

Removal Mechanisms of Illicit and Psychoactive Drugs during Wastewater Treatment

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Supporting Information

Table S1 Physicochemical properties of the illicit and psychoactive drugs studied.

Compound	MW (g/mol)	Log K _{ow}	Log D ^c	pK _a	K _{biol} (L g ⁻¹ ss day ⁻¹)
Diazepam	284.74	2.82	2.92	3.4	≤ 0.16 ^a
Flunitrazepam	309.45	2.06	2.03	1.8	^f
Venlafaxine	313.86	3.20	1.43	10.09	^f
Citalopram	405.30	3.74	1.27	9.78	3 ^c
Fluoxetine	309.33	4.05	1.75	9.8	9 ^c
Sertraline	342.69	5.51	3.14	9.16	^f
Aripiprazole	448.39	5.30	4.99	7.6	^f
Risperidone	410.48	3.49	1.81	8.76	^f
Ketamine	237.725	2.18	2.07	7.5	<0.1% ^b
Phentermine	149.23	1.90	-0.17	9.84	^f
Tramadol	263.38	3.01	0.52	9.41	≤ 0.13 ^a
Codeine	299.36	1.19	0.28	8.2	4.8 ± 0.6 ^a
Morphine	285.34	0.89	-0.13	8.21	13.5 ± 3.4 ^a
Oxycodone	315.36	0.66	0.45	8.53	0.21 ± 0.15 ^a
Methadone	309.45	3.93	2.8	9.2	0.18 ± 0.06 ^a
Fentanyl	336.471	4.05	3.01	8.99	^f
Methamphetamine	149.23	2.07	-0.57	9.87	4.8 ^b

Amphetamine	135.21	1.76	-0.62	9.9	19.4 ^b
Cocaine	303.35	2.30	1.22	8.61	f

a 1

b 2

c 3

d 4

^e Log D data from ChemSpider – Log D values are at pH 7.4

^f Data not available

Table S2 Summary of UHPLC-MS conditions used in the analytical method.

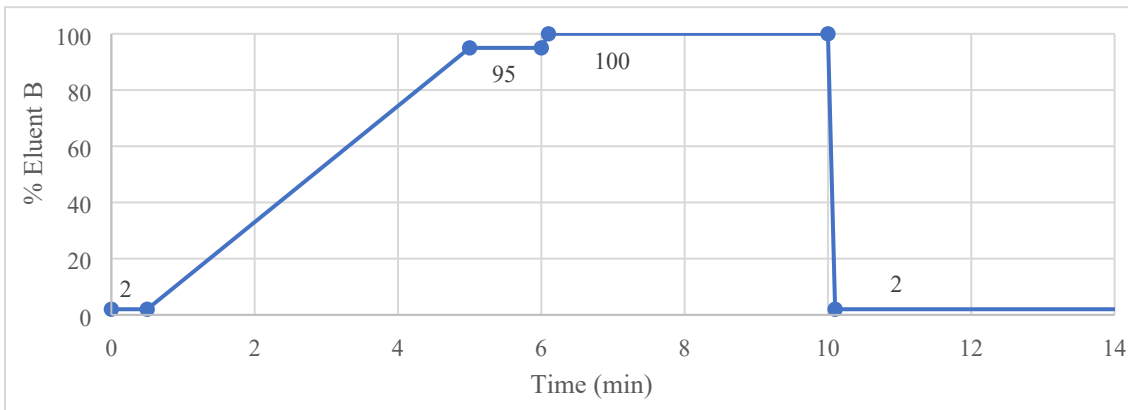
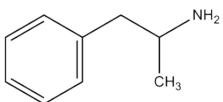
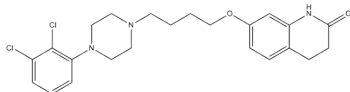
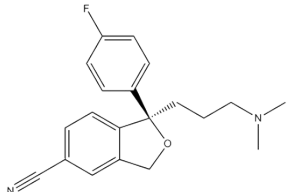
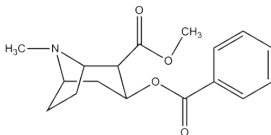
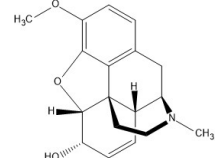
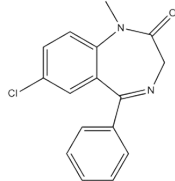
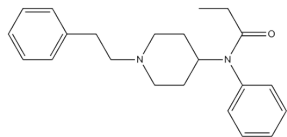
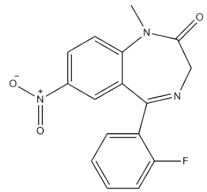
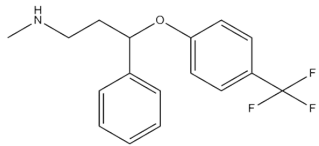
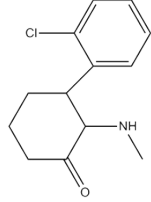
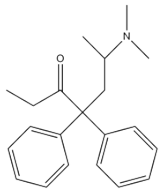
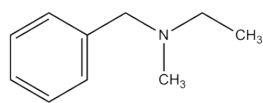
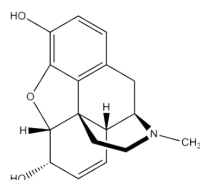
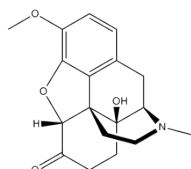
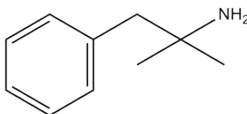
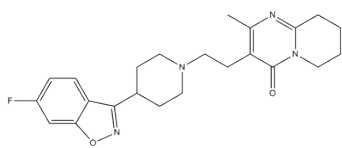
UHPLC Parameters	
Injection volume	25 µL raw and treated wastewater
Flow rate	0.4 mL/min
Column	Hypersil C18 column (100 x 2.1 mm, 1.9 µm)
Eluent A	Ammonium formate (10 mM) with 0.01% (v/v) formic acid
Eluent B	Methanol with 0.01% (v/v) formic acid
MS Parameters	
Ion Source Type	Max NG API source
Positive Ion (V)	3500
Vaporiser Temp (°C)	350
Ion Transfer Tube Temp (°C)	325
Mobile Phase Composition	
	

Table S3 Specific MS/MS parent and daughter ions monitored for each analyte, and the surrogate standard used for quantification.

Compound	Parent ion [m/z]	Daughter ions [m/z]	Surrogate standard
Amphetamine	136.2	119.2 91.2	Amphetamine-d6
Amphetamine-d6	142.1	125.2	N/A

Aripiprazole	450.1	287.1 176.1	Amphetamine-d6
Citalopram	325.2	262.1 109.2	Venlafaxine-d6
Cocaine	304.2	182.1 82.2	Cocaine-d3
Cocaine-d3	307.2	185.1	N/A
Codeine	300.3	215.3 183.2	Codeine-d6
Codeine-d6	306.3	218.1	N/A
Diazepam	285.1	154.1 257.1	Diazepam-d5
Diazepam-d5	290.1	262.1	N/A
Fentanyl	337.1	188.1 105.2	Fentanyl-d5
Fentanyl-d5	342.2	188.1	N/A
Flunitrazepam	314.3	268.1 239.1	Flunitrazepam-d7
Flunitrazepam-d7	321.1	275.1	N/A
Fluoxetine	310.1	44.3 148.1	Fluoxetine-d6
Fluoxetine-d6	316.2	44.3	N/A
Ketamine	238.1	125.0 220.1	Ketamine-d4
Ketamine-d4	242.1	129.0	N/A
Methadone	310.1	105.1 265.2	Methadone-d9
Methadone-d9	319.1	268.3	N/A
Methamphetamine	150.1	91.1 119.2	Methamphetamine-d9
Methamphetamine-d9	159.2	93.2	N/A
Morphine	286.2	201.2 183.1	Morphine-d6
Morphine-d6	292.3	181.1	N/A
Oxycodone	316.2	298.2 256.2	Oxycodone-d6
Oxycodone-d6	322.2	304.2	N/A
Phentermine	150.1	133.1 91.1	Phentermine-d5
Phentermine-d5	155.1	96.1	N/A
Risperidone	411.2	191.1 110.1	Fentanyl-d5
Sertraline	306.0	274.9 158.9	Fluoxetine-d6
Tramadol	264.1	58.2	Tramadol-d3
Tramadol-d3	268.1	58.2	N/A
Venlafaxine	278.1	58.2 260.1	Venlafaxine-d6
Venlafaxine-d6	284.2	64.2	N/A

Table S4 Elemental formula and structure of the illicit and psychoactive drugs investigated.

Amphetamine $C_9H_{13}N$		Aripiprazole $C_{23}H_{27}Cl_2N_3O_2$	
Citalopram $C_{20}H_{21}FN_2O$		Cocaine $C_{17}H_{21}NO_4$	
Codeine $C_{18}H_{21}NO_3$		Diazepam $C_{16}H_{13}ClN_2O$	
Fentanyl $C_{22}H_{28}N_2O$		Flunitrazepam $C_{16}H_{12}FN_3O_3$	
Fluoxetine $C_{17}H_{18}F_3NO$		Ketamine $C_{13}H_{16}ClNO$	
Methadone $C_{21}H_{27}NO$		Methamphetamine $C_{10}H_{15}N$	
Morphine $C_{17}H_{19}NO_3$		Oxycodone $C_{18}H_{21}NO_4$	
Phentermine $C_{10}H_{15}N$		Risperidone $C_{23}H_{27}FN_4O_2$	

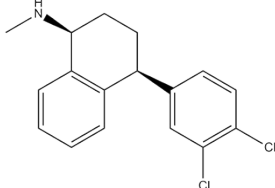
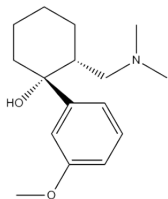
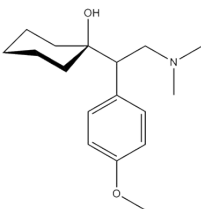
Sertraline $C_{17}H_{17}Cl_2N$		Tramadol $C_{16}H_{25}NO_2$	
Venlafaxine $C_{17}H_{27}NO_2$			

Table S5 LODs and LOQs for each analyte in raw and treated wastewater. LODs and LOQs were calculated from signal-to-noise ratios equal to 3 and 10, respectively, following the protocol used by Gros et al.⁵

Compound	LOD (ng L ⁻¹)		LOQ (ng L ⁻¹)	
	Raw	Treated	Raw	Treated
Amphetamine	18	11	59	38
Aripiprazole	9	4	30	12
Citalopram	5	1	15	3
Cocaine	5	1	18	5
Codeine	17	4	55	12
Diazepam	2	1	8	5
Fentanyl	2	1	6	2
Flunitrazepam	2	2	7	7
Fluoxetine	10	4	33	14
Ketamine	2	1	8	3
Methadone	4	3	14	9
Methamphetamine	3	10	11	3
Morphine	5	7	16	23
Oxycodone	14	8	47	25
Phentermine	4	3	14	11
Risperidone	6	1	20	2
Sertraline	7	4	25	13
Tramadol	3	2	9	8
Venlafaxine	2	1	5	5

Table S6 Frequency of detection of each drug above its LOD and LOQ in raw wastewater from each WWTP. n = number of raw samples analysed in each WWTP.

		OD WWTP (n = 22)	CAS WWTP (n = 14)	MBR WWTP (n = 3)
Class	Drug	(>LOD/>LOQ)%	(>LOD/>LOQ)%	(>LOD/>LOQ)%
Stimulants	Amphetamine	(100/9)	(100/100)	(100/100)
	Methamphetamine	(100/100)	(100/100)	(100/100)
	Cocaine	(0/0)	(71/71)	(33/0)
Opioids	Morphine	(100/100)	(100/100)	(100/100)
	Tramadol	(100/100)	(100/100)	(100/100)
	Oxycodone	(41/0)	(100/0)	(100/100)
	Codeine	(100/100)	(100/100)	(100/100)
	Methadone	(100/100)	(93/57)	(0/0)
	Fentanyl	(0/0)	(0/0)	(0/0)
Psychopharmaceuticals	Diazepam	(41/0)	(86/0)	(0/0)
	Flunitrazepam	(0/0)	(0/0)	(0/0)
	Citalopram	(100/100)	(100/100)	(100/100)
	Fluoxetine	(0/0)	(0/0)	(0/0)
	Venlafaxine	(100/100)	(100/100)	(100/100)

	Sertraline	(0/0)	(7/0)	(0/0)
	Aripiprazole	(0/0)	(0/0)	(0/0)
	Risperidone	(0/0)	(0/0)	(0/0)
Appetite suppressant	Phentermine	(100/100)	(100/100)	(100/100)
Hallucinogen	Ketamine	(14/0)	(86/86)	(100/100)

Table S7 Frequency of detection and quantification (>LOD/>LOQ)%, and median concentration (ng L⁻¹), of each analyte in treated wastewater in the three WWTPs, along with its relative standard deviation (%RSD). When the median concentration was between the LOD and LOQ, half the LOQ is reported, and no %RSD is reported. When the median concentration was lower than the LOD, then a concentration of 0 without %RSD is reported.

	OD WWTP (n=21)		CAS WWTP (n=14)		MBR WWTP (n=3)	
Drug	Concentration (%RSD)	Frequency (>LOD/>LOQ)	Concentration (%RSD)	Frequency (>LOD/>LOQ)	Concentration (%RSD)	Frequency (>LOD/>LOQ)
Amphetamine	0	(0/0)%	0	(0/0)%	0	(0/0)%
Methamphetamine	40 (97)	(100/100)%	74 (78)	(100/100)%	43 (44)	(100/100)%
Cocaine	0	(0/0)%	24 (42)	(93/93)%	0	(0/0)
Morphine	11	(81/0)%	11	(71/0)%	0	(0/0)%
Tramadol	776 (31)	(100/100)%	673 (39)	(100/100)%	46 (134)	(100/100)%
Oxycodone	13	(95/0)%	13	(100/21)%	0	(33/33)%
Codeine	114 (39)	(100/100)%	465 (45)	(100/100)%	0	(33/33)%
Methadone	19 (28)	(100/100)%	18 (24)	(100/100)%	0	(0/0)%
Diazepam	2	(100/0)%	2	(100/21)%	0	(0/0)%
Citalopram	99 (37)	(100/100)%	156 (40)	(100/100)%	0	(33/33)%
Venlafaxine	500 (30)	(100/100)%	577 (41)	(100/100)%	13 (158)	(100/100)%
Phentermine	61 (33)	(100/100)%	143 (48)	(100/100)%	51 (6)	(100/100)%
Ketamine	2	(71/10)%	41 (49)	(100/100)%	212 (72)	(100/100)%
Sertraline	0	(5/0)%	0	(21/0)%	0	(0/0)%
Risperidone	0	(0/0)%	0	(0/0)%	0	(0/0)%

Aripiprazole	0	(0/0)%	0	(0/0)%	0	(0/0)%
Fentanyl	0	(0/0)%	0	(21/0)%	0	(0/0)%
Flunitrazepam	0	(0/0)%	0	(0/0)%	0	(33/0)%
Fluoxetine	0	(0/0)%	7	(71/0)%	0	(0/0)%

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