Analysis of Microcystins by LC/MS

Microcystins are hepatotoxins that are produced by blue-green algae in eutrophied lakes, and they also act as carcinogenic promoters. Hence, it is highly necessary to monitor their concentrations in lakes, and according to the 2003 revised drinking water test methods, microcystin LR, which is produced by various blue-green algae, is one of the required tests. According to the WHO guideline established in 1998, the upper limit of microcystins in drinking water is 1000ng/L.

Because microcystins are peptides with ring-shaped structures, they have been measured by GC/MS or LC-UV following derivatization. However, due to the complex procedures and detection sensitivity, the 2001 revised drinking water test methods adopted LC/MS, which has a lower limit of quantitation of 10ng/L.

Here, the levels of microcystins LR, RR and YR were measured by LC/MS. With the drinking water test methods, pretreatment by solid-phase extraction concentrates samples 500-fold, and as a result, 10 ng/L samples are concentrated to 5µg/L. Figure 2 shows a chromatogram near the limit of quantitation, and detection was sufficient at concentrations of several µg/L. A calibration curve was prepared for each microcystin, and linearity was favorable at a concentration range of 2-50µg/L for LR and RR and 0.8-20µg/L for YR.

Figure 1. Chromatogram for each microcystin (LR, RR: 25µg/L; YR: 10µg/L)

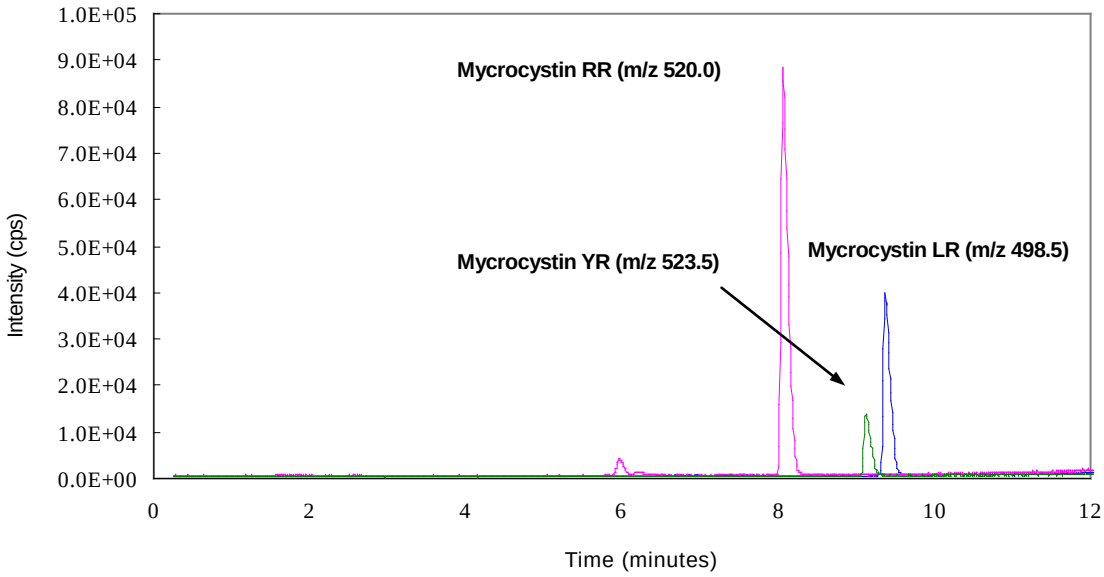

Figure 2. Chromatogram for each microcystin (LR, RR: 2μg/L; YR: 0.8μg/L)

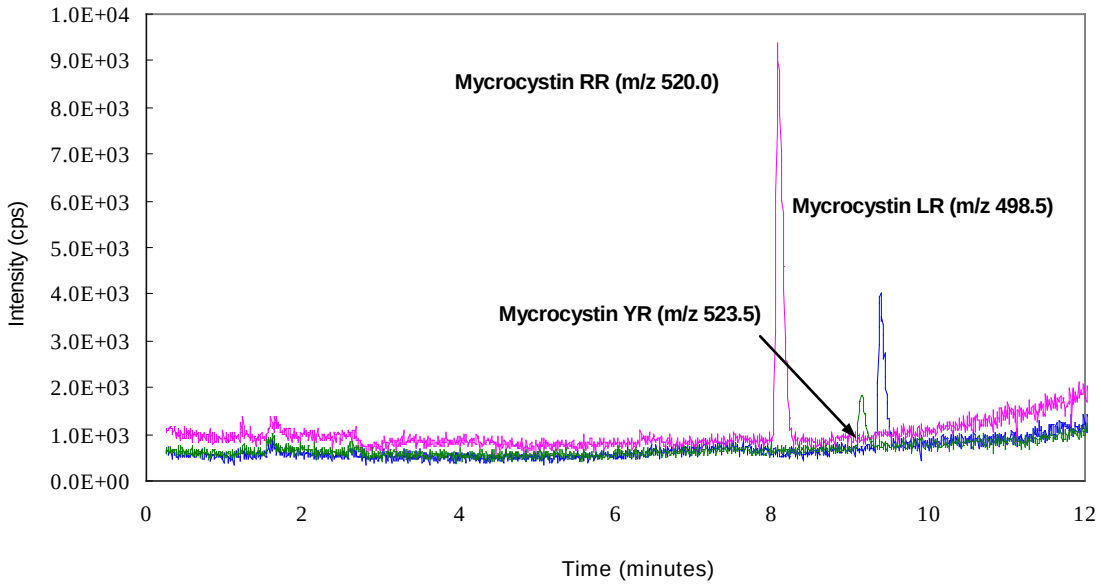

Figure 3. Calibration curve for each microcystin

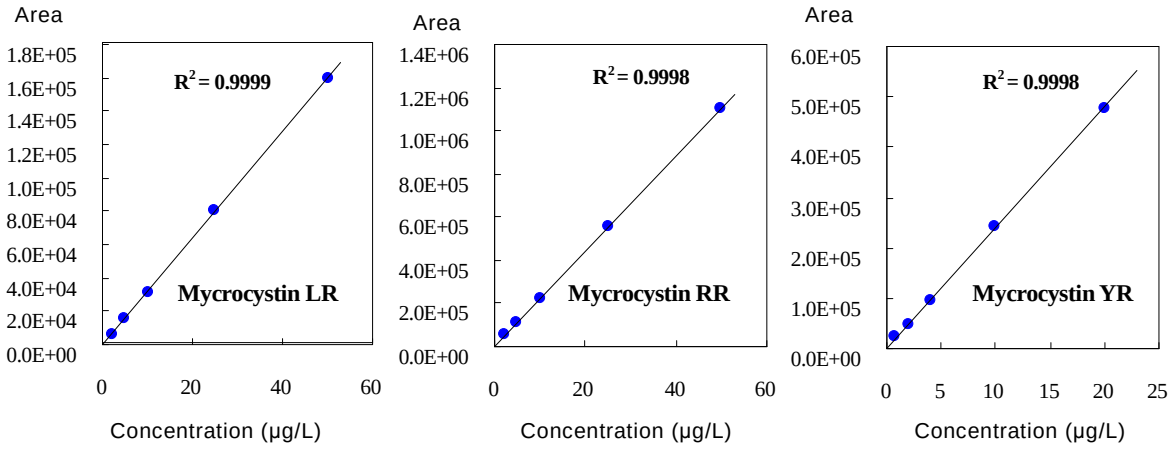

Table 1. Analysis conditions

Column:	**TSKgel ODS-100V, 3µm, 2.0mm ID x 15cm**
Mobile phase:	A: 0.1% formic acid in water B: 0.1% formic acid in acetonitrile
Gradient:	0min (10%B) → 10min (60%B) → 15min (60%B)
Flow rate:	0.2mL/min
Temperature:	40°C
Injection vol.:	5µL
Instrument:	QTRAP® (Applied Biosystems)
Ion Source:	ESI
Polarity:	Positive

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