Supplementary Information

Seasonal and Spatial Distribution of Caffeine, Atrazine, Atenolol and DEET in Surface and Drinking Waters from the Brazilian Federal District

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Method development and validation

Mass spectrometry parameters

In the AB Sciex 3200 triple quadrupole, substance-dependent parameters, with the exception of the collision-activated dissociation (CAD), were automatically optimized by the software Analyst® (Sciex, Toronto, Canada). They are declustering potential (DP), collision energy (CE), entrance potential (EP), collision cell entrance potential (CEP) and collision cell exit potential (CXP). For CAD optimization, as well as for source-dependent parameters, a mix solution containing 500 μg L^{-1} of each analyte was used. Figure S1 shows the response variation for CAD optimization.

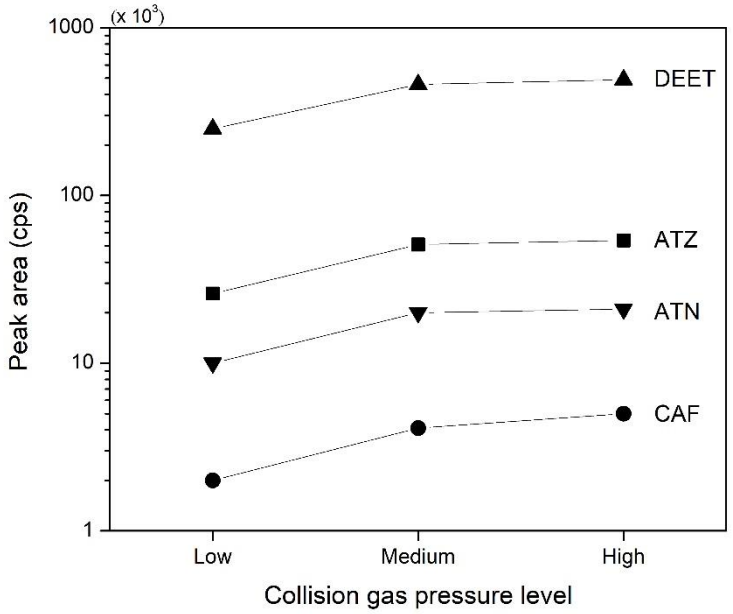

Figure S1. Optimization of the collision-activated dissociation (CAD) gas pressure. DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine. Low, medium and high-pressure levels are default conditions specified by the instrument.

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The high-pressure level for CAD produced higher peak areas for all substances. Figure S2 shows the optimization of the curtain gas pressure (CUR) between the curtain plate and the orifice plate.

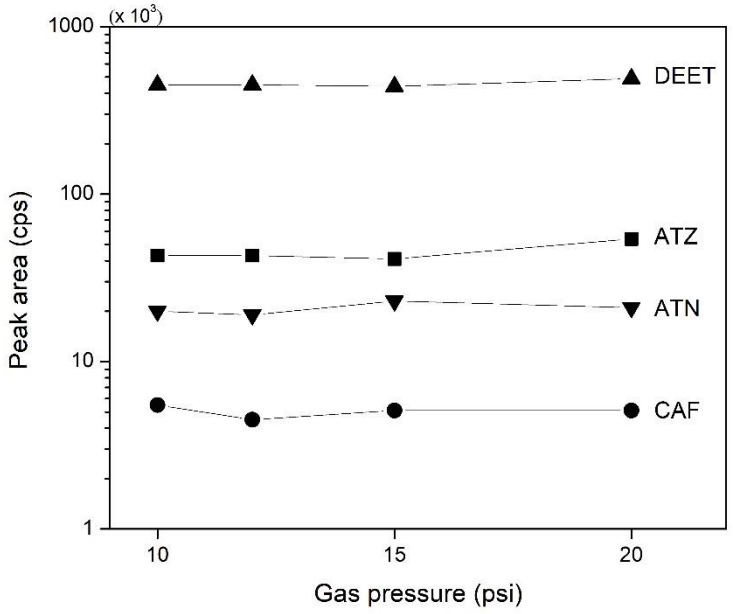

Figure S2. Optimization of the curtain gas pressure. DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine.

A CUR value of 20 psi produces the best response for atrazine and *N*,*N*-diethyl-*meta*-toluamide (DEET), whereas better results were obtained at 10 and 15 psi for caffeine and atenolol, respectively. Thus, a CUR of 20 psi was selected. Figure S3 shows the optimization for the source auxiliary gas pressure (GS1).

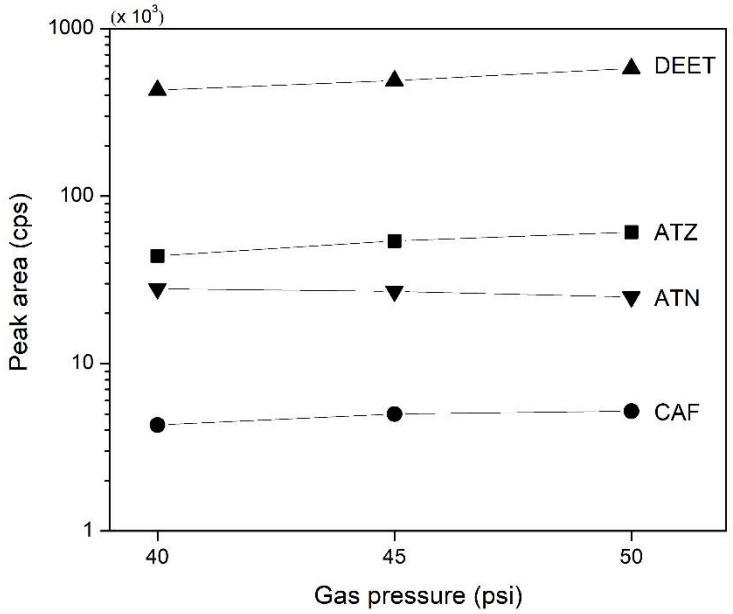

Figure S3. Optimization of the auxiliary gas pressure (GS1). DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine.

A pressure of 50 psi was selected for GS1 since produced higher peak areas for three substances. Figure S4 shows the optimization for the auxiliary drying gas pressure (GS2).

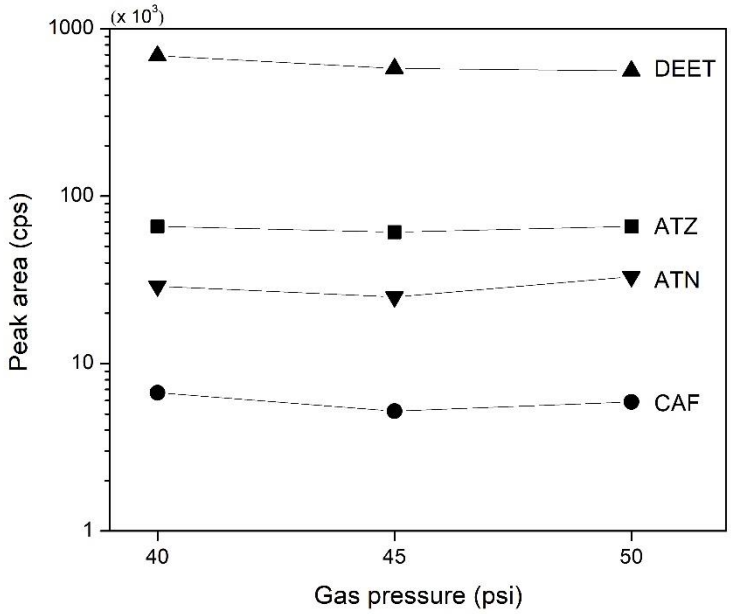

Figure S4. Optimization of the auxiliary gas pressure (GS2). DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine.

Better results for GS2 were obtained at 40 psi. Figure S5 portrayed the results obtained for the optimization of the potential applied to the electrospray ionization.

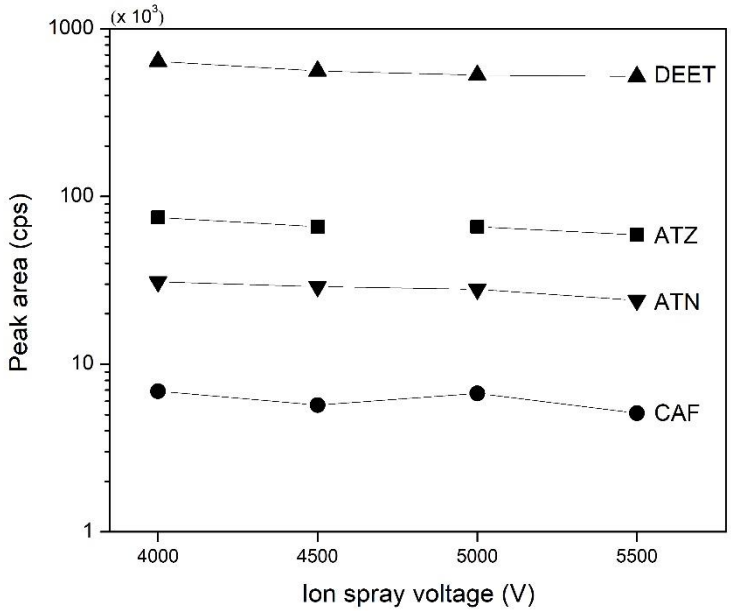

Figure S5. Optimization of the ion spray voltage. DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine.

It is possible to observe that all substances present the highest response at 4000 V. Figure S6 shows the variation of the gas temperature in the electrospray ionization source.

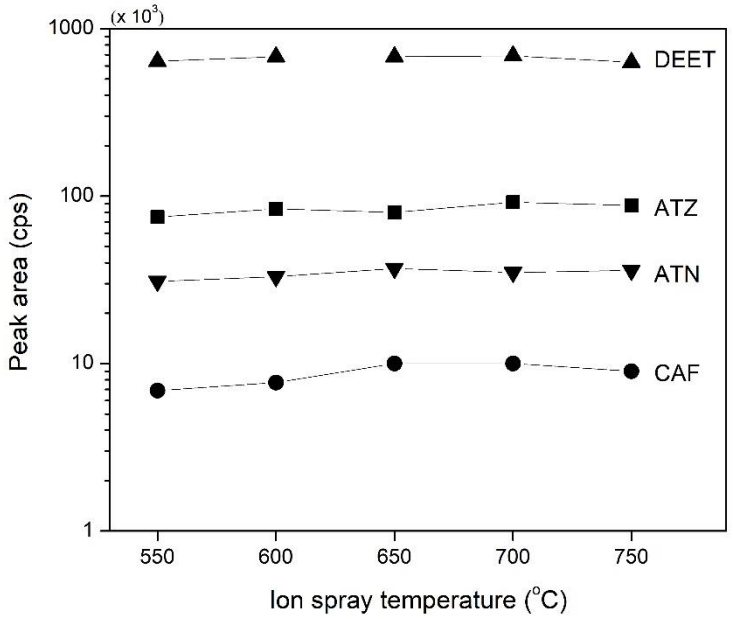

Figure S6. Optimization of the ion spray temperature. DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine.

An ion spray temperature of 700 °C produced the highest response for DEET and atrazine, whereas at 650 °C higher peak area was noticed for atenolol. Both temperatures produced the same response for caffeine. Thus, a temperature of 700 °C was selected.

Mobile phase conditions

Two buffer solutions were investigated as mobile phase additives: formic acid/ammonium formiate and acetic acid/ammonium acetate. Experiments were carried out using a standard solution prepared in water:acetonitrile (1:1 v/v) containing 5.0 μg L^{-1} of DEET and 50 μg L^{-1} of caffeine, atenolol and atrazine. A lower concentration of the former analyte was used due to its higher response, as can be seen in Figures S1 to S6. Successive chromatographic injections were performed in the isocratic mode (water:acetonitrile 1:1) varying the buffer employed as shown in Table S1.

Table S1. Additives evaluated in the mobile phase solvents

Condition	Deionized water / %		Acetonitrile / %
1	0.1 HCOOH	0.03 $HCOO^-$	1 HCOOH
2	0.1 HCOOH	0.03 $HCOO^-$	0.1 HCOOH
3	0.05 HCOOH	0.01 $HCOO^-$	0.05 HCOOH
4	0.05 HCOOH	0.03 $HCOO^-$	0.05 HCOOH
5	0.05 HCOOH	0.06 $HCOO^-$	0.05 HCOOH
6	0.1 H_3CCOOH	0.04 H_3CCOO^-	0.1 H_3CCOOH
7	1 H_3CCOOH	0.04 H_3CCOO^-	1 H_3CCOOH

Figure S7 shows the intensity of the analytical signal for the substances studied under each composition condition of the mobile phase tested. Through Figure S7 it is possible to notice that the condition 4 was the one that presented the highest values of areas for atenolol, atrazine and DEET. For caffeine the best results were obtained in

conditions 4 and 6. Thus, condition 4 was the one that presented the best results and was, therefore, the one chosen in this method.

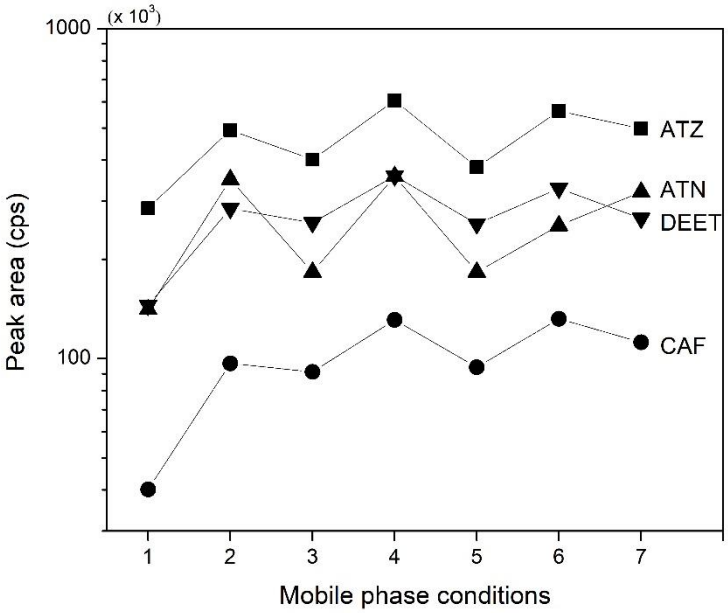

Figure S7. Optimization of the mobile phase condition under different additives composition. DEET: *N*,*N*-diethyl-*meta*-toluamide; ATZ: atrazine; ATN: atenolol; CAF: caffeine.

Working range and linearity

Determination was performed by external calibration using different analytical curves depending on the working range. Three analytical curves with eight points were constructed for each analyte except for caffeine. The latter presented higher levels in the samples leading us to use another analytical curve with a wider range. Table S2 shows curve parameters obtained for different working ranges.

Table S2. Analytical curve parameters obtained for each analyte in different working ranges

Working range / (μg L^{-1})	Caffeine			Atrazine			Atenolol			DEET		
	a	b	R^2	a	b	R^2	a	b	R^2	a	b	R^2
2.0-30	630	757	0.997	867	2740	0.999	874	2560	0.997	24900	46700	0.999
10-115	2440	1110	0.993	8890	3780	0.999	1160	2520	0.997	169000	52700	0.998
110-320	1990	1170	0.996	9320	2740	0.995	30100	2960	0.992	416000	39900	0.992
30-520	2470	1850	0.997	–	–	–	–	–	–	–	–	–

Concentration of each analyte on column. a: linear coefficient; b: angular coefficient; R^2: coefficient of determination.

Better sensitivities, expressed as the angular coefficients, were noticed for all analytes as the working ranges becomes higher, except for atrazine and DEET. For both analytes higher sensitivity was obtained at the working range from 10 to 115 μg L^{-1}. All analytical curves were homoscedastic and presented satisfactory correlation coefficients, i.e., higher than 0.992.

Precision

Repeatability was assessed at three different concentration levels by successive injections performed by the same analyst at the same day. Concentrations of the working solutions were 15, 90 and 250 µg L^{-1}, on column. Table S3 shows the intraday precision for each analyte obtained with ten replicates for each concentration level.

Table S3. Repeatability expressed by standard deviation and coefficient of variation obtained for each concentration level tested

	Caffeine			Atrazine			Atenolol			DEET		
Concentration / (µg L^{-1})	15	90	250	15	90	250	15	90	250	15	90	250
SD / (µg L^{-1})	1.48	2.45	5.57	0.44	1.46	2.72	3.69	2.92	3.53	0.58	1.35	10.5
CV / %	7.00	2.64	2.22	2.83	2.06	1.06	21.3	2.18	1.38	3.21	1.43	3.75

SD: standard deviation; CV: coefficient of variation; DEET: *N*,*N*-diethyl-*meta*-toluamide.

Coefficient of variation ranged from 1.06 to 3.75% except for atenolol and caffeine under the lowest concentration level. Caffeine presented a CV of 7.00% for a tested concentration of 15 µg L^{-1}, while atenolol presented a CV of 21.3% for the same tested concentration. Except for the latter situation, all intraday precision results were considered satisfactory, as determination were carried out by external calibration.

Intermediate precision was also obtained using the working solutions containing 15, 90 and 250 µg L^{-1} of each analyte, on column. However, in this case, the results were achieved by using ten replicates obtained in different days. Table S4 shows the standard deviation and the coefficient of variation obtained for the inter-day precision.

Table S4. Inter-day precision expressed by standard deviation and coefficient of variation obtained for each concentration level tested

	Caffeine			Atrazine			Atenolol			DEET		
Concentration / (µg L^{-1})	15	90	250	15	90	250	15	90	250	15	90	250
SD / (µg L^{-1})	2.35	11.0	33.3	0.64	5.49	23.7	4.41	14.2	29.3	0.52	5.26	17.6
CV / %	12.2	13.4	15.2	4.28	8.36	10.2	28.3	11.8	12.9	2.89	5.85	6.64

SD: standard deviation; CV: coefficient of variation; DEET: *N*,*N*-diethyl-*meta*-toluamide.

Higher CV were noticed for inter-day precision in comparison with the results obtained for intraday precision. Values ranged from 2.89 to 28.3%, being the higher CV also noticed for atenolol at the lowest concentration tested.

Accuracy

Recovery experiments were carried out in duplicates using samples obtained from Paranoá Lake previously filtered in 0.45 pore-sized cellulose acetate membranes and spiked with 40 ng L^{-1} of each analyte. Determination was carried out by external calibration and by standard addition in order to assess the matrix effect according to the parallelism test at 95% of significance. Table S5 shows the results obtained.

Table S5. Percentage of recovery for each analyte

	Caffeine	Atrazine	Atenolol	DEET
Recovery / %	80.2	64.1	64.7	78.9
SD / %	6.2	4.9	5.6	5.3

SD: standard deviation; DEET: *N*,*N*-diethyl-*meta*-toluamide.

Recovery for caffeine and DEET were satisfactory, but values obtained for atrazine and atenolol were lower than expected. The extraction procedure used in the present manuscript was originally development for a multi-residue study[1] carried out by the National Institute for Advanced Analytical Science and Technology (INCTAA) as pointed out in the main manuscript. As the method included the determination of other contaminants (not investigated in the samples from the Federal District), some of them may present less appreciable recoveries, but still with an acceptable standard deviation.

Reference

1. Machado, K. C.; Grassi, M. T.; Vidal, C.; Pescara, I. C.; Jardim, W. F.; Fernandes, A. N.; Sodré, F. F.; Almeida, F. V.; Santana, J. S.; Canela, M. C.; Nunes, C. R. O.; Bichinho, K. M.; Severo, F. J. R.; *Sci. Total Environ.* **2016**, *572*, 138.