

Nitrosamines Analysis in Drinking Water Using GC/MS/MS—Meeting Equivalence to EPA Method 521

Using Agilent 7010 and 7000 triple quadrupole GC/MS systems

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Abstract

The Eurofins Eaton Analytical-Agilent Method 521.1 (EEA-Agilent Method 521.1) is based on a multilaboratory study of nitrosamines in drinking water using triple quadrupole GC/MS (GC/MS/MS) in electron ionization (EI) mode¹. Currently, ion trap GC/MS (GC/IT) is the approved technology for the United States Environmental Protection Agency (EPA) Method 521, but GC-IT is being obsolete. The EPA was open to approval of alternate detection methods as long as the sample preparation step was unchanged. Analytes in EPA Method 521 were investigated with the addition of N-nitrosomorpholine (NMOR). The study was divided into two phases. In phase I, Lab A demonstrated that GC/MS/MS achieved lower lowest concentration minimum reporting levels (LCMRL) and detection limits (DL) than the approved GC/IT. Lower injection volume and shorter analysis times were accomplished with the GC/MS/MS. Good correlation between GC/MS/MS and GC/IT was observed when analyzing nitrosamines in numerous field samples. In phase II, Lab A extracts and splits LCMRL samples (32) to Lab B and Lab C for validation using Agilent 7010 and 7000 GC/MS/MS systems, respectively. Both 7010 and 7000 GC/MS/MS results were better than the LCMRL and DL requirements in Method 521. A linear calibration curve was achieved with $R^2 > 0.99$. Method performance was verified by three separate laboratories and the EPA has provided a letter of method equivalency.

Introduction

Nitrosamines, particularly nitrosodimethylamine (NDMA), are contaminants found in drinking water and of interest to the environmental community due to potential health concerns². NDMA is found in high concentrations and produced by industrial sources such as direct contamination from the manufacture of rocket fuel. In addition, NDMA is a disinfection by-product from the chlorination, chloramination, and ozonation of drinking water and wastewater³. EPA Method 521 (2005) is a procedure for determining nitrosamines in drinking water⁴. GC/IT is the approved instrumentation for Method 521, but the technology is being obsolete. The EPA Office of Groundwater and Drinking Water (OGWDW) considers alternate detection techniques without changing the guidelines for sample preparation and sensitivity criteria.

The EEA-Agilent Method 521.1 was developed using triple quadrupole GC/MS (GC/MS/MS) as an alternate technology using a two-part study. In study phase I, Lab A compared performance on GC/IT with GC/MS/MS. In phase II, Lab A split sample extracts to Lab B and Lab C for a three-laboratory validation study in accordance with EPA protocols using GC/MS/MS. The LCMRL and DL were evaluated using GC/MS/MS for nitrosamines listed in Method 521 with the addition of NMOR (Table 1)⁴.

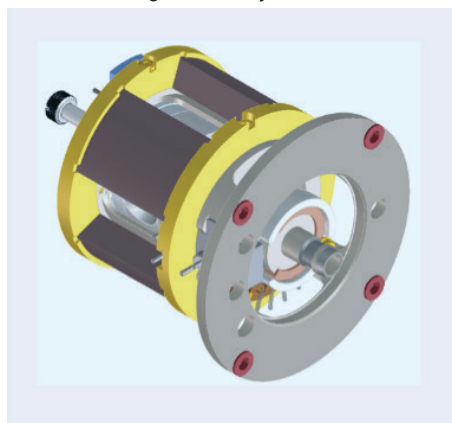
The 7010 and 7000 GC/MS/MS systems are equipped with the high-efficiency source (HES) and the extractor source, respectively. The 7010 HES is a source that is completely redesigned to produce 20-times more ions than the 7000 extractor source, achieving an increase in sensitivity (Figure 1). In phase II, these two sources were investigated to determine whether the HES is necessary to meet the LCMRL and DL requirements of Method 521.

This Application Note shows the development of EEA-Agilent Method 521.1 for determining nitrosamines in drinking water using GC/MS/MS technology in electron ionization (EI) mode instead of CI mode, as specified in Method 521. No changes were made to Method 521 procedures and guidelines related to sample preparation and quality control⁴.

Table 1. Nitrosamines investigated in EEA-Agilent Method 521.1.

Analyte	Chemical Abstract Services (CAS) registry number
N-nitrosodimethylamine (NDMA)	62-75-9
N-nitrosomethylethylamine (NMEA)	10595-95-6
N-nitrosodiethylamine (NDEA)	55-18-5
N-nitroso-di-n-propylamine (NDPA)	621-64-7
N-nitrosomorpholine (NMOR)	59-89-2
N-nitrosopyrrolidine (NPYR)	930-55-2
N-nitrosopiperidine (NPIP)	100-75-4
N-nitrosodi-n-butylamine (NDBA)	924-16-3

Lab A and Lab B
7010 GC/MS/MS
high-efficiency source



Lab C
7000 GC/MS/MS
extractor source

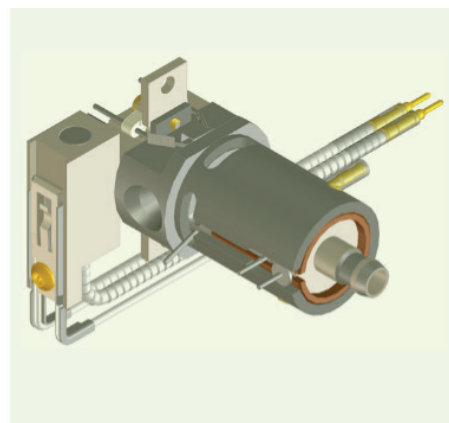


Figure 1. Source design of 7010 and 7000 GC/MS/MS systems. The extractor source is equipped in the 7000 GC/MS/MS. The high-efficiency source is equipped on the 7010 GC/MS/MS.

Experimental

Sample preparation

Detailed instructions on sample preparation are in EEA-Agilent Method 521.1^{1,4}. Analytes and surrogates were extracted by passing water samples through a solid phase extraction (SPE) cartridge containing coconut charcoal and eluted with methylene chloride. The methylene chloride extract is dried and concentrated before adding the internal standard. The sample components are separated, identified, and analyzed using GC/MS/MS or GC/IT.

GC/MS/MS systems

Agilent 7010 GC/MS/MS and 7000 GC/MS/MS systems consisted of an Agilent 7890 gas chromatograph (GC) with a 7010 and 7000 triple quadrupole mass spectrometer detector, respectively. Both systems were equipped with a 7693 automatic liquid sampler (ALS) and a multimode inlet (MMI). Inlet temperature and parameters vary between laboratories.

- Lab A used splitless programmed temperature injection.
- Lab B used splitless isothermal injection.
- Lab C used pulsed splitless isothermal injection.

Table 2–4 list injector, GC, and MSD parameters, respectively. Table 5 lists multiple reaction mode (MRM) transitions and optimal collision energies for all nitrosamines investigated.

Phase I: GC/IT compared to GC/MS/MS

Lab A extracted water samples and split sample extracts for comparison between Varian 4000 GC/IT and 7010 GC/MS/MS. EPA Method 521.4 describes GC-IT parameters in detail.

Phase II: Interlaboratory validation study using GC/MS/MS

Lab A extracted spiked water samples and split extracts to Labs B and C for method validation. Labs A and B validated the method using a 7010 GC/MS/MS. Lab C validated the method using a 7000 GC/MS/MS.

GC/MS/MS configuration

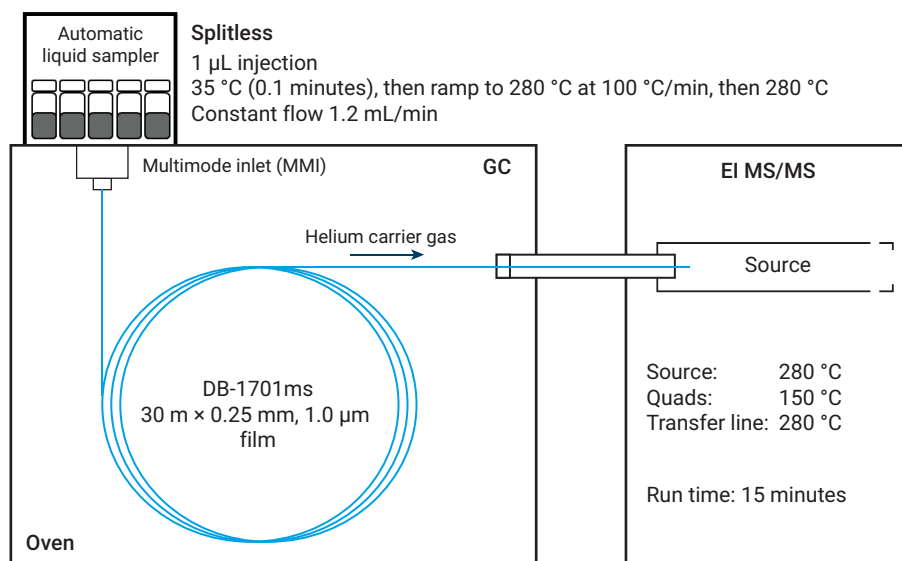


Figure 2. GC/MS/MS system configuration. Lab A injector parameters are shown. All labs followed the same GC/MS/MS configuration except for inlet parameters.

GC/MS/MS parameters

Table 2. Injector parameters.

Parameter	Setpoint
Inlet	Multimode Inlet
Liner	Splitless, double-tapered, Ultra Inert liner (p/n 5190-3983)*
Injection volume	1 µL
Viscosity delay	2 seconds
Carrier gas	Helium
Mode*	Splitless or pulsed splitless*
Temperature*	Programmed or isothermal*
Pressure	8.98 psi
Total flow	104.2 mL/min
Septum purge flow	3 mL/min
Purged flow to split vent	100 mL/min at 0.8 minutes
Lab A	
Liner	2 mm dimpled, splitless, ultra-inert (p/n 5190-2297)
Temperature	35 °C (hold 0.1 minutes) Rate 1: Ramp 100 °C/min to 280 °C
Mode	Splitless
Lab B	
Temperature	260– 280 °C
Mode	Splitless
Lab C	
Temperature	260–280 °C
Mode	Pulsed splitless
Injection pulsed pressure	40 psi until 0.75 minutes

* See Lab A, Lab B, and Lab C for liner, mode, and temperature settings.

Table 3. GC parameters.

Parameter	Setpoint
Column	DB-1701 (p/n 122-0733)
Column phase	14 % Cyanopropylphenyl 86 % dimethylpolysiloxane
Column dimensions	30 m length × 250 µm diameter, 1 µm film thickness
Column configuration	Inlet to mass spectrometer detector
Flow	1.2 mL/min
Oven temperature	Initial: 33 °C (hold 1 minute) Rate 1: Ramp 35 °C/min to 80 °C (hold 2 minutes) Rate 2: Ramp 10 °C/min to 140 °C Rate 3: Ramp 50 °C/min to 280 °C (hold 2 minutes)
Equilibration time	3–5 minutes

Table 4. MSD parameters.

Parameter	Setpoint
Ion source	EI
Source temperature	280 °C
Quadrupole 1 temperature	150 °C
Quadrupole 2 temperature	150 °C
Transfer line temperature	260–280 °C
Quench gas	Helium at 4 mL/min
Collision gas	Nitrogen at 1.5 mL/min
Solvent delay	5.5 minutes
Gain	3.0
Peak width	0.05
Electron energy	70 eV

Table 5. Retention times and suggested MRM transitions and collision energies for quantitation of method analytes in GC/MS/MS in EI mode.

Analyte	Retention time (min) ^a	Segment	Retention time window (min)	Precursor ion (m/z) ^b	Product ion (m/z) ^b	CE ^c	Resolution ^d	Dwell (ms) ^e
NDMA-d6 (SUR)	7.02	1	5.8–7.6	80	50	8	unit/wide	75
				80	46	25	unit/wide	75
NDMA	7.05	1	5.8–7.6	74	44	6	wide/wide	75
				74	42	22	wide/wide	75
NMEA	8.58	2	7.6–9.0	88	71	4	wide/wide	150
				88	42	23	wide/wide	150
NDEA	9.79	3	9.0–11.4	102	85	4	wide/wide	150
				102	44	12	wide/wide	150
NDPA-d14 (IS)	11.78	4	11.4–11.9	144	126	10	unit/unit	50
				144	50	20	unit/unit	50
NDPA	11.83	4	11.4–11.9	130	43	10	wide/wide	100
				101	70	10	wide/wide	100
NMOR	12.09	5	11.9–12.15	116	86	2	wide/wide	150
				116	56	15	wide/wide	150
NPYR	12.3	6	12.15–12.28	100	55	7	unit/wide	100
				100	70	7	unit/wide	100
				100	43	10	unit/wide	100
NPIP	12.59	7	12.28–12.62	114	84	7	wide/wide	150
				114	55	25	wide/wide	150
NDBA	12.89	8	12.62–13.8	158	141	10	wide/wide	100
				158	99	10	wide/wide	100
				116	99	10	wide/wide	100

^a Retention times obtained on a 7010 GC/MS/MS with parameters listed in Tables 2–4. Isothermal injection conditions were also used. Absolute retention times may vary slightly with different instrumentations.

^b Precursor and product ion transitions were chosen based on structural significance and ion abundance. The most abundant ion is selected as the quantifier ion.

^c Collision energy (CE) was optimized based on ion abundance and may vary slightly with different instrumentations.

^d MS Resolution for unit and wide consist of a full width half maximum of 0.7 and 1.2 amu, respectively.

^e Dwell times were adjusted to obtain approximately 3.3 cycles per second. Dwell times may be different between quantifier and qualifier ions, and vary between different instrumentations.

Results and discussion

Phase I: GC/IT compared to GC/MS/MS

GC/MS/MS provides advantages over GC/IT for the analysis of nitrosamines in drinking water. Faster analysis time is achieved with GC/MS/MS with the last compound, NDPA, eluting at 12.9 minutes compared to 30.5 minutes with GC/IT (Figure 3). The shorter analysis time allows labs to run 2–3 times more samples using GC/MS/MS compared to GC/IT. Baseline separation for NDPA, NPYR, and NMOR are accomplished using GC/MS/MS with a DB-1701 column while this presented challenges in the GC/IT. Matrix contamination is decreased with lower injection volume, from 10 to 20 μL in GC/IT to 1 μL in GC/MS/MS, allowing for less maintenance and more uptime. EI is used instead of CI, increasing the ease-of-use and reliability. The sensitivity of GC/MS/MS enabled low-level analysis of nitrosamines in extracted water samples (Figure 4). Excellent correlation was observed between GC/IT and GC/MS/MS for quantification of nitrosamines in water sample extracts from the field. Figure 5A shows an example of NDMA correlation in the analysis of field water samples. Figure 5B shows correlation in calibration of NDMA extracted from water samples. Lower LCMRL and DL could be achieved using GC/MS/MS (Figure 6).

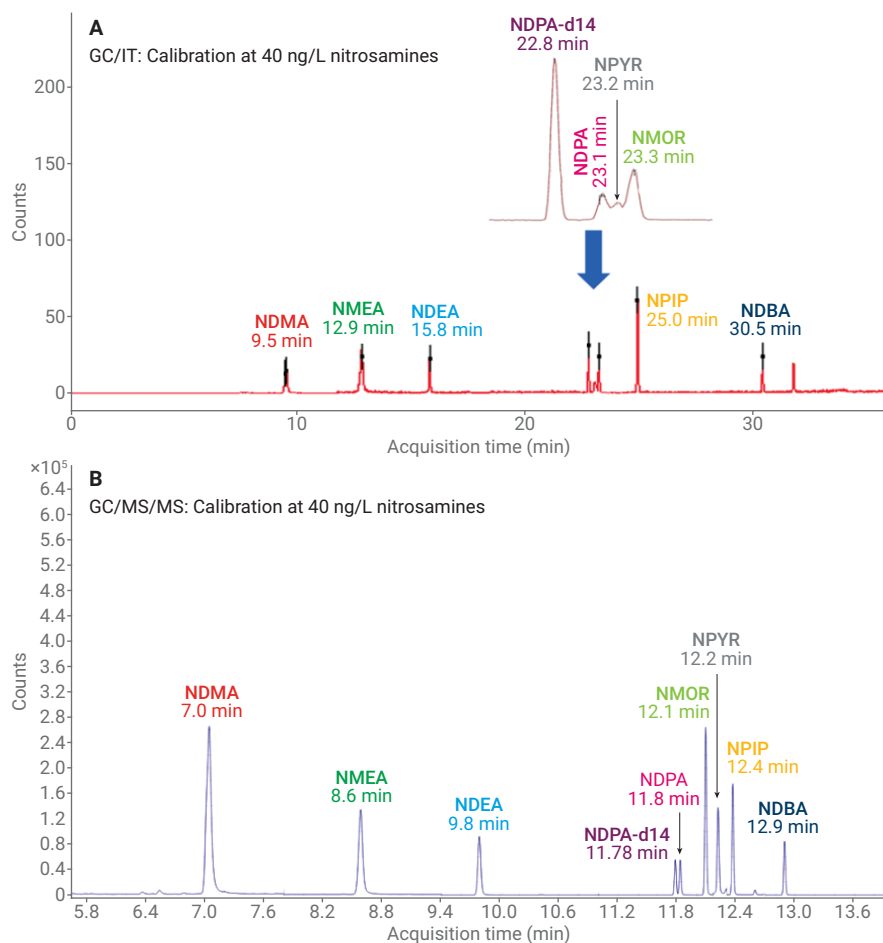


Figure 3. A) GC/IT: Calibration at 40 ng/L nitrosamines. B) GC/MS/MS: Calibration at 40 ng/L nitrosamines. GC/IT parameters follow Method 521⁴. GC/MS/MS parameters are shown in Tables 2–5.

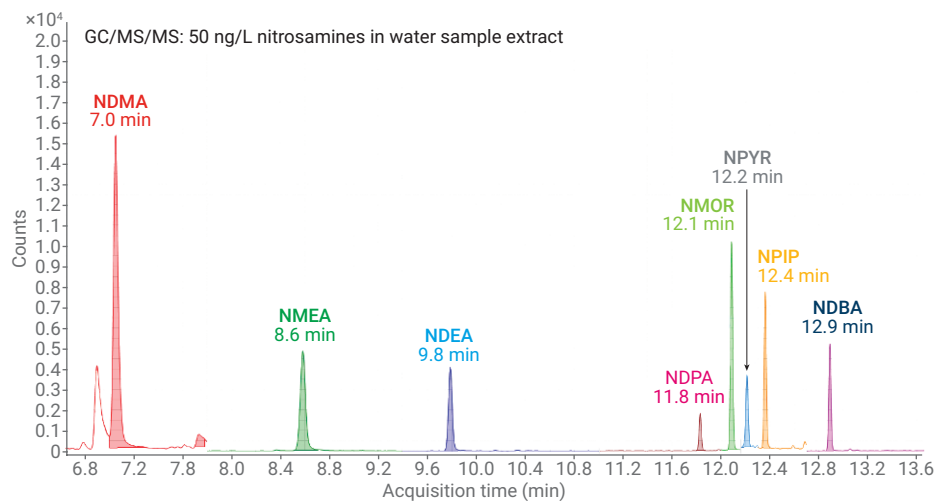


Figure 4. MRM of quantifier ion of 0.5 ng/L nitrosamines extracted from a water sample.

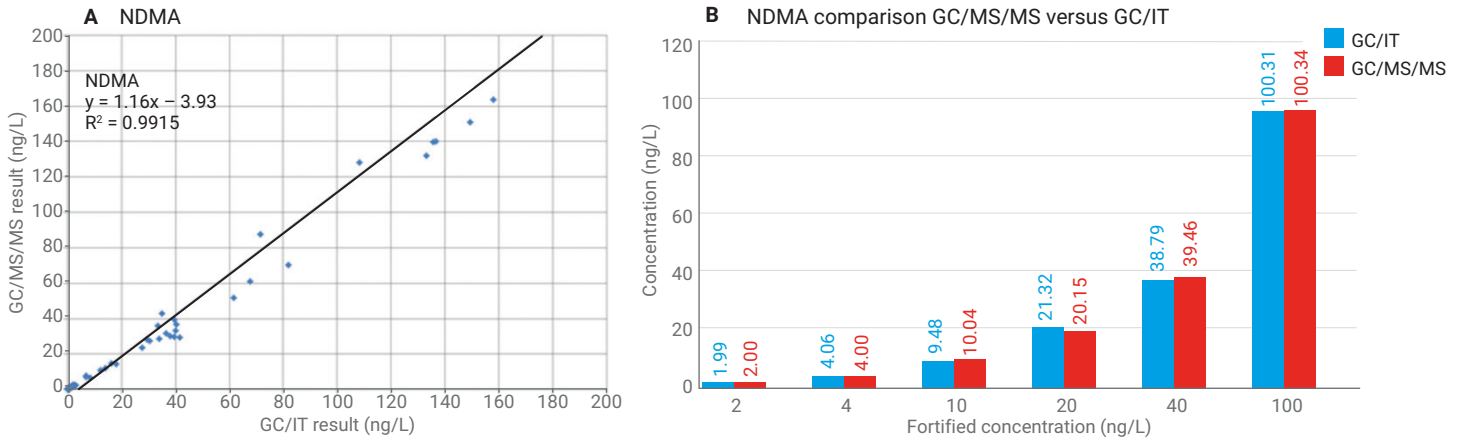


Figure 5. Correlation between GC/MS/MS and GC/IT analysis of NDMA in field water sample extracts (A) and calibration of an extracted water sample (B). Water samples were extracted and analyzed with GC/MS/MS and GC/IT using the same holding time, standards, extraction process, and mixes.

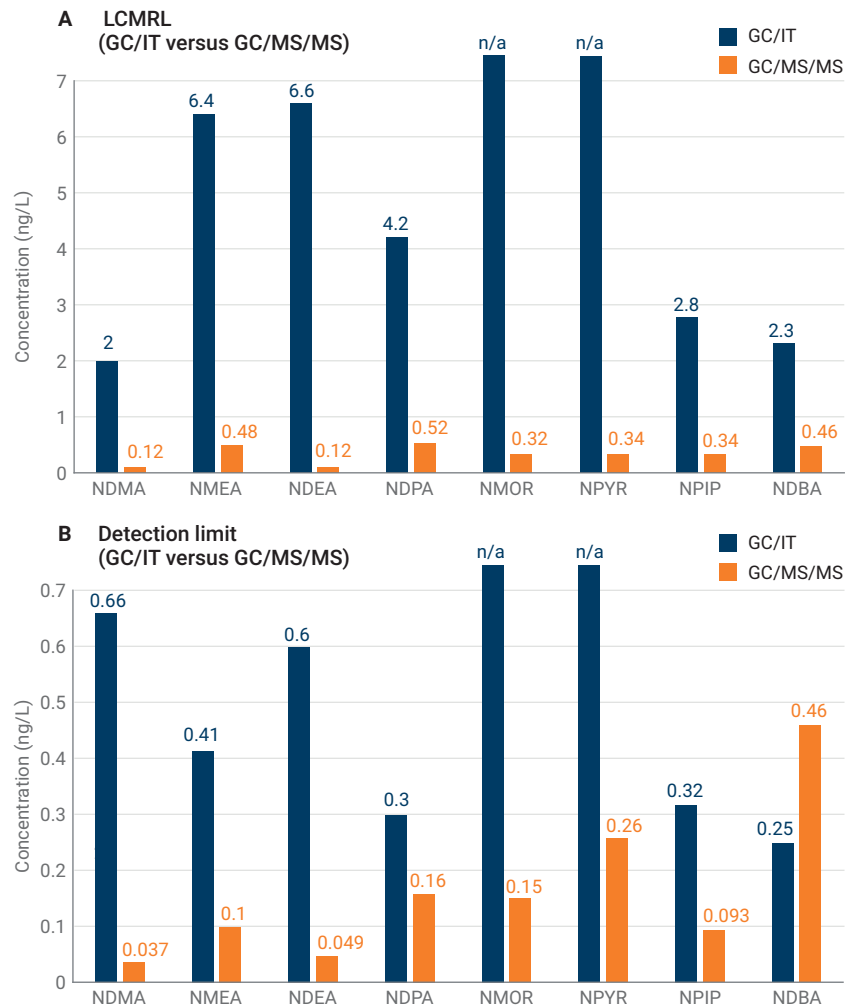


Figure 6. Lowest concentration minimum reporting level (A) and detection limit (B) comparison using a 7010 GC/MS/MS and a Varian 4000 GC/IT. Spiking levels used to calculate LCMRL and DL range 0.1–10 ng/L. n/a indicates that LCMRL and DL are above the highest spiking level or the spiking level exceeds the working range for NMOR and NPYR.

Phase II: Interlaboratory validation study

Both 7010 and 7000 GC/MS/MS systems meet and exceed the sensitivity requirements for nitrosamines analyzed in this study. For the interlaboratory validation study, Lab A extracted spiked reagent water samples and sent extract splits to Lab B for analysis using the 7010 GC/MS/MS and Lab C for analysis using the 7000 GC/MS/MS. Figures 7A and 7B are plots of LCMRL and DL data for each nitrosamine analyzed by the three laboratories. The LCMRL on 7010 GC/MS/MS ranged from 0.12 to 0.67 ng/L and the 7000 GC/MS/MS ranged from 0.33 to 1.2 ng/L (Figure 7A), which are both lower than the LCMRL of 1.2 to 2.1 ng/L in Method 521. The DL in the multilaboratory validation study ranged from 0.037 to 0.46 ng/mL on the 7010 GC/MS/MS systems and 0.12 to 0.6 ng/L on the 7000 GC/MS/MS systems, which are both lower than the DL of 0.26 to 0.66 ng/L in Method 521 (Figure 7B). Linearity was observed with $R^2 > 0.99$ using linear curve fit at 11 calibration points ranging 0.0625 to 100 ng/L on both 7010 and 7000 GC/MS/MS systems (Table 6). Both 7010 and 7000 GC/MS/MS systems meet the LCMRL and DL requirement. The 7010 GC/MS/MS can achieve lower LCMRL and DL than the 7000 GC/MS/MS.

EAA-Agilent Method 521.1 provides flexibility in injection parameters. Lab A, B, and C used temperature programmed splitless, isothermal splitless, and isothermal pulsed splitless injection, respectively. Lab A used a 2 mm dimpled splitless ultra inert liner, while Labs B and C used a 4 mm double-tapered splitless ultra inert liner. Although the three laboratories used different injection parameters and liners, all labs and both 7010 and 7000 GC/MS/MS systems were able to meet the LCMRL and DL requirements.

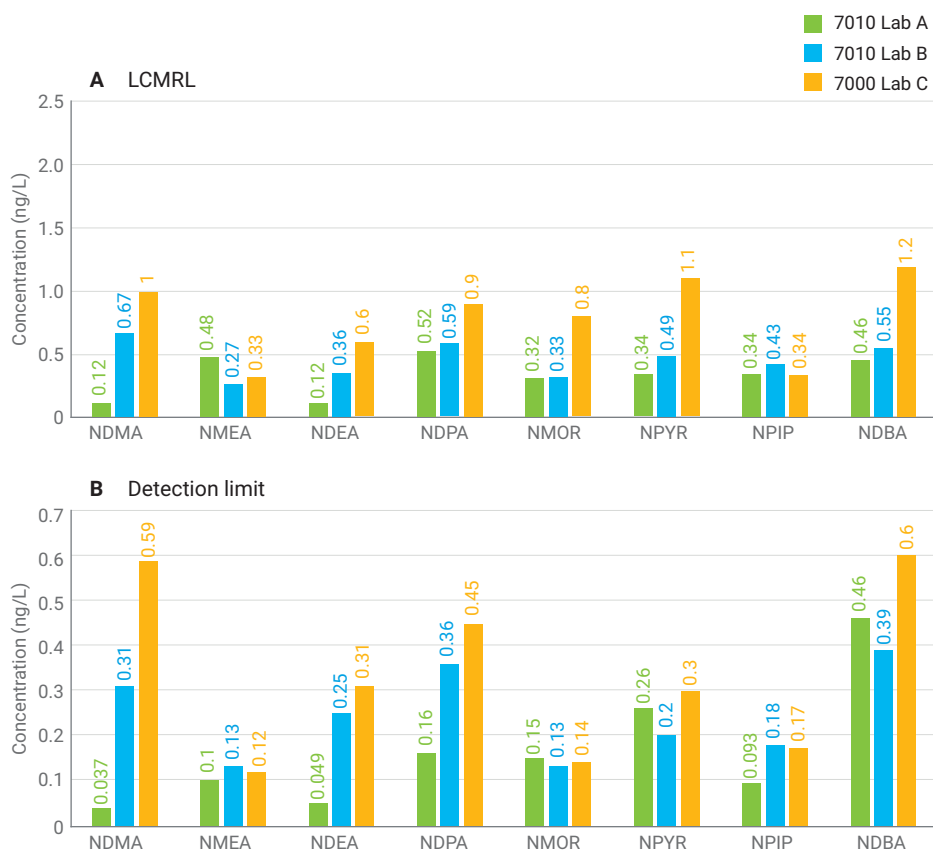


Figure 7. LCMRL and DL from interlaboratory validation studies. LCMRLs and DLs for all three laboratories were determined from laboratory fortified blanks (LFBs) at 0.1, 0.25, 0.5, 1.0, 2.0, 3.0, 4.0, 5.0, 8.0, and 10.0 ng/L analyte (n = 4 at each concentration) with the exception of NDMA for Lab C. NDMA for Lab C was determined from LFBs fortified at 1.0, 2.0, 3.0, 4.0, 5.0, 8.0, and 10.0 ng/L analyte (n = 4 at each concentration). LCMRL and DL data from Lab A and Lab B were obtained on a 7010 GC/MS/MS. Lab C data were obtained on a 7000 GC/MS/MS.

Table 6. Linearity of calibration curve from interlaboratory validation study*

Analyte	7010 Lab A	7010 Lab B	7000 Lab C
NDMA	0.9999	0.9979	0.9935
NMEA	0.9999	0.9983	0.9988
NDEA	0.9999	0.9993	0.9986
NDPA	0.9998	0.9987	0.9965
NMOR	1.0000	0.9993	0.9992
NPYR	0.9981	0.9994	0.9976
NPIP	0.9999	0.9993	0.9979
NDBA	0.9996	0.9990	0.9985

* R^2 is calculated using linear curve fit and 1/x weight at 11 calibration points (0.0625, 0.125, 0.25, 0.50, 1.0, 2.0, 4.0, 10, 20, 40, and 100 ng/L).

Conclusions

EPA-Agilent Method 521.1 provides a procedure to analyze nitrosamines in drinking water using GC/MS/MS as an alternate technology to the approved and obsoleted, GC/IT. Good correlation between the GC/MS/MS and GC-IT is observed for the analysis and calibration of water sample extracts from the field. Both 7010 and 7000 GC/MS/MS systems could easily meet the LCMRL and DL requirements with a much lower injection volume, shorter analysis time, and baseline separation of analytes, as demonstrated by the interlaboratory validation study with a split sample set. The GC/MS/MS method is robust in that LCMRL and DL can be met with the flexibility in liners and parameters. Based on these data, the EPA has formally approved the use of this method in lieu of GC/IT for nitrosamine analysis¹.

References

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