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Gas Chromatograph Tandem Mass Spectrometer GCMS-TQ™8050 NX

Application News

Enhancing Water Analysis: A Twin Line GC-MS/MS Approach to Pesticides and Nitrosamines Analysis

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User Benefits

◆ Avoid the use of flammable gas (for GCMS-PCI) and radioactive detector (ECD)
◆ Enhanced productivity while achieving high sensitivity for both pesticides and nitrosamines by utilizing Shimadzu GCMS-TQ8050 NX system with Twin Line MS Kit

■ Introduction

Water contamination by nitrosamines and pesticides can occur through various sources, including industrial runoff, agricultural practices, and wastewater discharges[1]. The US EPA recommends specific analytical methods for the analysis of nitrosamines and organochlorine pesticides. Traditionally, GCMS with Positive Chemical Ionization (PCI) mode is used for nitrosamines analysis (EPA 521), while GC with Electron Capture Detector (ECD) is employed for organochlorine pesticides analysis (EPA 8081) [2,3]. However, these techniques necessitate the use of flammable gas or radioactive detector, which requires special permits. Advancements in analytical technology have introduced an alternative approach using Multiple Reaction Monitoring (MRM) mode in a single GC-MS/MS system with the Twin Line MS kit. This innovative approach offers enhanced productivity by allowing 2 columns to be installed in the same instrument, while achieving high sensitivity. The method and quantitation detection limits are evaluated to ensure reliable results.
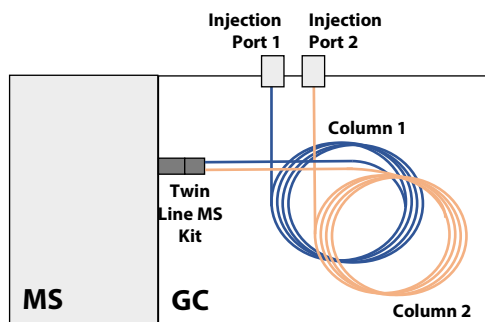

Figure 1: Shimadzu GCMS configuration with twin line MS kit

Shimadzu Twin Line MS Kit

Shimadzu Twin Line MS kit allows for the installation of two columns to the MS simultaneously (Figure 1). With this configuration, there is no need to uninstall and install columns when two different columns are required for various applications in a a single GC-MS/MS system, and thus avoiding downtime and the inconvenience of switching columns.

Additionally, Shimadzu MS is equipped with a high-capacity turbomolecular pump which is capable of accommodating flows from 2 columns without compromising the vacuum level and sensitivity.

■ Experimental

Instrumental and Analytical Conditions

A triple quadrupole GC-MS/MS system, GCMS-TQ8050 NX, (Shimadzu Corporation, Japan), with Twin Line MS Kit was employed in this work. The details of the instrumentation and analytical conditions for both pesticides and nitrosamines analysis are shown in Table 1.

For pesticides, The MRM transitions and collision energies (CE) were either taken from *Shimadzu Smart Pesticides Database* or developed using *Shimadzu MRM Optimization Tool*. For the analysis of nitrosamines, the *Shimadzu MRM Optimization tool was utilized* to develop an optimized MRM method.

Table 1. GC-MS/MS Instrumentation & Analytical Conditions

	Pesticides Analysis	Nitrosamines Analysis
Instrumentation		
GC-MS/MS System	GCMS-TQ8050 NX	
Auto Sampler	AOC-20i Plus & AOC-20s Plus	
Column	SH-I-5Sil MS (P/N 221-75954-30) (30 m x 0.25 mmID x 0.25 μm df)	SH-I-624Sil MS (P/N 221-75962-30) (30 m x 0.25 mmID x 1.40 μm df)
Accessories	Twin Line MS Kit (P/N: 225-20201-91)	
Gas Chromatograph		
Injection Port	SPL	PTV
Injection Volume	2 μL	
Carrier Gas	Helium	
Injection Mode	Splitless	Split
Injection Temperature	250 ºC	37 ºC (0.05 min) → 400 ºC/min to 250 ºC (17.42 min)
Sampling Cond.	Sampling Time = 1 min	Split Ratio =20
Flow Control	Constant Pressure	Linear Velocity
Injection Pressure	100.1 kPa	79.1 kPa
High Pressure Inj.	200.0 kPa	250.0 kPa
Purge Flow	3 mL/min	3 mL/min
Oven Temp. Program	50 ºC (1 min) → 25 ºC/min to 125 ºC → 10 ºC/min to 300 ºC (15 min)	37 ºC (1 min) → 12 ºC/min to 160 ºC → 5ºC/min to 175 ºC → 30ºC/min to 300 ºC (2 min)
Mass Spectrometer		
Ion Source Temp.	230 ºC	230 ºC
Interface Temp.	250 ºC	280 ºC
Detector Voltage	Relative to the Tuning Result + 0.6 kV	
MS Acquisition	MRM	

■ Pesticides Analysis Results

Detection and Separation

The target pesticides were introduced into the GC-MS/MS system through a Split/Splitless (SPL) injection port. The separation of these target pesticides was achieved using the SH-I-5Sil MS column. The MRM acquired mass chromatograms of all target pesticides at 10 ng/mL are shown in Figure 2.

Calibration Curve and Linearity

External standard calibration was utilized for pesticides quantitative analysis. The external standard calibration points of each pesticide prepared were 5, 10, 20, 50, 100, 150, and 200 ng/mL. Figure 3 shows the external standard calibration curves of all target pesticides, demonstrating excellent linearity with R^2 of at least 0.994 with %RSD of response factor (RF) of below 15%. Table 2 summarizes the retention time, R^2 value, and %RSD RF of all target pesticides.
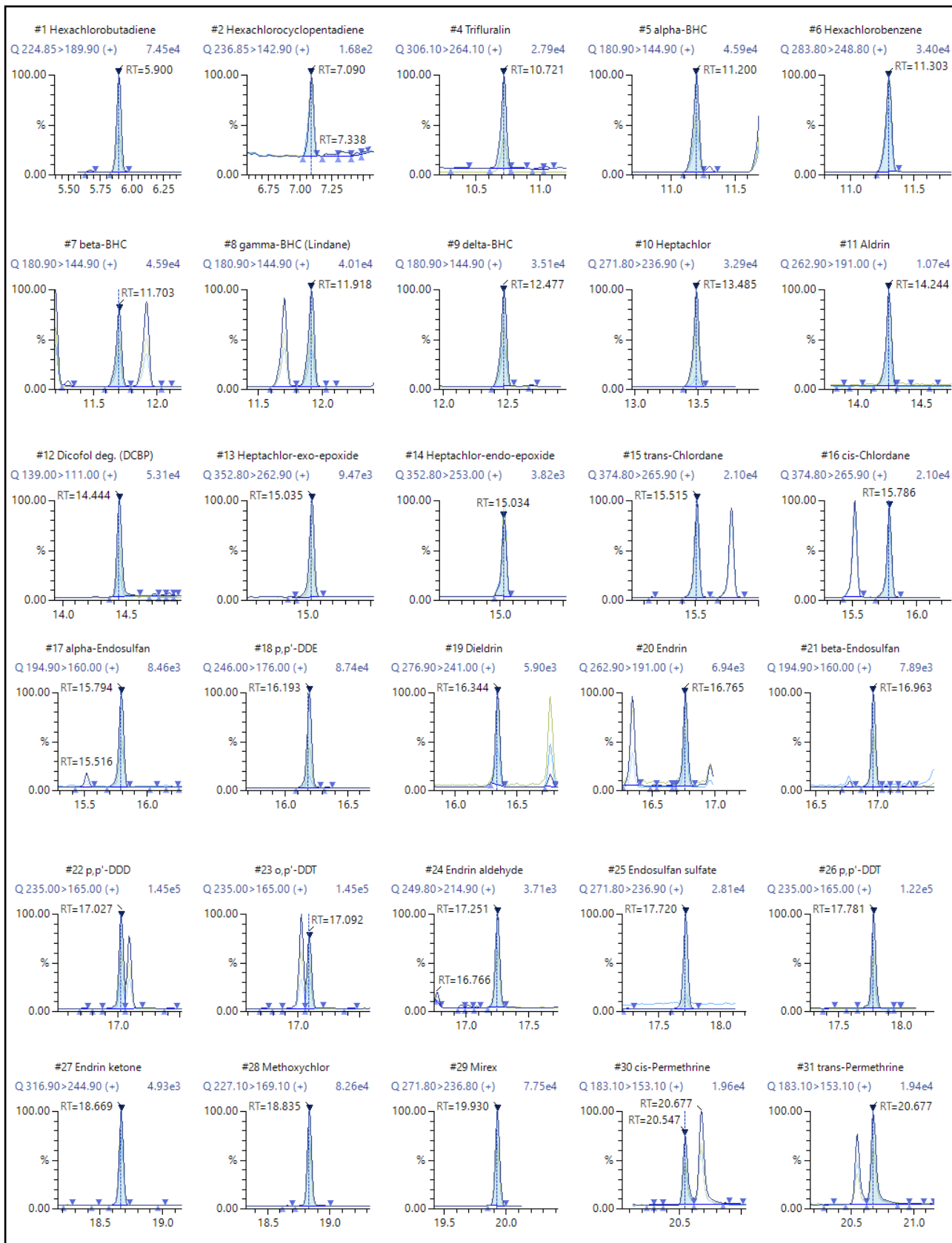

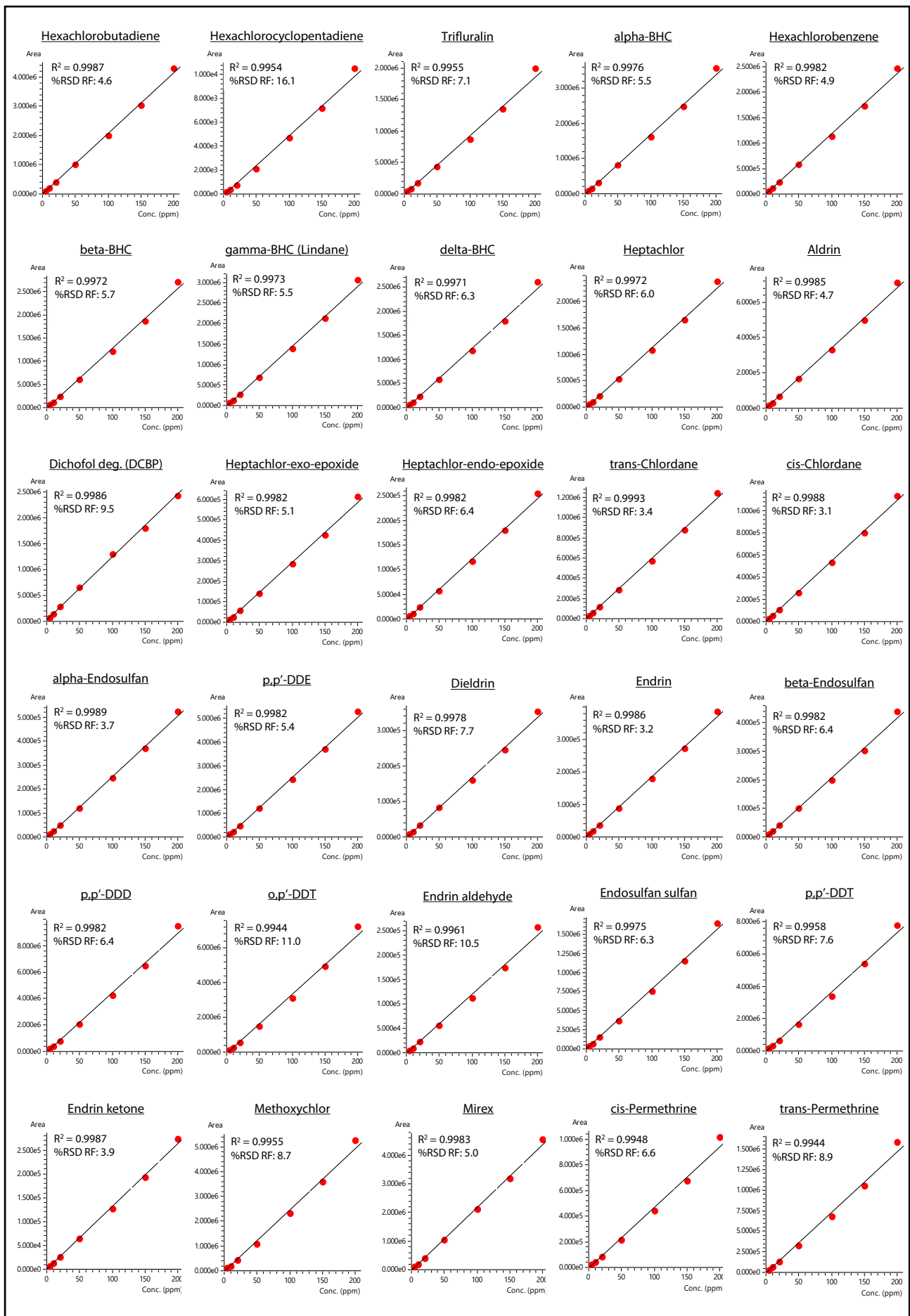Figure 2: Mass chromatograms of all target pesticides

Figure 3: External calibration curves of all target pesticides

Table 2. Summary of Retention Time, R², and %RSD of RF of All Target Pesticides

No	Compound Names	RT (min)	R²	%RSD RF
1	Hexachlorobutadiene	5.900	0.9987	4.6
2	Hexachlorocyclopentadiene	7.092	0.9967	14.4
3	Trifluralin	10.720	0.9955	7.1
4	alpha-BHC	11.198	0.9976	5.5
5	Hexachlorobenzene	11.302	0.9982	4.9
6	beta-BHC	11.703	0.9972	5.7
7	gamma-BHC (Lindane)	11.916	0.9973	5.5
8	delta-BHC	12.476	0.9971	6.3
9	Heptachlor	13.484	0.9972	6.0
10	Aldrin	14.243	0.9985	4.7
11	Dicofol deg. (DCBP)	14.442	0.9986	9.5
12	Heptachlor-exo-epoxide	15.034	0.9982	5.1
13	Heptachlor-endo-epoxide	15.034	0.9982	6.4
14	trans-Chlordane	15.515	0.9985	3.4
15	cis-Chlordane	15.786	0.9988	3.1
16	alpha-Endosulfan	15.792	0.9989	3.7
17	p,p'-DDE	16.192	0.9982	5.4
18	Dieldrin	16.338	0.9978	7.7
19	Endrin	16.767	0.9986	3.2
20	beta-Endosulfan	16.963	0.9978	3.8
21	p,p'-DDD	17.026	0.9965	8.3
22	o,p'-DDT	17.091	0.9944	11.0
23	Endrin aldehyde	17.249	0.9961	10.5
24	Endosulfan sulfate	17.719	0.9975	6.3
25	p,p'-DDT	17.780	0.9958	7.6
26	Endrin ketone	18.668	0.9987	3.9
27	Methoxychlor	18.835	0.9955	8.7
28	Mirex	19.930	0.9983	5.0
29	cis-Permethrine	20.548	0.9948	6.6
30	trans-Permethrine	20.679	0.9944	8.9

Table 3. Summary of SD and %RSD from the Quantitated Concentration of Seven Separate Vials of 10 ng/mL Pesticides Standard Solutions and the Calculated MDL and QDL of All Target Pesticides

No	Compound Names	SD	%RSD (Conc)	MDL (ng/mL)	QDL (ng/mL)
1	Hexachlorobutadiene	0.166	1.7	0.52	1.66
2	Hexachlorocyclopentadiene	0.536	5.0	1.68	5.36
3	Trifluralin	0.284	2.8	0.89	2.84
4	alpha-BHC	0.254	2.5	0.80	2.54
5	Hexachlorobenzene	0.298	2.9	0.94	2.98
6	beta-BHC	0.239	2.4	0.75	2.39
7	gamma-BHC (Lindane)	0.259	2.6	0.81	2.59
8	delta-BHC	0.265	2.6	0.83	2.65
9	Heptachlor	0.218	2.2	0.68	2.18
10	Aldrin	0.395	4.0	1.24	3.95
11	Dicofol deg. (DCBP)	0.240	2.8	0.75	2.4
12	Heptachlor-exo-epoxide	0.467	4.6	1.47	4.67
13	Heptachlor-endo-epoxide	0.380	3.7	1.19	3.8
14	trans-Chlordane	0.290	2.9	0.91	2.9
15	cis-Chlordane	0.239	2.4	0.75	2.39
16	alpha-Endosulfan	0.275	2.7	0.86	2.75
17	p,p'-DDE	0.254	2.6	0.80	2.54
18	Dieldrin	0.345	3.4	1.08	3.45
19	Endrin	0.485	5.4	1.52	4.85
20	beta-Endosulfan	0.407	4.1	1.28	4.07
21	p,p'-DDD	0.280	2.8	0.88	2.8
22	o,p'-DDT	0.164	1.7	0.51	1.64
23	Endrin aldehyde	0.392	3.8	1.23	3.92
24	Endosulfan sulfate	0.211	2.4	0.66	2.11
25	p,p'-DDT	0.273	3.0	0.86	2.73
26	Endrin ketone	0.230	2.4	0.72	2.3
27	Methoxychlor	0.261	2.9	0.82	2.61
28	Mirex	0.202	2.0	0.63	2.02
29	cis-Permethrine	0.188	1.9	0.59	1.88
30	trans-Permethrine	0.266	2.8	0.84	2.66

Method Detection Limit (MDL)

The calculated MDL provide the lowest concentration at which the method can accurately detect the pesticides, allowing for confident identification of trace amount of analyte in the sample.

In this application note, the MDL for pesticides were determined by analyzing seven separate vials of 10 ng/mL standard solutions (Pest MDL 1 – MDL 7). The quantitation result and concentration standard deviation (SD) of each compound were obtained to calculate the MDL based on the formula below:

$$MDL = 3.14 \times SD_{Conc.}$$

The SD determined for all target pesticides are tabulated in Table 3 together with their calculated MDL. All target pesticides have MDL below 2 ng/mL.

Quantitation Detection Limit (QDL)

The calculated QDL provides the lowest concentration at which the method can accurately quantify the pesticides in the sample with high level of confidence.

The QDL for pesticides were determined using the same data acquired for MDL determination, using the formula below:

$$QDL = 10 \times SD_{Conc.}$$

Table 3 tabulates the calculated QDL for all target pesticides in this study. Notably, all the target pesticides exhibited QDL values below 6 ng/mL. The low MDL and QDL indicate the capability of the analytical method employed in quantifying low concentration of these pesticides in water samples with high sensitivity.

Repeatability

In this study, the %RSD (Relative Standard Deviation) for the concentration quantitated for each pesticide in Pest MDL 1 to 7 standard solutions were less than 6% (Table 3). This signifies a high level of repeatability, demonstrating the precision of the analytical method employed, providing assurance in the reliability of the obtained data.

■ Nitrosamines Analysis Results

Detection and Separation

The target nitrosamines were injected through a Programmed Temperature Vaporization (PTV) unit and separated in the GC-MS/MS using SH-I-634Sil MS column. The MRM acquired mass chromatograms of all target nitrosamines at 2 ng/mL and internal standards (NDMA-d6 and NDPA-d14) at 20 ng/mL are shown in Figure 4.
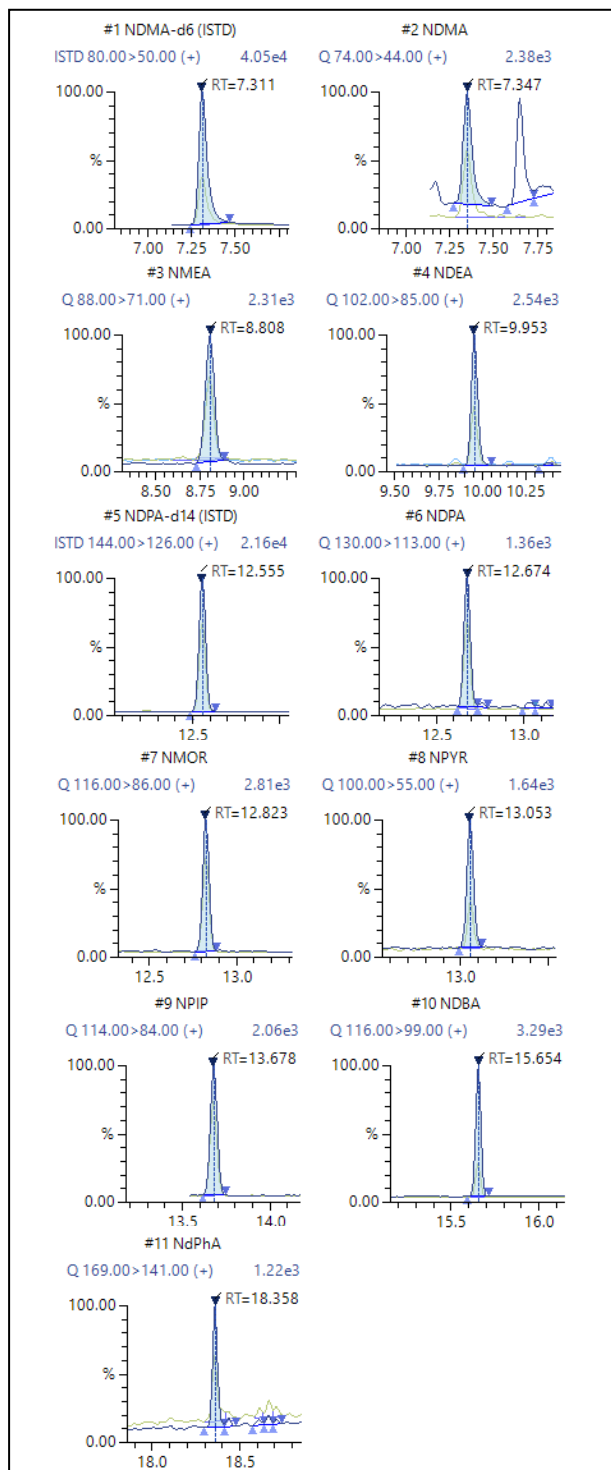
Figure 4: Mass chromatograms of all target nitrosamines (NdPhA was detected as diphenylamine).

Calibration Curve and Linearity

Internal standard calibration curve was utilized for the quantitative analysis. The internal standard calibration points of each nitrosamine prepared were 1, 2, 5, 10, 20, and 50 ng/mL with 20 ng/mL for both internal standards used (NDMA-d6 and NDPA-d14). Figure 5 shows the internal standard calibration curve of each nitrosamine, demonstrating excellent linearity with R^2 of at least 0.9991 with %RSD RF of below 10%. Table 4 summarizes the retention time, internal standard grouping, R^2 value, and %RSD RF of all target nitrosamines.
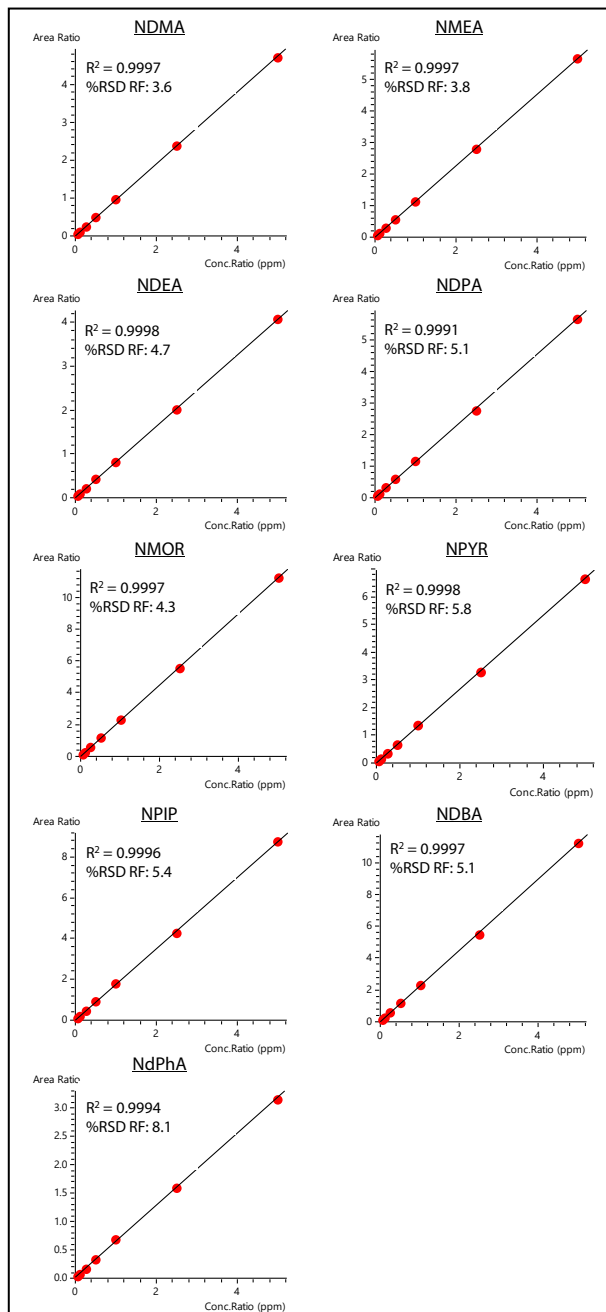

Figure 5: Internal standard calibration curves of all target nitrosamines (NdPhA was detected as diphenylamine).

Method Detection Limit (MDL)

MDL for nitrosamines were determined by analyzing seven separate vials of 2 ng/mL standard solutions (NSA MDL 1 to MDL 7). Quantitation result and concentration standard deviation (SD) of each compound were obtained to calculate the MDL.

The SD determined for all target nitrosamines are tabulated in Table 5 together with its calculated MDL. All target nitrosamines has excellent MDL of below 0.3 ng/mL.

Quantitation Detection Limit (QDL)

The QDL for nitrosamines were determined using the same data files acquired for MDL determination.

The QDL for all target nitrosamines are tabulated in Table 5. All the target nitrosamines exhibited QDL values below 1 ng/mL. This signifies that GCMS-TQ8050 NX with Twin Line MS Kit gives outstanding sensitivity in this analysis.

Table 4. Summary of Retention Time, Internal Standard Grouping, R^2, and %RSD of Response Factor of All Target Nitrosamines

No	Compound Names	ISTD Grp.	RT (min)	R^2	%RSD RF
1	N-nitroso-dimethylamine-d6 (NDMA-d6)	1	7.311	-	-
2	N-nitroso-dimethylamine (NDMA)	1	7.347	0.9997	3.6
3	N-nitroso-methylethylamine (NMEA)	1	8.808	0.9997	3.8
4	N-nitroso-diethylamine (NDEA)	1	9.953	0.9998	4.7
5	N-nitroso-dipropylamine-d14 (NDPA-d14)	2	12.555	-	-
6	N-nitroso-dipropylamine (NDPA)	2	12.674	0.9991	5.1
7	N-nitroso-morpholine (NMOR)	2	12.823	0.9997	4.3
8	N-nitroso-pyrrolidine (NPYR)	2	13.053	0.9998	5.8
9	N-nitroso-piperidine (NPIP)	2	13.678	0.9996	5.4
10	N-nitroso-di-n-butylamine (NDBA)	2	15.654	0.9997	5.1
11	N-nitroso-diphenylamine (NDPhA)*	2	18.358	0.9994	8.1

*NdPhA was detected as diphenylamine

Repeatability

Consistency and reliability are paramount in nitrosamine analysis to ensure accurate and trustworthy result. In this study, the %RSD for the concentration quantitated for each nitrosamine in NSA MDL 1 to 7 (2 ng/mL) standard solutions was less than 5% (Table 5). This indicates that the measured concentrations for each nitrosamine were consistently close to one another even at trace level, showcasing the high level of precision and reproducibility in our analysis.

Table 5. Summary of SD and %RSD from the Quantitated Concentration of Seven separate vials of 2 ng/mL Standard Solutions and the Calculated MDL and QDL of All Target Nitrosamines

No	Compound Names	SD	%RSD (Conc)	MDL (ng/mL)	QDL (ng/mL)
1	NDMA	0.088	4.1	0.28	0.88
2	NMEA	0.050	2.4	0.16	0.50
3	NDEA	0.038	1.9	0.12	0.38
4	NDPA	0.088	4.4	0.28	0.88
5	NMOR	0.061	3.0	0.19	0.61
6	NPYR	0.070	3.5	0.22	0.70
7	NPIP	0.077	3.9	0.24	0.77
8	NDBA	0.056	2.4	0.18	0.56
9	NDPhA*	0.093	4.8	0.29	0.93

*NdPhA was detected as diphenylamine

■ Conclusion

In conclusion, the Twin Line GC-MS/MS approach presents a viable alternative for the analysis of pesticides and nitrosamines in water. With this method, we have successfully separated and detected all pesticides and nitrosamines using the SH-I-5Sil MS and SH-I-624Sil MS columns, respectively, while demonstrating excellent linearity and low detection limits. Twin Line MS Kit reduced downtime by allowing two columns to be installed to the MS simultaneously, thus avoiding the inconvenience of switching columns between applications. In addition, the flows from both columns did not compromise on the GC-MS/MS sensitivity, as can be seen from the MDL and QDL results. These findings highlight the significance of this approach in ensuring the safety and quality of our water sources.

■ References

1. United States Environmental Protection Agency, Ground Water and Drinking Water, accessed 2 February 2024, https://www.epa.gov/ground-water-and-drinking-water

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3. Munch, J W. METHOD 521: Determination Of Nitrosamines In Drinking Water By Solid Phase Extraction And Capillary Column Gas Chromatography With Large Volume Injection And Chemical Ionization Tandem Mass Spectrometry (MS/MS). U.S. Environmental Protection Agency, Washington, DC, 2005.

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