

1 **Occurrence of pharmaceutical active compounds in sewage sludge**
2 **from two urban wastewater treatment plants and their potential**
3 **behaviour in agricultural soils**

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7 **Supplementary Material**

8 **Fig. S1.** Effect of dispersant agent in the extraction efficiency of PhACs by ASE-SPE-LC-
9 MS/MS. Chromatographic area of PhACs in two scales: (A) high scale and (B) short scale
10 (n = 3).

11 **Fig. S2** Recovery of PhACs exposed at different temperatures and analyzed by LC-MS/MS
12 (n = 3).

13 **Fig. S3.** Areas of PhACs extracted from sludges at different temperatures by ASE and
14 analyzed by SPE-LC-MS/MS. Chromatographic area of PhACs in two scales: (A) high scale
15 and (B) short scale (n = 3).

16 **Fig. S4.** Areas of PhACs extracted from sludges with two ratios ACN/H₂O (70/30 and
17 90/10, v/v) by ASE- SPE-LC-MS/MS. Chromatographic area of PhACs in two scales: (A)
18 high scale and (B) short scale (n = 3).

19 **Table S1.** Comparison of validated ASE-SPE-UPLC-MS/MS method with other methods
20 to quantify PhACs in sludge samples.

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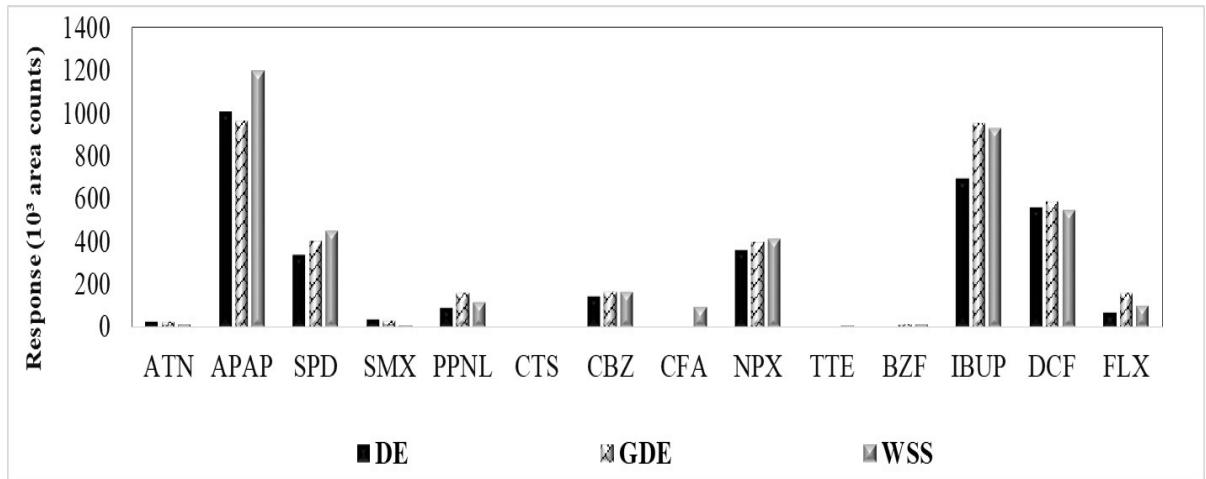
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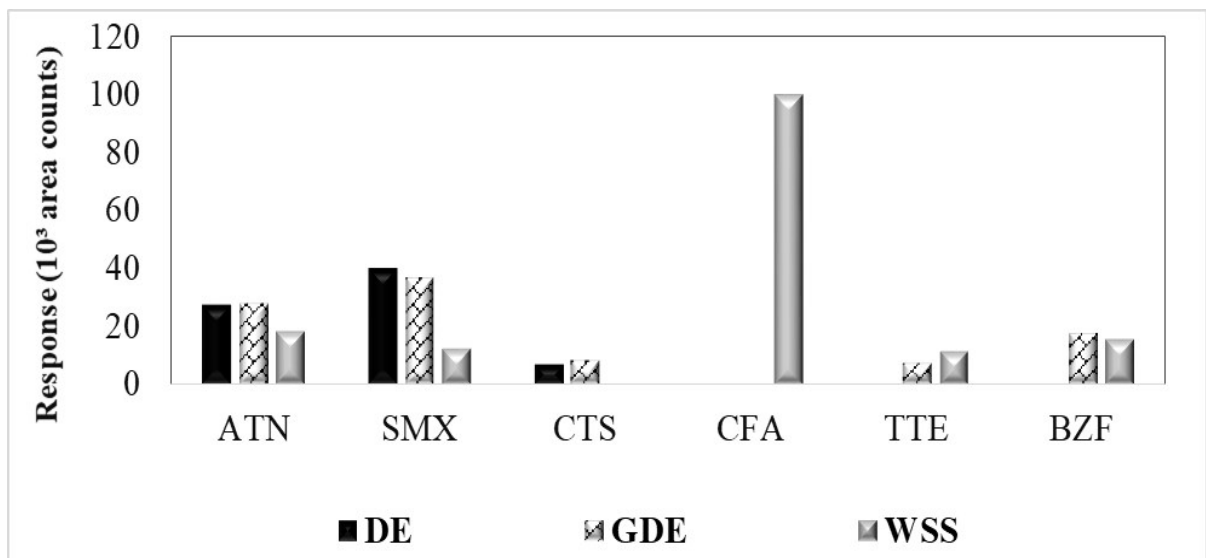
26 **Fig .S1.**

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(A)



(B)

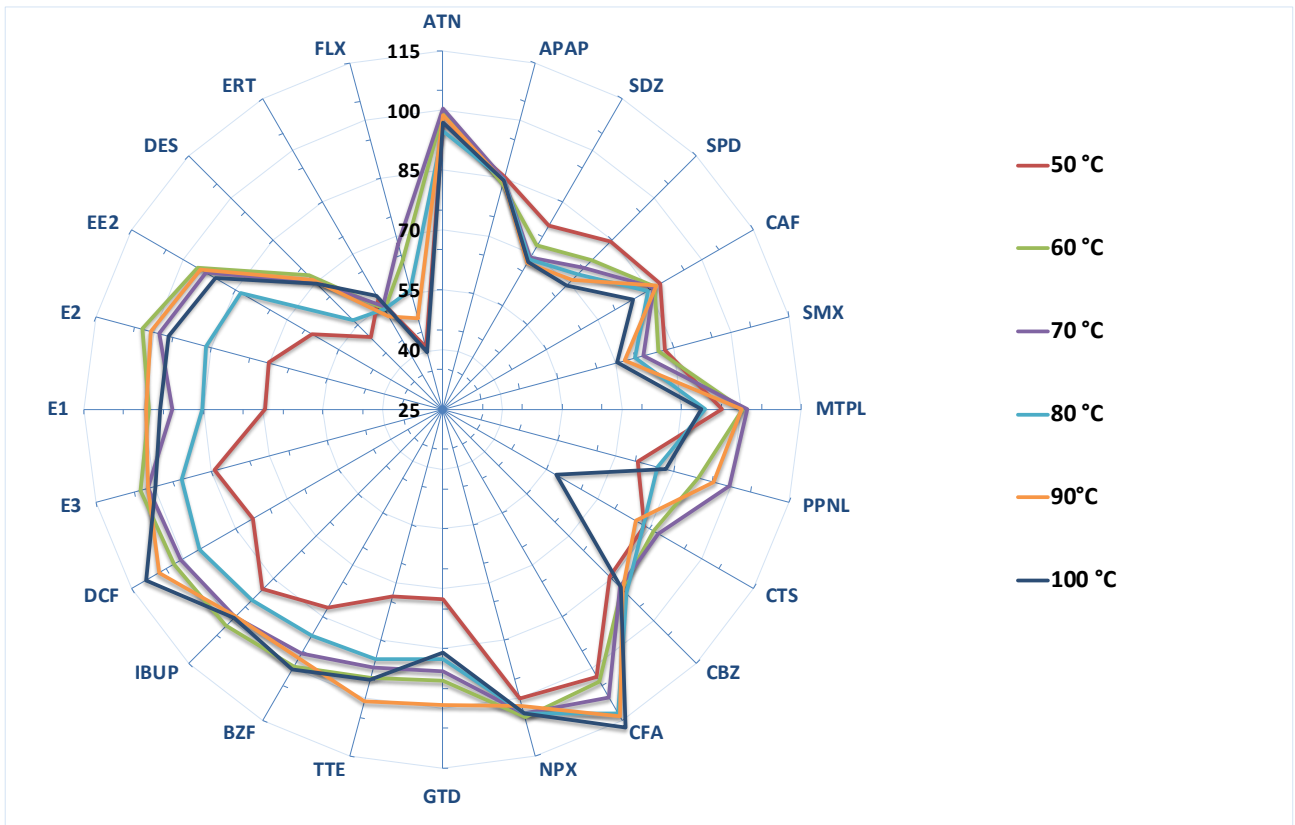


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30 Fig .S2.

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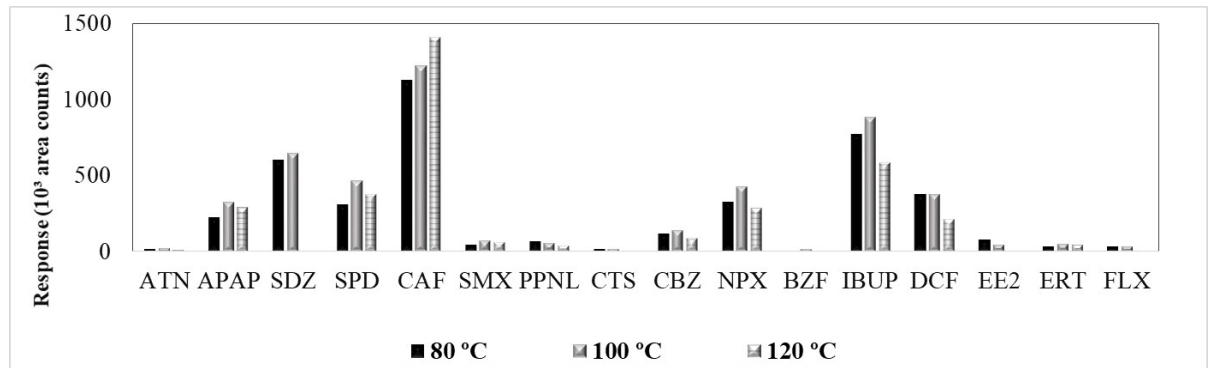
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34 **Fig. S3.**

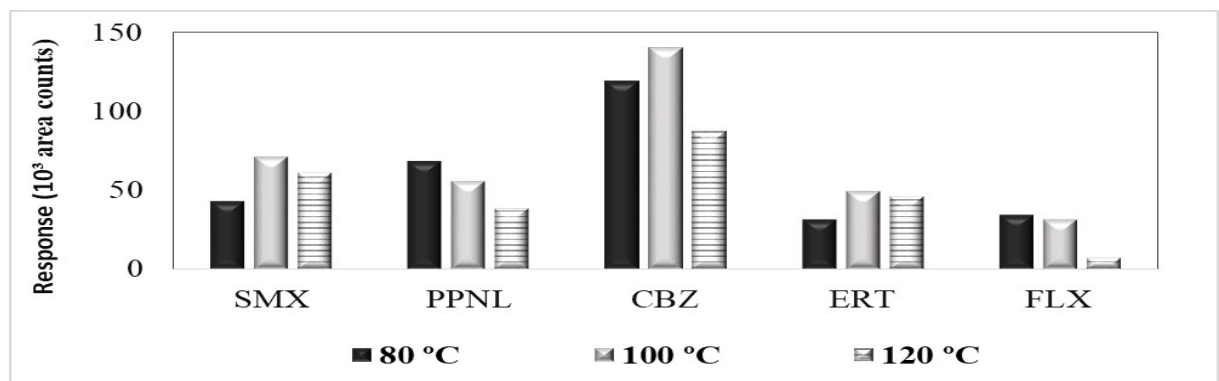
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(A)



(B)



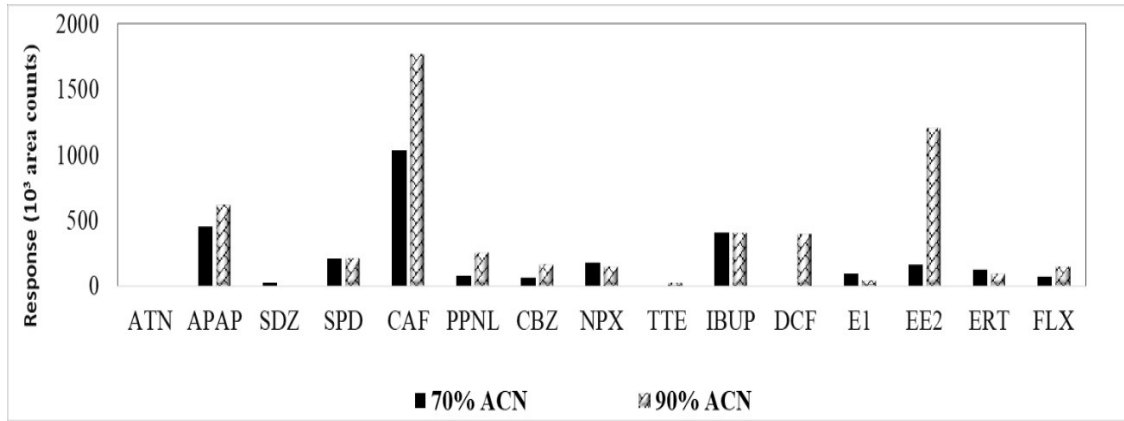
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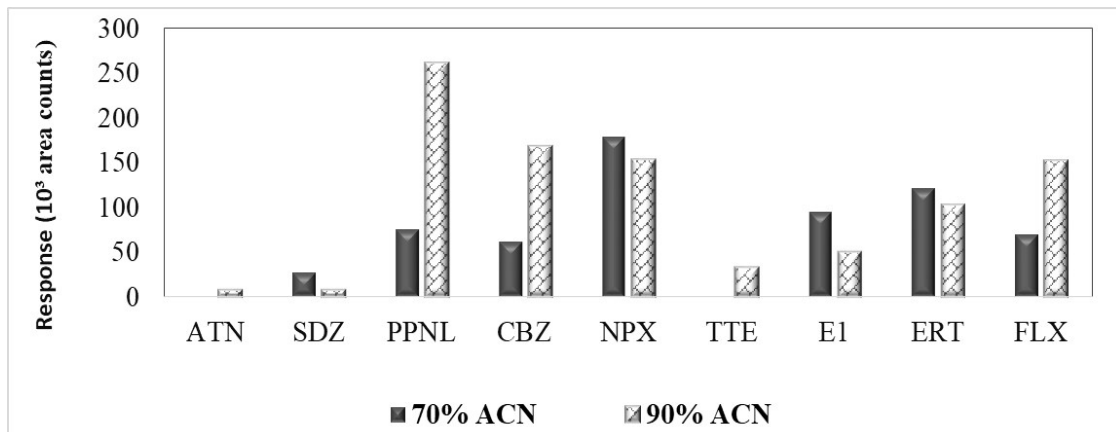
39 **Fig .S4.**

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(A)



(B)



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Matrix	Nº PhACs (Therapeutic classes)	Extraction configuration: Phase system	Instrumental configuration	Nº samples/ Nº WWTP	Cal., R ²	Precision	Recovery	LOQ (ng/g)	Ref.
Primary, mixed liquor (solid phase), and recirculated sludge	24 PhACs (analgesic, anti-dyslipidemics anticonvulsants, β-blockers psychostimulants, NSAIDs, corticosteroids, hormones antidepressants, antibiotics)	ASE/SPE Oasis HLB (200 mg)	UPLC-MS/MS	67/2	SAC, 0.9980- 0.9999	RSD _r 2.9-29%	30-110%	1.66-34.5	(A)
Treated sludge	129 PhACs (analgesics, antidepressants, antibiotics, benzodiazepines, antipsychotics, antiepileptics NSAIDs)	USE ---	UPLC-MS/MS	---/5	ISC, 0.99	RSD _r 3-20%	16-107%	2.7-66.3	27
Treated sludge	25 PhACs (fluoroquinolones, β-blockers, tetracyclines, macrolides, antiepileptics, lipid regulators, stimulants, sulfonamides,)	USE/SPE Oasis HLB (500 mg)	UPLC-MS/MS	---/1	ISC, > 0.995	RSD _r 3-13%	54-120%	0.02-1	22
Primary, secondary, anaerobically- digested, mixed, aerobically- digested, lagoon, and composted sludge	22 PhACs (NSAIDs, lipid regulators antibiotics, antiepileptics, stimulants, estrogens)	USE/SPE Oasis HLB (60 mg)	HPLC-MS/MS	28/10	ISC, >0.992	---	15-115%	0.05-26.4	24, 42
Primary, secondary, and digested sludge	16 PhACs (NSAIDs, antibiotics, beta- blockers, lipid regulators, antiepileptics, estrogens, stimulants)	USE/SPE Oasis HLB (60 mg)	HPLC-DAD	16/4	ISC, >0.992	---	<15- 115%	0.4-360	23, 42
Sewage and livestock sludge	24 PhACs (analgesics, stimulants, anti- seizures, NSAIDs, antibiotics)	USE/SPE Oasis HLB (200 mg)	UPLC-MS/MS	40/16	ISC >0.999	RSD _r 2-19%	---	0.001- 0.122	21
Mixed liquor (liquid phase)	5 PhACs (antidepressants, antibiotics)	---	HPLC-HRMS	---/1	ISC ---	RSD _r <20%	---	---	25
Digested sludge	42 PhACs (antidepressants, antibiotics, anticancer, antihypertensives, NSAIDs, analgesics, anti- asthmatics, cardiovasculars, diuretics, estrogens, antireflux agents, lipid regulators, erectile dysfunction agents)	ASE/SPE Oasis MCX (150 mg) Oasis HLB (60 mg)	HPLC-MS/MS	---/12	ISC, >0.998	RSD 0.8- 46.4%	53.4- 146%	0.12- 47.92	28
Untreated, and digested sludge, and compost	5 PhACs (NSAIDs, antidepressants)	USE/SPE SDB-RPS disk ---	HPLC-MS/MS	---/1	---	---	---	---	29

46 **Table S1**

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Legend:

(A) Obtained data; (---) data not available; (DAD) diode array detector; (HLB) hydrophilic-lipophilic-balance; (HPLC) high performance liquid chromatography; (HRMS) high resolution mass spectrometry; (ISC) internal standard calibration (isotopically labelled internal standard as surrogates); (LOQ) limit of quantification; (MS) mass spectrometry; (MCX) mixed-mode/cationic-exchange; (MS/MS) triple quadrupole; (NSAIDs) nonsteroidal anti-

51 inflammatory drugs; **(RSD_r)** relative standard deviation under repeatability conditions (intra-day precision); **(RSD_R)** relative standard deviation under
52 intermediation precision (inter-day precision); **(SAC)** standard addition calibration; **(SDB-RPS)** styrene divinylbenzene reversed phase sulfonate; **(SPE)**
53 solid phase extraction; **(UHPLC)** ultra-high-performance liquid chromatography; **(USE)** ultrasonic extraction.
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