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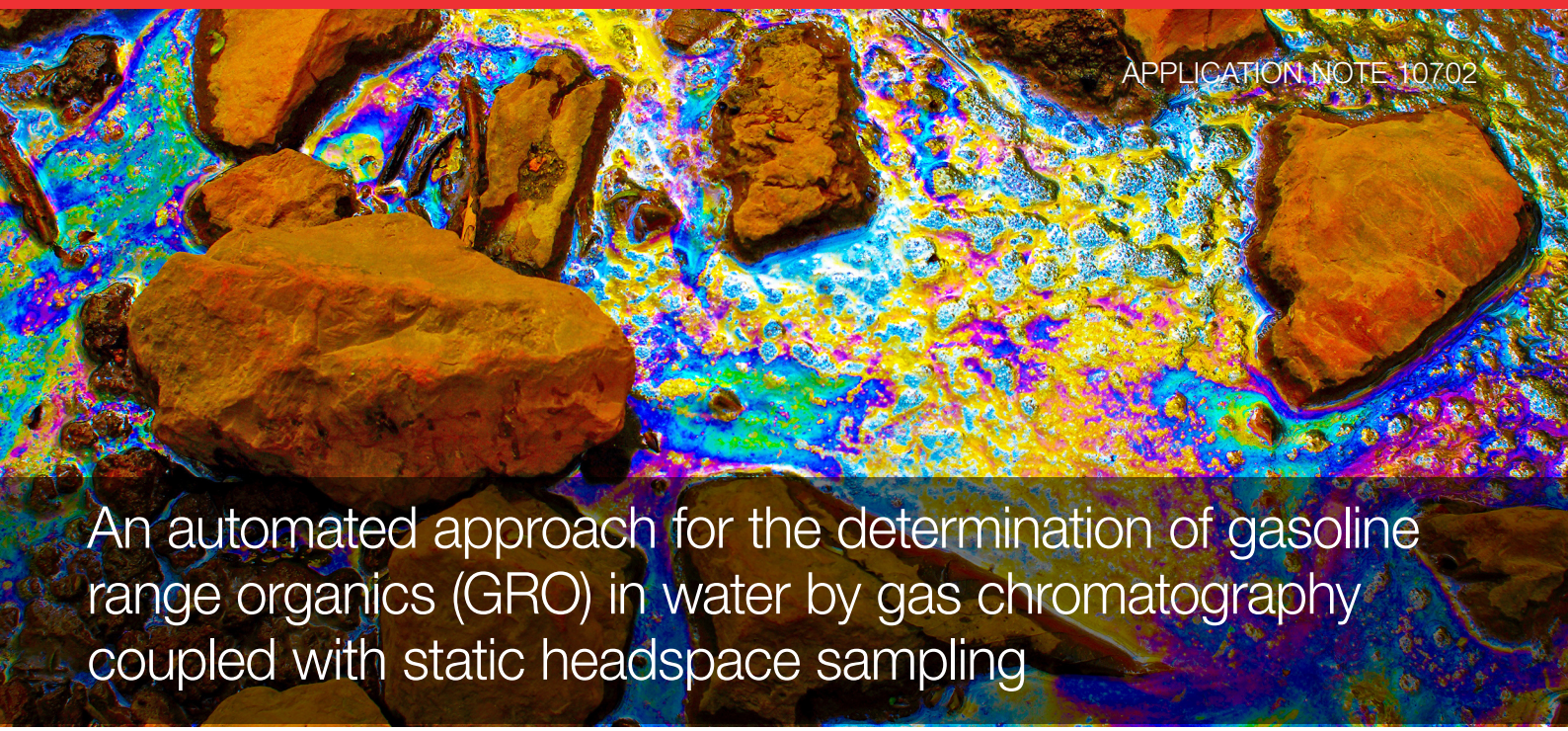

APPLICATION NOTE 10702

An automated approach for the determination of gasoline range organics (GRO) in water by gas chromatography coupled with static headspace sampling

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Keywords

Gasoline range organics, GRO, water, valve and loop, headspace-gas chromatography, HS-GC, flame ionization detector, FID, TriPlus 500 HS, environment

Goal

The aim of this application note is to demonstrate the quantitative performance of the Thermo Scientific™ TriPlus™ 500 Gas Chromatography Headspace (HS) Autosampler for the determination of gasoline range organics in water.

Introduction

Gasoline range organics (GRO) refer to hydrocarbons with a carbon range from C6 to C10 that have boiling points ranging from 60 °C to 170 °C. These chemicals are often present in the environment, especially in ground water and soil, mainly as a consequence of contamination incidents. The source of contamination can be human errors and accidents (such as oil spills) that occur when handling, storing, or transporting oil and oil products. If GRO are detected, the level of contamination needs to be determined by using quantitative analytical methods; therefore, this represents a routine application for environmental analysis laboratories. GRO are highly volatile compounds that can be easily extracted from the matrix without the need for time-consuming sample preparation. Therefore, the analytical technique of choice for this application is headspace sampling coupled to gas chromatography and mass spectrometry and/or flame ionization detection.

In this work, the headspace sampling technique coupled with gas chromatography-FID detection was employed to assess method sensitivity, precision, robustness, and linearity for quantitative assessment of GRO in water.

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Experimental

In all experiments, a TriPlus 500 HS autosampler was directly interfaced (without the need for an external transfer line) to a Thermo Scientific™ TRACE™ 1310 Gas Chromatograph equipped with a Thermo Scientific™ Instant Connect split/splitless SSL Injector and a Thermo Scientific™ Instant Connect Flame Ionization Detector (FID). Chromatographic separation was achieved on a Thermo Scientific™ TraceGOLD™ TG-1MS GC column, 30 m × 0.32 mm × 3.0 µm (P/N 26099-4840). Additional HS-GC-FID parameters are detailed in Table 1. The GC oven temperature program was optimized to reduce the analysis time and improve sample throughput; all peaks of interest elute in <13 minutes and the autosampler overlapping capability allows for long unattended sequences with automatic cycle time optimization.

Data acquisition, processing, and reporting

Data was acquired, processed, and reported using the Thermo Scientific™ Chromeleon™ Chromatography Data System (CDS) software, version 7.2. Integrated instrument control ensures full automation from instrument set-up to raw data processing, reporting, and storage. Simplified e-workflows deliver effective data management ensuring ease of use, sample integrity, and traceability.

Standard and sample preparation

GRO standard mix at 1000 µg/mL was purchased from Restek (P/N 30095) and serially diluted using tap water to obtain seven stock solutions ranging from 6.25 µg/L to 10,000 µg/L (ppb). An amount of these standard stock solutions (5 mL) was transferred into a 10 mL crimp cap headspace vial (vials P/N 10CV, caps P/N 20-MCBC-ST3) and used to assess method linearity, sensitivity, recovery, and repeatability.

Sample preparation

Unleaded petroleum was diluted with reagent water to produce a sample stock solution at 5% and kept refrigerated at 4 °C. The sample stock was used to evaluate the matrix recovery and the quantitative accuracy and precision.

Table 1. HS-GC-FID operating conditions for GRO determination in water

TRACE 1310 GC Parameters	
Inlet Module and Mode:	SSL, split
Split Ratio:	20:1
Septum Purge Mode, Flow (mL/min):	Constant, 5
Carrier Gas, Carrier Mode, Pressure (kPa):	He, constant pressure, 150
Oven Temperature Program	
Temperature 1 (°C):	50
Hold Time (min):	1
Temperature 2 (°C):	220
Rate (°C/min):	15
Hold Time 2 (min):	5
FID	
Temperature (°C):	300
Air Flow (mL/min):	350
H_2 Flow (mL/min):	35
N_2 Flow (mL/min):	40
Acquisition Rate (Hz):	25

TriPlus 500 HS Autosampler Parameters	
Incubation Temp. (°C):	85
Incubation Time (min):	30
Vial Shaking:	Fast
Vial Pressurization Mode:	Pressure
Vial Pressure (kPa) (Auxiliary Gas Nitrogen):	200
Vial Pressure Equilibration Time (min):	1
Loop Size (mL):	1
Loop/Sample Path Temp. (°C):	105
Loop Filling Pressure (kPa):	150
Loop Equilibration Time (min):	1
Needle Purge Flow Level:	5
Injection Mode:	Standard
Injection Time (min):	1

Results and discussion

Method linearity

Linearity was evaluated by injecting seven calibration levels at 6.25, 12.5, 25, 50, 1000, 2500, and 10,000 µg/L (ppb). A list of target compounds is reported in Table 2. Each concentration level was prepared and analyzed in triplicate (n = 3). The calculated correlation coefficients (R^2) were 1.000 for all the investigated gasoline organics. Moreover, the residual values (measured as % RSD of average response factors) were <6.5%, confirming an excellent linearity (Figure 1).

Detection limit and accuracy assessment (recovery)

The method detection limit is defined as the minimum concentration of a substance that can be measured and reported with 99% confidence that the analyte concentration is greater than zero.[2] According to the Wisconsin method[3] for GRO determination, the required limit of quantitation (LOQ) is 100 µg/L (ppb) or less for water samples and the method blank should not exceed a concentration of 50 µg/L (ppb). The method detection limit (MDL) was assessed analyzing n = 7 blank tap water samples (5 mL) and n = 7 tap water samples spiked at the concentration of 12.5 µg/L (ppb). MDL and LOQ were then calculated applying Equations 1 and 2, respectively.

The recovery was calculated using Equation 3 and was in the range 80% to 120%, with an average value of 105%. MDL, LOQ, and percent recovery results for the spiked samples are reported in Table 2. None of the investigated compounds could be detected in the tap water samples as shown in Figure 2.

(Equation 1)

$$MDL = t_{(n-1,1-\alpha=0.99)} * S$$

Where:
t = Student's *t*-value appropriate for the single-tailed 99th percentile t statistic and a standard deviation estimate with *n*-1 degrees of freedom, for
n = 7 injections: *t* = 3.143
S = standard deviation of the replicate analysis

(Equation 2)

$$LOQ = 10 * S$$

Where:
S = standard deviation of the replicate analysis

(Equation 3)

$$\text{Average } \%R = (C_{ave}/C_{sp}) * 100\%$$

Where:
C_{ave} = average concentration of the spiked samples
C_{sp} = initial spike concentration

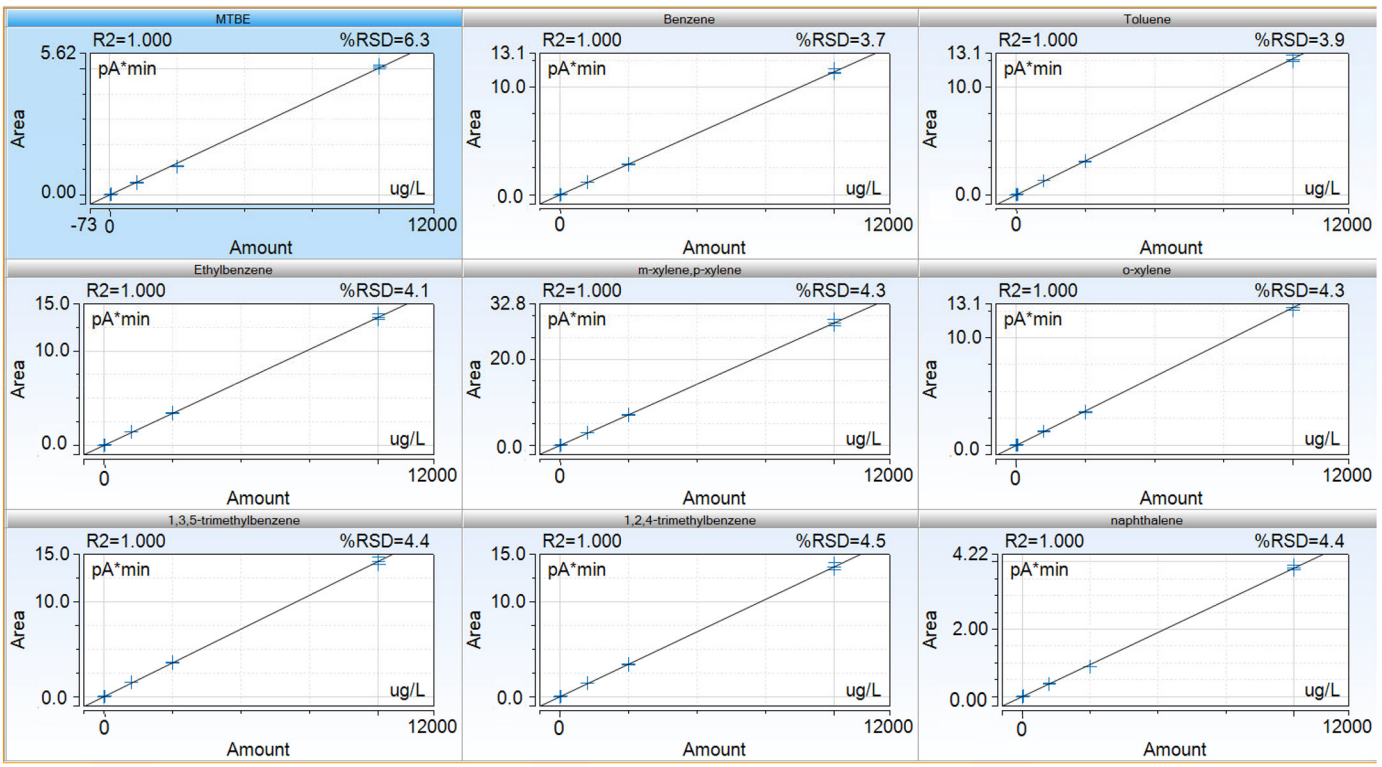


Figure 1. Calibration curves for GRO obtained by injecting seven concentration levels (6.25 to 10,000 µg/L). R^2 as well as response factors relative standard deviations (% RSD) are shown. Each calibration level was prepared and analyzed in triplicate (n = 3).

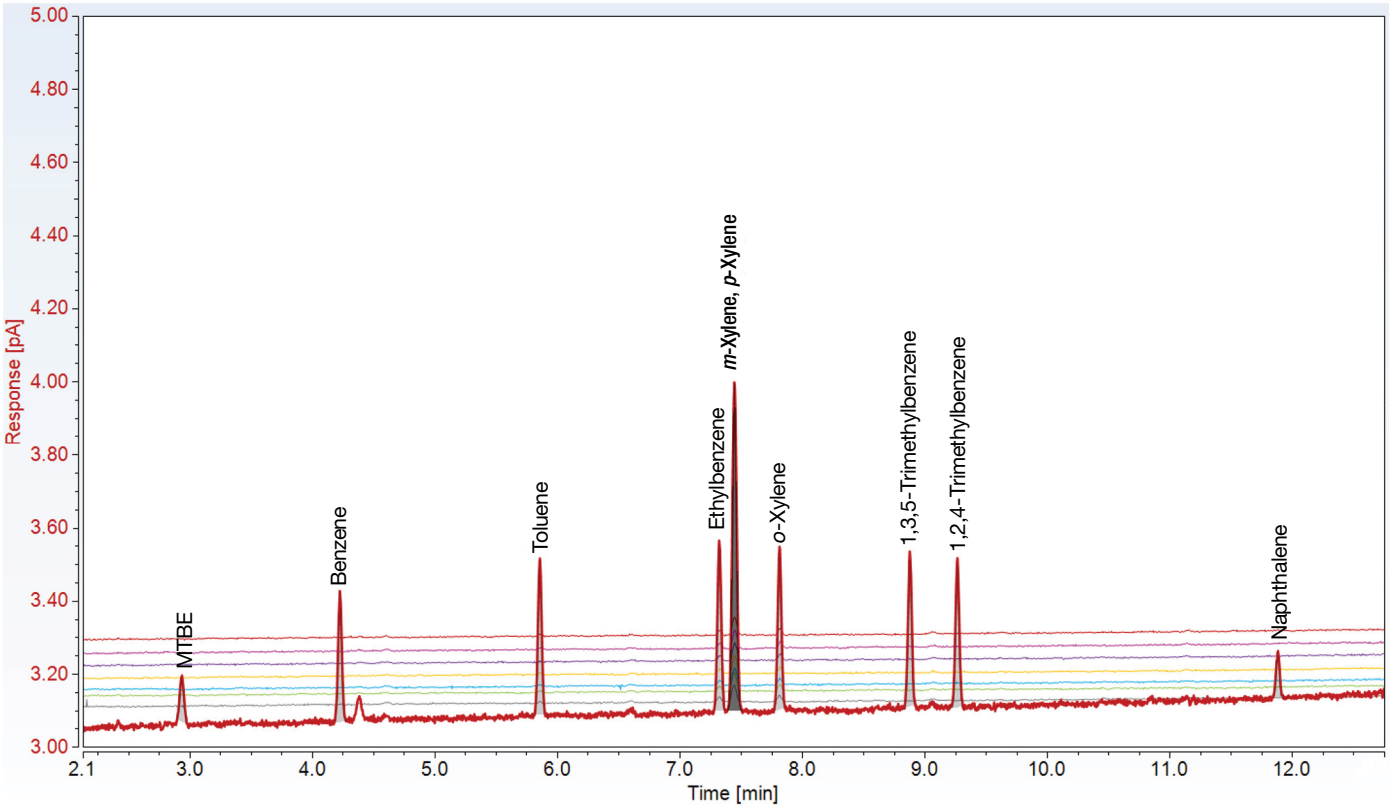


Figure 2. Comparison between chromatograms obtained analyzing n = 7 real tap water samples (unspiked) and a tap water sample spiked at 12.5 µg/L (ppb) (red trace). None of the investigated gasoline organics could be detected in the unspiked tap water samples.

Table 2. Calculated MDL, LOQ, and % recovery for n = 7 tap water samples spiked at a concentration level of 12.5 µg/L (ppb)

Gasoline Range Organics	Spiked Conc. (µg/L)	Average Measured Conc. (µg/L, n = 7)	Calculated MDL (µg/L)	Calculated LOQ (µg/L)	Average Recovery (%, n = 7)
Methyl *tert*-butyl ether (MTBE)	12.5	11.5	1.4	4.4	92
Benzene	12.5	12.8	1.2	3.9	103
Toluene	12.5	13.7	1.7	5.5	110
Ethylbenzene	12.5	12.8	1.3	4.0	102
m-Xylene, *p*-Xylene	12.5	12.8	0.8	2.7	103
o-Xylene	12.5	12.4	0.8	2.6	100
1,3,5-Trimethylbenzene	12.5	14.4	1.7	5.5	115
1,2,4-Trimethylbenzene	12.5	13.3	1.7	5.3	107
Naphthalene	12.5	14.6	2.2	7.1	117
Average		**13.1**	**1.4**	**4.6**	**105**

To assess the method accuracy (%) in tap water samples containing raw gasoline matrix, 30 µL of the sample stock solution (prepared as described in the sample preparation section) were diluted into two flasks previously filled with 30 mL of tap water and fortified with the standard solution at a concentration of 1000 µg/L (ppb) and 10,000 µg/L (ppb). A blank matrix solution was prepared by adding 30 µL of sample stock solution to 30 mL tap water. Then, 5 mL of each fortified solution and the blank matrix were transferred into 10 mL headspace vials (n = 5) and analyzed to assess the recovery. The average recoveries (%) for the spiked matrix samples were calculated using Equation 3 and confirmed to be within 80–120% of the spiked levels with an average value of 96.5% (Table 3). Chromeleon CDS matrix correction feature allowed for automated subtraction of the background leading to a precise quantitation of the spiked samples.

Precision

System repeatability was assessed using n = 10 consecutive injections of tap water samples spiked at a concentration of 50 µg/L (ppb) and n = 10 tap water samples spiked with the 5% raw gasoline solution. Peak area %RSDs obtained for both assessments are reported in Table 4. Excellent repeatability was obtained for both standard and matrix spiked samples with an average %RSD of 0.91 and 1.1, respectively.

Table 4. Peak area %RSDs obtained from n = 10 consecutive injections of tap water spiked with the standard solution at 50 µg/L (ppb) and n = 10 consecutive injections of tap water spiked with diluted raw gasoline. Average peak area %RSDs are 0.91 and 1.1 respectively.

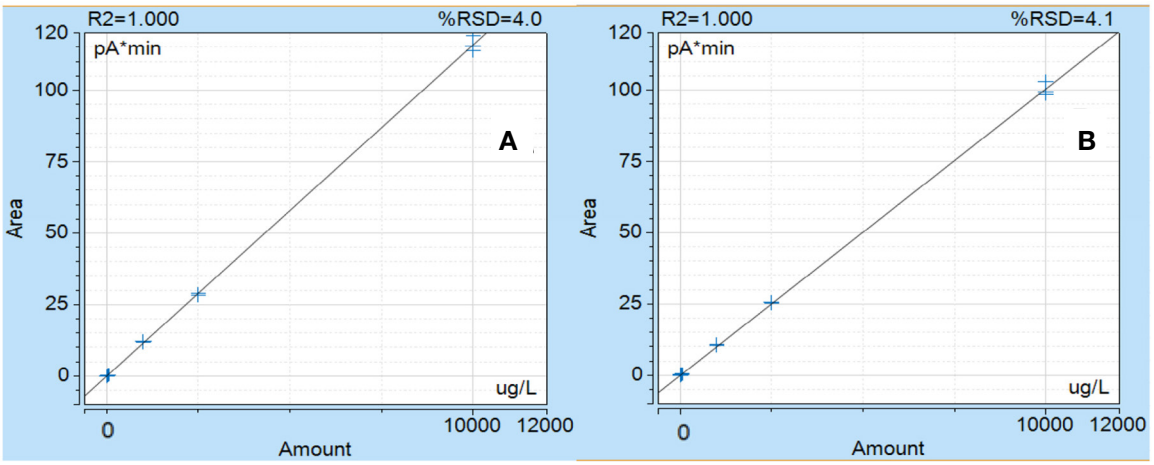
Gasoline Range Organics	Peak area %RSD	
	Tap Water Spiked with Stock Solution (n = 10)	Tap Water Spiked with Raw Gasoline (n = 10)
Methyl *tert*-butyl ether (MTBE)	1.0	1.0
Benzene	0.93	1.2
Toluene	0.87	1.1
Ethylbenzene	0.78	0.8
m-Xylene, *p*-Xylene	0.85	1.5
o-Xylene	0.92	1.2
1,3,5-Trimethylbenzene	0.98	1.2
1,2,4-Trimethylbenzene	0.99	1.1
Naphthalene	0.82	1.2
Average	**0.91**	**1.1**

Table 3. Calculated recoveries (%) for n = 5 tap water samples spiked with diluted raw gasoline and fortified with standard solution at a concentration of 1000 and 10,000 µg/L (ppb). Average concentrations are calculated subtracting the raw gasoline matrix.

Gasoline Range Organics	Average Blank Matrix Conc. (µg/L, n = 5)	Spiked Conc. 1 (µg/L)	Average Measured Conc. (µg/L, n = 5)	Average Recovery (%, n = 5)	Spiked Conc. 2 (µg/L)	Average Measured Conc. (µg/mL, n = 5)	Average Recovery (%, n = 5)
Methyl *tert*-butyl ether (MTBE)	7	1000	1,130	113	10,000	10,300	103
Benzene	4	1000	890	89	10,000	9,300	93
Toluene	142	1000	990	99	10,000	9,300	93
Ethylbenzene	25	1000	890	89	10,000	9,400	94
m-Xylene, *p*-Xylene	54	1000	900	90	10,000	9,300	93
o-Xylene	54	1000	920	92	10,000	9,300	93
1,3,5-Trimethylbenzene	8	1000	910	91	10,000	9,400	94
1,2,4-Trimethylbenzene	31	1000	920	92	10,000	9,200	92
Naphthalene	7	1000	1,160	116	10,000	10,500	105
Average			**970**	**97**		**9,600**	**96**

Quantitation of GRO in real water samples

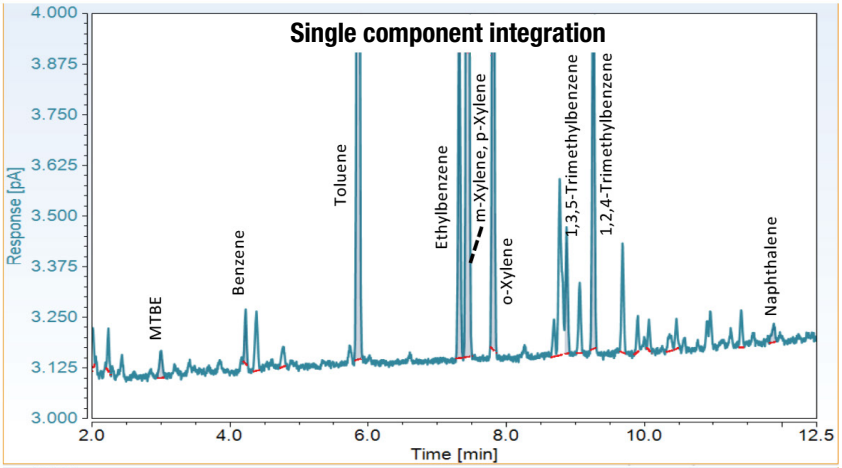
Tap water samples (5 mL, n = 10) were spiked with 1 µL of raw gasoline solution (5%) and analyzed. According to Wisconsin and EPA method 8015 C,[4] *GRO quantitation is based on a direct comparison of the total area within a defined retention time window to the total peak areas of the gasoline component standard.* Therefore, the calibration curves previously plotted using the single component peak integration were calculated integrating the total peak area and used to quantitate the spiked water samples. The total area was obtained integrating all the chromatographic peaks within the retention time window ranged from MTBE (RT = 2.92 min) to naphthalene (RT = 11.96 min) according to the Wisconsin method and from 2-methylpentane (RT = 2.62 min) to 1,2,4-trimethylbenzene (RT = 9.25 min) according to EPA 8015 C method. The *"baseline to baseline"* integration did not include the solvent peak. Calculated correlation coefficient (R^2) were 1.000 and the residual values (measured as % RSD of average response factors) were ~4% for both retention time windows confirming an excellent linearity. MDL, LOQ, and recovery were calculated for the total peak area calibration curves applying Equations 1, 2, and 3. Calibration curves and calculated MDL, LOQ, and percent recovery (total area integration applied) are shown in Figure 3. As an example, a chromatogram of a tap water sample (5 mL) spiked with raw gasoline solution (5%) (single component and EPA 8015 C total area integration) as well as the quantitation results obtained for the analyzed samples (single components and total area quantitation) are reported in Figure 4. A series of blank water vials (n = 5) was run after completing the sample sequence. No compound carry-over was detected in the blanks as demonstrated in Figure 5.



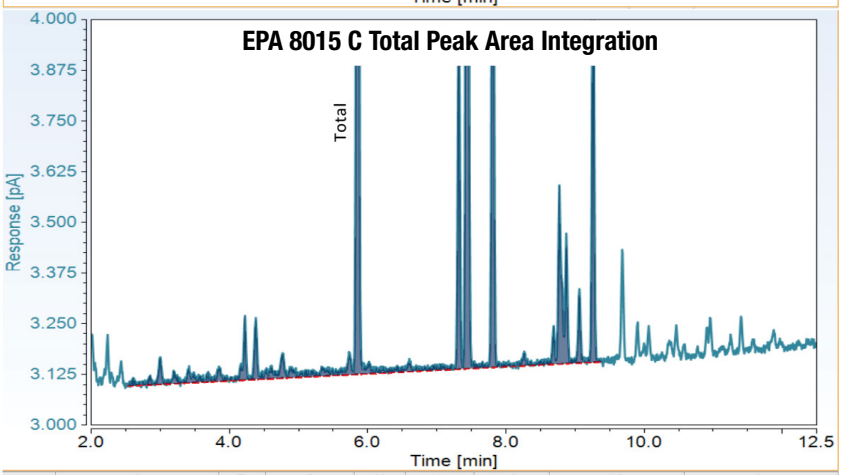
C

Gasoline Range Organics	Spiked Conc. (µg/L)	Average Measured Conc. (µg/L, n = 7)	Calculated MDL (µg/L)	Calculated LOQ (µg/L)	Average Recovery (%, n = 7)
Total Area Integration (Wisconsin method)					
Total	12.5	11.4	1.9	6.1	91
Total Area Integration (EPA 8015 C method)					
Total	12.5	13.0	2.2	7.0	105

Figure 3. Calibration curves were obtained integrating the total area within the gasoline range at each calibration level for Wisconsin (A) and EPA 8015 C (B). R^2, response factor relative standard deviations (% RSD) as well as calculated MDL, LOQ, and percent recovery (C) are shown.



A



B

	A	B	G	H	I	J	K	L
1	Peak Name		Rel Area		Amount		Channel	
2			%		ug/L			
4	MTBE	n.a.	0.61	n.a.	7.1	n.a.	FrontDetector	n.a.
5	Benzene	n.a.	0.73	n.a.	3.8	n.a.	FrontDetector	n.a.
6	Toluene	n.a.	31.02	n.a.	145.5	n.a.	FrontDetector	n.a.
7	Ethylbenzene	n.a.	5.72	n.a.	24.9	n.a.	FrontDetector	n.a.
8	m-xylene,p-xylene	n.a.	25.93	n.a.	54.0	n.a.	FrontDetector	n.a.
9	o-xylene	n.a.	11.51	n.a.	53.4	n.a.	FrontDetector	n.a.
10	1,3,5-trimethylbenzene	n.a.	1.97	n.a.	8.2	n.a.	FrontDetector	n.a.
11	1,2,4-trimethylbenzene	n.a.	7.17	n.a.	31.0	n.a.	FrontDetector	n.a.
12	naphthalene	n.a.	0.43	n.a.	6.6	n.a.	FrontDetector	n.a.
13	*n.a.*	*total*	*n.a.*	*100.00*	*n.a.*	*61.7*	*n.a.*	*FrontDetector_total*

Summary View | Peak Results | Library Search | System Suitability Tests | Calibration | Audit Trail

C

Gasoline Range Organics	Average Measured Conc. (µg/L, n = 10)	Average Measured Conc. (µg/L, n = 10)	
	Single Component Integration	Total Area Integration (Wisconsin)	Total Peak Area Integration (EPA 8015 C)
Methyl *tert*-butyl ether (MTBE)	7.1	53.3	56.0
Benzene	3.7		
Toluene	141.2		
Ethylbenzene	24.8		
m-Xylene, *p*-Xylene	53.1		
o-Xylene	53.7		
1,3,5-Trimethylbenzene	8.0		
1,2,4-Trimethylbenzene	31.1		

D

Figure 4. Example of tap water sample (5 mL) spiked with raw gasoline solution (5%) chromatogram applying single component integration (A) and total area integration (EPA 8015 C integration window), (B). Chromeleon "Peak Results" view (C) allows the display of the peak results for both integration types. Average quantitative results for n = 10 tap water samples spiked with raw gasoline and integrated using single components and total area are reported in the table (D).

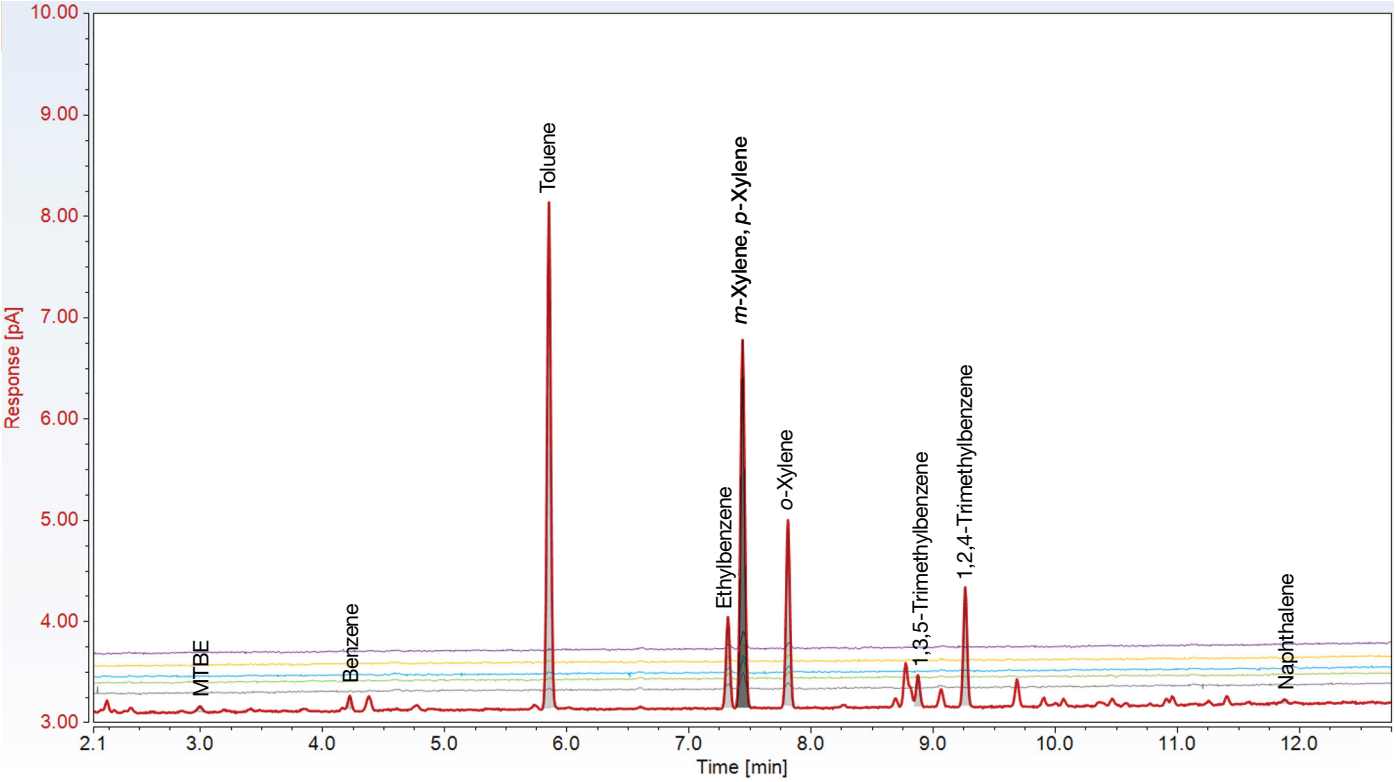


Figure 5. Comparison between chromatograms obtained analyzing n = 5 blank water vials after completing the sample sequence and a sample spiked with 1 µL of raw gasoline solution (5%) (red trace). None of the investigated gasoline organics or any residual matrix components could be detected in the blank water.

Conclusions

The results presented here demonstrate the suitability of the TriPlus 500 HS autosampler in combination with the Trace 1310 GC-FID for GRO analysis in environmental samples.

- Excellent linearity with correlation coefficient R^2 = 1.000 was obtained for all analytes. The Instant Connect Flame Ionization Detector (FID) allows sensitive detection of organic compounds as demonstrated by the calculated MDL and LOQ (average MDL = 1.4 µg/L (ppb) and average LOQ = 4.6 µg/L (ppb)).
- The advanced Quick Spin Shaking (QSS) feature of vials and direct column connection to the valve manifold ensure efficient analyte extraction. In the experiments performed here, the average compound recovery for matrix spiked samples was >96%.
- The low bleed and superior inertness of the TraceGOLD column allowed for highly reliable results. The high column efficiency allowed for a fast GC oven ramp supporting short analysis time (all analytes elute in <13 min) and high sample throughput to easily meet the needs of routine laboratories. Moreover, up to 240 sample vials can be accommodated into the trays for unattended 24-hour operations.
- The pneumatic control and the sample path inertness of the TriPlus 500 HS autosampler ensure reliable and reproducible analyte injection and transfer. Average peak area RSDs (n = 10 consecutive injections) were 0.91% for tap water samples spiked with the standard solution at 50 µg/L (ppb) and 1.1% for tap water spiked with diluted raw gasoline.
- The efficient purging of the pneumatic circuit of the TriPlus 500 HS autosampler eliminated potential for carry-over; no matrix components or gasoline organics were detected in the blank vials after a sequence of real samples contaminated with GRO chemicals.
- Quantitation of spiked samples is simplified with the Chromeleon CDS advanced reprocessing features allowing for easy single component and total peak area integration and compound quantitation.

Overall, the data shows that the TriPlus 500 gas chromatography static headspace autosampler provides a reliable analytical tool allowing environmental laboratories to produce consistent results with outstanding analytical performance for GRO quantitative analysis in water samples.

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