SUPPLEMENTARY MATERIAL

FLEXIBLE HIGH RESOLUTION-MASS SPECTROMETRY APPROACH FOR SCREENING NEW PSYCHOACTIVE SUBSTANCES IN URBAN WASTEWATER

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MATERIALS AND METHODS

Chemicals and materials

The deuterated analogues of illicit drugs including in the mixed solution were: cocaine-d3, benzoylecgonine-d3, cocaethylene-d8, amphetamine-d6, methamphetamine-d9, 3,4 methylenedioxyamphetamine-d5, 3,4-methylenedioxymethamphetamine-d5, 3,4 methylenedioxyethamphetamine-d5, 1,3-benzodioxolyl-N-methylbutanamine-d5, ketamine-d4, norketamine-d4, codeine-d6, oxycodone-d6, hydrocodone-d6, morphine-d3, morphine-3β-Dglucuronide-d3, 6-acetylmorphine-d6, methadone-d3, 2-ethylidene-1,5dimethyl-3,3diphenylpyrrolidine-d3. All analytical standards were purchased from Cerilliant Corporation (Round Rock, Texas, USA). Individual standards were available as solutions in methanol (MeOH) or acetonitrile (ACN) at concentrations of 1, 0.4 or 0.1 mg mL⁻¹. Each stock solution of 10 µg mL⁻¹ and the working solutions (1 and 0.1 μ g mL⁻¹) were prepared with MeOH and stored at -20° C in the dark. Mixed solutions were prepared daily at a concentration of 2 ng (reconstitution in 200 µL, 10% methanol (aq)) and injected before and after each sequence for assessing the sensitivity and selectivity of the instrument. The MS^2 spectra database is available on request.

Instrumental analysis

The specific parameters of each experiment are reported in the following lines:

1) Full MS experiment:

MS parameter	Value
Microscan	3
Resolution	70 000
AGC Target	1 e ⁶
Maximum IT	200 ms
Scan spectra	100 to 600 m/z

2) Data-dependant analysis (Full MS-ddMS² TOP5)

Full MS parameter	Value
Microscan	1
Resolution	70 000
AGC Target	1 e ⁶
Maximum IT	100 ms
Scan spectra	100 to 500 m/z

ddMS ² parameter	Value
Microscan	1
Resolution	17 500
AGC Target	1 e ⁵
Maximum IT	50 ms
Loop count	5
MSX count	1
Top N	5
Isolation window	3.0 m/z
Normalized Collision Energy	35, 50 eV

As we selected the "preliminary suspects" with the first Full MS experiment, an inclusion list was created for the data dependant analysis, in which a retention time (RT) window of ± 1 min was established for each suspect compound.

3) Data-independent analysis (DIA)

When no MS^2 scans were collected with Full MS-dd MS^2 , the Full MS scan was followed by a DIA experiment. Again, an inclusion list was created establishing a retention time (RT) window of ± 1 min for each suspect compound.

Full MS parameter	Value	
Microscan	1	
Resolution	70 000	
AGC Target	1 e ⁶	
Maximum IT	100 ms	
Scan spectra	100 to 500 m/z	

DIA parameter	Value
Microscan	1
Resolution	35 000
AGC Target	2 e ⁵
Maximum IT	auto
Loop count	2
MSX count	1
Isolation window	3.0 m/z
Normalized Collision Energy	35, 50 eV

RESULTS AND DISCUSSION

NPS spectral database

$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$	formulae) obtained for NPS fragmentation with Q-Exactive mass analyser.						
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$			DT	Theo	Fragment 1	Fragment 2	Fragment 3
Image Image <t< td=""><td>Compound</td><td>$[M+H]^+$</td><td>RT</td><td>m/z</td><td>Theo m/z</td><td>Theo m/z</td><td>Theo m/z</td></t<>	Compound	$[M+H]^+$	RT	m/z	Theo m/z	Theo m/z	Theo m/z
Synthetic camabinoids MDMB- CHMICA CHMICA $C_{23}H_{33}N_2O_3$ 25.1 385.2486 (240.1381 (C_1H_1NO) (144.0443 (C_1H_1NO) - 5Fpentyl-3- pyridinoindole $C_{10}H_{20}FN_2O$ 16.4 311.1555 232.1133 (C_1H_1NO) 291.1493 (C_1H_1NO) 144.0443 (C_1H_2NO) Methcathinone $C_{10}H_{14}NO$ 3.6 164.1071 131.0731 (C_4H_N) 133.0650 (C_4H_O) - Standard $C_{11}H_{16}NO$ 3.9 178.1226 105.0703 (C_4H_N) 133.0650 (C_4H_O) - 3.4-DMMC $C_{11}H_{16}NO$ 8.1 192.1384 159.1042 161.0960 - 3.4-DMMC $C_{11}H_{16}NO$ 6.1 178.1226 160.1121 145.0887 119.0858 (C_1H_1N) 3.4-DMC $C_{11}H_{16}NO$ 5.2 194.1176 161.0855 138.0683 145.0887 4.4-BEC $C_{12}H_{18}NO$ 6.8 192.1384 (C_{0}H_1N) (C_{0}H_1N) (C_{0}H_1N) 4-FMC $C_{10}H_{13}NO_3$ 5.7 222.1125 (C_{10}H_1NO) (C_{1H_2NO) (C_{10}H_1ON) <t< td=""><td>-</td><td></td><td>(min)</td><td>$[M+H]^+$</td><td>(Formula)</td><td>(Formula)</td><td>(Formula)</td></t<>	-		(min)	$[M+H]^+$	(Formula)	(Formula)	(Formula)
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$				Synthetic car	nnabinoids		
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	MDMB-	СЧМО	25.1	295 2496	240.1381	144.0443	
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	CHMICA	$C_{23}\Pi_{33}\Pi_{2}O_{3}$	23.1	363.2460	$(C_{16}H_{18}NO)$	(C_9H_6NO)	-
pyridinolindole Cl ₉ H ₂₀ H ₉ CO 10.4 311.155 (C ₁₄ H ₁₅ NOF) (C ₁₉ H ₁₉ N ₂ O) (C ₃ H ₆ NO) Methcathinone C ₁₀ H ₁₄ NO 3.6 164.1071 131.0731 133.0650 - Ethcathinone C ₁₁ H ₁₆ NO 3.9 178.1226 105.0703 133.0650 - 3,4-DMMC C ₁₂ H ₁₈ NO 8.1 192.1384 159.1042 161.0960 - 3-MMC C ₁₁ H ₁₆ NO 6.1 178.1226 (C ₀ H ₁₄ N) (C ₀ H ₁₄ N) (C ₁₀ H ₁₀ N) 3-MeOMC C ₁₁ H ₁₆ NO 6.1 178.1226 (160.0112) 145.0887 (C ₁₀ H ₁₁ N) 4-MEC C ₁₂ H ₁₈ NO 6.8 192.1384 145.0887 (C ₁₀ H ₁₁ N) (C ₁₀ H ₁₁ N) 4-FMC C ₁₀ H ₁₃ FNO 4.4 182.0976 (L ₁₀ H ₁₀ NO) (C ₁₀ H ₁₁ N) (C ₁₀ H ₁₁ N) Methylone C ₁₁ H ₁₆ NO ₃ 5.7 222.1125 (C ₁₁ H ₁₀ NO) (C ₁₀ H ₁₁ N) (C ₁₀ H ₁₁ N) Methylone C ₁₃ H ₁₈ NO ₃ 7.6 236.1281 188.1071 175.0630	5Fpentyl-3-	C. H. EN.O	16.4	311 1555	232.1133	291.1493	144.0443
Synthetic cathinones Methcathinone $C_{10}H_{14}NO$ 3.6 164.1071 (131.0731) (C_8H_8) (133.0650) (C_8H_9) - Ethcathinone $C_{11}H_{16}NO$ 3.9 178.1226 105.0703 133.0650 - 3.4-DMMC $C_{12}H_{18}NO$ 8.1 192.1384 (C_9H_9) (C_1H_9O) - 3-MMC $C_{11}H_{16}NO$ 6.1 178.1226 160.1121 145.0887 119.0858 3-MeOMC $C_{11}H_{16}NO$ 5.2 194.1176 (C_{10}H_{11}N) (C_{10}H_{11}N) 4-MEC $C_{12}H_{18}NO$ 6.8 192.1384 145.0887 147.0805 146.0965 4-FMC $C_{10}H_{13}FNO$ 4.4 182.0976 160.0757 132.0809 - Methylone $C_{11}H_{16}NO_3$ 5.7 222.1125 174.0914 175.0628 146.0965 Butylone $C_{12}H_{16}NO_3$ 5.7 222.1125 174.0914 175.0628 146.0965 Butylone $C_{14}H_{20}NO_3$ 8.0 250.1438 100.1126	pyridinolindole	C19112011102O	10.4	511.1555	$(C_{14}H_{15}NOF)$	$(C_{19}H_{19}N_2O)$	(C_9H_6NO)
Methcathinone $C_{10}H_{14}NO$ 3.6 164.1071 131.0731 (C_0H_0N) 133.0650 (C_0H_0N) - Ethcathinone $C_{11}H_{16}NO$ 3.9 178.1226 105.0703 (C_0H_0) 133.0650 (C_0H_0) - 3,4-DMMC $C_{12}H_{18}NO$ 8.1 192.1384 159.1042 (C_1H_13N) 161.0960 (C_0H_1N) - 3-MMC $C_{11}H_{16}NO$ 6.1 178.1226 160.1121 (C_0H_1N) 145.0887 (C_0H_1N) 119.0858 (C_0H_1N) 3-MeOMC $C_{11}H_{16}NO_2$ 5.2 194.1176 161.0835 (C_0H_1N) 132.06603 145.0887 (C_0H_1N) 4-MEC $C_{12}H_{18}NO$ 6.8 192.1384 145.0887 (C_0H_1N) 146.0965 4-FMC $C_{10}H_{13}FNO$ 4.4 182.0976 149.0636 123.0607 - Methylone $C_{11}H_{14}NO_3$ 4.0 208.0969 160.0757 132.0809 - Butylone $C_{12}H_{16}NO_3$ 5.7 222.1125 174.0914 175.0628 146.0965 Dipentylone $C_{12}H_{16}NO_3$ 7.6 236.1281 161.108				Synthetic ca	athinones		
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Methcathinone	$C_{10}H_{14}NO$	3.6	164 1071	131.0731	133.0650	_
Ethcathinone C ₁₁ H ₁₆ NO 3.9 178.1226 105.0703 (C ₆ H ₉) 133.0650 (C ₆ H ₉ O) - 3,4-DMMC C ₁₂ H ₁₈ NO 8.1 192.1384 159.1042 (C ₁₁ H ₁₃ N) 161.0960 (C ₁₁ H ₁₃ O) - 3-MMC C ₁₁ H ₁₆ NO 6.1 178.1226 160.1121 (160.1121 145.0887 119.0858 (C ₁₀ H ₁₁ N) 3-MeOMC C ₁₁ H ₁₆ NO ₂ 5.2 194.1176 161.0835 138.0683 145.0887 4-MEC C ₁₂ H ₁₈ NO 6.8 192.1384 145.0887 147.0805 146.0965 4-FMC C ₁₀ H ₁₃ FNO 4.4 182.0976 149.0636 123.0607 - Methylone C ₁₁ H ₁₄ NO ₃ 4.0 208.0969 160.0757 132.0809 - Butylone C ₁₂ H ₁₆ NO ₃ 5.7 222.1125 174.0914 175.0628 146.0965 C ₁₀ H ₁₂ NO ₃ 7.6 236.1281 188.1071 175.0630 135.0442 Dipentylone C ₁₄ H ₂₀ NO ₃ 8.0 250.1438 100.1126 175.0754 135.0442 <tr< td=""><td>Weddenholie</td><td></td><td>5.0</td><td>104.1071</td><td>(C_9H_9N)</td><td>(C_9H_9O)</td><td></td></tr<>	Weddenholie		5.0	104.1071	(C_9H_9N)	(C_9H_9O)	
3,4-DMMC $C_{12}H_{18}NO$ 8.1 192.1384 (C_3H_3) (C_3H_3O) 3-MMC $C_{11}H_{16}NO$ 6.1 178.1226 160.1121 145.0887 119.0858 3-MMC $C_{11}H_{16}NO$ 6.1 178.1226 160.1121 145.0887 119.0858 3-MeOMC $C_{11}H_{16}NO_2$ 5.2 194.1176 161.0835 138.0683 145.0887 4-MEC $C_{12}H_{18}NO$ 6.8 192.1384 145.0887 147.0805 146.0965 (C_{0}H_{11}N) (C_{0}H_{11}N) (C_{0}H_{11}N) (C_{0}H_{12}N) (C_{0}H_{12}N) 4-FMC $C_{10}H_{13}FNO$ 4.4 182.0976 149.0636 123.0607 - Methylone $C_{11}H_{16}NO_3$ 5.7 222.1125 174.0914 175.0628 146.0965 Butylone $C_{12}H_{16}NO_3$ 5.7 222.1125 174.0914 175.0630 135.0442 Dipentylone $C_{13}H_{18}NO_3$ 7.6 236.1281 188.1071 175.0630 135.0442 Methedrone $C_{11}H_{1$	Ethcathinone	$C_{11}H_{16}NO$	3.9	178.1226	105.0703	133.0650	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $					(C_8H_9)	(C_9H_9O)	
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	3,4-DMMC	$C_{12}H_{18}NO$	8.1	192.1384	159.1042	161.0960 (C. H. O)	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $					$(C_{11}H_{13}N)$ 160 1121	$(C_{11}H_{13}O)$	110.0959
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	3-MMC	$C_{11}H_{16}NO$	6.1	178.1226	$(\mathbf{C}_{11}\mathbf{H}_{11}\mathbf{N})$	$(C_{10}H_{11}N)$	(C_0H_{11})
3-MeOMC C ₁₁ H ₁₆ NO ₂ 5.2 194.1176 IC16H1NO3 (C ₀ H ₁₁ NO) IC36H0O3 (C ₀ H ₁₀ O) IC36H0O3 (C ₁₀ H ₁₁ O) 4-MEC C ₁₂ H ₁₈ NO 6.8 192.1384 145.0887 (C ₁₀ H ₁₁ N) 147.0805 (C ₁₀ H ₁₁ O) 146.0965 (C ₁₀ H ₁₁ O) 4-FMC C ₁₀ H ₁₃ FNO 4.4 182.0976 149.0636 (C ₉ H ₈ FN) 123.0607 (C ₈ H ₈ F) - Methylone C ₁₁ H ₁₄ NO3 4.0 208.0969 160.0757 (174.0914 175.0628 146.0965 Butylone C ₁₂ H ₁₆ NO3 5.7 222.1125 174.0914 175.0628 146.0965 Butylone C ₁₃ H ₁₈ NO3 7.6 236.1281 188.1071 (C ₁₂ H ₁₄ NO) 175.0630 135.0442 Dipentylone C ₁₄ H ₂₀ NO3 8.0 250.1438 100.1126 175.0754 135.0442 Methedrone C ₁₁ H ₁₆ NO 5.0 194.1176 161.0835 138.0663 145.0887 Methedrone C ₁₄ H ₁₀ NO 6.1 178.1226 160.1121 145.0887 - Methedrone C ₁₁ H ₁₆ NO 5.3 178.1226					161 0835	138 0683	145 0887
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	3-MeOMC	$C_{11}H_{16}NO_2$	5.2	194.1176	$(C_{10}H_{11}NO)$	$(C_{8}H_{10}O_{2})$	$(C_{10}H_{11}N)$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $			6.0	100 100 1	145.0887	147.0805	146.0965
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	4-MEC	$C_{12}H_{18}NO$	6.8	192.1384	$(C_{10}H_{11}N)$	$(C_{10}H_{11}O)$	$(C_{10}H_{12}N)$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $		C II ENO	4.4	192.0076	149.0636	123.0607	
Methylone $C_{11}H_{14}NO_3$ 4.0208.0969 $\frac{160.0757}{(C_{10}H_{10}NO)}$ $\frac{132.0809}{(C_{9}H_{10}N)}$ -Butylone $C_{12}H_{16}NO_3$ 5.7 222.1125 $\frac{174.0914}{(C_{11}H_{12}NO)}$ $\frac{175.0628}{(C_{11}H_{11}O_2)}$ $\frac{146.0965}{(C_{10}H_{12}N)}$ Pentylone $C_{13}H_{18}NO_3$ 7.6 236.1281 $\frac{188.1071}{(C_{12}H_{14}NO)}$ $\frac{175.0630}{(C_{11}H_{11}O_2)}$ $\frac{135.0442}{(C_{8}H_{7}O_2)}$ Dipentylone $C_{14}H_{20}NO_3$ 8.0 250.1438 $\frac{100.1126}{(C_{6}H_{14}N)}$ $\frac{175.0754}{(C_{11}H_{11}O_2)}$ $\frac{135.0442}{(C_{8}H_{7}O_2)}$ Methedrone $C_{11}H_{16}NO_2$ 5.0 194.1176 161.0835 138.0663 145.0887 Mephedrone $C_{11}H_{16}NO$ 6.1 178.1226 160.1121 145.0887 -Buphedrone $C_{11}H_{16}NO$ 5.3 178.1226 131.0731 132.0809 147.0805 MDPV $C_{16}H_{22}NO_3$ 8.7 276.1594 126.1279 135.0442 175.0754 $MDPV$ $C_{16}H_{22}NO_3$ 8.7 276.1594 126.1279 135.0442 175.0754 α -PVP $C_{15}H_{22}NO$ 8.2 232.1696 91.0548 126.1279 161.0691 α -PVP $C_{15}H_{22}NO$ 8.2 232.1696 91.0548 126.1279 161.0691	4-FIVIC	$C_{10}H_{13}FNO$	4.4	102.0970	(C_9H_8FN)	(C_8H_8F)	-
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	Methylone	CuHuNO	4.0	208 0969	160.0757	132.0809	_
Butylone $C_{12}H_{16}NO_3$ 5.7222.1125174.0914 (C_{11}H_{12}NO)175.0628 (C_{11}H_{11}O_2)146.0965 (C_{10}H_{12}N)Pentylone $C_{13}H_{18}NO_3$ 7.6236.1281188.1071 (C_{12}H_{14}NO)175.0630 (C_{11}H_{11}O_2)135.0442 (C_{8}H_{7}O_2)Dipentylone $C_{14}H_{20}NO_3$ 8.0250.1438100.1126 (C_{6}H_{14}N)175.0754 (C_{11}H_{11}O_2)135.0442 (C_{8}H_{7}O_2)Methedrone $C_{11}H_{16}NO_2$ 5.0194.1176161.0835 (C_{10}H_{11}NO)138.0663 (C_{8}H_{10}O_2)145.0887 (C_{10}H_{11}N)Mephedrone $C_{11}H_{16}NO$ 6.1178.1226160.1121 (C_{10}H_{14}N)145.0887 (C_{10}H_{11}N)-Buphedrone $C_{11}H_{16}NO$ 5.3178.1226131.0731 (C_{9}H_{9}N)132.0809 (C_{9}H_{10}N)147.0805 (C_{10}H_{11}O)Pentedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809 (C_{9}H_{10}N)161.0961 (C_{11}H_{13}O)-MDPV $C_{16}H_{22}NO_3$ 8.7276.1594 (C_{8}H_{16}N)126.1279 (C_{8}H_{16}N)175.0754 (C_{8}H_{7}O_2)161.0691 (C_{11}H_{11}O_2) α -PVP $C_{15}H_{22}NO$ 8.2232.169691.0548 (C_{12}H_{12}N)126.1279 (C_{14}H_{2}N)161.0691 (C_{11}H_{11}O_2)	Wiethylone	0111141003	4.0	200.0707	$(C_{10}H_{10}NO)$	$(C_9H_{10}N)$	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Butvlone	$C_{12}H_{16}NO_3$	57	222 1125	174.0914	175.0628	146.0965
Pentylone $C_{13}H_{18}NO_3$ 7.6236.1281188.10/1 (C_{12}H_{14}NO)175.0630 (C_{11}H_{10}O_2)135.0442 (C_{8}H_7O_2)Dipentylone $C_{14}H_{20}NO_3$ 8.0250.1438100.1126 (C_{6}H_{14}N)175.0754 (C_{11}H_{10}O_2)135.0442 (C_{8}H_7O_2)Methedrone $C_{11}H_{16}NO_2$ 5.0194.1176161.0835 (C_{10}H_{11}NO)138.0663 (C_{8}H_{10}O_2)145.0887 (C_{10}H_{11}N)Mephedrone $C_{11}H_{16}NO$ 6.1178.1226160.1121 (C_{11}H_{14}N)145.0887 (C_{10}H_{11}N)-Buphedrone $C_{11}H_{16}NO$ 5.3178.1226131.0731 (C_{9}H_{9}N)132.0809 (C_{9}H_{10}N)147.0805 (C_{10}H_{11}O)Pentedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809 (C_{9}H_{10}N)161.0961 (C_{11}H_{13}O)-MDPV $C_{16}H_{22}NO_3$ 8.7276.1594126.1279 (C_{8}H_{6}N)126.1279 (C_{8}H_{7}O_2)161.0691 (C_{11}H_{11}O_2) α -PVP $C_{15}H_{22}NO$ 8.2232.169691.0548 (C_{H})126.1279 (C_{H})161.0691 (C_{11}H_{10}O)	2 00 10000	01212101 (0)	0.17		$(C_{11}H_{12}NO)$	$(C_{11}H_{11}O_2)$	$(C_{10}H_{12}N)$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Pentylone	$C_{13}H_{18}NO_{3}$	7.6	236.1281	188.10/1	175.0630	135.0442
Dipentylone $C_{14}H_{20}NO_3$ 8.0250.1438100.1126175.0754155.0442Methedrone $C_{11}H_{16}NO_2$ 5.0194.1176161.0835138.0663145.0887Mephedrone $C_{11}H_{16}NO$ 6.1178.1226160.1121145.0887(C_{10}H_{11}N)Mephedrone $C_{11}H_{16}NO$ 6.1178.1226160.1121145.0887-Buphedrone $C_{11}H_{16}NO$ 5.3178.1226160.1121145.0897-Mephedrone $C_{11}H_{16}NO$ 5.3178.1226160.1121145.0897-Muphedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809161.0961-MDPV $C_{16}H_{22}NO_3$ 8.7276.1594126.1279135.0442175.0754 $(C_8H_{16}N)$ $(C_8H_{16}N)$ $(C_8H_{10}O_2)$ $(C_{11}H_{11}O_2)$ - α -PVP $C_{15}H_{22}NO$ 8.2232.169691.0548126.1279161.0691 $(C_{11}H_{10}O_2)$ $(C_{11}H_{10}O_2)$ $(C_{11}H_{10}O_2)$ $(C_{11}H_{10}O_2)$					$(C_{12}H_{14}NO)$	$(C_{11}H_{11}O_2)$	$(C_8H_7O_2)$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Dipentylone	$C_{14}H_{20}NO_3$	8.0	250.1438	(C, H, N)	1/5.0/54	155.0442
Methedrone $C_{11}H_{16}NO_2$ 5.0194.1176101.0833 (C_{10}H_{11}NO)138.0003 (C_{8}H_{10}O_2)143.0887 (C_{10}H_{11}N)Mephedrone $C_{11}H_{16}NO$ 6.1178.1226160.1121 (C_{11}H_{14}N)145.0887 (C_{10}H_{11}N)-Buphedrone $C_{11}H_{16}NO$ 5.3178.1226131.0731 (C_{9}H_{9}N)132.0809 (C_{9}H_{10}N)147.0805 (C_{10}H_{11}O)Pentedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809 (C_{9}H_{10}N)161.0961 (C_{11}H_{13}O)-MDPV $C_{16}H_{22}NO_3$ 8.7276.1594126.1279 (C_{8}H_{16}N)135.0442 (C_{8}H_{7}O_2)175.0754 (C_{11}H_{10}) α -PVP $C_{15}H_{22}NO$ 8.2232.169691.0548 (C_{14})126.1279 (C_{9}H_{9}N)161.0691 (C_{9}H_{9}N)					161 0835	138 0663	$(C_8\Pi_7O_2)$ 1/5 0887
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Methedrone	$C_{11}H_{16}NO_2$	5.0	194.1176	$(C_{10}H_{11}NO)$	$(C_{\circ}H_{10}O_{2})$	$(C_{10}H_{11}N)$
Mephedrone $C_{11}H_{16}NO$ 6.1178.1226100111111010101Buphedrone $C_{11}H_{16}NO$ 5.3178.1226131.0731132.0809147.0805Pentedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809161.0961-MDPV $C_{16}H_{22}NO_3$ 8.7276.1594126.1279135.0442175.0754 α -PVP $C_{15}H_{22}NO$ 8.2232.169691.0548126.1279161.0691 $(C_{2}H_{10}N)$ $(C_{2}H_{10}N)$ $(C_{2}H_{10}N)$ $(C_{11}H_{11}O_2)$		~ ~ ~ ~ ~ ~ ~			160.1121	145.0887	
Buphedrone $C_{11}H_{16}NO$ 5.3178.1226131.0731 (C ₉ H ₉ N)132.0809 (C ₉ H ₁₀ N)147.0805 (C ₁₀ H ₁₁ O)Pentedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809 	Mephedrone	$C_{11}H_{16}NO$	6.1	178.1226	$(C_{11}H_{14}N)$	$(C_{10}H_{11}N)$	-
Buphedrone $C_{11}H_{16}NO$ 5.3178.1226(C_9H_9N)(C_9H_10N)(C_{10}H_{11}O)Pentedrone $C_{12}H_{18}NO$ 7.2192.1384132.0809 (C_9H_10N)161.0961 (C_{11}H_{13}O)-MDPV $C_{16}H_{22}NO_3$ 8.7276.1594126.1279 (C_8H_16N)135.0442 (C_8H_7O_2)175.0754 (C_{11}H_{11}O_2) α -PVP $C_{15}H_{22}NO$ 8.2232.169691.0548 (C_7H_2)126.1279 (C_9H_10N)161.0691 (C_9H_10N)	Dunhadrona		5.2	178.1226	131.0731	132.0809	147.0805
$\begin{array}{ c c c c c c c c } \hline Pentedrone & C_{12}H_{18}NO & 7.2 & 192.1384 & \begin{array}{c} 132.0809 & 161.0961 & & \\ \hline (C_{9}H_{10}N) & (C_{11}H_{13}O) & & \\ \hline MDPV & C_{16}H_{22}NO_3 & 8.7 & 276.1594 & \begin{array}{c} 126.1279 & 135.0442 & 175.0754 & \\ \hline (C_{8}H_{16}N) & (C_{8}H_{7}O_2) & & \\ \hline (C_{11}H_{11}O_2) & & \\ \hline & & & & \\ \hline & & & & \\ \hline & & & &$	Bupnedrone	$C_{11}H_{16}NO$	5.3		(C_9H_9N)	$(C_9H_{10}N)$	$(C_{10}H_{11}O)$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Pentedrone	C.H.NO	7.2	102 1384	132.0809	161.0961	
$\begin{array}{ c c c c c c c c } \hline MDPV & C_{16}H_{22}NO_3 & 8.7 & 276.1594 & \begin{array}{c c c c c c c c c c c c c c c c c c c $	Tentedrone		1.2	192.1384	$(C_9H_{10}N)$	$(C_{11}H_{13}O)$	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	MDPV	$C_{16}H_{22}NO_3$	8.7	276.1594	126.1279	135.0442	175.0754
α -PVP $C_{15}H_{22}NO$ 8.2 232.1696 91.0548 126.1279 161.0691 (C_2H_2) (C_2H_2) (C_2H_2N) (C_2H_2N)				_, 0.107 f	$(C_8H_{16}N)$	$(C_8H_7O_2)$	$(C_{11}H_{11}O_2)$
$(C_{2}H_{2}) = (C_{2}H_{2}) = (C_{2}H_{2})$	α-PVP	$C_{15}H_{22}NO$	8.2	232.1696	91.0548	126.1279	161.0691
$\frac{(C/11/)}{10(1000)} = \frac{(C/11/)}{10(1000)} = \frac{(C/11/)}{10(1000)}$					(C7H7)	$(C_8H_{16}N)$	$(C_{11}H_{13}O)$
α -PVT C ₁₃ H ₂₀ NOS 6.8 238.1258 $\begin{pmatrix} 120.12/8 \\ C_{c}H_{c}N \end{pmatrix}$ $\begin{pmatrix} 9/.0111 \\ C_{c}H_{c}S \end{pmatrix}$ $\begin{pmatrix} 16/.0524 \\ C_{c}H_{c}OS \end{pmatrix}$	α-PVT	$C_{13}H_{20}NOS$	6.8	238.1258	$120.12/\delta$ (C _c H ₁ N)	97.0111 (C-H-S)	107.0524

Table S1. Retention time (RT), parent and product ions (theoretical accurate m/z values and chemical formulae) obtained for NPS fragmentation with Q-Exactive mass analyser.

$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	3,4-DMeO- α-	CurllaNOa	Q /	202 1007	151.0754	126.1279	221.1173
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	PVP	$C_{17}H_{261}NO_3$	8.4 292.190	292.1907	$(C_9H_{11}O_2)$	$(C_8H_{16}N)$	$(C_{13}H_{17}O_3)$
$\begin{array}{ c c c c c c c } \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2}N}) \\ \hline (c_{c H_{2}N}) & (c_{c H_{2}N}) & (c_{c H_{2$	$A_{-}C1_{-}\alpha_{-}PPP$	CuHurCINO	8.0	238 0993	98.0969	139.0310	167.0258
Phenethylamines Phenethylamines 25B-NBoMe $C_{18}H_{23}BrNO_3$ 14.7 380.0860 (CaH-O) (CrH-O) (CrH-O) 25C-NBoMe $C_{18}H_{22}CINO_3$ 14.2 336.1361 121.0650 91.0548 - 25I-NBoMe $C_{18}H_{23}INO_3$ 15.5 428.0717 121.0650 91.0548 - 25IP-NBoMe $C_{21}H_{09}NO_3$ 17.6 344.2219 (CaH-O) (CrH-O) (CrH-O) 2C1-4.5- $C_{11}H_{15}CINO_2$ 9.0 230.0942 (CaH-O) (CrH-O) (CrH-O) PMA $C_{10}H_{16}NO$ 5.2 166.1226 121.0650 149.0962 - PMMA $C_{10}H_{18}NO$ 5.8 180.1383 121.0650 (CaH-O) (C	4-01-0-111		0.0	230.0773	$(C_6H_{12}N)$	(C_8H_8Cl)	(C ₉ H ₈ OCl)
$\begin{array}{c c c c c c c c c c c c c c c c c c c $			r	Phenethy	lamines	1	
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	25B-NBoMe	C10H22BrNO2	147	380 0860	121.0650	91.0548	_
$\begin{array}{c c c c c c c c c c c c c c c c c c c $		0181123011(03	11.7	500.0000	(C_8H_9O)	(C ₇ H ₇)	
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	25C-NBoMe	C18H23ClNO3	14.2	336.1361	121.0650	91.0548	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $		- 1023			(C_8H_9O)	(C ₇ H ₇)	
$\begin{array}{ c c c c c c } \hline \hline C(2\pi h_3 0) & (C\pi h_7) & (C\pi h_7$	25I-NBoMe	$C_{18}H_{23}INO_3$	15.5	428.0717	121.0650	91.0548	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $					(C_8H_9O)	(C_7H_7)	
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	25iP-NBoMe	$C_{21}H_{30}NO_3$	17.6	344.2219	121.0650	91.0548	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	2 C1 4 5				<u>(C8П9О)</u> 195 0262	$(C_7\Pi_7))$	179.0097
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	2-CI-4,3-	$C_{11}H_{15}ClNO_2$	9.0	230.0942	185.0303	(C H O C)	1/8.098/
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	IVIDIVIA				121.0650	$(C_{10}\Pi_{11}O_{2}C_{1})$	$(C_{11}\Pi_{14}O_{2})$
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	PMA	$C_{10}H_{16}NO$	5.2	166.1226	$(C_{2}H_{2}O)$	$(C_{14}H_{12}O)$	-
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$					121.0650	1/0.0062	
$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$	PMMA	$C_{11}H_{18}NO$	5.8	180.1383	$(C_{0}H_{0}O)$	$(C_{10}H_{12}O)$	-
$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$					181 1010	166.0776	103 0546
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	NEDPA	$C_{16}H_{20}N$	10.9	226.1590	$(C_{14}H_{12})$	$(C_{13}H_{10})$	$(C_{8}H_{7})$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $					105 0703	79 0549	(0,11/)
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	2-PEA	$C_8H_{12}N$ 3	3.2	122.0966	(C_8H_9)	$(C_{6}H_{7})$	-
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	<i>6</i> JT	C U N	1.0	6 175.1229	158.0963	130.0652	117.0575
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	5-11	$C_{11}H_{15}N_2$	4.6		$(C_{11}H_{12}N)$	(C_9H_8N)	(C_8H_7N)
6-APDBC11H16NO3.3178.1220 (C_9H_9O) $(C_{11}H_{13}O)$ -MT-45 $C_{24}H_{33}N_2$ 15.2349.2633181.1010166.0776-MT-45 $C_{24}H_{33}N_2$ 15.2349.2633181.1010 $(C_{14}H_{13})$ $(C_{13}H_{10})$ -AMT $C_{11}H_{15}N_2$ 5.9175.1229158.0963130.0652-5-MeO-MIPT $C_{15}H_{22}N_2O$ 7.2247.1805174.0914121.0650203.10675-MeO-DALT $C_{17}H_{22}N_2O$ 9.3271.1805110.0966174.0912-6-Minorex $V_{11}H_{12}N_2O$ 9.3271.1805110.0966174.0912-4- $C_{10}H_{13}N_2O$ 6.0177.1022160.119145.0885134.0964methylaminorex $C_{10}H_{13}N_2O$ 8.2191.1179148.1121131.0857-4,4' - DMAR $C_{11}H_{15}N_2O$ 8.2191.1179148.1121131.0857- V_{4} $V_{11}H_{15}N_2O$ V_{21} $V_{11}H_{14}N_1$ $V_{11}H_{15}N_2$ - V_{21} V_{21} V_{21} V_{21} V_{21} - V_{21} V_{21} V_{22} V_{21} V_{22} - V_{22} V_{21} V_{22} V_{21} V_{22} - V_{22} V_{21} V_{22} V_{22} V_{22} V_{22} V_{22} V_{22} V_{22} V_{21} V_{22} V_{22} <		C II NO	5 5	179 1006	133.0648	161.0959	
$\begin{array}{ c c c c c c } \hline Opioids \\ \hline MT-45 & C_{24}H_{33}N_{2} & 15.2 & 349.2633 & \frac{181.1010}{(C_{14}H_{13})} & \frac{166.0776}{(C_{13}H_{10})} & - \\ \hline \\$	0-APDD	$C_{11}\Pi_{16}NO$	5.5	178.1220	(C_9H_9O)	$(C_{11}H_{13}O)$	-
$\begin{array}{c c c c c c c c c c c c c c c c c c c $				Opio	ids		
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	MT_45	CarHanNa	15.2	3/10 2633	181.1010	166.0776	
$\begin{tabular}{ c c c c c c c } \hline $Tryptation $ISR.0963$ & 130.0652 & $C_{11}H_{12}N$ & $C_{11}H_{15}N_2$ & 2.9 & 175.1229 & 158.0963 & $(C_{9}H_8N)$ & $-$$$$C_{9}H_8N$ & $C_{9}H_8N$ & $C_{9}H_8N$ & $C_{12}H_{12}N_2$ & $C_{11}H_{12}N_3$ & $(C_{9}H_8N)$ & 121.0650 & 203.1067 & $(C_{12}H_{15}N_2O)$ & $C_{12}H_{12}N_2$ & $(C_{11}H_{12}NO)$ & $(C_{8}H_9O)$ & $(C_{12}H_{15}N_2O)$ & $C_{12}H_{15}N_2O$ & 2.1067 & $(C_{11}H_{12}NO)$ & $(C_{8}H_9O)$ & $(C_{12}H_{15}N_2O)$ & $C_{12}H_{15}N_2O$ & 2.1067 & $(C_{11}H_{12}NO)$ & $(C_{11}H_{12}NO)$ & $(C_{11}H_{12}NO)$ & $(C_{11}H_{12}NO)$ & $(C_{11}H_{12}NO)$ & $C_{11}H_{15}N_2O$ & 9.3 & 271.1805 & 110.0966 & 174.0912 & $C_{11}H_{12}NO$ & $-$$ & V	111-45	C2411331V2	13.2	547.2055	$(C_{14}H_{13})$	$(C_{13}H_{10})$	
$\begin{array}{c c c c c c c c c c c c c c c c c c c $				Tryptar	nines	1	1
$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	AMT	$C_{11}H_{15}N_{2}$	59	175 1229	158.0963	130.0652	_
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$			5.5	175.1227	$(C_{11}H_{12}N)$	(C_9H_8N)	
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	5-MeO-MIPT	C15H22N2O	7.2	247.1805	174.0914	121.0650	203.1067
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$			=	2	$(C_{11}H_{12}NO)$	(C ₈ H ₉ O)	$(C_{12}H_{15}N_2O)$
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	5-MeO-DALT	$C_{17}H_{22}N_2O$	9.3	271.1805	110.0966	174.0912	-
Aminorex derivatives 4- methylaminorex $C_{10}H_{13}N_2O$ 6.0 177.1022 160.119 ($C_{10}H_{12}N_2$) 145.0885 ($C_{10}H_{11}N$) 134.0964 ($C_{9}H_{12}N$) 4,4' - DMAR $C_{11}H_{15}N_2O$ 8.2 191.1179 148.1121 ($C_{10}H_{14}N$) 131.0857 ($C_{10}H_{11}$) - Ketamine analogues			l	A · 1	$(C_7H_{12}N)$	$(C_{11}H_{12}NO)$	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Aminorex derivatives						
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	4-	$C_{10}H_{13}N_2O$	6.0 177.102	177.1022	160.119	145.0885	134.0964
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	metnyiaminorex	-			$(C_{10}H_{12}N_2)$	$(U_{10}H_{11}N)$	$(C_9H_{12}N)$
Ketamine analogues	4,4' – DMAR	$C_{11}H_{15}N_2O$	8.2	191.1179	148.1121	131.085/	-
	Katamina analoguas						
				Ketannile a	121 0650	175 1117	203 1067
Methoxetamine $C_{15}H_{22}NO_2$ 7.3 248.1640 $(C_{2}H_{2}O)$ $(C_{15}H_{12}O)$ $(C_{15}H_{12}O)$ $(C_{15}H_{12}O)$ $(C_{15}H_{12}O)$	Methoxetamine	$C_{15}H_{22}NO_2$	7.3	248.1640	$(C_{\circ}H_{\circ}O)$	$(C_{12}H_{12}\Omega)$	$(C_{12}H_{12}O_{2})$

HRMS ions are ordered according to their relative abundance in the spectrum, from the most (Fragment 1) to the least abundant (Fragment 3).

Fig S1. Chromatograms and MS² spectra for 3,4-DMeO-alpha-PVP in A) wastewater and B) reference standard. Identification at confidence level 1.



Fig S2. Chromatograms and MS² spectra for 2-phenetylamine (2-PEA) in A) wastewater and B) reference standard. Identification at confidence level 1.





Fig S3. Chromatograms and MS² spectra for para-methoxyamphetamine (PMA) in A) wastewater and B) reference standard. Identification at confidence level 1.

Fig S4. Chromatograms and MS² spectra for (A) 2-methoxyamphetamine (2-MA) in wastewater and B) analytical standard of *para*-methoxyamphetamine (PMA). Identification at confidence level 2.



Fig. S5. Chromatogram and MS^2 spectra for the DOiP in wastewater. Identification at confidence level 3.











Fig. S8. Chromatogram and MS^2 spectra for the diphenidine in wastewater. Identification at confidence level 3.





Fig. S9. Chromatogram and MS^2 spectra for the AMB-FUBINACA in wastewater. Identification at confidence level 3.