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CEE 697z
Organic Compounds in Water and Wastewater

Cyanotoxins
WQ Modeling & Degradation in Lakes

Lecture #32

Australian Study

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Determination of rate constants and half-lives for the simultaneous biodegradation of several cyanobacterial metabolites in Australian source waters

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Lake Burragorang

- Microcystin (MCLR)
- Cylindrospermopsin (CYN)
- Saxitoxin (STX)

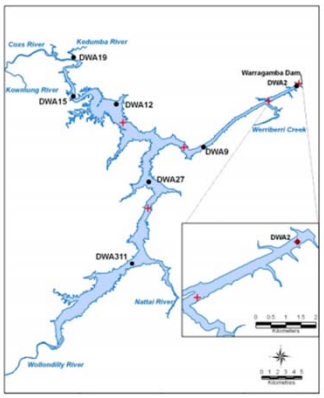
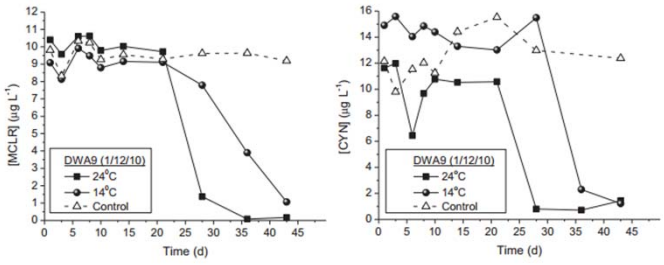


Fig. 1 – Map of Lake Burragorang. Water was sampled from DWA2 (dam wall) and DWA9 for the laboratory experiments.

Lab Tests



- Controls were autoclaved

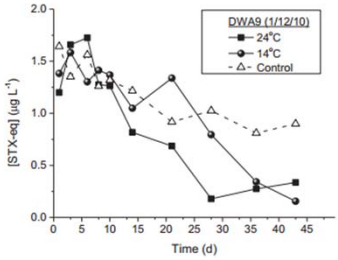


Fig. 2 – Examples of cyanobacterial metabolite biodegradation in water sampled from Lake Burragorang at location DWA9 during summer.

Inoculated Tests

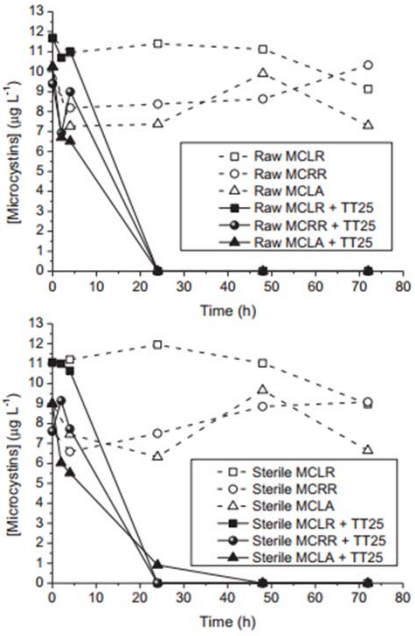


Fig. 4 – Inoculation of microcystin-degrading bacterium, TT25, into raw and sterile DWA2 water spiked with MCLR, MCRR and MCLA.

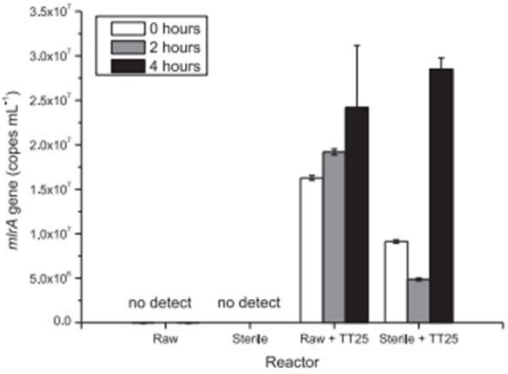


Fig. 5 – Abundance of *mlrA* gene during the inoculation of microcystin-degrading bacterium, TT25, into raw and sterile DWA2 water spiked with MCLR, MCRR and MCLA. Error bars represent 95% confidence intervals from duplicate measurements.

Ontario Lake Study

- The study site is a small (~13.2 ha) and shallow (depth: mean 2.4 m, max 4.5 m)
- privately owned lake, south of the City of Ottawa, Canada
- The lake is polymictic and has 4–5 months of ice cover.
- The site was originally a stone quarry, has no inflow or outflow channel, and the principal water input and output are precipitation and avapotranspiration.
- The water residence time was estimated to be approximately 1.8 years using precipitation and evapotranspiration rates obtained from the National Climate Data and Information Archive and The Hydrological Atlas of Canada. The sediments are composed of gravel and sand with little organic matter accumulation

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Fate and Persistence of Particulate and Dissolved Microcystin-LA from *Microcystis* Blooms

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ABSTRACT

Few studies have estimated fate and persistence of the hepatotoxic microcystins (MCs) *in situ*, making ecological and human health risk assessments challenging. We determined fate and persistence of MC congeners during 2 years of *Microcystis* blooms in a small, shallow, closed-basin lake in Ontario, Canada. *In situ* half-lives were compared to estimates obtained *in vitro* under controlled temperature and light. The blooms produced elevated microcystin-LA (MC-LA) (maximum ~4.2 mg L^{-1}) with minor concentrations of MC-LR, -RR, and -YR. Dissolved MC-LA declined more slowly and persisted longer than particulate MC-LA with *in situ* half-lives (total 1.5–8.5 days) shorter than *in vitro* (total 6.8–60.0 days). Half-lives in 2010 were two to eight times shorter compared to 2009, likely due to differences in bloom phenology and species/strain composition. *In vitro*, higher temperature (4°C → 25°C in dark), and irradiance (dark → 45 → 260 μE $m^{-2}s^{-1}$ at 25°C) accelerated particulate and dissolved MC-LA decline, respectively. MC-RR accumulated in surface sediments while MC-LA was near detection despite elevated surface water concentrations. MC-LA appears to persist longer in surface waters than the equally toxic MC-LR, requiring almost the entire recreational season (9.5 weeks) to reach guideline concentrations (20 μg L^{-1}).

MC Congeners

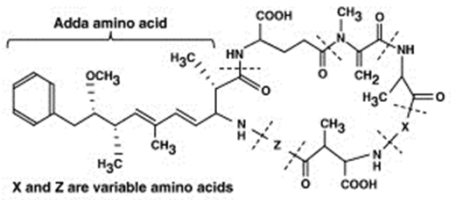

Table 1. Comparison of the properties of microcystin congeners.

	MC-LA	MC-LR	MC-YR	MC-RR
Toxicity (LD_{50} μg/kg)	50	50	70	600
Net charge (pH 7)	−2	−1	−1	0
Molecular weight	909	994	1044	1037
Amino acid substituents	Leu, Ala	Leu, Arg	Tyr, Arg	Arg, Arg
Hydrophobicity	Decreasing → → → →			

Adapted from Newcombe *et al.* (2003).

Fate and Persistence of Microcystin-LA

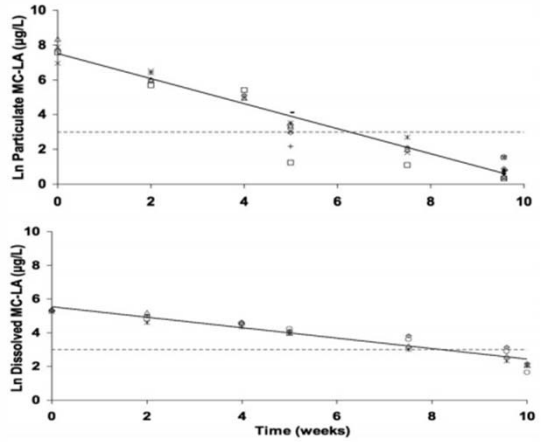

Figure 1. *In situ* (whole lake) decline of particulate (cell-bound) ($R^2 = 0.98$, $p < .001$) and dissolved (extracellular) ($R^2 = 0.97$, $p < .001$) MC-LA ($\mu g\ L^{-1}$). Each symbol represents a different station of the lake sampled through time in 2009. Dashed horizontal line denotes recreational guideline of 20 $\mu g\ L^{-1}$, which corresponds to a moderate probability of adverse health effects during recreational exposure (WHO 2006; Chorus and Bartram 1999; Codd *et al.* 2005).

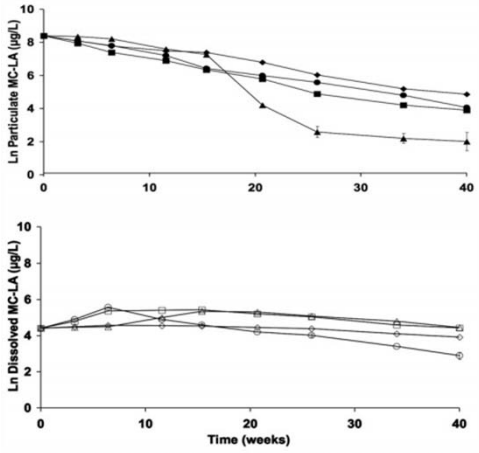

Figure 2. The decline of particulate (cell-bound; filled symbols) and dissolved (extracellular; open symbols) MC-LA ($\mu g\ L^{-1}$) under *in vitro* conditions of 4°C dark (diamonds), 25°C dark (triangles), 25°C low light (squares), and 25°C high light (circles). Duplicate incubations were carried out using bloom material collected in 2009.

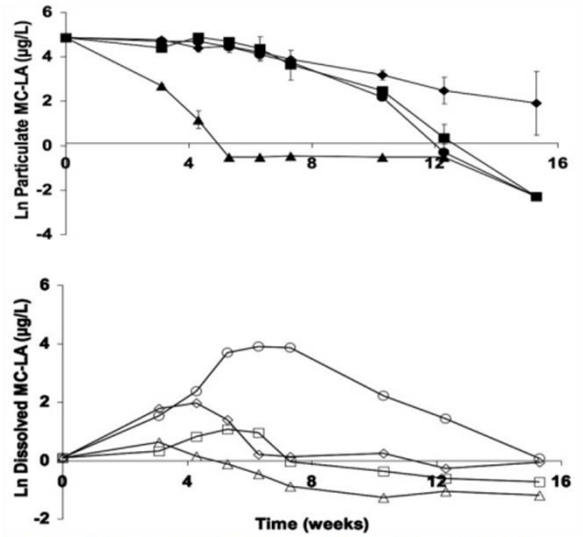

Figure 3. Decline of particulate (cell-bound; filled symbols) and dissolved (extracellular; open symbols) MC-LA ($\mu g\ L^{-1}$) under *in vitro* conditions of 4°C dark (diamonds), 25°C dark (triangles), 25°C low light (squares), and 25°C high light (circles). Duplicate incubations were carried out using bloom material collected in 2010.

- Half lives in the 2 blooms

Table 2. Estimates of MC-LA half-life based on the May 2009 *Microcystis* bloom (±SD).

	Half life of microcystin-LA (days)		
Conditions	Particulate	Dissolved	Total
In situ (n = 5)	6.5 ± 0.4	15.8 ± 1.0	8.5 ± 0.5
In vitro (n = 2)			
25°C (260 μE m^{-2} s^{-1})	44.9 ± 0.7	63.5 ± 5.3	47.4 ± 1.0
25°C (45 μE m^{-2} s^{-1})	42.8 ± 0.7	120.4 ± 1.0	55.6 ± 0.7
25°C (Dark)	23.8 ± 2.4	131.5 ± 7.5	41.6 ± 0.2
4°C (Dark)	54.6 ± 0.5	251.0 ± 35.9	60.0 ± 0.1

Table 3. Estimates of MC-LA half-life based on the August 2010 *Microcystis* bloom (±SD).

	Half life of microcystin-LA (days)		
Conditions	Particulate	Dissolved	Total
In situ (n = 5)	1.5 ± 0.03	2.8 ± 0.3	1.5 ± 0.1
In vitro (n = 2)			
25°C (260 μE m^{-2} s^{-1})	9.2 ± 0.7	10.9 ± 0.3	11.6 ± 0.3
25°C (45 μE m^{-2} s^{-1})	10.5 ± 0.9	26.5 ± 0.9	9.4 ± 1.0
25°C (Dark)	5.0 ± 0.1	33.8 ± 2.2	6.8 ± 0.04
4°C (Dark)	24.2 ± 1.3	31.3 ± 1.8	25.7 ± 4.2

- To next lecture