Supplementary information

Determination of pharmaceuticals in freshwater sediments using ultrasonic-assisted extraction with SPE clean-up and HPLC–DAD or LC-ESI-MS/MS detection

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Table S1 Physico-chemical properties of the study sediments.

Sediment	Coordinates	Texture	Silt %	Clay %	Sand%	OC %	pH $CaCl_2$	CEC
Buttercrambe (BTC), YO, UK	54.017012, -0.881074	Sandy loam	35.48	34.25	62.92	2.83	6.88	13.45
Bishop Wilton (BW), YO, UK	53.982712, -0.790092	Loam	45.92	4.73	49.35	9.9	8.1	35.58
Millington (MIL), YO, UK	53.964920, -0.719305	Sandy clay	0.88	37.25	61.87	8.02	7.15	37.08
German beck (GER),YO, UK	53.935850, -1.054470	Sandy clay loam	1.22	30.97	67.81	5.69	7.1	24.26
Helmsley (HLM), YO, UK	54.242978, -1.055166	Sandy	10.08	0.12	89.8	0.98	6.65	5.85
Moors (MOR), YO, UK	54.371324, -0.965524	Loamy sand	21.05	0.35	78.6	3.52	6.35	11.26
Harborough (HAB), LT, UK	52.626226, -0.890155	Loamy sand	26.7	1.12	72.18	1.12	7.45	11.34
Skeffington (SKF), LT, UK	52.620847, -0.905779	Sandy clay loam	0.38	36.52	63.1	7.92	7.02	28.39
Tigris River (BGD), Baghdad, Iraq	33.361904, 44.370943	Silt loam	58.15	2.04	39.81	3.42	7.1	12.99
Alhussainya River (HUS), Karbala, Iraq	32.623024, 44.027632	Silt loam	71.15	2.91	25.94	3.51	7.3	19.07

Table S2 Optimized HPLC-DAD and LC-MS/MS conditions: recovery %, instrumental limit of detection (quantification) IDL (IQL) and for the analysis of the target pharmaceuticals

Compound	Retention time (min)	LC-MS/MS						HPLC-DAD	
		Ionization Mode and acquisition	Precursor ion *(m/z)*	Product ion *(m/z)*	Collision energy (eV)	Recovery% (RSD%) at 20 ng mL^{-1} LC-MS/MS	IDL (IQL) ng L^{-1} LC-MS/MS	Recovery% (RSD%) at 2.0 µg mL^{-1} HPLC-DAD	IDL (IQL) ng mL^{-1} HPLC-DAD
Amitriptyline	14.35	+ve MRM	278.2	105.1	35	112.2 (2.7)	10 (30.0)	104.1 (1.1)	9 (30.0)
Atenolol	2.55	+ve MRM	267	145	40	98.5 (3.1)	22 (70.0)	99.2 (3.4)	14 (46.5)
Cimetidine	2.77	+ve MRM	253	95	35	100.1 (2.2)	15 (47.5)	101.5 (2.6)	10 (33.3)
Diltiazem	14.02	+ve MRM	415	178	35	102.2 (1.3)	6 (20.3)	100.1 (1.2)	7 (23.3)
Mefenamic acid	14.64	+ve MRM	242.1	224.1	20	106.3 (1.0)	6 (19.0)	99.9 (0.3)	6 (20.0)
Ranitidine	3.01	+ve MRM	315	176	30	99.6 (3.1)	9 (30.3)	101.5 (3.0)	12 (40)

Section S1: Recoveries calculation

Recoveries were calculated based on the obtained response (concentration) of the analytes in the spiked sediment at different steps of the extraction process. For calculation of the total recovery (RECtotal) we calculated the ratio of the response of the analyte from the sediment sample spiked with the target compounds at 1µg g^{-1} (X) to the response obtained after spiking the reconstituted extract (Z) as shown in Equation (S1).

$$RECtotal = \left(\frac{X}{Z}\right) \times 100 \quad \text{(S1)}$$

Recoveries of the SPE step (REC_{SPE}) were calculated as the ratio of the response of the analyte obtained from the extracted sample spiked with analyte (Y) to the post-extracted spiked sample (obtained from spiked reconstituted extract (Z) (Equation S2).

$$REC_{SPE} = \left(\frac{Y}{Z}\right) \times 100 \quad \text{(S2)}$$

Table S3. Pharmaceuticals recoveries, limits of detection (LODs) and limits of quantitation (LOQs) in sediments. Standard deviation (±RSD %) is presented in parentheses for three replicates using HPLC-DAD

Sediment	Spiking level µg g^{-1}	Atenolol		Cimetidine		Amitriptyline		Diltiazem		Mefenamic acid		Ranitidine	
		Recovery% (±RSD%)	LOD (LOQ) (ng g^{-1})	Recovery% (±RSD%)	LOD (LOQ) (ng g^{-1})	Recovery% (±RSD%)	LOD (LOQ) (ng g^{-1})	Recovery% (±RSD%)	LOD (LOQ) (ng g^{-1})	Recovery% (±RSD%)	LOD (LOQ) (ng g^{-1})	Recovery% (±RSD%)	LOD (LOQ) (ng g^{-1})
BTC	1.0	100.24 (3.2)	37.1 (122.5)	50.18 (3.6)	31.3 (103.3)	93.97 (2.5)	34.0 (112.2)	75.81 (7.3)	20.4 (67.3)	96.94 (3.6)	23.3 (76.9)	51.72 (3.2)	20.2 (66.7)
	0.5	99.3 (9.9)		50.05 (7.8)		95.50 (5.4)		70.25 (5.6)		93.25 (2.8)		45.2 (4.5)	
	0.2	95.21 6.2)		48.32 (10.4)		85.52 (6.3)		75.88 (4.3)		90.41 (4.1)		50.3 (6.4)	
BGD	1.0	84.68 (5.1)	55.6 (183.5)	46.31 (4.2)	15.4 (50.8)	78.29 (5.8)	20.5 (67.7)	79.43 (5.9)	36.8 (121.5)	100.1 (2.8)	15.0 (49.5)	34.63 (5.6)	13.2 (43.6)
	0.5	86.32 (5.5)		42.65 (4.1)		72.36 (4.3)		80.52 (9.8)		92.35 (3.1)		38.25 (6.9)	
	0.2	79.22 (11.0)		45.22 (5.4)		72.52 (4.5)		77.30 (7.5)		99.25 (2.4)		33.4 (4.2)	
HUS	1.0	74.55 (2.6)	32.2 (106.0)	42.34 (4.7)	26.7 (88.1)	80.96 (6.6)	28.2 (93.1)	70.09 (3.7)	34.1 (112.5)	105.04 (6.1)	21.3 (70.3)	31.28 (1.6)	31.4 (103.6)
	0.5	75.22 (3.5)		42.38 (7.7)		81.65 (3.8)		69.47 (9.5)		105.8 (7.2)		31.14 (2.8)	
	0.2	79.64 (6.2)		40.52 (9.9)		80.02 (5.5)		63.21 (8.3)		102.52 (6.8)		30.4 (4.3)	
SKF	1.0	87.99 (5.3)	23.0 (76.0)	43.47 (3.2)	15.4 (50.8)	124.94 (9.2)	25.5 (84.2)	62.40 (4.3)	35.2 (116.2)	84.02 (1.3)	14.0 (46.2)	30.24 (4.4)	12.4 (40.9)
	0.5	80.35 (4.8)		40.62 (1.6)		111.20 (7.5)		62.58 (8.7)		79.32 (2.4)		33.25 (1.7)	
	0.2	81.46 (4.5)		42.56 (5.7)		111.85 (3.7)		60.21 (9.3)		85.28 (2.5)		32.1 (3.8)	
HAB	1.0	93.58 (2.5)	28.0 (92.5)	68.45 (4.9)	19.0 (62.7)	82.30 (1.8)	20.5 (67.7)	102.05 (4.1)	30.8 (101.6)	103.21 (4.1)	19.5 (64.4)	50.98 (7.5)	30.2 (99.7)
	0.5	99.25 (2.9)		67.50 (2.5)		80.52 (2.7)		101.10 (5.2)		101.20 (3.1)		44.6 (6.8)	
	0.2	94.4 (4.7)		67.33 (4.5)		80.46 (6.5)		99.35 (4.9)		101.32 (3.2)		48.5 (9.6)	
MIL	1.0	108.86 (6.1)	34.6 (114.2)	55.09 (2.9)	17.8 (58.7)	75.77 (8.8)	25.3 (84.0)	68.16 (5.3)	39.1 (129.0)	88.78 (3.5)	20.8 (68.6)	32.78 (4.1)	21.3 (70.3)
	0.5	107.62 (8.8)		50.42 (4.8)		76.65 (6.2)		66.52 (6.5)		85.25 (4.1)		30.2 (4.8)	

	0.2	97.7 (5.6)		52.38 (5.7)		70.45 (8.0)		69.25 (12.5)		87.46 (6.6)		29.5 (6.8)	
HLM	1.0	114.64 (6.7)	25.5 (84.2)	57.45 (8.2)	30.2 (100.0)	104.42 (5.1)	38.0 (125.5)	74.98 (2.9)	12.6 (42.0)	97.21 (3.2)	24.0 (79.2)	45.20 (3.2)	27.8 (91.7)
	0.5	107.32 (8.6)		56.55 (8.1)		103.50 (5.8)		70.52 (2.8)		92.35 (3.9)		41.2 (5.6)	
	0.2	113.2 (8.1)		49.32 (9.6)		101.65 (12.1)		69.54 (2.5)		96.52 (7.6)		45.5 (8.8)	
MOOR	1.0	94.17 (2.5)	25.8 (85.1)	56.81 (4.5)	28.7 (94.7)	123.79 (11.8)	25.3 (83.5)	80.14 (3.3)	20.5 (67.7)	90.65 (5.6)	23.2 (106.3)	54.87 (8.2)	32.1 (105.9)
	0.5	88.66 (5.1)		50.87 (3.8)		107.54 (9.8)		82.36 (2.8)		90.50 (4.5)		55.2 (9.5)	
	0.2	85.35 (8.1)		55.54 (8.2)		99.28 (7.9)		77.25 (4.1)		89.52 (4.1)		52.1 (6.3)	
GER	1.0	88.17 (5.8)	28.3 (93.3)	50.18 (2.5)	19.7 (56.0)	97.84 (3.7)	17.3 (57.1)	70.67 (6.6)	45.2 (149.1)	82.19 (3.1)	18.2 (60.1)	53.86 (7.5)	24.8 (81.1)
	0.5	82.12 (9.1)		54.50 (4.7)		95.85 (6.6)		65.35 (5.1)		75.32 (4.4)		50.1 (4.2)	
	0.2	88.62 (5.1)		50.88 (6.2)		88.74 (5.5)		71.85 (11.6)		80.84 (5.8)		52.3 (7.8)	
BW	1.0	77.85 (11.5)	58.5 (193.1)	42.61 (9.9)	31.2 (103.0)	100.82 (2.9)	56.9 (187.8)	60.32 (4.8)	29.6 (97.7)	76.51 (4.4)	16.2 (53.5)	37.49 (8.2)	28.5 (94.1)
	0.5	77.65 (9.8)		40.20 (7.2)		96.52 (2.8)		60.88 (6.2)		77.2 (2.3)		33.5 (5.3)	
	0.2	74.98 (12.4)		40.52 (15.8)		90.31 (10.1)		58.22 (5.7)		76.4 (5.1)		36.4 (9.1)	

Table S4. Recoveries and limits of detection (LODs) and limits of quantitation (LOQs) for each pharmaceuticals in sediment. Relative standard deviation (RSD %) is presented in parentheses for three replicates using LC-MS/MS

		Amitriptyline		**Atenolol**		**Cimetidine**		**Diltiazem**		**Mefenamic acid**		**Ranitidine**	
Sediment	Conc. ngg^{-1} (d.w)	Recover% (± RSD%)	LOD (LOQ) (ng g^{-1})	Recover% (± RSD%)	LOD (LOQ) (ng g^{-1})	Recover% (± RSD%)	LOD (LOQ) (ng g^{-1})	Recover% (± RSD%)	LOD (LOQ) (ng g^{-1})	Recover% (± RSD%)	LOD (LOQ) (ng g^{-1})	Recover% (± RSD%)	LOD (LOQ) (n gg^{-1})
BTC	100	99.6 (10.3)	0.3 (1.0)	93.1 (5.9)	1.9 (6.0)	50.3 (2.1)	0.7 (2.3)	80.2 (10.2)	0.1 (0.4)	82.5 (2.3)	2.3 (8.0)	45.2 (7.8)	0.3 (1.0)
	200	110.5 (15.8)		94.7 (6.4)		57.6 (4.3)		75.8 (16.5)		83.8 (5.0)		43.9 (5.8)	
	500	105.4 (10.5)		98.2 (8.1)		58.5 (1.9)		80.1 (3.8)		90.5 (6.7)		51.0 (2.5)	
BW	100	70.3 (8.1)	0.14 (0.5)	77.1 (6.1)	1.9 (6.0)	48.0 (4.6)	0.6 (1.8)	60.2 (6.3)	0.05 (0.17)	70.0 (4.4)	0.1 (0.4)	40.1 (9.1)	0.7 (2.2)
	200	74.2 (7.3)		80.9 (7.4)		48.4 (3.1)		63.4 (8.5)		72.3 (5.7)		43.0 (14.4)	
	500	76.8 (5.2)		83.3(2.3)		51.4 (3.4)		70.8 (4.9)		72.8 (3.4)		49.3 (4.8)	
MIL	100	81.8 (5.6)	0.07 (0.25)	70.5 (6.4)	1.3 (5.0)	44.2 (8.2)	0.6 (1.9)	65.1 (5.2)	0.03 (0.1)	70.2 (5.1)	0.3 (1.0)	45.2 (8.1)	0.6 (1.9)
	200	84.8 (3.4)		76.4 (5.4)		48.7 (11.5)		69.0 (4.6)		77.1 (13.0)		47.0 (11.5)	
	500	87.3 (5.1)		77.1 (3.8)		49.0 (5.8)		72.0 (10.2)		79.5 (9.4)		47.2 (5.1)	
GER	100	69.1 (10.4)	0.2 (0.7)	80.2 (6.8)	2.5 (8.0)	50.4 (15.5)	1.2 (4.0)	72.1 (3.4)	0.02 (0.07)	83.1 (5.8)	2.0 (6.0)	52.5 (6.9)	0.5 (1.6)
	200	73.2 (11.3)		88.1 (9.2)		55.1 (15.8)		76.0 (3.5)		84.7 (5.4)		53.0 (8.4)	
	500	79.2 (3.8)		87.6 (4.2)		56.8 (9.2)		76.3 (4.8)		90.3 (11.2)		58.2 (4.3)	
HLM	100	80.0 (5.9)	0.09 (0.3)	73.2 (6.2)	1.8 (6.0)	48 .1 (14.3)	0.6 (1.9)	91.5 (8.6)	0.04 (0.16)	87.8 (5.9)	0.1 (0.3)	44.3 (5.1)	0.2 (0.6)
	200	80.7 (5.8)		73.0 (7.9)		48.8 (8.9)		96.2 (5.3)		89.0 (3.5)		46.7 (3.6)	
	500	85.3 (7.2)		77.8 (5.4)		45.3 (5.2)		99.0 (4.3)		92.3 (6.1)		50.2 (3.6)	
MOR	100	83.6 (9.1)	0.13 (0.5)	75.6 (12.3)	2.5 (8.0)	41.2 (6.5)	0.8 (2.5)	65.3 (7.7)	0.05 (0.17)	70.1 (5.1)	0.2 (0.6)	48.0 (5.8)	0.3 (0.9)
	200	90.0 (7.0)		76.4 (10.4)		46.1 (10.3)		76.2 (8.2)		73.6 (8.4)		48.2 (4.9)	

	500	92.4 (11.5)		80.2 (6.8)		55.1 (7.8)		77.0 (4.1)		77.0 (2.0)		55.6 (7.0)	
HAB	100	102 (7.3)	0.14 (0.5)	73.9 (5.2)	2.2 (7.0)	51.9 (5.5)	0.7 (2.3)	92.2 (9.1)	0.06 (0.2)	74.1 (8.0)	2.0 (6.0)	42.3 (6.6)	0.6 (1.9)
	200	120.6 (5.9)		78.7 (8.7)		55.4 (12.4)		97.3 (7.4)		76.5 (6.7)		45.0 (11.8)	
	500	115.7 (7.6)		78.2 (6.9)		57.8 (7.1)		96.3 (5.6)		76.8 (10.1)		49.6 (4.3)	
SKF	100	95.5 (5.7)	0.18 (0.6)	83.0 (9.8)	3.5 (12.0)	49.6 (2.8)	0.5 (1.7)	60.3 (5.3)	0.04 (0.13)	90.1 (5.3)	2.5 (8.0)	45.5 (4.5)	0.6 (1.8)
	200	100.7 (9.5)		82.5 (13.2)		53.0 (7.5)		67.4 (7.1)		92.5 (8.4)		48.2 (6.3)	
	500	102.2 (3.8)		88.5 (7.2)		60.2 (5.7)		70.1 (4.9)		100.1 (8.2)		49.3 (5.1)	
BGD	100	106.6 (9.8)	0.2 (0.7)	75.3 (6.2)	1.5 (5.0)	42.5 (8.1)	0.5 (1.7)	75.6 (2.8)	0.07 (0.2)	75.8 (4.1)	0.1 (0.4)	50.3 (2.3)	0.8 (2.4)
	200	120.0 (11.2)		80.1 (5.7)		42.7 (5.9)		78.3 (10.3)		76.8 (5.8)		43.6 (15.2)	
	500	109.8 (6.2)		82.2 (7.1)		45.3 (2.9)		77.8 (6.6)		82.3 (6.9)		46.2 (4.1)	
HUS	100	70.0 (9.3)	0.13 (0.5)	72.2 (10.5)	2.7 (9.0)	52.1 (3.6)	0.7 (2.4)	77.8 (7.2)	0.04 (0.13)	80.0 (3.6)	0.15 (0.5)	40.3 (7.2)	0.4 (1.3)
	200	68.8 (6.9)		73.3 (11.5)		50.0 (8.9)		80.1 (6.0)		82.5 (5.9)		45.3 (8.1)	
	500	74.5 (3.6)		73.5 (5.6)		50.3 (5.2)		88.1 (5.6)		85.5 (3.1)		46.3 (1.1)	

Section S2: Matrix effects

The matrix effects were studied by evaluating the signal suppression or enhancement for each pharmaceutical. To assess the influence of matrix components, signals of final sediment extracts spiked with analytes were compared with signals observed from solvent dissolved pharmaceuticals. A value of greater or less than zero indicates signal enhancement or suppression; respectively. The equation used for the signal suppression calculation was (Eq. S3):

$$Matrix\ effect\ \% = \left(\frac{(Area\ sediment - Area\ blank)}{Area\ standard} - 1\right) \times 100 \quad \text{(S3)}$$

Where: Area $_{sediment}$ is the peak area of the analyte(s) recorded for the sediment spiked with the target compound(s) after extraction, Area $_{blank}$ is the peak area of analytes recorded for blank samples and Area $_{standard}$ is peak area of the analyte(s) recorded for the standard solution.

Table S5 Matrix effect of pharmaceuticals in LC-ESI-MS/MS analysis at concentration of 100 ng g^{-1} in all sediment samples

Compound	Matrix effect %									
	BTC	**BW**	**MIL**	**GER**	**HLM**	**MOR**	**HAB**	**SKF**	**BGD**	**HUS**
Amitriptyline	3.1	-22.5	-18.8	-20.3	12.4	-12.5	8.1	-20.1	-7.0	5.1
Atenolol	-10.5	-42.5	-22.5	-20.5	-18.5	-20.5	-15.3	-22.0	-13.6	-6.1
Cimetidine	-12.1	-38.0	-25.1	-18.8	-20.5	-13.8	-15.4	-12.5	-18	-10.0
Diltiazem	-12.8	-20.3	-18.5	-15.2	8.6	-6.0	-22.3	-12.8	12.4	-6.0
Mefenamic acid	-12.5	-20.1	-12.0	-19.3	5.0	-4.1	8.2	-16.8	-13.2	-10.7
Ranitidine	-16.2	-20.1	-18.5	-8.5	-20	-16.4	-5.0	-13.1	-18.3	-10.0

(-) for signal suppression, (+) for signal enhancement