

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

Mihoko Yamaguchi, Thermo Fisher Scientific, Yokohama, Japan

## 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.

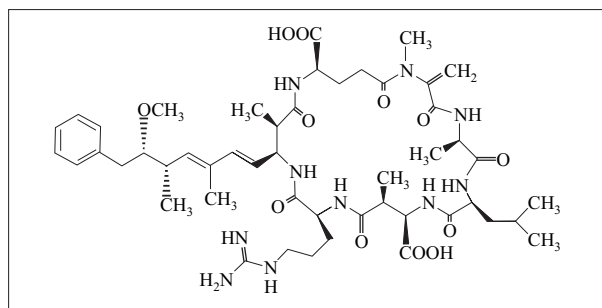


Figure 1: Microcystin-LR

## 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.

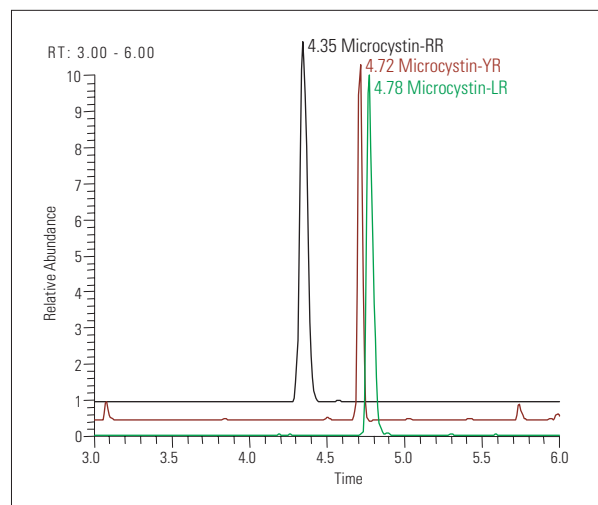


Figure 2: SRM Chromatogram (RT 4.35: Microcystin-RR, RT 4.72: Microcystin-YR, RT 4.78: Microcystin-LR)

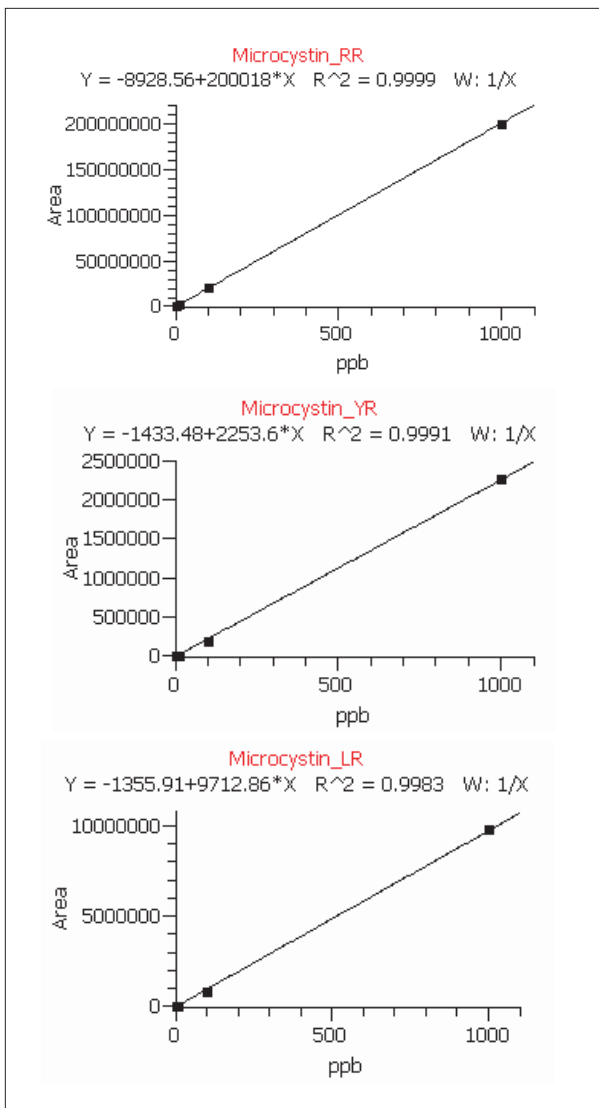


Figure 3: Calibration Curves 0.1 ppb – ~1.0 ppm

## 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.

In addition to these offices, Thermo Fisher Scientific maintains a network of representative organizations throughout the world.

**Australia**  
+61 2 8844 9500

**Austria**  
+43 1 333 50340

**Belgium**  
+32 2 482 30 30

**Canada**  
+1 800 532 4752

**China**  
+86 10 5850 3588

**Denmark**  
+45 70 23 62 60

**France**  
+33 1 60 92 48 00

**Germany**  
+49 6103 408 1014

**India**  
+91 22 6742 9434

**Italy**  
+39 02 950 591

**Japan**  
+81 45 453 9100

**Latin America**  
+1 608 276 5659

**Netherlands**  
+31 76 587 98 88

**South Africa**  
+27 11 570 1840

**Spain**  
+34 91 657 4930

**Sweden/Norway/Finland**  
+46 8 556 468 00

**Switzerland**  
+41 61 48784 00

**UK**  
+44 1442 233555

**USA**  
+1 800 532 4752

[www.thermo.com](http://www.thermo.com)



Thermo Finnigan LLC,  
San Jose, CA USA is ISO Certified.

©2007 Thermo Fisher Scientific Inc. All rights reserved. All trademarks are the property of Thermo Fisher Scientific Inc. and its subsidiaries.

Specifications, terms and pricing are subject to change. Not all products are available in all countries. Please consult your local sales representative for details.

AN62272\_E 01/07S

View additional Thermo Scientific LC/MS application notes at: [www.thermo.com/appnotes](http://www.thermo.com/appnotes)