

Analysis of Microcystins from Blue-green Algae Using the TSQ Quantum Ultra LC-MS/MS System

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Key Words

- TSQ Quantum Ultra™
- Blue-green algae
- LC-MS/MS
- Microcystin
- SRM

Introduction

Overgrowth of algae is a common problem in many wetlands with advanced stages of eutrophication (the enrichment of chemical nutrients containing nitrogen or phosphorus in an ecosystem). This often results in a thick, colored layer on the water's surface, known as an algal bloom. Some of the algae that grow in these bodies of water, known as Cyanobacteria or blue-green algae, produce toxic compounds known as microcystins.

Microcystins have a ring peptide structure consisting of seven amino acids, and more than 80 homologs are known. One of the most widely studied of the microcystins is known as Microcystin-LR, and is shown in Figure 1. Many of the microcystins are particularly toxic to the liver. (See References.) Among them are Microcystin-LR, YR and RR, which have been detected in wetlands in Japan. This application note reports on the analysis of these microcystins by using LC-MS/MS.

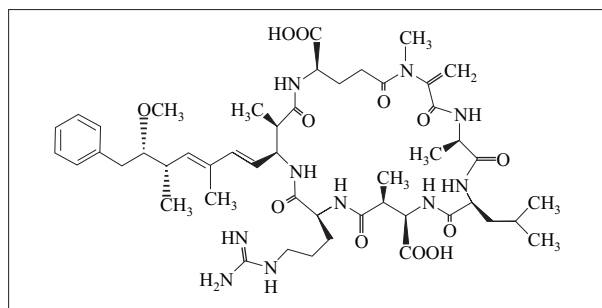


Figure 1: Microcystin-LR

Method

HPLC: HTC PAL Autosampler and Surveyor™ MS pump

Column: HyPURITY™ C18 2.1×50 mm, 5 μ (Thermo Scientific)

Mobile Phase A: Water with 0.1% Formic Acid

Mobile Phase B: Acetonitrile

Gradient: 30%B (0.5 min) → 80%B (in 3 min) → 80%B (2 min hold) → 30%B (7 min hold)

Injection Volume: 20 μL

Flow: 0.2 mL/min

Column temperature: Room temperature

MS: TSQ Quantum Ultra

Ionization: Positive ESI

Spray voltage: 5000 V

Sheath gas: 45 arbitrary units

Auxiliary gas: 15 arbitrary units

Sweep gas: 2 arbitrary units

Capillary T: 350 °C

Source CID: Off

Collision gas: Ar, 1.2 mTorr

Scan Time: 0.15 sec

SRM setting: 519.9 → 135.0 @ 32 V (RR)

995.7 → 135.0 @ 65 V (LR)

1045.8 → 135.0 @ 70 V (YR)

SRM Chromatogram (STD 1.0 ppb)

The SRM chromatograms for 1.0 ppb standards are shown in Figure 2. The linear calibration curves of the standards (0.1 ppb–1.0 ppm) are shown in Figure 3.

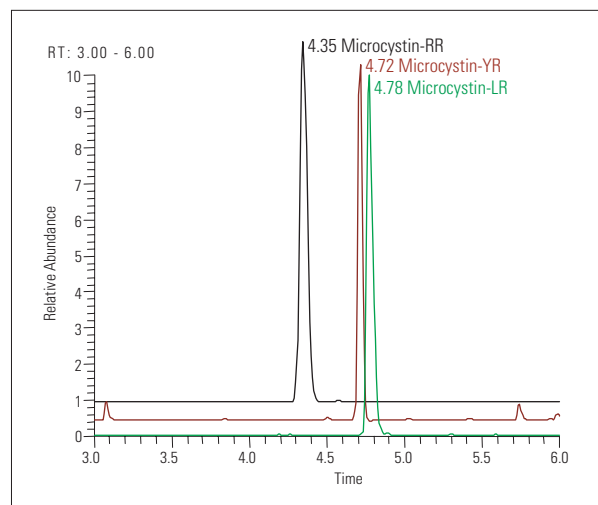


Figure 2: SRM Chromatogram (RT 4.35: Microcystin-RR, RT 4.72: Microcystin-YR, RT 4.78: Microcystin-LR)

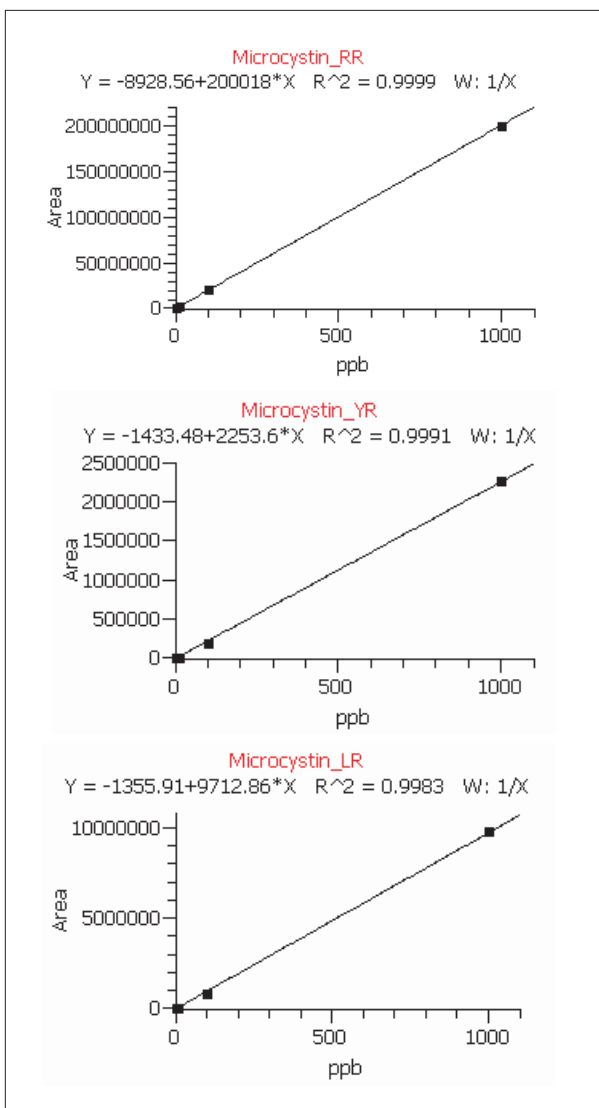


Figure 3: Calibration Curves 0.1 ppb – ~1.0 ppm

Conclusion

Microcystin-LR, YR and RR can be quantitatively analyzed over four orders of dynamic range (0.1 ppb–1.0 ppm) by using the TSQ Quantum Ultra triple quadrupole LC-MS/MS system from Thermo Fisher Scientific.

References

- T Ohta; R Nishiwaki; M Suganuma; J Yatsunami; A Komori; S Okabe; M Tatematsu; H Fujiki. 1993. Significance of the Cyanobacterial Cyclic Peptide Toxins, the Microcystins and Nodularin, in Liver-Cancer. *Mutation Research*, 292:286-287.
- JG Pace; NA Robinson; GA Miura; CF Matson; TW Geisbert; JD White. 1991. Toxicity and Kinetics of [H-3] Microcystin-Lr in Isolated Perfused Rat Livers. *Toxicology and Applied Pharmacology*, 107:391-401.
- R Nishiwaki; T Ohta; E Sueoka; M Suganuma; K Harada; MF Watanabe; H Fujiki. 1994. Two significant aspects of microcystin-LR: Specific binding and liver specificity. *Cancer Lett*, 83:283-289.
- I Falconer; A Jackson; J Langley; M Runnegar. 1980. Liver Pathology of a Toxin from the Bloom-Forming Blue-Green Alga Microcystis Aeruginosa. *Proceedings of the Australian Biochemical Society*, 13:41-41.

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