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Analysis of Psychoactive Cathinones and Tryptamines by Electrospray Ionization Atmospheric Pressure Ion Mobility Time-of-Flight Mass Spectrometry

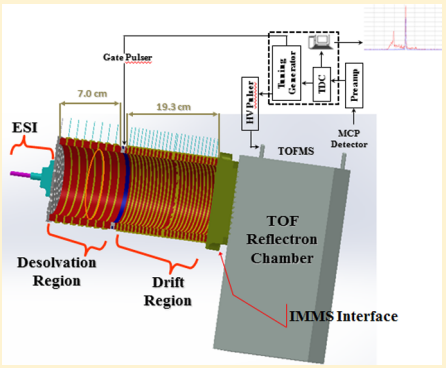
A Bakarr Kanu,*[,†] Simon D. Brandt,[‡] Mike D. Williams,[§] Nancy Zhang,[§] and Herbert H. Hill[§]

[†]Department of Chemistry, Winston-Salem State University, Winston-Salem, North Carolina 27110, United States

[‡]School of Pharmacy & Biomolecular Sciences, Liverpool John Moores University, Byrom Street, Liverpool, L3 3AF, U.K.

[§]Department of Chemistry, Washington State University, Pullman, Washington 99164-4630, United States

ABSTRACT: The ability to use positive ion monitoring mode with an atmospheric pressure ion mobility time-of-flight mass spectrometer (APIM(tof)-MS) to detect psychoactive cathinones and tryptamines from aqueous phase samples was evaluated. The study used a traditional electrospray ionization (ESI) source for sample introduction and ionization. A total of four cathinones (mephedrone, butylone, 4-Me-PPP, and 4-MEC) and five tryptamines (5-EtO-DPT, 5-EtO-DALT, 5-EtO-MIPT, 5-EtO-ALCHT, and 5-EtO-2MALET) were investigated, and we report on parent ions, collision induced dissociation (CID) fragment ions, reduced mobility (K_o), mass flight times, and detection limits obtained from a single instrument run for the psychoactive substances. Detection limits reported ranged from 3 to 11 μM concentration for the compounds studied. This detection limit range corresponded to 1−5 ng of material needed for improved detection on the instrument. This article demonstrates that it was possible to use a single instrument platform for the separation, detection, and identification of cathinones and tryptamines in less than 1 min. The application holds great promise for detecting and identifying a new class of drugs often referred to as "bath salts" or "legal highs" distributed over the Internet.



Structural modifications of molecules form the basis of any form of drug discovery in the attempt to understand their medicinal and pharmacological properties. Clearly, the manipulation of structural templates and the ability to understand the interactions involved between drugs and their molecular targets is important for the development of new drugs. While a large proportion of potential drug candidates are abandoned within the industrial context, it has become apparent that a variety of drug derivatives described in patents and the scientific literature are being increasingly detected on the recreational drug market.[1,2] Commonly used terms for these particularly diverse groups of compounds may range from "designer drugs" and "new psychoactive substances" to "legal highs" or "bath salts", respectively. One of the compound classes encountered recently as emerging psychoactive "legal highs" are synthetic cathinone analogues structurally derived from the mild stimulant 2-amino-1-phenylpropan-1-one. With the increasing availability of pharmacological data obtained for a number of recreationally used cathinones (e.g., mephedrone, methylone, and MDPV), researchers hope to shed more light on their properties. It has become clear that there may be some overlap but also differences in their modes of action.[3,4] Bupropion (Wellbutrin, Zyban) is also a cathinone derivative which is used for the treatment of nicotine dependence, and a large number of other cathinone derivatives have been prepared to evaluate their potential for treating depression, nicotine, cocaine addiction, or obesity.[5−9]

Some of the triptan analogues used for the treatment of migraine are characterized by the presence of *N*,*N*-dimethyltryptamine (DMT), and closely related substances have also been explored as valuable receptor probes.[10−12] Some *N*,*N*-dimethylated derivatives are found in nature,[13,14] but so far higher homologues have been products of research laboratories and are reported to display a range of psychoactive effects in humans.[15] A number of these derivatives have also been available over the Internet as "research chemicals".[16]

Developing effective and efficient responses to new psychoactive substances is a challenge to public health, and this is particularly apparent in a context in which the Internet is playing a pivotal role in diffusion.[17,18] The European Monitoring Centre for Drugs and Drug Addiction (EMCDDA) reported that the number of new psychoactive substances that were notified for the first time in the European Union rose to a total of 73 substances in 2012, and since 2005, over 200 notifications have been reported. Cathinones and tryptamines are frequently represented drug classes, but others are increasingly detected as well.[19] It is becoming ever clearer that the usage of novel psychoactive compound classes is a worldwide phenomenon.

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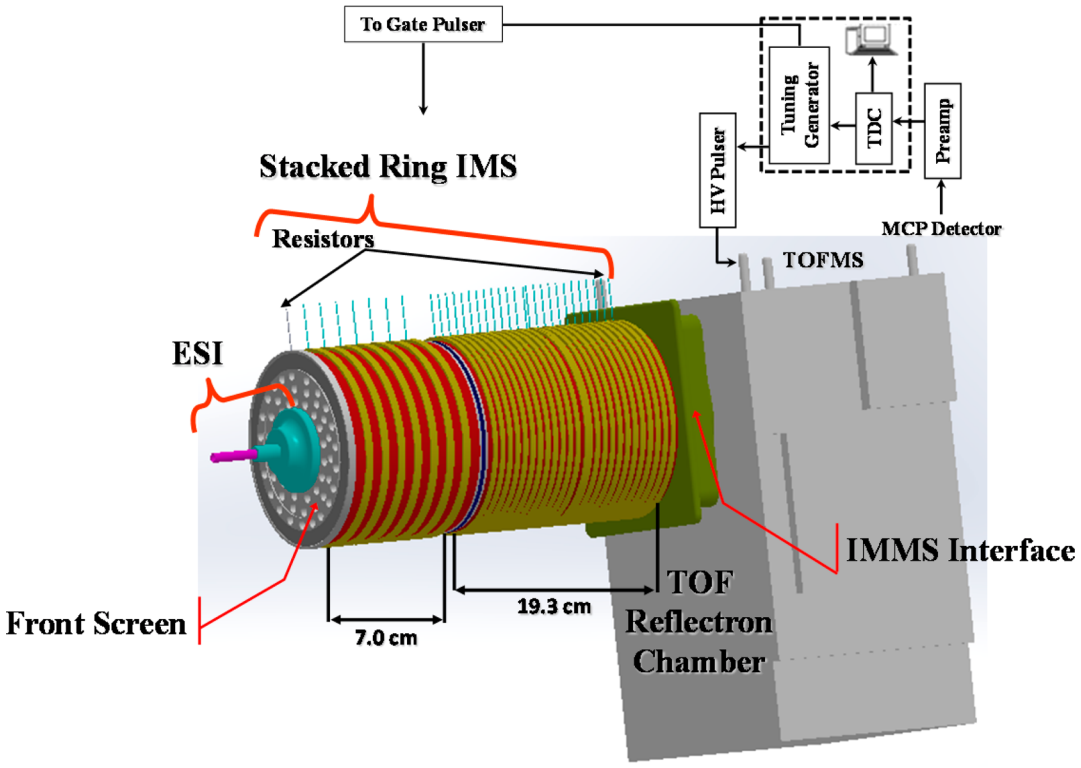

Figure 1. Schematic representation of the APIM(tof)MS used in this investigation. Ions from the electrospray process are injected into the desolvation region of the atmospheric pressure ion mobility tube. These ions are gated and periodically injected into the drift region where they are separated according to their size/charge ratio and pulsed orthogonally into the time-of-flight mass spectrometer; there they are separated according to mass/charge ratio. Two-dimensional (2-D) IMMS data were acquired, and for every ion mobility measurement there were 1000 mass measurements.

The gaining interest in the topic of newly emerging psychoactive substances is reflected in an extensive growth of research publications concerned with the implementation and applications of instrumental analysis to areas important for clinical and forensic scientists. As expected, the most commonly used approaches include the coupling of mass spectrometry (MS) detection to gas chromatography (GC) and liquid chromatography (LC) as a key feature of separation. In cases where unambiguous compound identification is required, especially when reference materials are unavailable, additional spectroscopic tools such as nuclear magnetic resonance (NMR) spectroscopy are of key importance. The range of these methodologies applied to the analysis of substituted cathinone analogues included the characterization of Internet products or synthesized material (e.g., refs 20−31), metabolism studies (e.g., refs 32 and 33), or detection in biofluids (e.g., refs 34−36). In addition, the application of X-ray crystallography,[37] Raman spectroscopy,[38] and capillary electrophoresis[39] have also been reported, thus broadening the areas of analysis. A review of the literature on profiling and analysis of psychoactive synthetic tryptamines also revealed that HPLC-, GC-, and CE-based approaches were most frequently used.[40,41]

An instrumental approach that has yet to be employed is atmospheric pressure ion mobility time-of-flight mass spectrometry (APIM(tof)MS). Indeed, the coupling of the two techniques complemented and instrumentally matched one another so well that they seemed to fuse into one analytical measurement.[42] The APIM separates ions in the millisecond time scale and gives size/charge information of the data. The MS separates ions in the microsecond time scale and gives mass/charge information of the data.[42] The two techniques when coupled together provide three-dimensional data of intensity, drift time, and mass. This information after processing will give two-dimensional mobility−mass spectra. Several publications have described the use of ion mobility mass spectrometry (IMMS) for the analysis of drugs of abuse,[42−47] but IMMS or specifically APIM(tof)MS applications for cathinones and tryptamines is currently absent in the literature. When the literature was reviewed, it was noted that several techniques have to be combined to properly evaluate and provide all the information needed for adequately analyzing the different forms of cathinones and tryptamines derivatives purchased from the Internet. The capability of IMMS as a single tool may well be the answer to this challenge.

For the first time, data for four tryptamines and five cathinones are presented following analysis by APIM(tof)MS in positive ion mode. The positive ion mode was chosen for this study due to the facile formation of positive ions[43−47] when drugs of abuse are analyzed. Drift times, reduced mobility, and detection limits were determined.

EXPERIMENTAL SECTION

Instrumentation. The fundamental parts of the instrumentation used to perform the experiments consisted of an electrospray ionization (ESI) source, an atmospheric pressure ion mobility spectrometer (APIMS), and a time-of-flight mass spectrometer (tof)MS. The APIMS was constructed in-house at Washington State University (Pullman, WA), and the (tof)MS was obtained from Ionwerks (Houston, TX). The two instruments were coupled at Washington State University.

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The ESI-APIM(tof)MS configuration has been previously described in considerable detail.[47−50] Figure 1 is a schematic representation of the APIM(tof)MS design used for these experiments, and all operating conditions for positive mode operation are summarized in Table 1.

Table 1. APIM(tof)MS Operating Conditions Summary

Electrospray Ionization		
ESI voltage	3000	V
ESI flow rate	3	μL min^{-1}
sample concentration	50	μM
Atmospheric Pressure Ion Mobility		
desolvation region length	7	cm
drift region length	19.3	cm
first ring voltage	10 000	V
ion gate voltage	8590	V
last ring voltage	770	V
drift flow	1000	mL min^{-1}
drift gas	nitrogen	
temperature	200	°C
pressure	680−705	Torr
IMS scan time		ms
Pressure Interface		
pressure	2.1	Torr
nozzle lens	200	V
focus lens	700	V
skimmer lens	28	V
Time-of-Flight Mass Spectrometry		
pressure	2×10^{-6}	Torr
lens 1	12.20	V
lens 2	32.30	V
lens 3	−80.40	V
deflector up	−0.50	V
deflector down	0.50	V
reflector back plane	−361.50	V
reflector grid plane	969.50	V
MCP front	4500.00	V
MCP bias	2320.00	V
Acquisition Timing		
(tof)MS resolution	600	ns
APIM gate pulse frequency	5000	Hz
APIM gate pulse width	200	μs
(tof)MS extraction frequency	16.667	kHz
(tof)MS extraction pulse width	3	μs

Electrospray Ionization. The ESI solution was delivered with the use of a syringe pump (KD Scientific, Holliston, MA). The flow rates were set at 3 μL min^{-1} and delivered from a 250 μL syringe (Hamilton gastight syringe, Reno, NV) during the entire set of experiments. The syringe was connected to a 360 μm o.d. × 20 μm i.d. × 25 cm long fused silica capillary (Polymicro Technologies, Phoenix, AZ) using a zero dead volume needle to capillary connector (Upchurch Scientific, Oak Harbor, WA). In trying to optimize the flow rate, it was necessary to use a large tip aperture and connect this aperture to another fused silica capillary of smaller internal diameter (i.d.). To achieve the flow rate, this tip was then connected to an emitter using another zero dead volume stainless steel union (Upchurch Scientific, Oak Harbor, WA). The emitter used was a 150 μm o.d. × 50 μm i.d. × 18 cm long fused silica capillary (Polymicro Technologies, Phoenix, AZ). The electrical contact was applied through the stainless steel zero dead volume union (Upchurch Scientific, Oak Harbor, WA) that connected the emitter with the fused silica capillary transfer line (360 μm o.d. × 75 μm i.d., ∼25 cm long).

Atmospheric Pressure Ion Mobility. The high resolution APIM consisted of the conventional stack ring interlocking design for a total length of 26.3 cm[51,52] using stainless steel rings and Macor. The instrument consisted of two sections, and the division was achieved using a Bradbury-Nielson ion gate. The desolvation and drift regions were 7.0 and 19.3 cm long, respectively. To generate a uniform electric field down the tube, the stainless steel guard rings were connected to each other through 0.5 MΩ (desolvation region) and 1 MΩ (drift region) high-temperature resistors for the reaction and drift regions, respectively (±1%; Caddock Electronics Inc., Riverside, CA). A 10 kV voltage was normally applied to the first ring electrode, and the last ring electrode voltage was adjusted by a variable resistor to be ∼770 V referenced to ground. The drift voltage was dropped gradually across the drift tube via the resistor chain to form an electric field of 380 V cm^{-1} in the desolvation region and 445 V cm^{-1} in the drift region. The lower electric field in the desolvation region allowed solvated ions to spend more time in the heated drift gas for more efficient desolvation prior to introduction into the drift region. Ions were introduced into the drift region at a gate pulse width of 0.2 ms and a frequency of 5000 Hz. The temperature of the APIM was set to 200 °C and the buffer gas (nitrogen) flow rate was 1 L min^{-1} throughout the tube. Reduced mobility (K_o) was calculated using the dimensions of the APIM. To ensure consistent K_o values, caffeine (1.50−1.54 cm^2 V^{-1} s^{-1}) was used to standardize the instrument.[53]

Time-of-Flight Mass Spectrometry. A low-pressure interface (∼2.1 Torr) was used to interface the APIM (680−705 Torr for Pullman, WA) to the (tof)MS (∼10^{-6} Torr). The interface consisted of three nozzle, focus, and skimmer lenses. Ions were guided from the interface by a series of lenses and orthogonally pulsed into the 1 m reflectron flight tube and detected by a multichannel plate. Acquisition of data for the experimental sequence consisted of a timing mechanism that composed of a real-time three-dimensional (3-D) matrix obtained simultaneously of intensity, mobility drift, and mass flight time. The APIM gate pulse width of 0.2 ms at a frequency of 5000 Hz used allowed for a maximum of 30 ms APIM spectra to be acquired. The (tof)MS extraction frequency was set to 16.667 kHz, which provided a mass spectrum that consisted of ions with flight times up to 60 μs. Within each 30 ms time window 2.0×10^3 (tof) extractions were obtained. The APIM, (tof) extractor, and (tof) digital-to-time converter were all triggered by a personal computer (PC) based timing controller. Synchronization of the electronic hardware was facilitated by the use of a Dell Precision T3400 model with a Core 2 Quad Processor workstation running the 3-D Ionwerks data acquisition software. Six replicate measurements were made to provide adequate ion statistics during data analysis. Acquired data were then transported to another personal computer and analyzed using a program created by Ionwerks that ran on IDL Virtual Machine Version 6.3 (ITT Visual Information Solutions, Boulder, CO). Two-dimensional spectra for APIM-(tof)MS data were acquired using m/z, drift time, and intensity for each ion.

Safety Considerations. Parts of the instrument, the ESI source and IMS tube, were operated at high voltages; thus only trained personnel had the privilege to operate the instrument.

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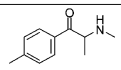
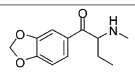
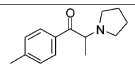
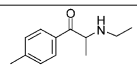
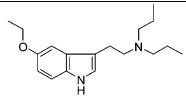
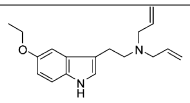
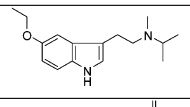
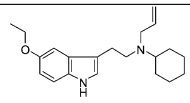
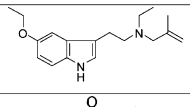
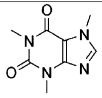
Table 2. Psychoactive Cathinones and Tryptamines Investigated in This Study: Structure, Mass[a] (Da), Approximate Mass Detected[b] (Da), Ions,[c] and K_o[d]

Compound	Structure	Mass (Da)[a]	Approx. Mass Detected (Da)[b]	[c]Ions (M+1)$^+$[c]	K_o[d]
Mephedrone		177.27	178.26	$[C_{11}H_{15}NO + H]^+$	1.43 ± 0.01
Butylone		221.28	222.12	$[C_{12}H_{15}NO_3 + H]^+$	1.34 ± 0.02
4-Me-PPP		217.34	218.39	$[C_{14}H_{19}NO + H]^+$	1.33 ± 0.02
4-MEC		191.30	192.22	$[C_{12}H_{17}NO + H]^+$	1.41 ± 0.01
5-EtO-DPT		288.48	289.22	$[C_{18}H_{28}N_2O + H]^+$	1.14 ± 0.02
5-EtO-DALT		284.44	285.19	$[C_{18}H_{24}N_2O + H]^+$	1.16 ± 0.01
5-EtO-MIPT		260.42	261.19	$[C_{16}H_{24}N_2O + H]^+$	1.21 ± 0.02
5-EtO-ALCHT		326.53	327.24	$[C_{21}H_{30}N_2O + H]^+$	1.06 ± 0.01
5-EtO-2MALET		286.46	287.21	$[C_{18}H_{26}N_2O + H]^+$	1.15 ± 0.02
Caffeine		194.22	195.22	$[C_7H_{10}N_4O_2 + H]^+$	1.52 ± 0.02

[a]Mass calculated using atomic masses from the periodic table. [b]Accurate mass detected by (tof)MS. [c]Approximate m/z for $[M + H]^+$ expected. [d]K_o values are in $cm^2\ V^{-1}\ s^{-1}$.

Warning signs were posted to keep site clearance whenever the instrument was in operation mode.

Chemicals and Reagents. A total of four cathinones and five tryptamines, available as their hydrochloride salts, were analyzed. All compounds were available from previous studies.[27,28,54] The four cathinones were mephedrone (2-(methylamino)-1-*p*-tolylpropan-1-one), butylone (1-(benzo-[*d*][1,3]dioxol-5-yl)-2-(methylamino)butan-1-one, 2-(pyrrolidin-1-yl)-1-*p*-tolylpropan-1-one (4-Me-PPP), and 2-(ethylamino)-1-*p*-tolylpropan-1-one (4-MEC). The tryptamines investigated were 5-ethoxy-*N*,*N*-dipropyltryptamine (5-EtO-DPT), 5-ethoxy-*N*,*N*-diallyltryptamine (5-EtO-DALT), 5-ethoxy-*N*-methyl-*N*-isopropyltryptamine (5-EtO-MIPT), 5-ethoxy-*N*-allyl-*N*-cyclohexyltryptamine (5-EtO-ALCHT), and 5-ethoxy-*N*-ethyl-*N*-(2-methylallyl)tryptamine (5-EtO-2MALET). All compounds were prepared in an ESI solvent before analysis, and the ESI solvent used in the positive ion mode was a mixture of 49.5% MeOH/49.5% H_2O/1% HOAc.[54] Methanol was HPLC grade and was obtained from J.T. Baker (Phillipsburg, NJ). Water was 18 MΩ deionized, prepared by Barnstead Nanopure Water Systems.

Calculations. All reduced mobility constants (K_o) were calculated from experimentally determined drift times (t_d) using eq 1.[51,55,56]

$$K_o = \frac{L^2}{Vt_d}\frac{273.15}{T}\frac{P}{760} \quad (1)$$

where L (cm) is the length of the IMS drift cell, V is the voltage drop across L, T is the drift gas temperature (K), and P is the pressure (Torr) of the drift region where the reduced mobility, K_o ($cm^2\ V^{-1}\ s^{-1}$), was determined. Under these standard conditions of temperature (273.15 K) and pressure (760 Torr), the number density of the drift gas molecules is normalized.

The analyte detection limit (dl) was determined from the spectra.[56,57] By assuming a linear response, the minimum concentration closer to the detection limit whose signal was greater than the instrument noise was electrosprayed into the instrument. The detection limits at 3σ were estimated using statistics as follows:

$$\text{analyte}_{dl} = \frac{t_{\alpha,n}\text{RSD}_x(\text{amount electrosprayed})}{100\%} \quad (2)$$

where $t_{\alpha,n}$ is the t-table value for $n - 1$ degrees of freedom, α is the confidence interval, n is the number of replicate measurements minus 1, and RSD_x is the relative standard deviation.[58]

The relative standard deviation was determined using eq 3.

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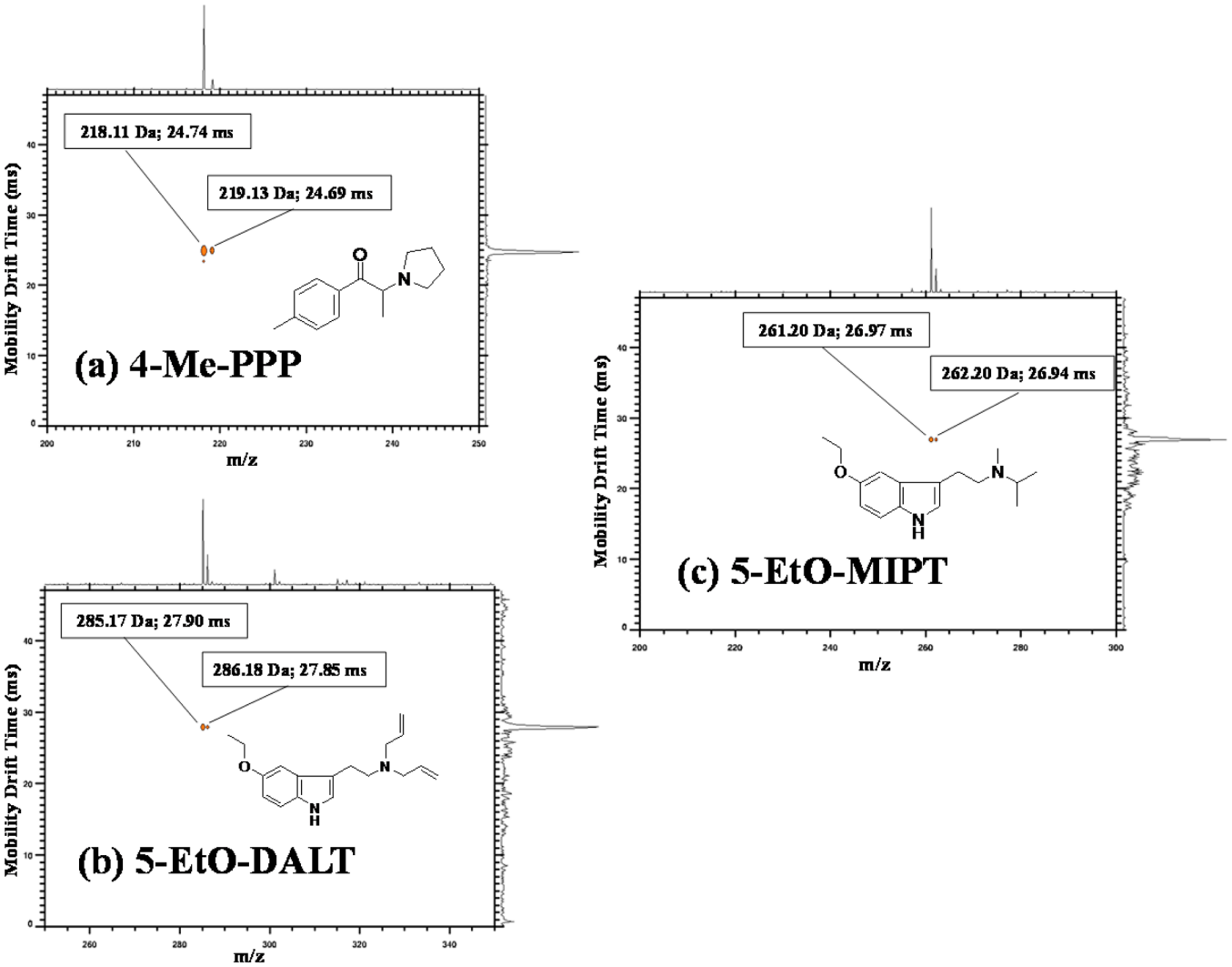

Figure 2. ESI-APIM(tof)MS 2-D spectra of selected cathinones and tryptamines for (a) 4-Me-PPP, (b) 5-EtO-DALT, and (c) 5-EtO-MIPT. Each spectrum shows the $[M + {}^{1}H]^{+}$ and $[M + 2]^{+}$ ions. The +2 ion can be accounted for by the presence of a ^{13}C isotope. It should be noted that the drift times in this figure differ from those presented under Results and Discussion. This was because a second drift tube was interfaced to the same (tof)MS instrument in order to test the reproducibility of the reduced mobility values. The new instrument conditions and associated drift times produced reduced mobility that matched the values presented in Table 2 to 0.01 $cm^2\ V^{-1}\ s^{-1}$.

$$\mathrm{RSD}_x = \frac{S_x}{\mathrm{mean}} \tag{3}$$

where S_x is the standard deviation at 3σ. For six replicate measurements ($n - 1 = 5$), $\alpha = 0.01$ (i.e., 99% confidence that the value of analyte$_{dl}$ was greater than 0, $t_{\alpha,n} = 4.03$) from the Student t-table. With the use of this approach all detection limits of psychoactive cathinones and tryptamines were estimated.

RESULTS AND DISCUSSION

Reduced Mobility and Detection Limit. The cathinone and tryptamine analogues were introduced into the APIM-(tof)MS to determine their reduced mobility (K_o) in nitrogen. Data of K_o for the cathinones and tryptamines studied were not previously available in the literature. To ensure the K_o values reported were accurate and to prevent day-to-day variations of temperature and pressure, a calibration standard was used. This investigation used caffeine as a standard in the positive ion mode. All reduced mobility calculations were obtained using eq 1, and the value obtained for caffeine was $1.52 \pm 0.02\ cm^2\ V^{-1}\ s^{-1}$. The literature K_o values reported for caffeine ranged from 1.50 to 1.54 $cm^2\ V^{-1}\ s^{-1}$.[53,59−61] The drift times for the four cathinones recorded for mephedrone, butylone, 4-Me-PPP, and 4-MEC were 16.05, 17.09, 17.31, and 16.24 ms, respectively. Butylone gave a smaller drift time than 4-Me-PPP even though it had a higher molecular mass. One possible reason for this may have been because it is more sterically hindered. The drift time for the five tryptamines, i.e., 5-EtO-DPT, 5-EtO-DALT, 5-EtO-MIPT, 5-EtO-ALCHT, and 5-EtO-2MALET, were 20.17, 19.73, 19.03, 21.68, and 19.93 ms, respectively. Table 2 provides a summary of all the K_o values for the cathinones and tryptamines studied in this investigation for the first time. Figure 2 shows representative spectra obtained from some single component cathinones and tryptamines studied. One special feature of the spectra obtained with an APIM(tof)MS is that they exhibited a mass−mobility correlation for classes of ions.[42] Such features are usually displayed on a 2-D plot, and these same correlations were demonstrated in the spectra reported in this study. In Figure 2a, for example, the spectrum of 4-Me-PPP shows the drift time in millisecond(s) obtained

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from the ion mobility part of the instrument and the mass/charge (m/z) ratio obtained from the mass spectrometry part of the instrument. Using a 2-D plot, data from both instruments can be displayed. The plot shows two ions detected for 4-Me-PPP: the $[M + {}^{1}H]^{+}$ ion at 218.10 Da and the $[M + 2]^{+}$ ion at 219.13 Da. The major ion was the $[M + {}^{1}H]^{+}$ ion with a much greater intensity than that of the $[M + 2]^{+}$ ion. All +2 ions were accounted for by the presence of a ^{13}C isotope. Similar interpretations can be deduced for the spectra in Figure 2b,c.

The detection limit for each of the psychoactive cathinones and tryptamine was estimated by injecting a 5 μM concentration for each compound. This was the concentration that was discernible from the instrument noise for all analytes. Table 3 summarizes the data for the detection limits estimated

Table 3. Detection Limits for Psychoactive Cathinones and Tryptamines

compound	intensity	detection limit (μM)	RSD (%)	detectable mass (ng)
mephedrone	1.43	8.24 ± 0.03	2.04	2.20
butylone	1.34	5.28 ± 0.02	1.31	1.75
4-Me-PPP	1.88	3.15 ± 0.01	0.78	1.03
4-MEC	1.23	7.06 ± 0.02	1.75	2.03
5-EtO-DPT	1.46	6.20 ± 0.02	1.54	2.68
5-EtO-DALT	0.93	11.11 ± 0.03	2.76	4.74
5-EtO-MIPT	1.96	2.91 ± 0.01	0.72	1.14
5-EtO-ALCHT	1.95	3.88 ± 0.02	0.96	1.90
5-EtO-2MALET	1.46	7.11 ± 0.03	1.76	3.06
caffeine	2.02	4.72 ± 0.02	1.17	1.38

using eqs 2 and 3. The lowest detection limit was 2.91 μM recorded for 5-EtO-MIPT, and the highest detection limit was recorded as 11.11 μM for 5-EtO-ALCHT. The micromolar detection limits were converted to mass using the ESI flow rate (3 μL min^{-1}) and the sample run time of 30 s. Using the molecular mass for each material, the masses required for better detection were calculated. The lowest detectable mass was 1.03 ng recorded for 4-Me-PPP, and the highest detectable mass of 4.74 ng was recorded for 5-EtO-DALT. The detectable masses for the other substances are summarized in Table 3.

ESI-APIM(tof)MS Responses for Individual Components and Mixtures. The background spectrum of the ESI solvent revealed major m/z peaks at 60.03, 73.66, 80.06, 83.64, and 99.38 Da. The background m/z ions were identified as $HC_2H_3O_2^{+}$ (~60 Da), $(H_2O)_4H^{+}$ (~73 Da), $H^{12}C^{13}CH_3O_2(H_2O)H^{+}$ (~80 Da), $(^{13}C^{12}CH_3O_2Na)^{+}$ or $(CH_3OH)_2(H_2O)H^{+}$ (~83 Da), and $^{2}H^{12}C^{13}CH_3O_2(H_2O)_2H^{+}$ (~99 Da). All four tryptamines and five cathinones produced a proton parent ion, $[M + H]^{+}$, and functional group collision induced dissociation (CID) fragments upon transitioning the APIM(tof)MS pressure interface. Most CID fragments resulting from pressure interface differences will normally have the same mobility because of the pressure differences between the pressure interface (~2.1 Torr) and the vacuum of the (tof)MS. This same observation was reported in previous studies with similar instrumentation.[62] For example, 5-EtO-MIPT produced a parent ion at $[5\text{-EtO-MIPT} + H]^{+}$ corresponding to 261 Da and a CID fragment $[5\text{-EtO-MIPT}-C_4H_{11}N]^{+}$ corresponding to 188 Da, possibly representing the $[5\text{-EtO-3-vinylindole}]^{+}$-type species.[41] Both ions showed different mass flight times but identical mobility times (26.97 ms and 26.94 ms, respectively) in the spectra. Likewise, 5-EtO-DALT produced a parent ion at $[5\text{-EtO-DALT} + H]^{+}$ corresponding to 285 Da and two CID fragments $[5\text{-EtO-DALT}-C_6H_{10}N]^{+}$ and $[5\text{-EtO-DALT}-C_8H_{14}N]^{+}$ corresponding to 188 and 160 Da, respectively. The $C_6H_{10}N$ corresponds to one $N(CH_2)_2(CH)_2(CH_2)_2$ group (96 Da), attached as a side chain to the structure of 5-EtO-DALT. Similarly, the $C_8H_{14}N$ corresponds to one $N\text{-}(CH_2)_4(CH)_2(CH2)_2$ group (124 Da), attached as a side

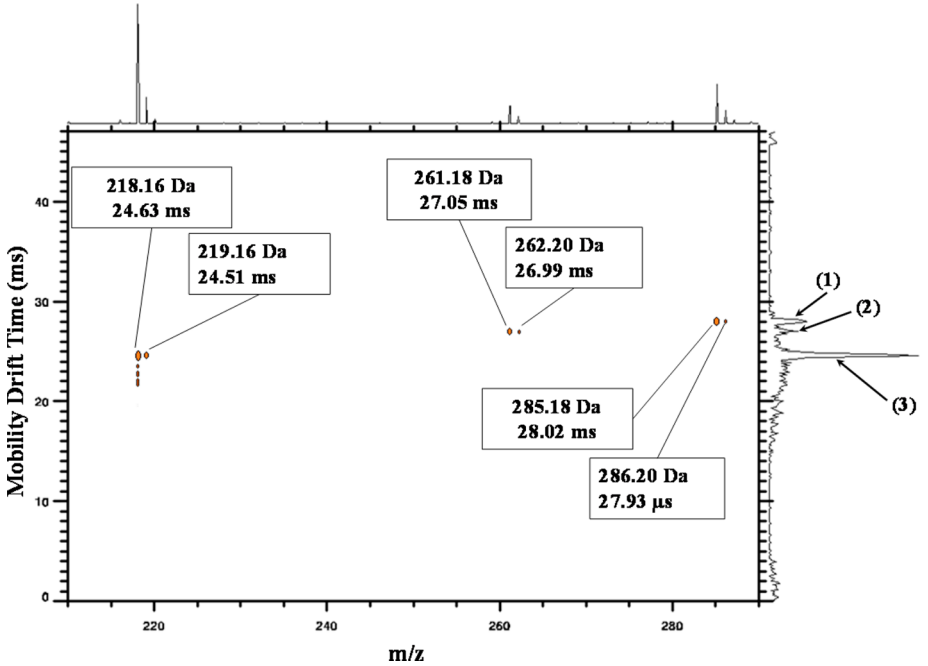

Figure 3. APIM(tof)MS of a 50 μM liquid phase mixture for 4-Me-PPP (3), 5-EtO-DALT (1), and 5-EtO-MIPT (2). The ions detected were the following: $[5\text{-EtO-DALT} + {}^{1}H]^{+}$ (285 Da); $[5\text{-EtO-DALT} + 2]^{+}$ (286 Da); $[5\text{-EtO-MIPT} + {}^{1}H]^{+}$ (261 Da); $[5\text{-EtO-MIPT} + 2]^{+}$ (262 Da); $[4\text{-Me-PPP} + {}^{1}H]^{+}$ (218 Da); $[4\text{-Me-PPP} + 2]^{+}$ (219 Da). The +2 ions were accounted for by the presence of a ^{13}C isotope.

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chain to the structure of 5-EtO-DALT. These three ions only differed by their respective mass flight times (drift times = 27.90, 27.86, and 27.90 ms, respectively). This trend was observed for all cathinones and tryptamines studied in this investigation. All spectra also showed a $[M + 2]^+$ ion which can be explained by the formation of a $[M + 2]^+$ ion having a ^{13}C isotope. Figure 3 is a spectrum showing the mixture of 4-Me-PPP, 5-EtO-DALT, and 5-EtO-MIPT. Mass flight times and K_o values similar to those observed with the single component compounds were detected. The resulting K_o values for all psychoactive cathinones and tryptamines studied are to our knowledge the first ever published results for these compounds. Moreover, the fact that the caffeine standard produced a K_o value compared to literature values reported so far helped to not only validate the use of APIM(tof)MS as an instrument for the accurate detection and identification of psychoactive cathinones and tryptamines but also to provide us with an insight as to how these compounds behave as they move through an APIM drift space.

CONCLUSIONS

The instrument used in this investigation offers a unique feature over other approaches of analysis. An APIM(tof)MS instrument produces a mass−mobility spectrum correlating classes of ions. This spectrum is a rapid 2-D data acquisition display with the capability of electronically coupling and decoupling CID fragment ions facilitating the identification of mobility selected parent ions. In this study, the employment of APIM(tof)MS in the positive ion mode has shown the detection capability of this instrument for analyzing psychoactive cathinones and tryptamines. This study clearly and conclusively determined the parent ions, CID fragment ions, K_o values, mass flight times, and detection limits produced from within a single experimental run using a single instrument platform for psychoactive cathinones and tryptamines. This study demonstrates that IMS can be used to detect and identify a new class of drugs referred to as "designer drugs", "bath salts", or "legal highs" that are increasingly distributed over the Internet. The use of APIM(tof)MS to detect these substances will undoubtedly increase the reliability of the identification and decrease the possibility of false positive responses. While this study was limited to the detection of psychoactive cathinones and tryptamines, because APIM(tof)MS detects both positive and negative ions, the analytical approach demonstrated in this study is expected to be applicable to other classes of drugs.

AUTHOR INFORMATION

Corresponding Author

*Tel.: 336-750-3199. Fax: 336-750-2549. E-mail: kanuabb@wssu.edu, abu_kanu_01@yahoo.co.uk.

Notes

The authors declare no competing financial interest.

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