

Supporting information

A multi-residue analytical method for extraction and analysis of pharmaceuticals and other selected emerging contaminants in sewage sludge

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Mass-spectrometry parameters

Tables S1-4 report the analytical parameters for the analysis of pharmaceuticals, the mass to charge ratio (m/z) of the precursor and product ions together with the optimized collision energies (CE) and the average MRM ratio. The pharmaceuticals are divided in the tables according to the extraction method (Oasis MCX or HLB cartridges) and the ionisation mode. Table S5 reports the analytical parameters of the labelled substances used as internal standards (IS) for pharmaceuticals, and Table S6 reports the analytical parameters used for the perfluorinated compounds and their respective IS.

Table S1: Analytes extracted with MCX cartridges and analysed in positive ionization mode.

Analytes	Precursor ion (m/z)	Declustering potential (DP; V)	Product ions (m/z)	Collision energy (CE; eV)	Collision Cell Exit Potential (CXP; eV)	MRM ratio
clarithromycin	748.2	100	158.1	36	10	3.72
			590.1	27		
dehydro-erythromycin	716.2	80	158.1	38	10	1.60
			558.1	23		
lincomycin	407.1	60	126.1	34	9	1.27
			359.1	25		
vancomycin	725.1	70	144.1	19	10	3.03
			100.1	23		
diclofenac	296.1	45	215.1	24	15	1.47
	298.1		250.1	17		
			252.1	17		
ketoprofen	255.1	120	209.1	19	14	1.51
			105.1	30		
			194.1	31		
paracetamol	152.1	90	110.1	22	9	2.66
			65.1	38		
salbutamol	240.1	60	148.1	25	14	2.54
			166.1	17		
atenolol	267.1	70	145.1	34	14	0.95
			190.1	25		
carbamazepine	237.1	70	194.1	26	13	4.53
			192.1	30		
10.11-dihydro-10.11-dihydroxy carbamazepine	271.1	50	180.1	50	13	1.45
			236.0	16		
desmethyldiazepam	271.1	100	140.1	37	12	1.47
			165.1	37		
diazepam	285.1	100	154.1	35	13	0.97
			193.1	43		
paroxetine	330.2	90	192.1	29	12	1.97
			70.1	50	11	

ranitidine	315.1	80	176.1	22	12	1.88
			130.1	31	9	
sildenafil	475.2	90	100.1	36	11	1.69
			283.1	52	20	

Table S2: Analytes extracted with MCX cartridges and analysed in negative ionization mode.

Analytes	Precursor ion (m/z)	Declustering potential (DP; V)	Product ions (m/z)	Collision energy (CE; eV)	Collision Cell Exit Potential (CXP; eV)	MRM ratio
ibuprofen	205.1	-50	161.1	-10	-11	N.D.
naproxen	229.2	-50	170.0	-20	-12	1.08
			169.1	-39		
			185.1	-9		
17-β-estradiol	271.2	-130	183.1	-53	-12	1.04
			145.1	-52		
			239.2	-52		
estrone	269.2	-140	145.1	-49	-12	7.75
			159.1	-48		
17-α-ethinylestradiol	295.2	-140	145.1	-52	-12	1.54
			159.1	-46		
			183.1	-51		
bezafibrate	360.2	-80	274.2	-24	-14	2.13
			154.1	-38	-10	
clofibric acid	213.1	-45	127.1	-20	-9	3.83
			85.1	-13		
gemfibrozil	249.2	-70	121.1	-20	-9	1.53
			127.1	-14		

Table S3: Analytes extracted with HLB cartridges and analysed in positive ionization mode.

Analytes	Precursor ion (m/z)	Declustering potential (DP; V)	Product ions (m/z)	Collision energy (CE; eV)	Collision Cell Exit Potential (CXP; eV)	MRM ratio
cefazolin	455.1	60	323.1	15	13	1.61
			156.1	21		
ciprofloxacin	332.1	70	288.1	23	15	1.64
			245.1	32		
ofloxacin	362.1	70	318.1	26	20	1.33
			261.1	36		
sulphamethoxazole	254.1	60	156.1	21	10	1.12
			92.1	33		
cyclophosphamide	261.1	60	140.1	29	12	2.55
			106.1	24		
methotrexate	455.2	90	308.1	27	20	2.15
			175.1	49	12	
enalapril	377.1	80	234.1	26	15	3.48
			160.1	36	10	
enalaprilat	349.1	60	206.1	25	14	3.60
			160.1	34	9	
irbesartan	429.1	80	207.1	34	15	4.75
			195.1	31		
losartan	423.2	70	207.1	32	14	3.23
			377.1	21		
valsartan	436.1	70	235.1	25	15	1.04
			291.1	24		
ramipril	417.2	90	234.1	30	14	2.86
			117.1	53	12	
ramiprilat	389.1	80	206.1	28	13	1.86
			156.1	27		
simvastatin	419.1	70	199.1	20	14	1.06

			285.1	15	18	
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Table S4: Analytes extracted with HLB cartridges and analysed in negative ionization mode.

Analytes	Precursor ion (m/z)	Declustering potential (DP; V)	Product ions (m/z)	Collision energy (CE; eV)	Collision Cell Exit Potential (CXP; eV)	MRM ratio
furosemide	329.2	-80	285.2	-21	-14	1.23
			205.2	-30		
hydrochlorothiazide	269.2	-100	269.2	-26	-14	0.94
			205.1	-31		
atorvastatin	557.2	-70	278.2	-59	-23	1.19
			397.2	-40		
rosuvastatin	480.2	-100	418.2	-19	-20	2.18
			340.2	-30		
			298.2	-39		

Table S5: Parameters for analysis of the labelled internal standards in positive and negative ionization modes.

Analytes	Precursor ion (m/z)	Declustering potential (DP; V)	Product ions (m/z)	Collision energy (CE; eV)	Collision Cell Exit Potential (CXP; eV)
Internal Standards used in positive ionization mode					
atenolol-d7	274.1	70	145.1	35	14
carbamazepine-d10	247.1	50	204.1	29	14
ciprofloxacin-d8	340.1	70	296.1	25	18
ketoprofen-d3	258.1	100	212.1	19	14
ofloxacin-d3	365.1	70	321.0	27	21
ramipril-d5	422.1	80	239.1	30	14
salbutamol-d3	234.1	60	151.1	26	14
simvastatine-d6	425.1	70	199.1	20	13
sulphamethoxazole-d4	258.1	60	160.1	22	12
valsartan-d3	439.1	70	235.1	27	15
Internal Standards used in negative ionization mode					

bezafibrate-d6	366.2	-80	274.1	-24	-17
17-β-estradiol-d3	274.2	-130	185.2	-54	-12
ibuprofen-d3	208.2	-50	164.1	-10	-11
ketoprofen-d3	256.2	-50	212.2	-10	-13

Table S6: Parameters for the analysis of perfluorooctanoate (PFOA) and perfluorooctane sulfonate (PFOS).

Analytes	Precursor ion (m/z)	Declustering potential (DP; V)	Product ions (m/z)	Collision energy (CE; eV)	Collision Cell Exit Potential (CXP; eV)	MRM ratio
PFOA	413.2	-40	169.1	-26	-12	0.42
			369.1	-26		
PFOS	499.2	-70	80.1	-80	-12	6.28
			99.1	-65		
¹³C₄-PFOA	417.2	-40	172.1	-40	-12	-
¹³C₄-PFOS	503.2	-70	80.1	-80	-12	-

Calculation of recoveries

Table S7 and S8 report, respectively for MCX and HLB cartridges, the raw data used to calculate the recovery of each of the three replicates. The pharmaceuticals are grouped according to their therapeutic classes.

Table S7: Concentrations of analytes in blanks and spiked samples and percentage recoveries calculated from MCX cartridges extraction.

Analytes	Concentrations in blanks (ng/g)			Δ blank matrix-procedural blank (ng/g)		Mean conc. in matrices (ng/g)	Measured concentration in recoveries (ng/g)			Δ Concentrations in recoveries – average matrix concentration (%)			Mean recoveries (%)
	Procedural Blank	Matrix Blank 1	Matrix Blank 2	Matrix blank 1	Matrix blank 2		Rec 1	Rec 2	Rec 3	Rec 1	Rec 2	Rec 3	
ANTIBIOTICS													
cefazolin	0	0.00	0.00	0.00	0.00	0.00	143.07	37.38	39.31	71.54	18.69	19.65	36.6
ciprofloxacin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
clarithromycin	0.00	30.71	17.01	30.71	17.01	23.86	210.01	179.80	187.54	93.07	77.97	81.84	84.3
dehydro-erythromycin	0.00	3.93	4.88	3.93	4.88	4.40	236.65	183.17	203.69	116.13	89.38	99.64	101.7
lincomycin	0.00	0.87	0.86	0.87	0.86	0.87	169.89	183.01	171.55	84.51	91.07	85.34	87.0
ofloxacin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
sulphamethoxazole	0.00	5.30	4.98	5.30	4.98	5.14	212.46	187.79	185.99	103.66	91.32	90.43	95.1
vancomycin	0.00	60.90	77.37	60.90	77.37	69.13	201.59	165.49	160.84	66.23	48.18	45.85	53.4
ANTICANCER AGENTS													
cyclophosphamide	0.00	0.00	0.00	0.00	0.00	0.00	124.73	108.22	121.03	62.36	54.11	60.52	59.0
methotrexate	0	0.00	0.00	0.00	0.00	0.00	157.74	160.81	98.89	78.87	80.41	49.44	69.6
ANTIHYPERTENSIVES													
enalapril	0.00	0.00	0.00	0.00	0.00	0.00	151.12	154.08	172.98	75.56	77.04	86.49	79.7
enalaprilat	0.00	0.00	0.00	0.00	0.00	0.00	125.67	121.86	109.12	62.83	60.93	54.56	59.4
irbesartan	0.00	134.92	167.50	134.92	167.50	151.21	249.96	344.39	285.36	49.38	96.59	67.08	71.0
losartan	0.00	21.25	27.85	21.25	27.85	24.55	152.73	193.90	164.29	64.09	84.68	69.87	72.9
ramipril	0.00	0.00	0.00	0.00	0.00	0.00	170.88	172.22	172.09	85.44	86.11	86.04	85.9
ramiprilat	0.00	1.01	1.18	1.01	1.18	1.10	120.21	119.99	95.88	59.56	59.45	47.39	55.5
valsartan	0.00	40.15	68.98	40.15	68.98	54.57	204.98	214.63	204.32	75.21	80.03	74.88	76.7
ANTI INFLAMMATORY AND ANALGESICS													
diclofenac	7.37	54.35	54.85	46.97	47.47	47.22	197.88	188.26	201.44	75.33	70.52	77.11	74.3
ibuprofen	0.00	0.00	0.00	0.00	0.00	0.00	150.25	174.12	165.66	75.13	87.06	82.83	81.7

ketoprofen	4.79	6.07	6.14	1.28	1.35	1.32	176.59	179.49	193.62	87.64	89.09	96.15	91.0
naproxen	0.52	1.16	1.01	0.64	0.49	0.56	196.72	199.54	196.71	98.08	99.49	98.07	98.5
paracetamol	0	0.00	0.00	0.00	0.00	0.00	106.26	101.39	121.85	53.13	50.70	60.92	54.9
ANTI-ASTHMATICS													
salbutamol	0.00	0.00	0.00	0.00	0.00	0.00	153.66	168.02	158.66	76.83	84.01	79.33	80.1
CARDIOVASCULARS													
atenolol	0.00	2.36	2.53	2.36	2.53	2.44	186.35	177.03	178.14	91.95	87.29	87.85	89.0
ANTIDEPRESSANTS													
carbamazepine	0.00	9.88	11.89	9.88	11.89	10.89	189.93	194.39	209.08	89.52	91.75	99.10	93.5
10.11-dihydro-10.11-dihydroxycarbamazepine	0.00	3.93	4.88	3.93	4.88	4.40	236.65	183.17	203.69	116.13	89.38	99.64	101.7
desmethyldiazepam	0.00	0.53	0.58	0.53	0.58	0.56	166.93	203.56	190.54	83.47	101.78	95.27	93.5
diazepam	0.00	0.21	0.26	0.21	0.26	0.23	138.54	168.57	161.59	69.27	84.28	80.79	78.1
paroxetine	0.00	11.73	15.72	11.73	15.72	13.72	290.86	312.52	285.28	138.57	149.40	135.78	141.2
DIURETICS													
furosemide	0.00	4.18	4.16	4.18	4.16	4.17	75.15	84.17	84.70	35.49	40.00	40.27	38.6
hydrochlorothiazide	0.00	8.59	7.77	8.59	7.77	8.18	67.42	63.88	64.61	29.62	27.85	28.21	28.6
ESTROGENS													
17-β-estradiol	0.00	0.00	0.00	0.00	0.00	0.00	228.08	207.48	213.50	114.04	103.74	106.75	108.2
estrone	0.00	2.98	2.12	2.98	2.12	2.55	270.93	285.28	331.68	134.19	141.36	164.56	146.7
17-α-ethinylestradiol	0.00	0.00	0.00	0.00	0.00	0.00	166.15	239.91	260.90	83.07	119.96	130.45	111.2
ANTIREFLUX AGENTS													
ranitidine	0.00	22.57	26.90	22.57	26.90	24.74	142.52	170.28	139.93	58.89	72.77	57.60	63.1
LIPID REGULATORS													
atorvastatin	0.00	3.43	4.07	3.43	4.07	3.75	130.11	138.66	135.19	63.18	67.45	65.72	65.5
bezafibrate	0.00	0.00	0.00	0.00	0.00	0.00	171.46	175.73	168.28	85.73	87.87	84.14	85.9
clofibrac acid	0.00	0.00	0.00	0.00	0.00	0.00	142.90	150.04	143.90	71.45	75.02	71.95	72.8
gemfibrozil	0.00	0.00	0.00	0.00	0.00	0.00	135.59	125.27	127.45	67.80	62.63	63.73	64.7
rosuvastatin	0.00	0.00	0.00	0.00	0.00	0.00	117.18	113.27	118.21	58.59	56.63	59.10	58.1
simvastatin	0.00	0.00	0.00	0.00	0.00	0.00	186.45	170.71	158.15	93.23	85.35	79.07	85.9
ERECTILE DYSFUNCTION AGENTS													

sildenafil	0.00	0.00	0.00	0.00	0.00	0.00	261.49	287.97	276.45	130.75	143.99	138.22	137.7
PERFLUOROALKYLPHENOLS													
PFOA	0.00	2.10	2.00	2.10	2.00	2.00	203.1	198.3	192.7	100.6	98.2	95.3	98.0
PFOS	0.00	11.90	11.20	11.90	11.20	11.5	224.3	214.6	215.4	106.4	101.6	102.0	103.3

Table S8: Concentrations of analytes in blanks and spiked samples and percentage recoveries calculated from HLB cartridge extraction.

Analytes	Concentrations in blanks (ng/g)			Δ blank matrix-extraction blank (ng/g)		Mean conc. in matrices (ng/g)	Measured concentration in recoveries (ng/g)			Δ Concentrations in recoveries – average matrix concentration (%)			Mean recoveries (%)
	Extracti on Blank	Matrix Blank 1	Matrix Blank 2	Matrix blank 1	Matrix blank 2		Rec 1	Rec 2	Rec 3	Rec 1	Rec 2	Rec 3	
ANTIBIOTICS													
cefazolin	0.00	0.00	0.00	0.00	0.00	0.00	191.63	137.23	107.04	95.82	68.62	53.52	72.6
ciprofloxacin	9.83	890.54	903.00	880.71	893.17	886.94	1046.79	1092.87	1073.32	79.92	102.96	93.19	92.0
clarithromycin	0.00	98.06	121.56	98.06	121.56	109.81	313.21	297.41	242.84	101.70	93.80	66.51	87.3
dehydro-erythromycin	0.00	0.28	1.15	0.28	1.15	0.72	374.30	395.27	353.60	186.79	197.28	176.44	186.8
lincomycin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
ofloxacin	0.00	961.78	1108.70	961.78	1108.70	1035.24	1185.30	1252.45	1222.97	75.03	108.60	93.86	92.5
sulphamethoxazole	0.00	4.61	4.46	4.61	4.46	4.54	214.37	196.27	194.07	104.91	95.87	94.77	98.5
vancomycin	0.00	28.60	53.12	28.60	53.12	40.86	153.28	158.79	114.92	56.21	58.97	37.03	50.7
ANTICANCER AGENTS													
cyclophosphamide	0.00	0.00	0.00	0.00	0.00	0.00	167.96	163.51	149.34	83.98	81.75	74.67	80.1
methotrexate	0.00	0.00	0.00	0.00	0.00	0.00	212.94	206.68	151.91	106.47	103.34	75.96	95.3
ANTIHYPERTENSIVES													
enalapril	0.00	0.00	0.00	0.00	0.00	0.00	139.73	154.48	201.57	69.86	77.24	100.78	82.6
enalaprilat	0.00	0.00	0.00	0.00	0.00	0.00	161.17	180.43	227.49	80.59	90.22	113.75	94.8
irbesartan	0.00	127.08	107.15	127.08	107.15	117.12	241.86	312.40	322.87	62.37	97.64	102.88	87.6
losartan	0.00	24.22	22.96	24.22	22.96	23.59	145.14	176.73	170.78	60.78	76.57	73.60	70.3
ramipril	0.00	0.00	0.00	0.00	0.00	0.00	158.44	160.30	172.24	79.22	80.15	86.12	81.8
ramiprilat	0.00	1.87	1.66	1.87	1.66	1.76	186.68	216.24	264.20	92.46	107.24	131.22	110.3

valsartan	0.00	40.05	68.98	40.05	68.98	54.51	200.80	204.75	196.01	73.14	75.12	70.75	73.0
ANTI INFLAMMATORY AND ANALGESICS													
diclofenac	7.04	21.13	40.71	14.09	33.67	23.88	211.13	120.32	71.34	93.62	48.22	23.73	55.2
ibuprofen	0.00	0.00	0.00	0.00	0.00	0.00	155.94	168.23	165.88	77.97	84.12	82.94	81.7
ketoprofen	7.03	14.59	12.96	7.55	5.92	6.74	173.81	173.55	196.32	83.54	83.41	94.79	87.2
naproxen	0.70	1.21	1.45	0.00	0.75	0.38	184.05	152.68	146.72	91.84	76.15	73.17	80.4
paracetamol	0.00	0.00	0.00	0.00	0.00	0.00	68.55	94.50	71.46	34.28	47.25	35.73	39.1
ANTI-ASTHMATICS													
salbutamol	4.78	0.00	0.00	0.00	0.00	0.00	173.03	202.44	206.36	86.52	101.22	103.18	97.0
CARDIOVASCULARS													
atenolol	3.679	0.00	0.00	0.00	0.00	0.00	206.97	183.85	178.69	103.48	91.92	89.35	94.9
ANTIDEPRESSANTS													
carbamazepine	0.00	8.86	12.70	8.86	12.70	10.78	206.54	195.01	197.72	97.88	92.11	93.47	94.5
10.11-dihydro-10.11-dihydroxy carbamazepine	0.00	4.53	5.88	4.53	5.88	5.21	228.41	249.09	255.09	111.60	121.94	124.94	119.5
desmethyldiazepam	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
diazepam	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
paroxetine	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
DIURETICS													
furosemide	0.00	9.26	6.41	9.26	6.41	7.84	81.74	153.56	212.91	36.95	72.86	102.54	70.8
hydrochlorothiazide	0.00	27.54	15.59	27.54	15.59	21.56	81.89*	177.43	295.98	30.16*	77.93	137.21	107.2
ESTROGENS													
17-β-estradiol	0.00	0.00	0.00	0.00	0.00	0.00	203.95	188.57	181.93	101.98	94.28	90.96	95.7
estrone	0.00	0.00	0.00	0.00	0.00	0.00	352.17	258.76	209.16	176.09	129.38	104.58	136.68
17-α-ethinylestradiol	0.00	0.00	0.00	0.00	0.00	0.00	258.59	201.77	171.14	129.29	100.88	85.57	105.2
ANTIREFLUX AGENTS													
ranitidine	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
LIPID REGULATORS													
atorvastatin	0.00	3.28	4.45	3.28	4.45	3.86	208.08	154.04	159.21	102.11	75.09	77.68	85.0
bezafibrate	0.00	0.00	0.00	0.00	0.00	0.00	166.21	174.54	165.60	83.11	87.27	82.80	84.4
clofibric acid	0.00	0.00	0.00	0.00	0.00	0.00	157.06	296.40	456.47	78.53	148.20	228.23	151.7

gemfibrozil	0.28	0.59	0.68	0.32	0.41	0.36	137.95	131.40	82.04	68.79	65.52	40.84	58.4
rosuvastatin	0.00	0.00	0.00	0.00	0.00	0.00	141.94	264.38	371.92	70.97	132.19	185.96	129.7
simvastatin	0.00	0.00	0.00	0.00	0.00	0.00	186.10	171.26	166.61	93.05	85.63	83.30	87.3
ERECTILE DYSFUNCTION													
sildenafil	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
PERFLUOROALKYLPHENOLS													
PFOA	0.00	2.1	2.0	2.1	2.0	2.0	208.61	214.95	234.04	103.3	106.5	116.0	108.59
PFOS	0.00	11.9	11.2	11.9	11.2	11.5	218.88	226.71	234.85	103.7	107.6	111.7	107.65

* Excluded from the calculation.