

Comparative Assessment of Physical and Chemical Cyanobacteria Cell Lysis Methods for Total Microcystin-LR Analysis

Katherine E. Greenstein, Arash Zamyadi and Eric C. Wert

MATERIALS AND METHODS

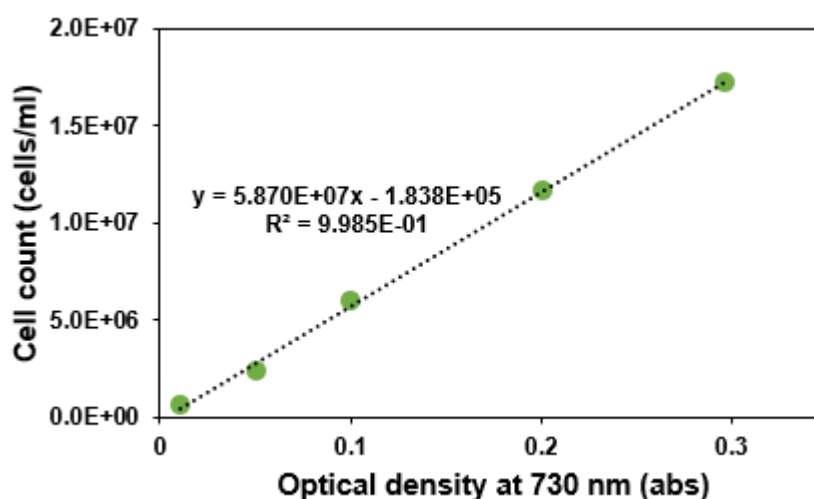


Figure S1. Correlation of cell count (cells/mL) versus optical density at 730 nm (OD730). Laboratory-cultured *M. aeruginosa* cells were counted using digital flow cytometry.

RESULTS AND DISCUSSION

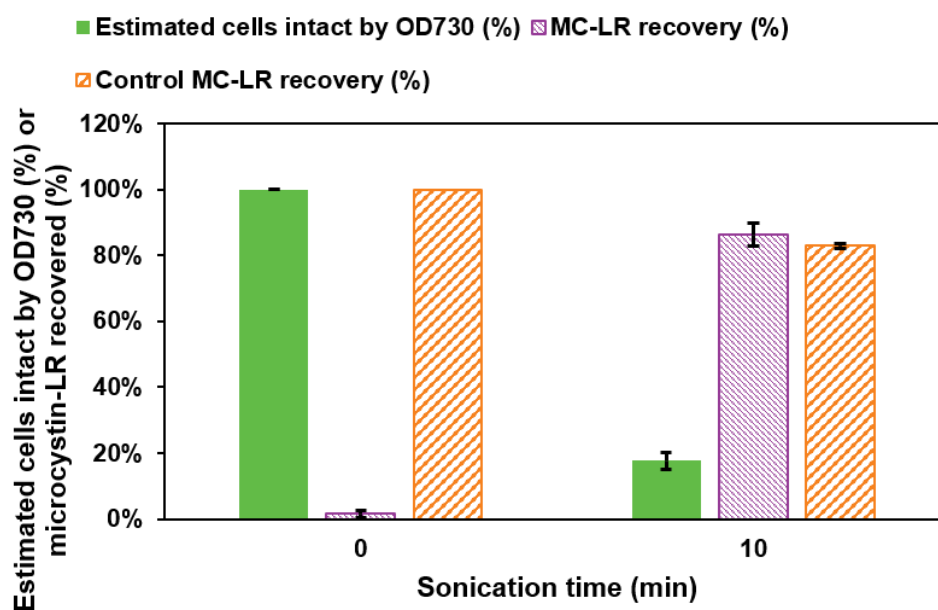


Figure S2. Estimated cells intact by OD730 (%; solid green) or MC-LR recovered (%; striped purple) versus sonication time (minutes). Error bars represent standard deviation from duplicate experiments. Sonication was tested on laboratory-cultured *M. aeruginosa* suspended in Colorado River water (CRW) at high ($\sim 10^7$ cells/mL) cell density. Recovery of MC-LR from controls in CRW (no cells present) is also shown (%; striped orange).

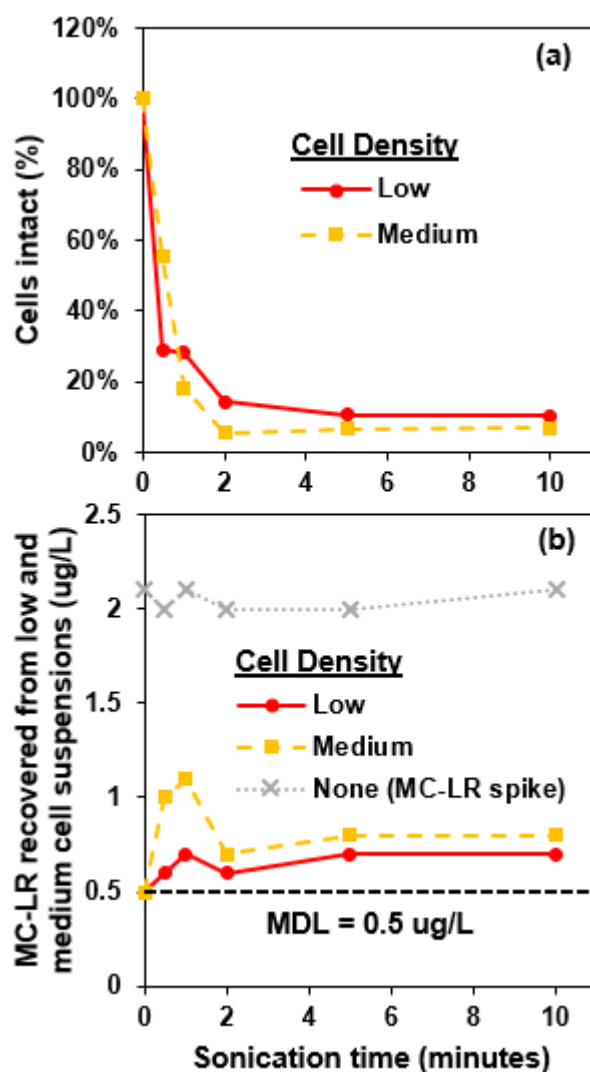


Figure S3. (a) Estimated cells remaining intact (%) and (b) microcystin-LR (MC-LR) recovery (ug/L) versus sonication time (minutes) for laboratory-cultured *M. aeruginosa* in a Canadian laboratory (Polytechnique Montréal) for cross-laboratory validation. Data are shown for low ($\sim 10^5$ cells/mL, red circles) and medium ($\sim 10^6$ cells/mL, yellow squares) density suspensions of laboratory-cultured *M. aeruginosa* in phosphate buffer. The MC-LR-spiked control solution (with no cells) is represented by the grey dotted line.

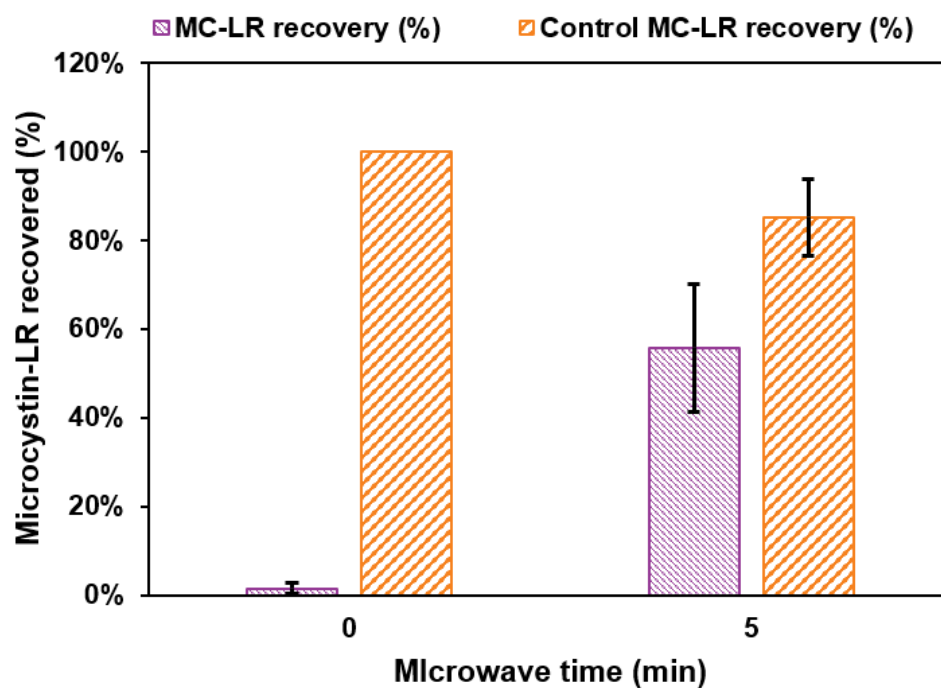


Figure S4. MC-LR recovered (%) versus microwave time (minutes). Error bars represent standard deviation from duplicate experiments. Sonication was tested on laboratory-cultured *M. aeruginosa* suspended in Colorado River water (CRW) at high ($\sim 10^7$ cells/mL) cell density. Recovery of MC-LR from controls in CRW (no cells present) is also shown (% , striped orange).

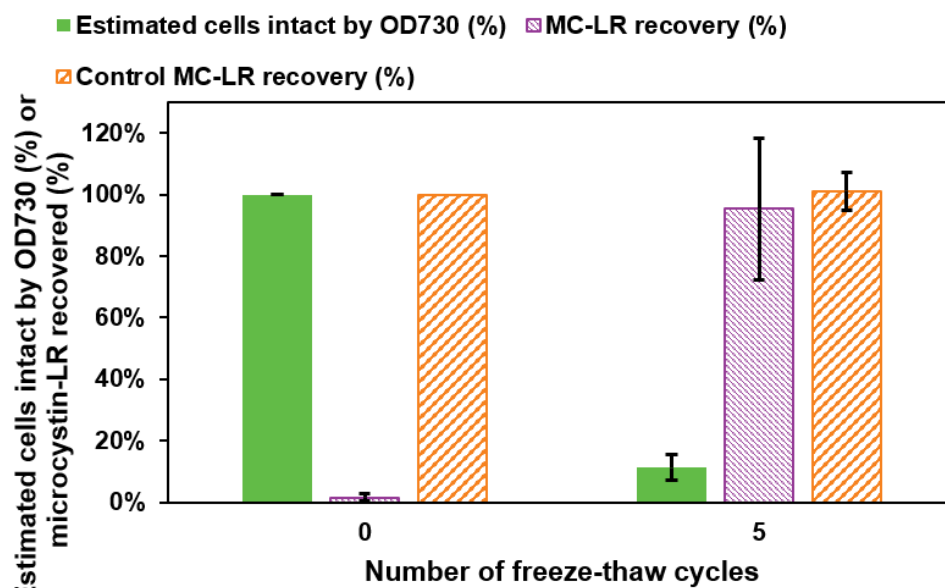


Figure S5. Estimated cells intact by OD730 (%; solid green) or MC-LR recovered (%; striped purple) versus sonication time (minutes). Error bars represent standard deviation from duplicate experiments. Sonication was tested on laboratory-cultured *M. aeruginosa* suspended in Colorado River water (CRW) at high ($\sim 10^7$ cells/mL) cell density. Recovery of MC-LR from controls in CRW (no cells present) is also shown (%; striped orange).

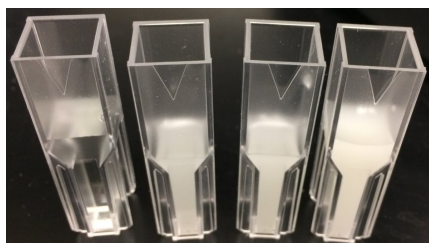


Figure S6. Image of cuvettes containing solution with no Abraxis QuikLyse™ reagents (left) with increasing amounts of reagents as cuvettes progress to the right. A white precipitate formed with the addition of reagents, which necessitated allowing samples to settle, even after filtering with Abraxis kit-enclosed filters, prior to MC-LR analysis via liquid chromatography-tandem mass spectrometry (LC-MS/MS).

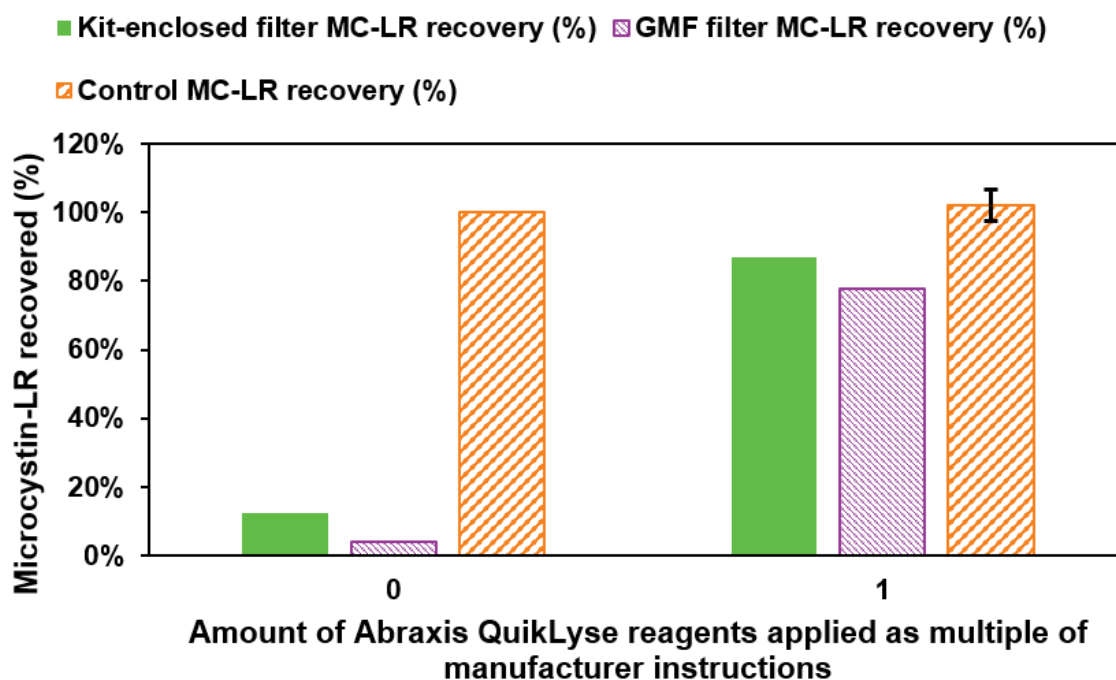


Figure S7. MC-LR recovered (%) versus use of Abraxis QuikLyse™ reagents. Both kit-enclosed filters (solid green) and glass microfiber (GMF) filters (striped purple) were separately examined. The chemical lysis was tested on laboratory-cultured *M. aeruginosa* suspended in Colorado River water (CRW) at high ($\sim 10^7$ cells/mL) cell density. Recovery of MC-LR from controls in CRW (no cells present) is also shown (%; striped orange).