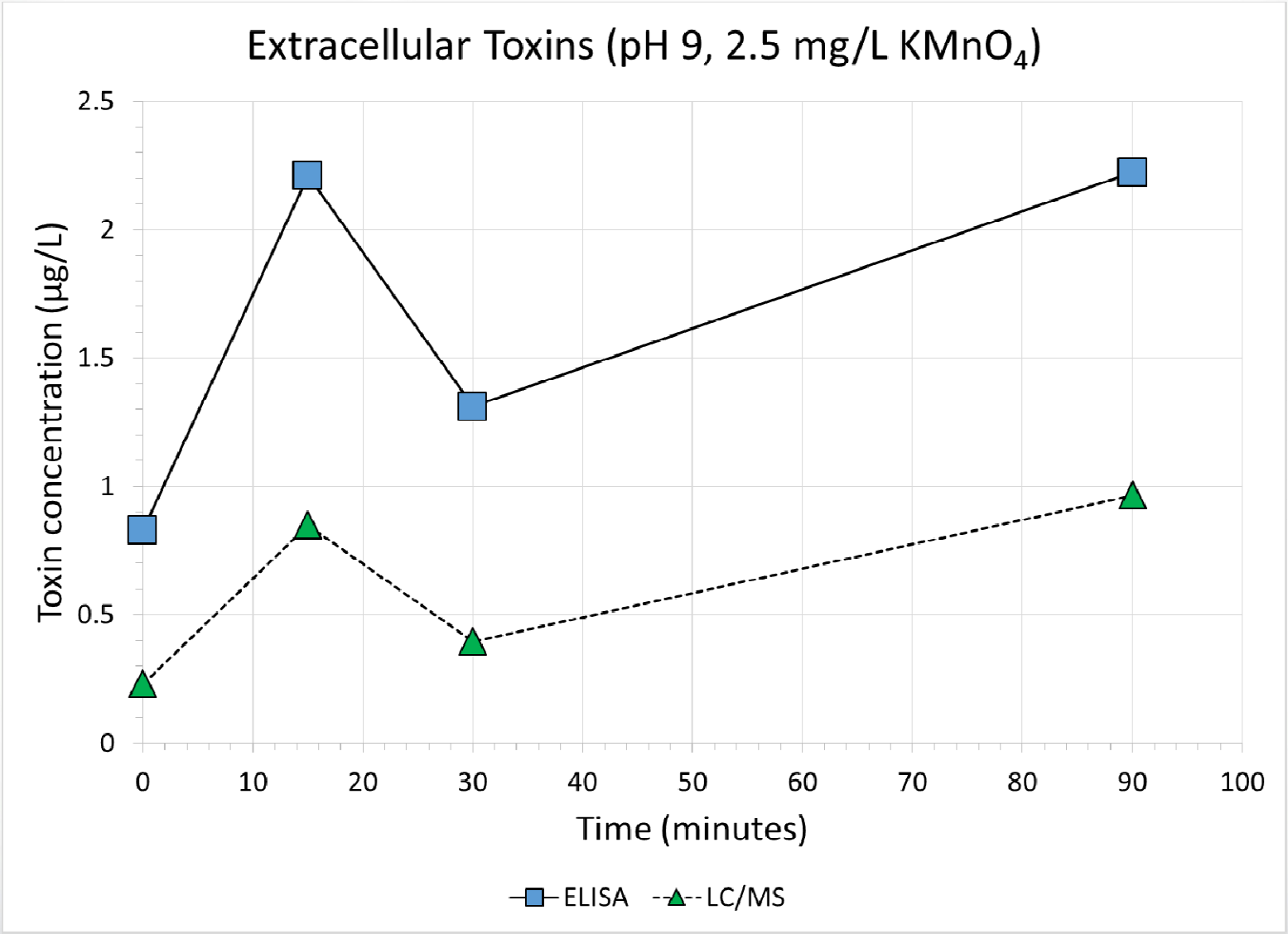
Jar Test Results



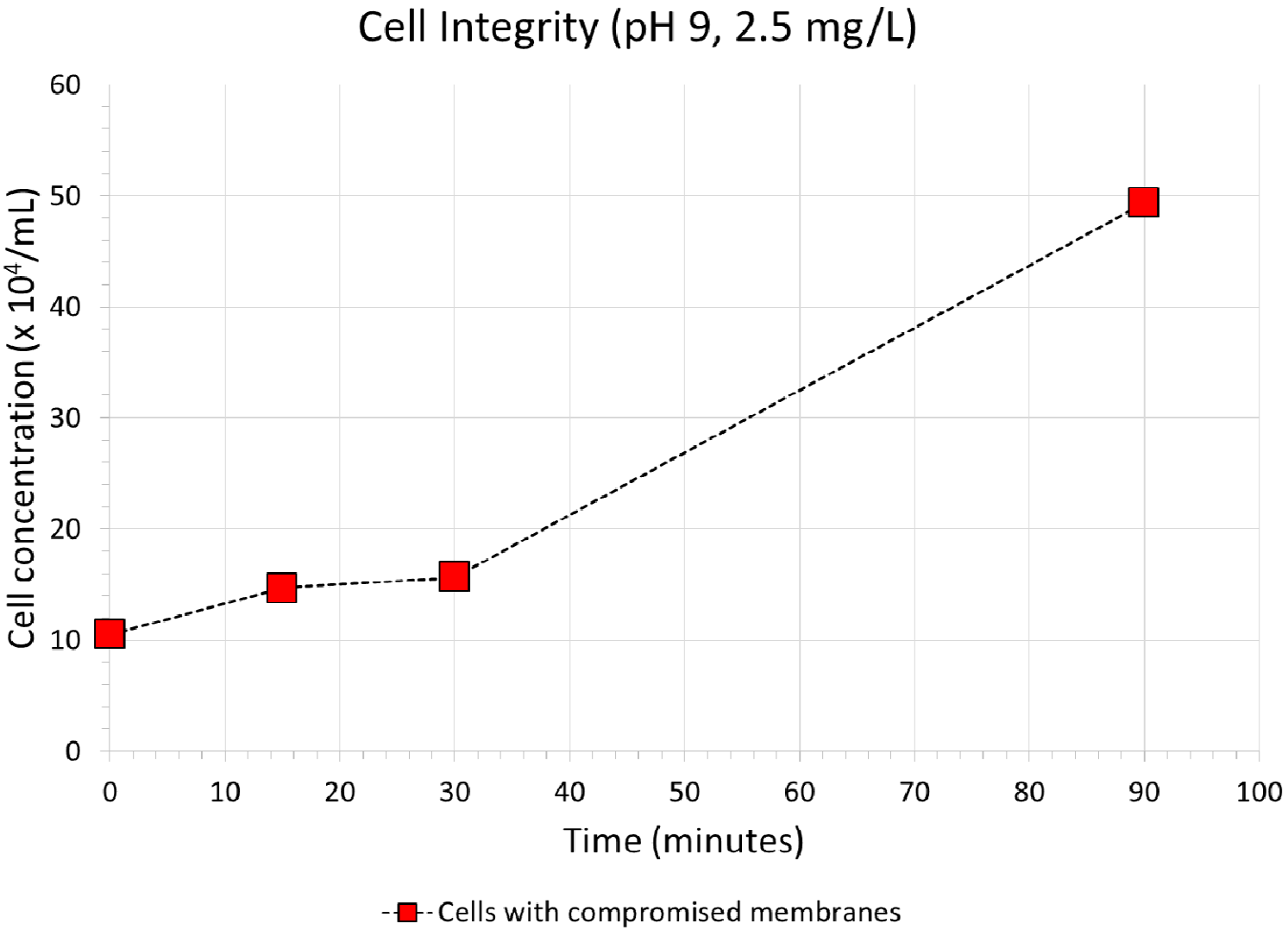
Jar Test Results



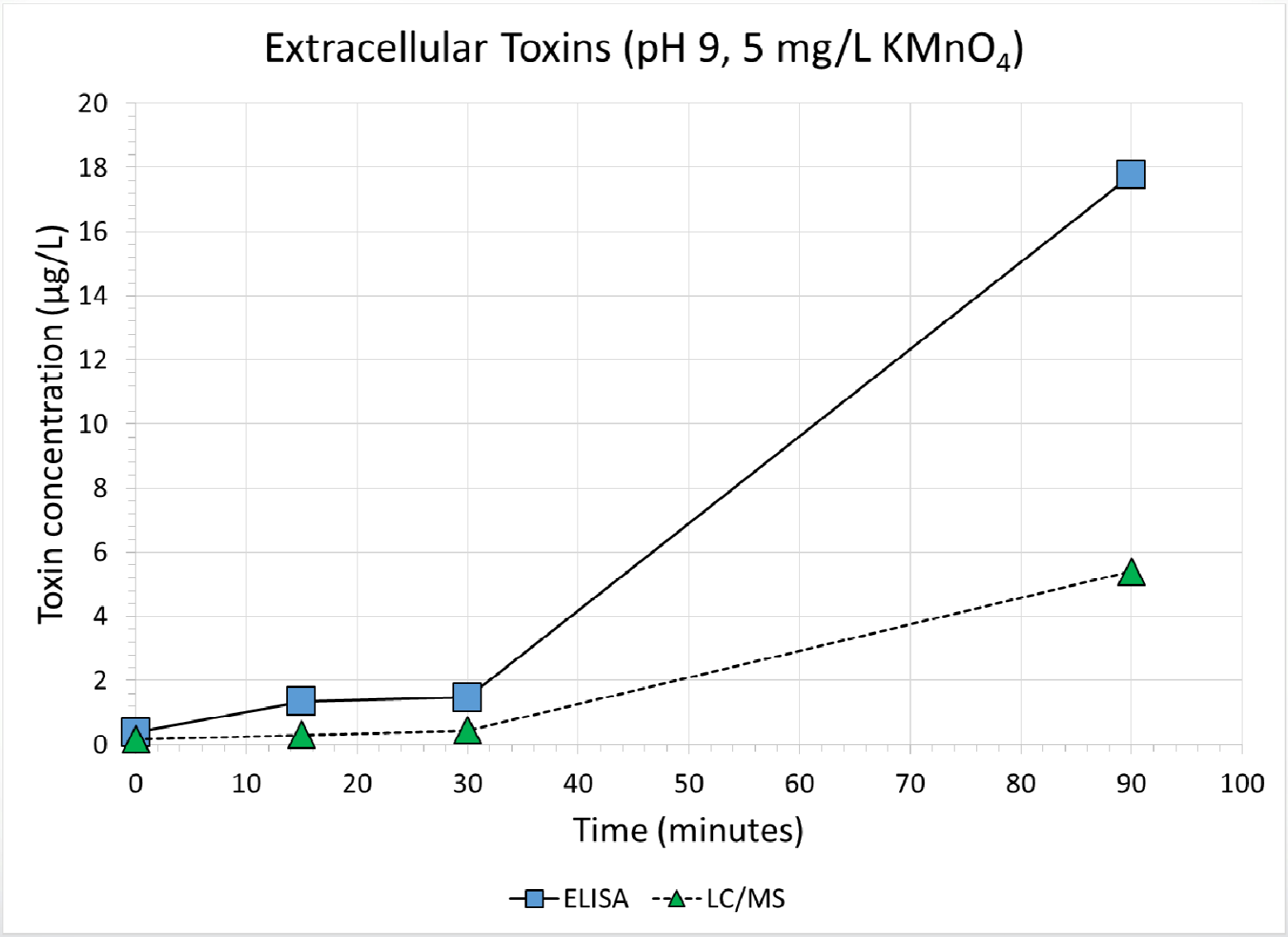
Jar Test Results



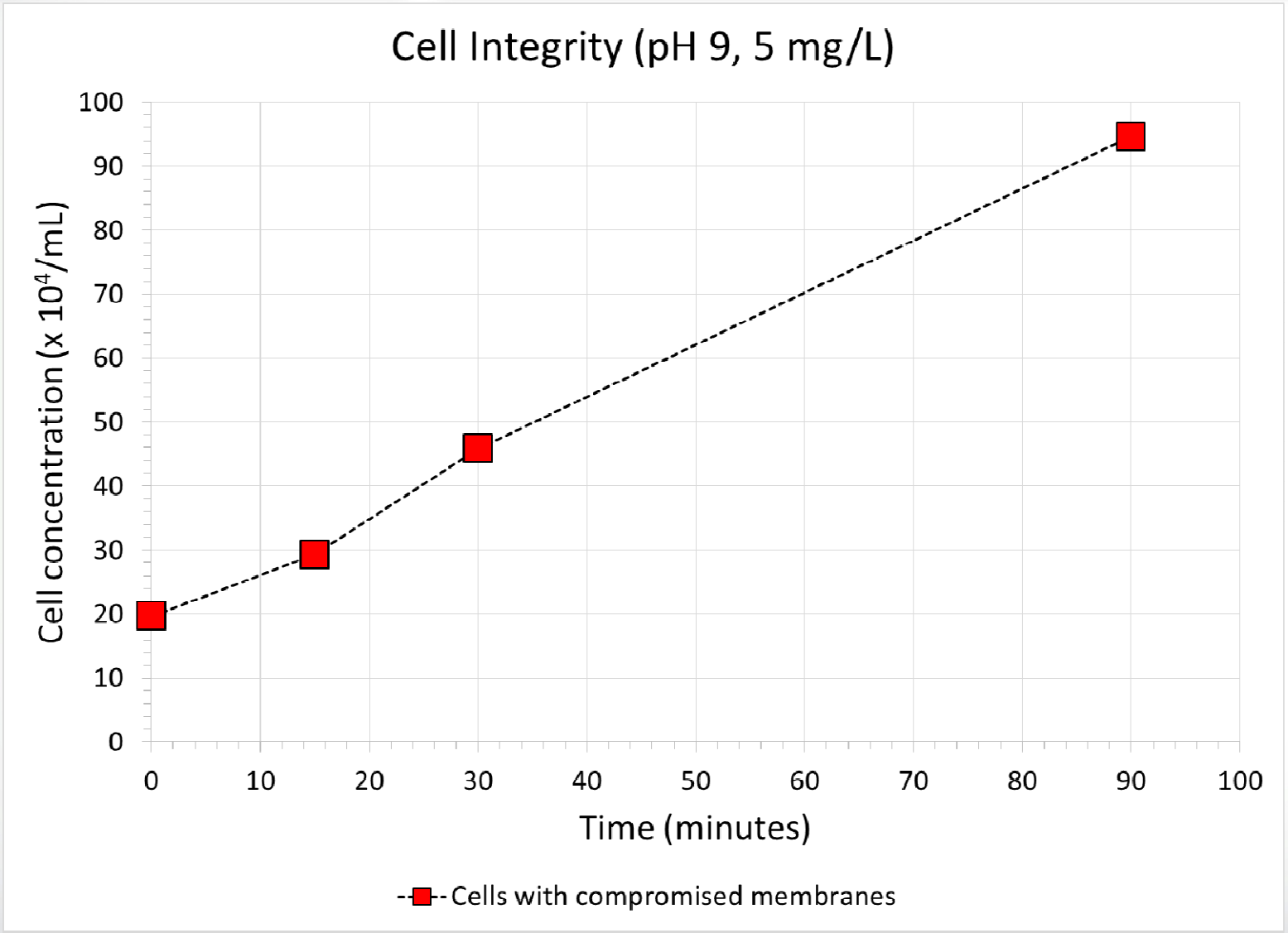
Jar Test Results



Jar Test Results



Jar Test Results



Jar Test Result Highlights

- Increases in extracellular toxin concentrations observed at all dose and pH combinations
- Increased concentrations of membrane-compromised cells observed at all dose and pH combinations

Jar Test Water Quality Parameters

	pH 7 1 mg/L	pH 7 2.5 mg/L	pH 7 5 mg/L	pH 9 1 mg/L	pH 9 2.5 mg/L	pH 9 5 mg/L
Toxin increase (ELISA)	21	15	3.7	55	2.7	47
Toxin increase (LC/MS)	10	11	4.6	51	4.2	32
Total toxin, t = 0 ELISA, µg/L	48	36	10	53	53	33
Total toxin, t = 0 LCMS, µg/L	9.1	8.3	3.8	19	15	9.5
Chloro-A T = 0 µg/L	190	200	150	390	320	300

Toxin Speciation in Lake Erie and US EPA Jar Tests

- Lake Erie blooms produce MYCs-RR, -LR, -YR, -WR, -LY, and -LA
- Jar test microcystins produce primarily MYCs-LR and -LA in a ~83:17 ratio
- Extracellular toxin mixture following release matches total toxin ratio

MYC Cogeners	Lake Erie Sampling, Average Over Season	US EPA Jar Tests
MYC-LA	2.1	17.0
MYC-LR	39.2	83.0
MYC-LY	2.6	0.0
MYC-RR	43.7	0.0
MYC-YR	9.5	0.0
MYC-WR	2.9	0.0

Conclusions and Recommendations

- Majority of toxins in DWTP influents were intracellular
- Increases in extracellular toxin concentrations, along with moderate decreases in total toxin levels were observed following $KMnO_4$ addition
- Follow-up jar tests support observation of toxin release following $KMnO_4$
- Jar test LC/MS/MS and ELISA results tend to show comparable relative changes in toxin concentrations.
- Implementation of increased permanganate concentrations at the full-scale should be undertaken with caution:
  - Toxin sampling around point of permanganate application
  - Contingency plan in case increased extracellular toxin concentrations are observed

Planned Work – 2015 Bloom Season

- Results from 2013 and 2014 suggest HAB activity focused on the Western Basin
- Cincinnati lab is committed to supporting monitoring efforts in both Lake Erie and US EPA Region 8 during the 2015 bloom season and continuing bench-scale studies on effects of permanganate on toxin release
- Increase in influent sampling frequency to catch and monitor bloom activity
  - Monthly monitoring in May and June, with bi-weekly sampling in July and beyond. Based on chlorophyll-a and phycocyanin results sampling will increase to weekly
  - When bloom activity appears to be peaking sampling frequencies will intensify

Disclaimer

U.S. Environmental Protection Agency, through its Office of Research and Development, funded and managed, or partially funded and collaborated in, the research described herein. It has been subjected to the Agency's peer and administrative review and has been approved for external publication. Any opinions expressed in this paper are those of the author(s) and do not necessarily reflect the views of the Agency, therefore, no official endorsement should be inferred. Any mention of trade names or commercial products does not constitute endorsement or recommendation for use.

Thank You!

Nicholas Dugan
513-569-7239
dugan.nicholas@epa.gov

Toby Sanan
513-569-7667
sanan.toby@epa.gov