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Identification of pyrolysis products of the new psychoactive substance 2-amino-1-(4-bromo-2,5-dimethoxyphenyl)ethan-1-one (bk-2C-B) and its iodo analog bk-2C-I

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Abstract:	2-Amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrochloride (bk-2C-B) has recently emerged as a new psychoactive substance (NPS). It is most commonly consumed orally although there are indications that it might also be ingested by inhalation or 'smoking'. Information about the stability of bk-2C-B when exposed to heat is unavailable and the potential for pyrolytic degradation and formation of unknown substances available for inhalation prompted an investigation using a simulated 'meth pipe' scenario. Twelve products following pyrolysis of bk-2C-B were detected and verified by organic synthesis of the corresponding standards. In addition, 2-amino-1-(4-iodo-2,5-dimethoxyphenyl)ethanone hydrochloride (bk-2C-I) has been characterized for the first time and subjected to pyrolysis as well. Similar products were formed, which indicated that the replacement of the bromo with the iodo substituent did not affect the pyrolysis pattern under the conditions used. Two additional products were detected in the bk-2C-I pyrolates, namely 1-(2,5-dimethoxyphenyl)-ethanone and 1-iodo-4-ethenyl-5-methoxyphenol. The potential ingestion of pyrolysis products with unknown toxicity adds an element of concern.

Identification of pyrolysis products of the new psychoactive substance 2-amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrochloride (*bk*-2C-B) and its iodo analog *bk*-2C-I

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ABSTRACT

2-Amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrochloride (*bk*-2C-B) has recently emerged as a new psychoactive substance (NPS). It is most commonly consumed orally although there are indications that it might also be ingested by inhalation or 'smoking'. Information about the stability of *bk*-2C-B when exposed to heat is unavailable and the potential for pyrolytic degradation and formation of unknown substances available for inhalation prompted an investigation using a simulated 'meth pipe' scenario. Twelve products following pyrolysis of *bk*-2C-B were detected and verified by organic synthesis of the corresponding standards. In addition, 2-amino-1-(4-iodo-2,5-dimethoxyphenyl)ethanone hydrochloride (*bk*-2C-I) has been characterized for the first time and subjected to pyrolysis as well. Similar products were formed, which indicated that the replacement of the bromo with the iodo substituent did not affect the pyrolysis pattern under the conditions used. Two additional products were detected in the *bk*-2C-I pyrolates, namely 1-(2,5-dimethoxyphenyl)-ethanone and 1-iodo-4-ethenyl-5-methoxyphenol. The potential ingestion of pyrolysis products with unknown toxicity adds an element of concern.

INTRODUCTION

Many ring-substituted phenethylamines show psychoactive and psychedelic effects in humans[1] and these are based on the now considered prototypical set of substances published in *PIHKAL*.[2] The extent of their control varies across the globe and depends on the nature of legislative control mechanisms that are in place. A number of phenethylamines not specifically controlled internationally or those exemplifying a development beyond the traditional *PHKAL* compounds[3] are sometimes captured under the term new psychoactive substances (NPS).[4] In the last decade, the number of unregulated psychoactive substances available for purchase *via* the Internet and high street shops has grown significantly, which created challenges to policy makers, health care professionals, law enforcement and researchers across disciplines.[5-7]

One of the phenethylamine analogs emerging on the NPS market in more recent years was 2-amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrochloride (*bk*-2C-B) (Figure 1), the *beta*-keto analog of the popular psychedelic 2-(4-bromo-2,5-dimethoxyphenyl)ethan-1-amine (2C-B). The detection of *bk*-2C-B on the European market was first reported to the European Monitoring Centre for Drugs and Drug Addiction (EMCDDA) in December 2013[8] and it has since been detected in other European countries including e.g. Italy.[9] The synthesis of *bk*-2C-B was first published in 2004 where it served as an intermediate for the preparation of 2-(4-bromo-2,5-dimethoxyphenyl)morpholine.[10] The synthesis and extensive analytical characterization of *bk*-2C-B, verifying the identification of this substance obtained from a test purchase from an Internet vendor, was reported more recently. Furthermore, it was reported that *bk*-2C-B was

prone to GC-induced degradation when analyzed without derivatization.[11] Another report complementing the analytical data related to *bk*-2C-B is also available.[12]

Recreational use of *bk*-2C-X has been reported to be closer to MDMA than those of the analogous 2C-X. Users report that the effects begin within 15-20 minutes and increase over 60-90 minutes. The general consensus is that *bk*-2C-B lasts about 10 hours, noticeably longer than 2C-B. Negative effects of snorting include severe burning and purple mucus. Gastrointestinal effects including severe diarrhea are also common.[13] The few user reports on the forums indicate that *bk*-2C-I also lasts longer, but does not produce as intense a high as *bk*-2C-I. Concerns were expressed about the toxicity of iodine.

Drugs may be ingested by various routes of administration including 'smoking.' The most common route of administration associated with ring-substituted phenethylamines is the oral route and this appears to apply to *bk*-2C-B as well although there is some indication however that this substance may also be smoked. Mixing *bk*-2C-B with marijuana, either as a solid dose or smoked, has been reported by some users to intensify the effects of both drugs.[14]

A key concern associated with the conditions encountered during pyrolysis is the formation of degradation products that may be formed during smoking and knowledge about the behavior of drugs under these harsh conditions provide important information on potential biomarkers and toxicity.[15] In the present study, *bk*-2C-B was subjected to pyrolysis conditions for analysis of condensed residues formed after vaporization. Furthermore, the analytical characterization and pyrolysis of 2-amino-1-(4-iodo-2,5-dimethoxyphenyl)ethanone hydrochloride(*bk*-2C-I) (Figure 1), a substance that has not yet been described in the literature, has also been included in this investigation.

EXPERIMENTAL

Materials

A sample of *bk*-2C-B as the hydrogen chloride salt was obtained as a test purchase from an internet vendor and 1-(4-Bromo-2,5-dimethoxyphenyl)-2-bromoethanone **(11)** and 1-(4-bromo-2,5-dimethoxyphenyl)ethanone **(6)** were prepared as reported previously.[11] 1-Bromo-2,5-dimethoxy-4-methylbenzene **(1a)**, 1,4-dibromo-2,5-dimethoxybenzene **(5)**, and all reagents were obtained from Sigma-Aldrich (Arklow, Ireland). A sample of *bk*-2C-I has been donated by an Internet retailer in 2015.

Syntheses

1-(4-Bromo-2,5-dimethoxyphenyl)-2-chloroethanone (**9**)

This was prepared via the sulfuryl chloride method previously reported for 2-chloro-1-(4-methylphenyl)propanone[16] to afford yellow crystals with a 36% yield: m.pt. 116-118 ^{o}C; ^{1}H NMR ($CDCl_3$) δ 7.28 (s, 1H, Ar-H), 7.26 (s, 1H, Ar-H), 4.79 (s, 2H, CH_2), 3.95 (s, 3H, OMe), 3.92 (s, 3H, OMe); ^{13}C NMR ($CDCl_3$) δ 190.99 (C=O), 153.23 (Ar-C), 150.57 (Ar-C), 123.96 (Ar-C), 119.04 (Ar-C), 117.44 (Ar-CH), 113.20 (Ar-CH), 56.79 (OMe), 56.47 (OMe), 51.03 (CH_2); HR-ESIMS found *m/z* 292.9267 (theory $[M+H]^+$ $C_{10}H_{11}{}^{79}Br^{35}ClO_3$, *m/z* 292.9575).

5-(4-Bromo-2,5-dimethoxyphenyl)oxazole (**10**)

A mixture of 2-amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrobromide (177 mg, 0.5 mmol), *p*-toluenesulfonic acid (5 mg) and triethyl orthoformate (2 mL) was heated at 140 ^{o}C for 3 h. The volatiles were removed under vacuum and the residue was purified by preparative TLC (silica gel; cyclohexane/EtOAc, 1/1) to afford a light brown solid (22 mg, 0.08 mmol, 16%): ^{1}H NMR ($CDCl_3$) δ 9.06 (s, 1H, O-CH=N), 7.63 (s, 1H, C=CH-N), 7.21 (s, 1H, Ar-H), 7.20 (s, 1H, Ar-H), 3.95 (s, 3H, OMe), 3.93 (s, 3H, OMe); ^{13}C NMR ($CDCl_3$) δ 156.28 (O-CH=N), 150.23 (Ar-C), 149.96 (Ar-C), 138.49 (Ar-C-O), 117.85 (C=CH-N), 116.51 (Ar-CH), 116.22 (Ar-CH), 108.90 (Ar-C), 106.95 (Ar-C), 56.90 (OMe), 56.17 (OMe). HR-ESIMS found *m/z* 283.9926 (theory $[M+H]^+$ $C_{11}H_{11}{}^{79}BrNO_3$, *m/z* 283.9917).

1-Bromo-4-ethyl-2,5-dimethoxybenzene (**2**)

Triethylsilane (351 µL, 2.2 mmol) was added to a solution of 1-(4-bromo-2,5-dimethoxyphenyl)ethanone (259 mg, 1 mmol) in trifluoroacetic acid (2 mL) and the mixture was stirred at room temperature for 1 h. The volatiles were removed under vacuum and the residue was purified by preparative TLC (silica gel; cyclohexane/EtOAc, 7/3) to afford a colorless solid (61 mg, 0.25 mmol, 25%): m.pt. 34-36^{o}C; ^{1}H NMR ($CDCl_3$) δ 7.03 (s, 1H, Ar-H), 6.78 (s, 1H, Ar-H), 3.87 (s, 3H, OMe), 3.80 (s, 3H, OMe), 2.62 (q, *J* = 7.5 Hz, 2H, CH_2), 1.20 (t, *J* = 7.5 Hz, 3H, CH_3); ^{13}C NMR ($CDCl_3$) δ 151.69 (Ar-C), 149.80 (Ar-C), 132.88 (Ar-C), 115.55 (Ar-CH), 113.63 (Ar-CH), 108.02 (Ar-C), 56.86 (OMe), 55.96 (OMe), 23.25 (CH_2), 14.00 (CH_3). For this compound, sufficient ionization under the employed HR-ESI-MS conditions was not observed for high accuracy mass analysis.

1-Bromo-4-(2-bromoethyl)-2,5-dimethoxybenzene (**8**)

A mixture of 2-(4-bromo-2,5-dimethoxyphenyl)ethanol (130 mg, 0.5 mmol, prepared by the reduction of 2-(4-bromo-2,5-dimethoxyphenyl)acetic acid with borane-THF) and thionyl bromide (2 mL) was refluxed for 2 h. The volatiles were removed under vacuum and the residue was recrystallized from cyclohexane/ethyl acetate to afford a beige solid (126 mg, 0.39 mmol, 78%): m.pt. 66-68 ^{o}C; ^{1}H NMR ($CDCl_3$) δ 7.06 (s, 1H, Ar-H), 6.78 (s, 1H, Ar-H), 3.88 (s, 3H, OMe), 3.81 (s, 3H, OMe), 3.57 (t, *J* = 7.5 Hz, 2H, CH_2), 3.14 (t, *J* = 7.5 Hz, 2H, CH_2); ^{13}C NMR ($CDCl_3$) δ 151.72 (Ar-C), 149.69 (Ar-C), 127.02 (Ar-C), 115.71 (Ar-CH), 115.01 (Ar-CH),

109.83 (Ar-C), 56.85 (OMe), 55.86 (OMe), 34.39 (CH_2), 31.51 (CH_2). For this compound, sufficient ionization under the employed HR-ESI-MS conditions was not observed for high accuracy mass analysis.

1-Bromo-4-(2-chloroethyl)-2,5-dimethoxybenzene (**7**)

A mixture of 2-(4-bromo-2,5-dimethoxyphenyl)ethanol (130 mg, 0.5 mmol, prepared by the reduction of 2-(4-bromo-2,5-dimethoxyphenyl)acetic acid with borane-THF) and thionyl chloride (2 mL) was refluxed for 2 h. The volatiles were removed under vacuum and the residue was recrystallized from cyclohexane/ethyl acetate to afford colorless solid (89 mg, 0.32 mmol, 64%): m.pt. 52-54°C; ^{1}H NMR ($CDCl_3$) δ 7.06 (s, 1H, Ar-H), 6.79 (s, 1H, Ar-H), 3.88 (s, 3H, OMe), 3.81 (s, 3H, OMe), 3.72 (t, J = 7.5 Hz, 2H, CH_2), 3.06 (t, J = 7.5 Hz, 2H, CH_2); ^{13}C NMR ($CDCl_3$) δ 151.83 (Ar-C), 149.97 (Ar-C), 124.22 (Ar-C), 115.86 (Ar-CH), 115.31 (Ar-CH), 110.26 (Ar-C), 57.01 (OMe), 56.04 (OMe), 43.46 (CH_2), 30.33 (CH_2). For this compound, sufficient ionization under the employed HR-ESI-MS conditions was not observed for high accuracy mass analysis.

1-Bromo-2,5-dimethoxy-4-vinylbenzene (**3**)

Sodium hydride (180 mg, 7.5 mmol) was added to methyltriphenylphosphonium bromide (1.34, 3.75 mmol) in diethyl ether (25 mL) at 0 °C and the mixture was stirred for 1 h. 4-Bromo-2,5-dimethoxybenzaldehyde (612 mg, 2.5 mmol) in diethyl ether (10 mL) was then added and the mixture was stirred for 3 h at room temperature. Saturated aqueous sodium carbonate was then added, the organic layer collected, dried (anhydrous magnesium sulfate), evaporated to dryness and purified by preparative TLC (silica gel; cyclohexane/EtOAc, 7/3) to afford a colorless solid (178 mg, 0.73 mmol, 29%): m.pt. 50-52 °C; ^{1}H NMR ($CDCl_3$) δ 7.09 (s, 1H, Ar-H), 7.04 (s, 1H, Ar-H), 6.99 (dd, J = 17.7, 11.2 Hz, 1H, =CH) 5.76 (dd, J = 17.7, 1.1 Hz, 1H, one H from =CH_2), 5.33 (dd, J = 11.2, 1.1 Hz, 1H, one H from =CH_2), 3.90 (s, 3H, OMe) and 3.82 (s, 3H, OMe); ^{13}C NMR ($CDCl_3$) δ 151.11 (Ar-C), 150.04 (Ar-C), 130.86 (=CH), 126.53 (Ar-C), 116.49 (Ar-CH), 114.83 (Ar-CH), 110.99 (Ar-C), 110.03 (=CH_2), 56.75 (OMe) and 55.23 (OMe); HR-ESIMS found m/z 243.0020 (theory $[M+H]^+$ $C_{10}H_{12}{}^{79}BrO_2$, m/z 243.0015).

Instrumentation

All samples were run on an Agilent Technologies gas chromatograph-mass spectrometer (6890 N GC and 5975 inert mass selective detector) fitted with a DB-5 MS column (30 m x 0.25 mm, 0.25 μm film thickness) with the following parameters: injector port, 260 °C, transfer line, 250 °C; EI, 70 eV; TIC mode, m/z 35-550; ionization source, 230 °C; quadrupole, 150 °C. Helium was used as the carrier gas (1.1 mL/min). The temperature program started at 100 °C with a ramp rate of 15 °C per minute up to 300 °C followed by a 34.7 min hold for a total run time of 48 min. The solvent delay was set for 4 min.

NMR spectra ^{1}H (600 MHz) and ^{13}C (150 MHz) were recorded on a Bruker AV600 NMR spectrometer using a 5 mm TCI cryoprobe.

HR-ESI mass spectra were recorded by direct injection into a LTQ Orbitrap Discovery (Thermo Fisher, UK). Samples were dissolved in acetonitrile/water (1:1, containing 0.1% formic acid) and infused at a rate of 5 µl/min. Full accurate high-resolution (30000) mass scans were performed in positive electrospray mode. Measured accurate masses were within ± 5ppm of the theoretical masses. The following conditions were used: drying gas (N_2) 10L/min, capillary temperature 310 ^{o}C, spray voltage 4 V, capillary voltage 22 V and tube lens 77 V.

Pyrolysis

A sample of the drug (5-9 mg) was loaded into an aluminum foil boat, placed in the bottom of a 20 mL crimp vial and heated with a disposable lighter for approximately 30 s until yellow vapors formed in the vial and a brown tar had formed in the boat. The foil cup was removed and the flask was rinsed with 1 mL of acetonitrile to dissolve the residues for GC-MS analysis. Acetylation was performed using 100 µL of acetic anhydride and heating at 70 °C for 3 h. The vial was cooled to room temperature and the contents analyzed by GC-MS. For TMS derivatization, the residue was taken up in *N*,*O*-bis(trimethylsilyl)trifluoroacetamide (BSTFA)/toluene (1/1) and analyzed by GC-MS. Ethylation was performed by dissolving the residue in acetone (200 µL). Iodoethane (50 µL) and anhydrous potassium carbonate (approx. 20 mg) were added and the mixture was heated at 90 ^{o}C for 20 min. The mixture was allowed to cool to room temperature, evaporated to dryness and the residue was reconstituted in acetonitrile (200 µL) for GC-MS analysis. For NMR studies, 100 mg of *bk*-2C-B was pyrolyzed in a similar manner, using a 500 mL round bottom flask. The pyrolysate was dissolved in deuterated chloroform.

RESULTS AND DISCUSSION

The *bk*-2C-B and *bk*-2C-I were both obtained from online vendors. The identity of the *bk*-2C-B was confirmed in a previous study.[11] While analysis by LC-MS and NMR indicated the sample was not contaminated with by-products or adulterants, 1-(4-bromo-2,5-dimethoxyphenyl)ethanone and a pyrazine dimer were detected when the sample was run on GC-MS without derivatization. A potential reason for this formation might have included a catalytic conversion in the injection port liner due to the presence of active sites, which suggests that the extent of product formation might potentially vary across GC systems. In addition, it was determined that the purchased *bk*-2C-B was composed of mixture of chloride and bromide salts.

The GC-MS of the *bk*-2C-B and *bk*-2C-I are shown in Figure 2. In addition to 1-(4-bromo-2,5-dimethoxyphenyl)ethanone and the pyrazine dimer reported by Power et al.[11], 5-(4-bromo-2,5-dimethoxyphenyl)oxazole was formed upon injection of the *bk*-2C-B on the GC. Whereas both

1-(4-iodo-2,5-dimethoxyphenyl)ethanone and 5-(4-iodo-2,5-dimethoxyphenyl)oxazole were present in the chromatogram of the *bk*-2C-I, the iodo dimer was not detected.

Pyrolysis products of *bk*-2C-B products were produced under conditions comparable to those employed by drug users using a 'meth pipe'.[15] The sample was heated with a butane lighter until yellow fumes appeared and an orange residue was deposited on the sides of the glass vial. The pyrolysis was repeated several times in order to determine which product were representative of those inhaled under the conditions used in drug abuse. A final pyrolysis was run with 9 mg in order to obtain sufficient product for the interpretation of the mass spectra. The substances reported here appeared in multiple pyrolysis runs.

Twelve pyrolysis products, shown in Figure 3, were identified through interpretation of the mass spectra and confirmed by comparison to synthesized standards. The chromatograms of the pyrolates, standards and mass spectra for products (**1a)**-(**5)** are shown in Figure 4. Interpretation of the mass spectra for products (**6**)-(**12**) are shown in Figure 5 was aided by the characteristic isotope patterns of bromine and chlorine (Supporting Information).

The stability of the enol radical resulted in a distribution of products from homolytic reactions. Most of the pyrolysis products underwent homolytic cleavage of the C-N, although some C-C bond cleavage was observed. The mechanism for the hemolytic cleavage is shown in Figure 6. The stereochemistry of the halogen addition also supports a hemolysis. Products (**1a**)**,** (**1b**)**,** (**2**)**,** (**3**)**,** (**5**)**,** (**7**) and (**8**) also lost the *beta* ketone. The first peak detected at 6.18 min revealed co-elution of two pyrolates. One, 1-bromo-2,5-dimethoxy-4-methylbenzene (**1a**), was an example of *beta* C-O bond cleavage. The second was consistent with one or both of the phenols, 3-bromo-4-methoxyphenol and 2-bromo-4-methoxyphenol (**1b**), which illustrated activity at the benzene ring, resulting in both C-C cleavage and demethylation of the methoxy group. In comparison, the C-C cleavage was not observed in the pyrolysis of mephdrone.[17] The phenols were confirmed by derivatization with acetic anhydride where a gain in mass of 42 amu resulted in a retention time shift to 6.75 min. The mass spectrum of the derivatized phenol is shown in Figure 5.

The formation of 1-bromo-4-ethyl-2,5-dimethoxy-benzene (**2**) was an example of deamination, as well as loss of the loss of oxygen in the ketone. The mechanism might have been similar to the formation of iso-mephedrone observed during mephedrone pyrolysis, with the formation of a cyclic amine intermediate that would result in the formation of an iso-2C-B intermediate, followed by deamination.[16] The peak eluting at 7.01 min was confirmed as 1-bromo-4-ethenyl-2,5-dimethoxy-benzene (**3**)

The fragmentation pattern representing the peak at 7.38 min was initially thought to represent 4-bromo-2,5-dimethoxy-benzaldehyde but this was not confirmed when compared to a standard. Instead, an alternative structure was proposed, 1-(4-bromo-2-hydroxy-5-

methoxyphenyl)ethanone (**4**), which was confirmed by comparison with a standard and by NMR provided in the Supplemental Information. Other pyrolysis products that retained the ketone group included pyrolates (**6**), (**9**), and (**11**). 1-(4-Bromo-2,5-dimethoxyphenyl)ethanone was formed by loss of the amine group, also seen in the pyrolysis of mephedrone.[16] In mephedrone, however, halogen substitution similar to 1-(4-bromo-2,5-dimethoxyphenyl)-2-chloro-ethanone (**9**) or 2-bromo-1-(4-bromo-2,5-dimethoxyphenyl)-ethanone (**11**) was not detected. In the later example, chlorination rather than bromination would occur in the case of mephedrone. Of the remaining products, (**5**), (**7**) and (**8**) were halogenated analogs of (**1a**), and (**2**). Two halogenated products were detected in the pyrolysis of mephedrone and identified as α-chloro ketones, 1-chloro-1-(4-methylphenyl)- 2-propanone and 2-chloro-1-(4-methylphenyl)-1-propanone[16].

The pyrolysis products of *bk*-2C-B differed significantly from those observed with other phenethylamines[15] including methiopropamine and the ring-substituted cathinone, mephedrone.[15,16] Methylation[18] and *N*,*O* rearrangement did not occur.[19] Product (**1b**) was unique in that the C-C bond cleavage appeared to occur at the benzene ring and the corresponding product was not detected in either the pyrolysis of methamphetamine or mephedrone.[16,18] The (**1b**) and (**4**) represented reactions at the benzene ring, presumably through the formation of an alcohol from the associated methoxy group. Products (**5**), (**7**-**9**), and (**11**) were the result of halogenation. Several products resulted from bond cleavage of either the α-β C-C bond (**1a**) or the C-N bond (**2**-**4**, **6**) and similar products were detected in the pyrolysis of both methamphetamine and methiopropamine.[17,18] Products (**1a**, **2**, and **3**) were analogous to the oxidative degradation products reported for methamphetamine.[18]

Only two products retained the amine nitrogen: 5-(4-bromo-2,5-dimethoxyphenyl)oxazole (**10**) and a dimer (**12**). These substances were present in the chromatograms of *bk*-2C-B as received and may be formed either during the pyrolysis or in the GC injector by a reaction of *bk*-2C-B as reported by Power et al.[11] A proposed mechanism for the fragmentation of product (**10**) is shown in Figure 7, with formaldehyde being formed in-situ from breakdown of the *bk*-2C-B.

Formation of the pyrazine dimer (**12**) may have formed through a condensation reaction of the β-ketone as seen in the report by Powers et al., although thermal rearrangements were also observed during mephedrone and methiopropamine pyrolysis.[16,17]

The difference in the products may have reflected the activating nature of the methoxy groups located on the benzene ring. Several products originating from oxidative degradation or bond cleavage showed a halogenated counterpart: compounds (**7**) and (**8**) were the chlorinated and brominated counterparts of (**2**). Products (**9** and **11**) were the halogenated counterparts of (**6**). The halogenated counterparts for the phenols (**1b** and **4**) were not detected.

While some level of toxicity is predicted for the α-chloro ketones,[20] no toxicity data are available for the substances formed during *bk*-2C-B pyrolysis. Products (**1a**, **2**, and **5**) have been

studied as fungicides.[21] The ethenyl compound (**3**) has been researched for activity as a 2-photon chromophore.[22] Products (**6** and **11**) were used as precursors in the preparation of serotonin 5-HT_{2A} receptor agonists[10] and (**1b**) was identified as a precursor for 1,2-cyclized phenylethylamines studied as agonists for both 5-HT_{2A} and 5-HT_{2B} receptors.[23] A literature structure search for product (**7**) gave no results, whereas the analogous bromoethyl product (**8**) appeared in a study of phenylethylamine analogs for use in treating glaucoma.[24]

As summarized in the Supporting Information, similar products were identified following the pyrolysis of *bk*-2C-I. The retention times increased as a reflection of increased mass of the iodine substituent. Products analogous to *bk*-2C-B (**1**-**4**), (**6**-**8**) and (**10**) were also formed during the pyrolysis of *bk*-2C-I. Two additional pyrolysis products were detected and tentatively identified through interpretation of their EI mass spectra. 1-(2,5-Dimethoxyphenyl)-ethanone (**13**) might have been formed by cleavage at the nitrogen and the decomposition product 1-iodo-4-ethenyl-5-methoxyphenol (**14**) might have been a decomposition product similar to 1-iodo-4-ethenyl-2,5-dimethoxy-benzene (**3**) and 1-(4-iodo-2-hydroxy-5-methoxyphenyl)-ethanone (**4**). The only *bk*-2C-I pyrolysis product mentioned in a biological study was the product (**4**) mentioned in a patent for P2X3 and P2X2/3 antagonists.[25]

Conclusion

Inhalation of a vaporized drug commonly produces a quick onset of effects but also shorter duration. However, the potential ingestion of pyrolysis products with unknown toxicity adds an element of concern. Twelve products resulting from the pyrolysis of *bk*-2C-B have been identified and similar chemicals were detected for *bk*-2C-I. The pyrolysis conditions resulted primarily in oxidative degradation and C-C or N-C bond cleavage, often accompanied by halogenation.

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References

[1] A.T. Shulgin, T. Manning, P.F. Daley, *The Shulgin Index - Volume One: Psychedelic Phenethylamines and Related Compounds*, Transform Press, Berkeley, CA, USA, **2011.**

[2] A. Shulgin, A.T. Shulgin, *Phenethylamines I Have Known and Loved: A Chemical Love Story*, Transform Press, Berkeley, CA, USA, **1991.**

[3] L.A. King. New phenethylamines in Europe. *Drug Test. Anal.* **2014**, *6*, 808.

[4] S.D. Brandt, L.A. King, M. Evans-Brown. The new drug phenomenon. *Drug Test. Anal.* **2014**, *6*, 587.

[5] United Nations Office on Drugs and Crime (UNODC). The challenge of new psychoactive substances. A Report from the Global SMART Programme March 2013. United Nations Publication, Vienna, 2013. Available at: https://www.unodc.org/documents/scientific/NPS_2013_SMART.pdf [18 September 2016].

[6] European Monitoring Centre for Drugs and Drug Addiction (EMCDDA). European Drug Report 2016: Trends and Developments. EMCDDA, Lisbon, **2016**. Available at: http://www.emcdda.europa.eu/system/files/publications/2637/TDAT16001ENN.pdf [09 September 2016].

[7] M. Evans-Brown, R. Sedefov. New psychoactive substances: driving greater complexity into the drug problem. *Addiction* **2017**, *112*, 36.

[8] European Monitoring Centre for Drugs and Drug Addiction (EMCDDA)-Europol. Europol 2013 Annual Report on the implementation of Council Decision 2005/387/JHA. EMCDDA, Lisbon, **2014**. Available at: http://www.emcdda.europa.eu/system/files/publications/814/TDAN14001ENN_475519.pdf [18 September 2016].

[9] S. Odoardi, F.S. Romolo, S. Strano-Rossi. A snapshot on NPS in Italy: distribution of drugs in seized materials analysed in an Italian forensic laboratory in the period 2013-2015. *Forensic Sci. Int.* **2016**, *265*, 116.

[10] R.A. Glennon, M.L. Bondarev, N. Khorana, R. Young. β-Oxygenated analogues of the 5-HT_{2A} serotonin receptor agonist 1-(4-bromo-2,5-dimethoxyphenyl)-2-aminopropane. *J. Med. Chem.* **2004**, *47*, 6034.

[11] J.D. Power, P. Kavanagh, J. O'Brien, M. Barry, B. Twamley, B. Talbot, G. Dowling, S.D. Brandt. Test purchase, identification and synthesis of 2-amino-1-(4-bromo-2, 5-dimethoxyphenyl)ethan-1-one (*bk*-2C-B). *Drug Test. Anal.* **2015**, *7*, 512.

[12] G. Frison, S. Odoardi, S. Frasson, R. Sciarrone, G. Ortar, F.S. Romolo, S. Strano Rossi. Characterization of the designer drug *bk*-2C-B (2-amino-1-(bromo-dimethoxyphenyl)ethan-1-one) by gas chromatography/mass spectrometry without and with derivatization with 2,2,2-

trichloroethyl chloroformate, liquid chromatography/high-resolution mass spectrometry, and nuclear magnetic resonance. *Rapid Commun. Mass Spectrom.* **2015**, *29*, 1196.

[13] Drugs-Forum *bk*-2C-B Drug Info. https://drugs-forum.com/forum/showthread.php?t=228051 [09 January 2017].

[14] http://www.rollitup.org/t/*bk*-2c-b-thread.824039/ [09 January 2017].

[15] S. Bell, C. Nida. Pyrolysis of drugs of abuse: a comprehensive review. *Drug Test. Anal.* **2015**, *7*, 445.

[16] P. Kavanagh, J. O'Brien, J.D. Power, B. Talbot, S.D. McDermott. 'Smoking' mephedrone: the identification of the pyrolysis products of 4-methylmethcathinone hydrochloride. *Drug Test. Anal.* **2013**, *5*, 291.

[17] E.D. Bouso, E.A. Gardner, J.E. O'Brien, B. Talbot, P.V. Kavanagh. Characterization of the pyrolysis products of methiopropamine. *Drug Test. Anal.* **2014**, *6*, 676.

[18] M. Sato, M. Hida, H. Nagase. Analysis of pyrolysis products of methamphetamine. *J. Anal. Toxicol.* **2004**, *28*, 638.

[19] C.L. Stevens, R.D. Elliott, B.L. Winch. Aminoketone rearrangements. II. The rearrangement of phenyl α-aminoketones. *J. Am. Chem. Soc.* **1963**, *85*, 1464.

[20] A.K. Nigam, M.V. Suryanarayana, P.K. Gutch, S.P. Sharma, L.N. Tomar, R. Vijayaraghavan. Thermal decomposition studies of riot control agent ω-chloroacetophenone (CN) by pyrolysis-gas chromatography-mass spectrometry. *J. Hazard Mater.* **2010**, *184*, 506.

[21] R.M. Scribner, E.J. Soboszenski. Bodenfungicid. DE1274398. E. I. du Pont de Nemours and Company, Wilimgton, Delaware, USA. **1968**.

[22] K. Kamada, Y. Iwase, K. Sakai, K. Kondo, K. Ohta. Cationic two-photon absorption chromophores with double-and triple-bond cores in symmetric/asymmetric arrangements. J. Phys. Chem. C **2009**, *113*, 11469.

[23] V. Isberg, J. Paine, S. Leth-Petersen, J.L. Kristensen, D.E. Gloriam. Structure-activity relationships of constrained phenylethylamine ligands for the serotonin 5-HT_2 receptors. *Plos One* **2013**, *8*, e78515.

[24] M. Hellberg, A. Namil, Z. Feng, J. Ward. Phenylethylamine analogs and their use for treating glaucoma. US 2007/0072920 A1. Alcon Research Ltd. Fort Worth, Texas, USA. **2007**.

[25] C. Broka, D. Carter, M. Dillon, R. Hawley, A. Jahangir, C. Lin, D. Parish. Diaminopyrimidines as P2X3 and P2X2/3 antagonists. US2005/0209260 A1. Roche Palo Alto LLC, Palo Alto, CA, USA. **2005**.

Figure Captions

Figure 1. Chemical structures of 2-(4-bromo-2,5-dimethoxyphenyl)ethan-1-amine hydrochloride (2C-B), 2-amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrochloride (*bk*-2C-B), and 2-amino-1-(4-iodo-2,5-dimethoxyphenyl)ethanone hydrochloride(*bk*-2C-I) respectively.

Figure 2. Chromatograms of a. *bk*-2C-B and b. *bk*-2C-I.

Figure 3. Schematic of *bk*-2C-B pyrolysis products.

Figure 4. (a) Total ion chromatogram for pyrolysis products of 2-amino-1-(4-bromo-2,5-dimethoxyphenethyl)ethanone chloride (*bk*-2C-B); (b) comparison with standards; and (c) mass spectra for products (**1a**)-(**5**).

Figure 5. Electron ionization mass spectra of the products (**6**)-(**12**) formed during pyrolysis of 2-amino-1- (4-bromo-2,5-dimethoxyphenethyl)ethanone hydrochloride (*bk*-2C-B).

Figure 6. Schematic of the hemolytic cleavage of *bk*-2C-Br.

Figure 7. Proposed mechanism for the fragmentation of product 10.

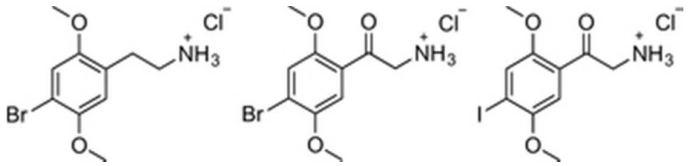

FIgure 1. Chemical structures of 2-(4-bromo-2,5-dimethoxyphenyl)ethan-1-amine hydrochloride (2C-B), 2-amino-1-(4-bromo-2,5-dimethoxyphenyl)ethanone hydrochloride (bk-2C-B), and 2-amino-1-(4-iodo-2,5-dimethoxyphenyl)ethanone hydrochloride(bk-2C-I) respectivelyChromatograms of a. bk-2C-B and b. bk-2C-I.

Figure 1
30x6mm (300 x 300 DPI)

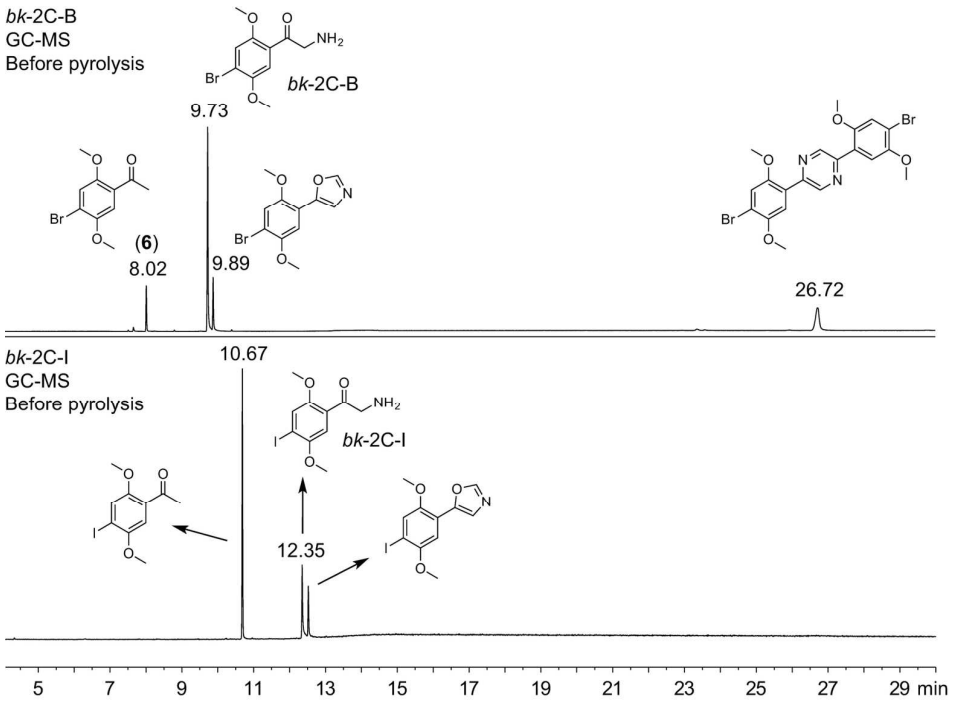

Figure 2. Chromatograms of a. bk-2C-B and b. bk-2C-I.
Figure 2
150x111mm (300 x 300 DPI)

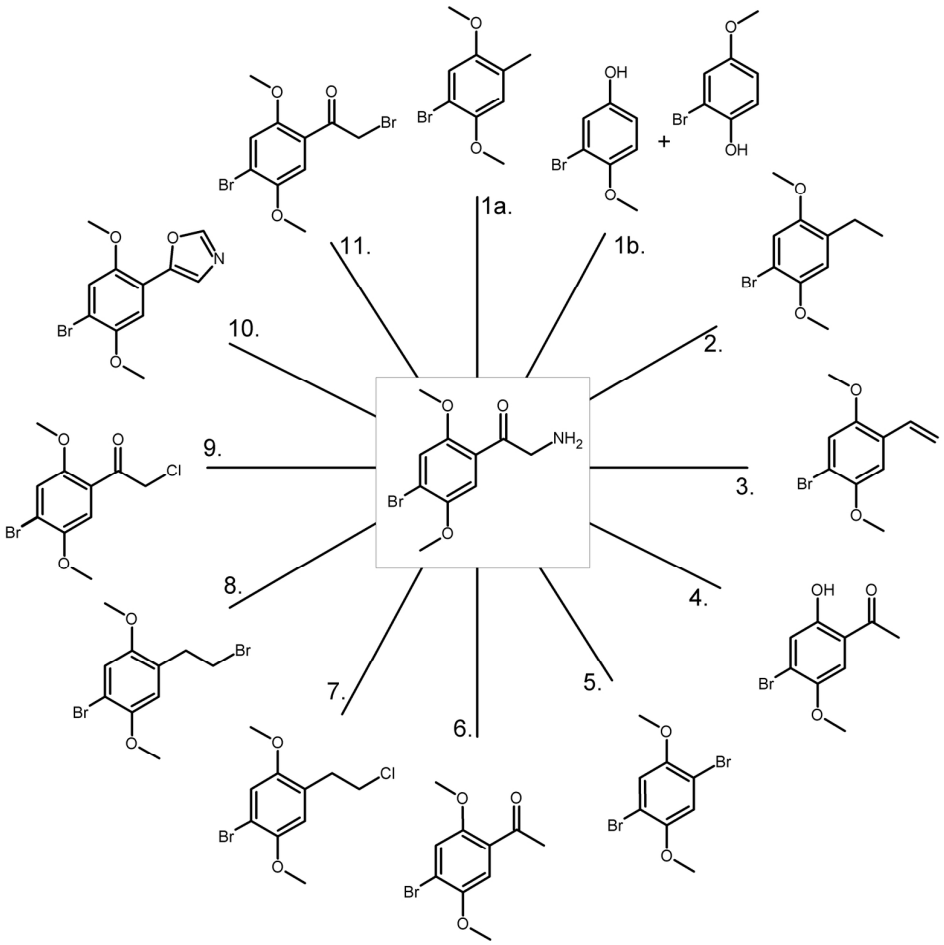

Figure 3. Schematic of bk-2C-B pyrolysis products.
Figure 3
166x164mm (300 x 300 DPI)

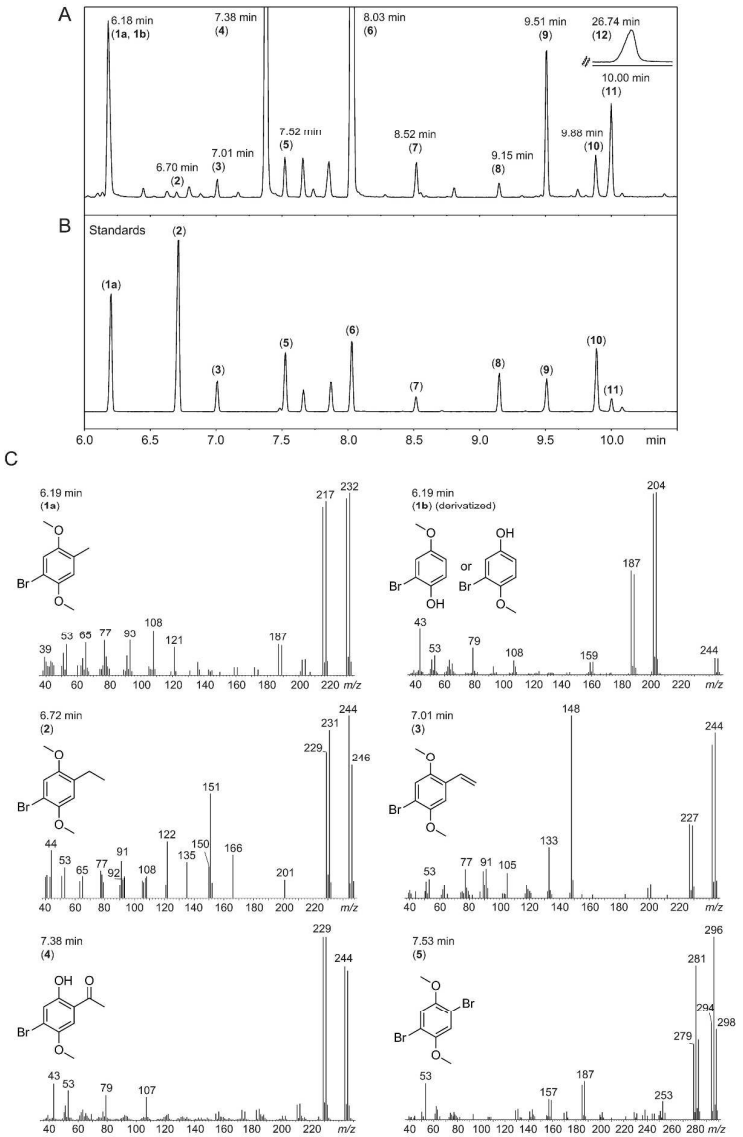

Figure 4. (a) Total ion chromatogram for pyrolysis products of 2-amino-1-(4-bromo-2,5-dimethoxyphenethyl)ethanone chloride (bk-2C-B); (b) comparison with standards; and (c) mass spectra for products (1a)-(5).

Figure 4
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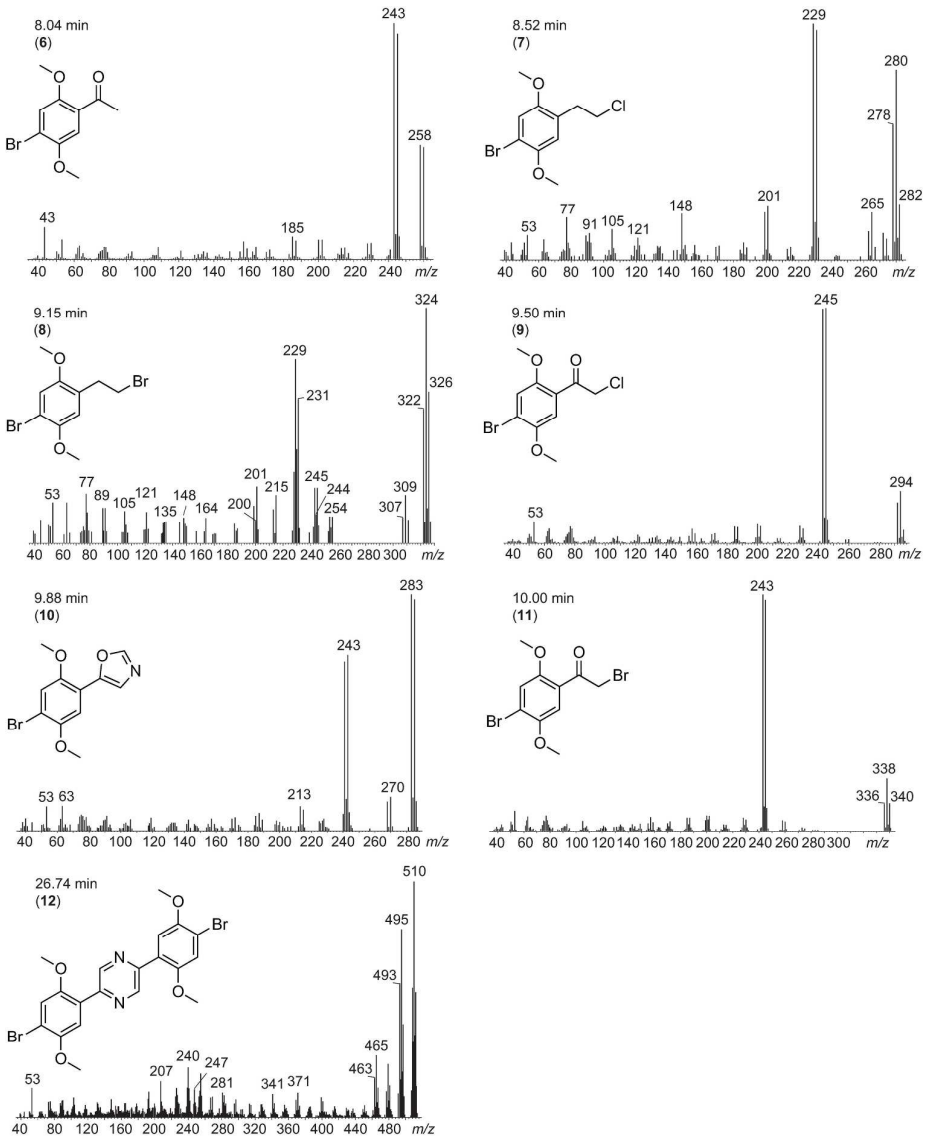

Figure 5. Electron ionization mass spectra of the products (6)-(12) formed during pyrolysis of 2-amino-1-(4-bromo-2,5-dimethoxyphenethyl)ethanone hydrochloride (bk-2C-B).

Figure 5
225x277mm (300 x 300 DPI)

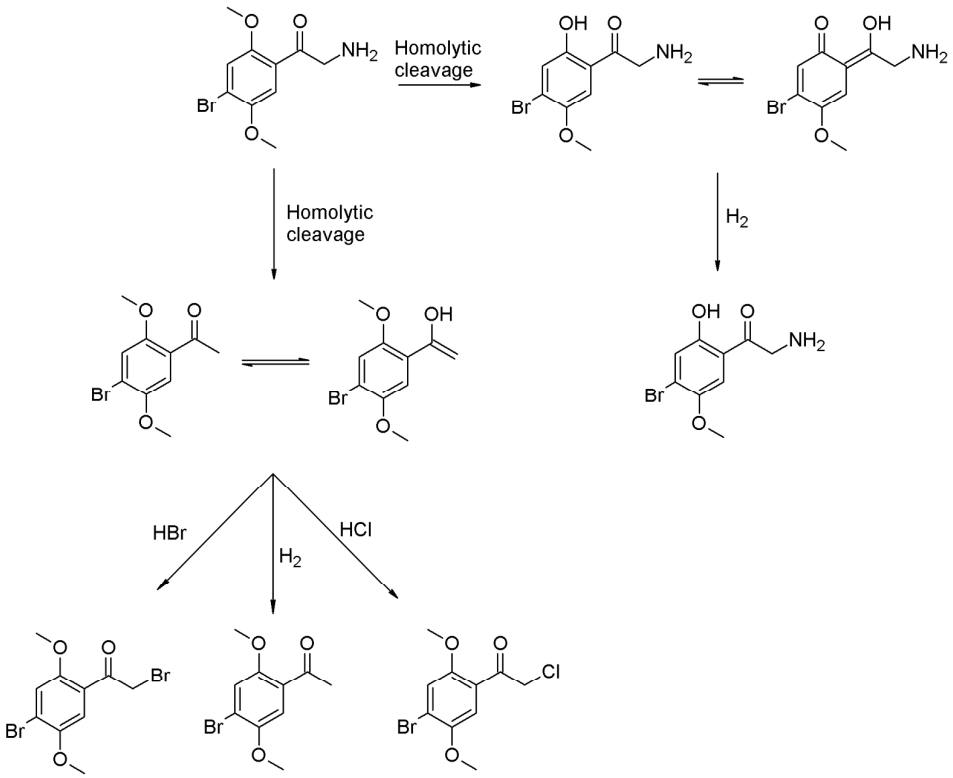

Figure 6. Schematic of the hemolytic cleavage of bk-2C-Br.

171x139mm (300 x 300 DPI)

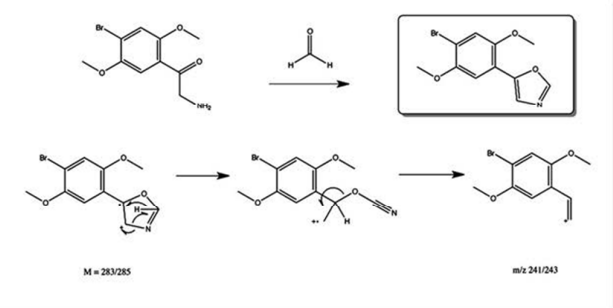

Figure 7. Proposed mechanism for the fragmentation of product 10.
Figure 7
254x190mm (96 x 96 DPI)