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ORIGINAL ARTICLE

Postmortem distribution of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in body fluids and solid tissues in a fatal poisoning case: usefulness of adipose tissue for detection of the drugs in unchanged forms

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Abstract We encountered an autopsy case in which the cause of death was judged as poisoning by multiple drugs, including AB-CHMINACA, 5-fluoro-AMB, and diphenidine. The deceased was a 30-year-old man. The postmortem interval to autopsy was estimated to be 3.5 days. Femoral vein blood, right heart blood, left heart blood, urine, and eight solid tissues including adipose tissue were collected and frozen until analysis. Extraction of the three drugs, and internal standards phencyclidine and 5-fluoro-AB-PINACA was performed by a modified QuEChERS method, followed by analysis by liquid chromatography–tandem mass spectrometry. Because this study dealt with various kinds of human matrices, we used the standard addition method to overcome the matrix effects. Quantitation of all three compounds was only achieved for the adipose tissue, whereas the levels of 5-fluoro-AMB were below the lower limit of quantitation (about 1 ng/g or ml) in all other samples. AB-CHMINACA was quantitated for all solid tissues, but not for all body fluid specimens. Diphenidine showed high concentrations in all specimens; it was highest in the adipose tissue (11,100 ± 1,120 ng/g), an order of magnitude lower in other solid tissue specimens, and two orders of magnitude lower in body fluid samples. The results suggest that adipose tissue is the best specimen for detection of lipophilic drugs, such as AB-CHMINACA, 5-fluoro-AMB, and diphenidine, in their unchanged forms. In this poisoning case, diphenidine appeared to have played the major role in the cause of death, because the concentrations of diphenidine were much higher than those of the synthetic cannabinoids in all specimens tested. To our knowledge, this is the first report to document the presence of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in actual postmortem specimens in a fatal poisoning case.

Keywords AB-CHMINACA · 5-Fluoro-AMB · Diphenidine · Synthetic cannabinoid · Adipose tissue · Postmortem distribution

Introduction

In recent years, synthetic cannabinoids [1–5] and cathinones [6–9] have become widely distributed and now cause social problems throughout many parts of the world. Most of the new emerging drugs have appeared as alternatives to controlled substances such as narcotics and designated substances. It is obvious that the types of designer drugs and their combinations in illegal products are increasing in diversity [5].

Recently, we encountered a fatal case of drug poisoning, in which newly emerging synthetic cannabinoids AB-CHMINACA and 5-fluoro-AMB were found together with diphenidine, a known *N*-methyl-D-aspartate (NMDA) receptor channel blocker. The three compounds were recently identified as designer drug components of illegal products [5, 10], although the present report is the first to document the presence of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in postmortem human specimens obtained in a fatal poisoning case.

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Case history

The deceased was 30-year-old man. He was found dead in his car, which was parked in a corner of a supermarket parking lot. He was lying in the prone position with his head on the front passenger seat and with his left leg on the driver's seat. He wore trousers, but was naked from the waist up. He held a disposable lighter in his right hand. Just below his chest, an opened package of herbal blend labeled as "Herbal Incense. The Super Lemon" was found. A battered empty coffee can was found on the driver's seat; on the surface of the flattened can, fire debris and herb ash were observed, suggesting that he inhaled the smoke of the herbal blend after using the lighter to burn the material on the flattened part of the empty coffee can. Another small package of herbal blend without any label or brand name, which was also open, was found on the middle part of back seat of the car. Upon medical examination at the scene, no criminality was suspected for the deceased; the cadaver was relatively fresh and no injuries were found on the body surface. The cadaver was stored in a refrigerated morgue at 3–4 °C for about 2 days until autopsy. At the beginning of the autopsy, the postmortem interval was estimated to be about 3.5 days. The autopsy was performed at our Department. As macroscopic observations, there was livor mortis associated with vibices for wide areas of the body surface. There was extensive postmortem epidemic desquamation. There were no serious injuries related to the death of the victim. Internally, there were a few subcutaneous hemorrhages in the left lower leg.

The routine analysis of blood alcohol by gas chromatography showed negative results. Immunochemical drug screening tests with a Triage DOA kit (Alere, Waltham, MA, USA) and Instant-View M-1 (Alfa Scientific Designs, Poway, CA, USA) for urine specimens also showed negative results. NAGINATA screening for conventional drugs and toxic compounds in whole blood using gas chromatography–mass spectrometry (GC–MS) [11] showed a negative result.

We analyzed the herbal specimens found at the scene to determine their drug contents. Each herbal debris specimen was immersed in a centrifuge tube containing 1.0 ml of acetonitrile. It was sonicated for 10 min, and centrifuged at 10,000 rpm for 2 min. The supernatant layer was decanted into a test tube. Because herbal products usually contain large amounts of drugs, each supernatant layer was diluted more than 1,000-fold with acetonitrile. The extract solution prepared from the package without a brand name was subjected to our routine GC–MS analysis for drugs; two peaks appeared in the total ion chromatogram (TIC) and we obtained mass spectra from both peaks. We consulted the Cayman Spectral Library [12]; it strongly suggested two synthetic cannabinoids, AB-CHMINACA and 5-fluoro-AMB. For the extract solution obtained from the package labeled "Herbal Incense. The Super Lemon", our routine GC–MS analysis showed an intense single peak. The corresponding mass spectrum agreed well with the known spectrum of diphenidine, which we described in our previous report [10], thus confirming the identity of a third major component. Therefore, we focused our attention on the identification and quantitation of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in body fluids and solid tissues obtained from the victim.

Materials and methods

AB-CHMINACA, 5-fluoro-AMB, and 5-fluoro-AB-PINACA (internal standard, IS for AB-CHMINACA and 5-fluoro-AMB) were purchased from Cayman Chemical (Ann Arbor, MI, USA); diphenidine-HCl was purchased from Wako Pure Chemical Industries (Osaka, Japan). Phencyclidine-HCl (IS for diphenidine) was a gift from Dr. T. Nishikawa, National Center of Neurology and Psychiatry, Kodaira, Tokyo, Japan. Other common chemicals used were of the highest purity commercially available. Plastic centrifuge tubes with caps (5-ml capacity, 6 × 1.5 cm external diameter) and stainless beads for crushing solid tissues were purchased from TAITEC, Saitama, Japan; the QuEChERS dispersive-SPE centrifuge tubes with caps (2-ml capacity), each of which contained 25 mg of primary secondary amine, 25 mg of end-capped octadecylsilane (C_{18EC}), and 150 mg of magnesium sulfate, and Captiva ND Lipids cartridges (3-ml capacity) were purchased from Agilent (Santa Clara, CA, USA).

Blood specimens from the right and left heart atria and from the femoral vein, urine, and solid tissue specimens such as brain, heart muscle, lung, liver, spleen, kidney, pancreas, and adipose tissue were collected at autopsy and kept frozen at −80 °C until analysis; the adipose tissue specimen was obtained from the abdominal subcutaneous area of the deceased.

Extraction procedure for fluidal specimens

The first step of the extraction procedure for fluidal specimens, such as whole blood and urine, was different from that for solid tissue specimens. To 0.10 ml of whole blood or urine in a 1.5-ml plastic tube with a cap, 1.0 ml of acetonitrile and 100 ng each of 5-fluoro-AB-PINACA and phencyclidine (ISs) with or without appropriate amounts of 5-fluoro-AMB, AB-CHMINACA, and diphenidine all dissolved in 10 µl of acetonitrile were added, vortex-mixed for 10 s, sonicated for 5 min, and centrifuged at 10,000 rpm for 2 min. All supernatant fraction was decanted to the QuEChERS dispersive-SPE centrifuge tube

(2 ml) containing 25 mg of C_{18EC}, 25 mg of primary and secondary amine (PSA), and 150 mg of magnesium sulfate [13], followed by vortexing for 30 s, and centrifuging at 10,000 rpm for 2 min. The upper acetonitrile layer was passed through a Captiva ND Lipids cartridge. A 3.5-µl aliquot of the eluate was analyzed by liquid chromatography–tandem mass spectrometry (LC–MS–MS).

Extraction procedure for solid tissue specimens except adipose tissue

One gram of each solid tissue specimen was placed in a 5-ml plastic tube with a cap containing 4 ml of acetonitrile and 1,000 ng each of 5-fluoro-AB-PINACA and phencyclidine (ISs) dissolved in 10 µl of acetonitrile. The tissue specimens were minced with clean surgical scissors. Five stainless beads (5 mm external diameter) were added to the mixture. The tube was capped, held to a bead beater-type shaking machine (Beads Crusher µT-12; TAITEC), and vigorously shaken at 3,200 rpm for 5 min. All suspension solution was transferred to a large test tube, and 5 ml of acetonitrile was added to it and shaken gently. Six 1-ml portions were taken from the 10-ml homogenate suspension solution and were prepared with and without addition of different amounts of 5-fluoro-AMB, AB-CHMINACA, and diphenidine all dissolved in 10 µl of acetonitrile in 1.5-ml plastic centrifuge tubes with caps, vortexed for 30 s, and centrifuged at 10,000 rpm for 2 min. The supernatant was decanted into the QuEChERS dispersive-SPE centrifuge tube containing the three compounds, vortexed, and centrifuged as described above. The upper acetonitrile layer was passed through a Captiva ND Lipids cartridge. A 3.5-µl aliquot of the eluate was analyzed by LC–MS–MS.

Extraction procedure for the adipose tissue specimen

One gram of solid adipose tissue specimen was placed in a 5-ml plastic tube with a cap containing 4 ml of acetonitrile and 1,000 ng each of 5-fluoro-AB-PINACA and phencyclidine (ISs) dissolved in 10 µl of acetonitrile. The specimen was minced with clean surgical scissors. The 5-ml plastic tube containing the mixture was heated at 80 °C for 10 min, and five stainless beads (5 mm external diameter) were added to the mixture. The tube was capped, held to a bead beater-type shaking machine (Beads Crusher µT-12; TAITEC), and vigorously shaken at 3,200 rpm for 5 min. Despite vigorous shaking, the liquefied fat layer and the acetonitrile layer did not mix well. All of the mixture except the stainless beads was transferred to a scaled 50-ml conical flask, and the total volume was made up to 50 ml by addition of acetonitrile, followed by gentle shaking. At this stage, the liquid fat and acetonitrile were mixed completely. Six 1-ml portions taken from the 50-ml mixture solution were prepared with and without the addition of different amounts of 5-fluoro-AMB, AB-CHMINACA, and diphenidine all dissolved in 10 µl of acetonitrile in 1.5-ml plastic centrifuge tubes with caps, vortexed for 30 s, and centrifuged at 10,000 rpm for 2 min. The supernatant fraction was decanted into the QuEChERS dispersive-SPE centrifuge tube containing the three compounds, vortexed, and centrifuged as described above. The upper acetonitrile layer was passed through a Captiva ND Lipids cartridge. A 3.5-µl aliquot of the eluate was analyzed by LC–MS–MS.

LC–MS–MS conditions

LC–MS–MS with electrospray ionization (ESI) was conducted on an Agilent 1200 LC-SL system connected to a 6460 Triple Quad LC/MS tandem MS instrument (Agilent). The Agilent LC-SL system contained a microdegasser and a high-performance autosampler. For LC separation, a ZORBAX Eclipse Plus C18 column (100 × 2.1 mm i.d., particle size 1.8 µm, Agilent) was used. The LC conditions were: injection volume, 3.5 µl; flow rate, 0.25 ml/min; elution mode, gradient with 10 mM ammonium formate/0.1 % formic acid in distilled water (A) and acetonitrile (B) from 90 % A/10 % B to 100 % B over 20 min followed by isocratic elution with the final solvent composition for 10 min. The column and autosampler operated at room temperature.

The tandem MS conditions were: interface, ESI mode; polarity, positive ion mode; ion source temperature, 320 °C; ion source voltage, 500 V; quantitation, selected reaction monitoring (SRM) mode using the peak area; ion transitions: m/z 357 → 241 for AB-CHMINACA, m/z 364 → 304 for 5-fluoro-AMB, m/z 266 → 181 for diphenidine, m/z 349 → 304 for 5-fluoro-AB-PINACA (IS), m/z 244 → 91 for phencyclidine (IS); fragmentor voltage and collision energy were: 120 and 21 V, respectively, for AB-CHMINACA, 120 and 9 V for 5-fluoro-AMB, and 100 and 13 V for diphenidine.

Data acquisition, peak integration, and calculation were performed with a computer workstation (Agilent Masshunter, Revision Acquision B. 02. 01, Qualification B. 03. 01SP2 and Quantification B. 04. 00).

Standard addition method

The standard addition method is frequently used for analysis by atomic absorption spectroscopy and gas chromatography to overcome the matrix effect [14]. In our previous study, we had to use the standard addition method to measure glycols in human whole blood, because the compounds were found to be present in whole blood specimens collected from healthy subjects [15]. During our

use of the method, we have found that it has great advantages. First, the standard addition method can completely overcome the matrix effects, especially in cases investigating the distribution of a xenobiotic compound in different types of matrices. The method becomes particularly useful in the absence of a suitable stable isotopic IS. Second, the method does not require blank human body fluids or solid tissues that are negative for target compounds; generally, the collection of nonessential human tissue samples is avoided because of ethical reasons. The only disadvantage of the standard addition method is that it is somewhat laborious due to necessity of constructing a calibration curve with not less than six plot points to obtain a single concentration value. The details of using the standard addition method are explained in our previous report [16].

Matrix effects and recovery rates

Although the standard addition method can overcome matrix effects and even recovery rates, it is useful to consider the matrix effects that are present, especially when LC–MS–MS is used and lipid-rich matrices are encountered. The principle and the calculation method for matrix effects and recovery rates are described in our previous reports [8, 9, 16]. The main difference in the present study is the use of the Captiva ND Lipids cartridge at the final stage of extraction, just before LC–MS–MS analysis. The matrix effects and recovery rates for AB-CHMINACA, 5-fluoro-AMB, and diphenidine were tested only for adipose tissue at their preexisting concentrations as an example.

Results and discussion

Homogenization of tissue specimens

Before the extraction step, the solid tissue specimens should be homogenized. According to the modified QuEChERS method describing by Usui et al. [13], 0.5 g of liver tissue was mixed with 2.5 ml of aqueous solution containing five stainless beads (5 mm diameter) and salts, and crushed by vigorous shaking with a bead beater-type homogenizer at 2,500 rpm for 2 min. In previous studies [8, 16], we tried this homogenization method. However, with only the bead beater-type homogenization, some solid tissues obtained from human cadavers could not be completely crushed, and so we also used the conventional Polytron homogenizer for incompletely crushed tissue samples. In the present study, such bead beater-type homogenization was employed in the acetonitrile solution, which enabled complete crushing of relatively hard human tissues without the aid of the Polytron homogenizer.

Identification of AB-CHMINACA, 5-fluoro-AMB, and diphenidine by LC–MS–MS

Figure 1 shows the product ion mass spectra of the reference standards of diphenidine, AB-CHMINACA, and 5-fluoro-AMB, and those of the extract obtained from the adipose tissue as an example. Each mass spectrum obtained from the extract coincided with that of the corresponding reference standard compound, confirming the preliminary results obtained from examination of the two herbal packages by GC–MS. This result suggests that the subject consumed both herbal products simultaneously or with a short interval between use of the two packages. The mass spectrum of 5-fluoro-AMB was recorded only for the adipose tissue. For AB-CHMINACA, its fragmentation pattern was observed in spectra for solid tissues such as brain, heart muscle, liver, kidney, and pancreas without any impurity. The spectrum of diphenidine was identified in the spectra of all specimens.

Validation of the method

Figure 2 shows an example of the SRM chromatograms for the target compounds and ISs extracted from the adipose tissue. In the adipose tissue, the concentration of diphenidine was far higher than those of AB-CHMINACA and 5-fluoro-AMB, as shown in the total ion chromatogram (TIC) (the locations of the other two compounds are shown with arrows). The ISs were also not recognized in the TIC. In the SRM chromatograms, both AB-CHMINACA and 5-fluoro-AMB appeared as sharp symmetrical peaks, and no impurity peaks were observed for both chromatograms with low backgrounds; this was also true for the ISs (Fig. 2, bottom chromatograms).

Table 1 shows the standard addition calibration equations for the three compounds in the specimens of the present deceased. The correlation coefficient values obtained for all specimens examined were greater than 0.99. By extensive dilution of some specimens with acetonitrile, the detection limit (signal-to-noise ratio $\geq$3) for the three compounds by this method was around 0.1 ng/ml or g. The lower quantitation limit (signal-to-noise ratio $\geq$10) was around 1 ng/ml or g.

Because we employed the standard addition method for quantitation without the use of blank specimens, it was impossible to present the usual accuracy and precision data. Instead, as shown in Table 2, we repeated intraday and interday determinations of the three compounds in the adipose tissue specimen as an example.

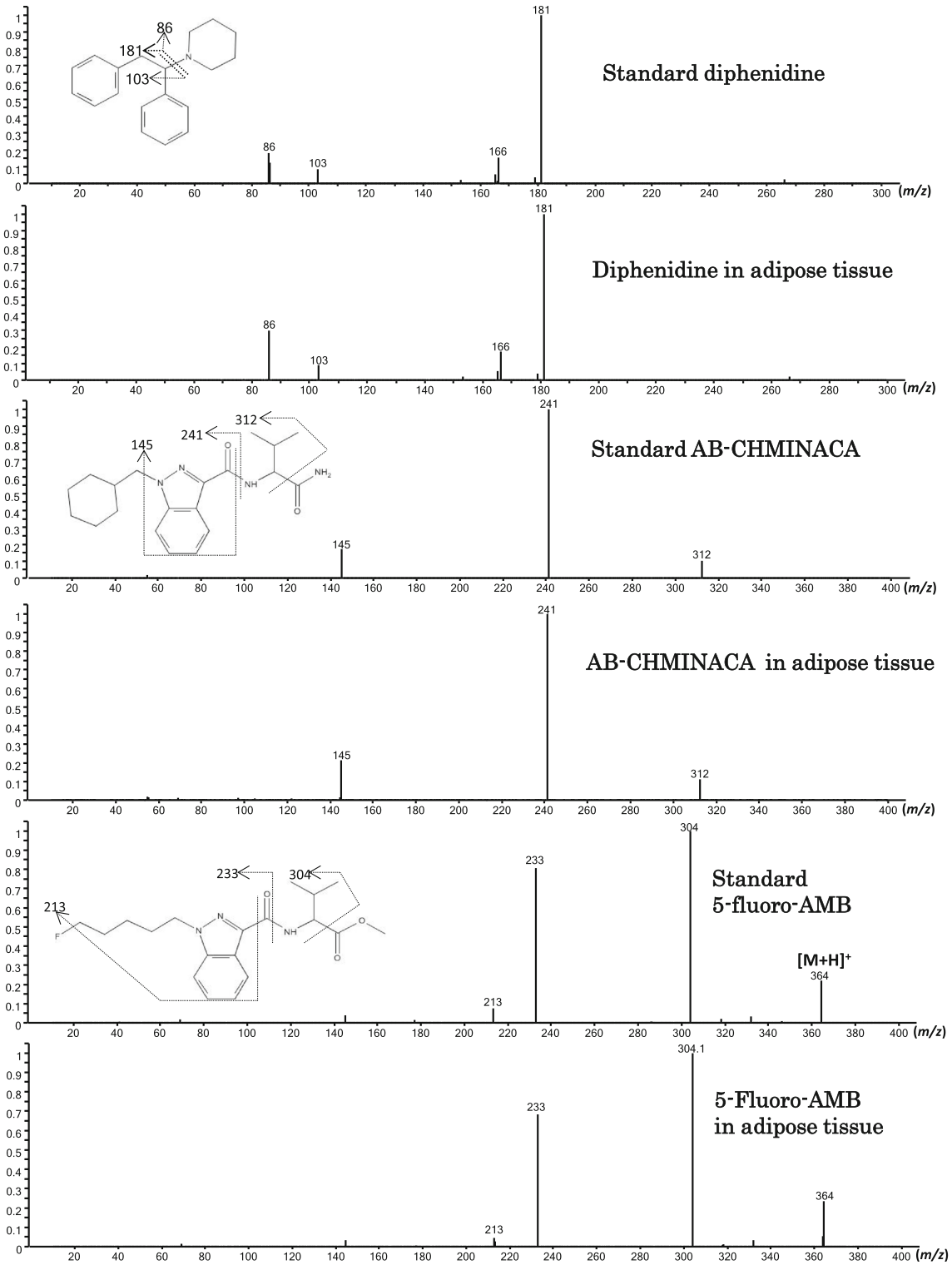

Fig. 1 Product ion mass spectra of the reference standards of diphenidine, AB-CHMINACA, and 5-fluoro-AMB in comparison with those obtained from the extract of the adipose tissue specimen recorded by liquid chromatography–tandem mass spectrometry together with the probable fragmentation mode

The repeatability expressed as relative standard deviations was not greater than 16.9 %. Although the standard addition method can overcome matrix effects and low recovery rates, the difference in the matrix effects according to extraction method or to kinds of matrices are of great interest. In a study on the postmortem distribution

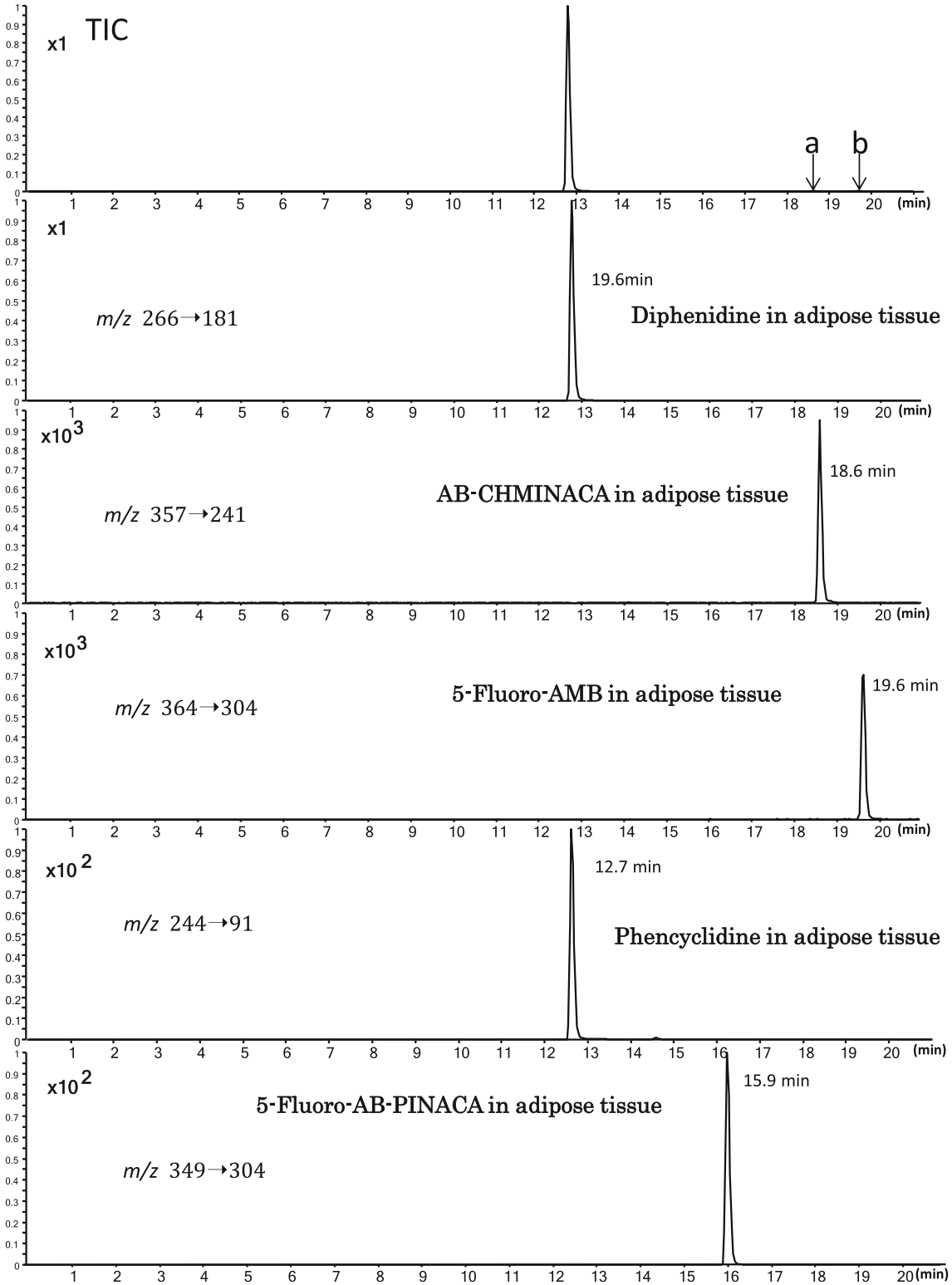

Fig. 2 Total ion chromatogram (TIC) and selected reaction monitoring chromatograms for diphenidine, AB-CHMINACA, 5-fluoro-AMB, and two internal standards in the extract of the adipose tissue specimen. *a*, expected retention time for AB-CHMINACA on TIC; *b*, expected retention time for 5-fluoro-AMB on TIC

Table 1 Standard addition calibration equations for diphenidine, AB-CHMINACA, and 5-fluoro-AMB in body fluids and solid tissues of the deceased

Analyte	Specimen	Equation[a]	Correlation coeffcient (r)
Diphenidine	Femoral vein blood	$y = 0.00167x + 1.20$	0.997
	Right heart blood	$y = 0.00149x + 1.05$	0.997
	Left heart blood	$y = 0.00144x + 1.33$	0.996
	Urine	$y = 0.00162x + 0.610$	0.993
	Brain	$y = 0.000266x + 0.413$	0.998
	Heart muscle	$y = 0.000342x + 0.706$	0.996
	Lung	$y = 0.000183x + 0.292$	0.996
	Liver	$y = 0.000296x + 0.877$	0.997
	Spleen	$y = 0.000227x + 0.296$	0.998
	Kidney	$y = 0.000256x + 0.640$	0.999
	Pancreas	$y = 0.000270x + 0.514$	0.997
	Adipose tissue	$y = 0.000582x + 5.29$	0.999
AB-CHMINACA	Brain	$y = 0.000337x + 0.00525$	0.997
	Heart muscle	$y = 0.000328x + 0.00658$	0.998
	Lung	$y = 0.000331x + 0.00266$	0.992
	Liver	$y = 0.000328x + 0.00697$	0.999
	Spleen	$y = 0.000472x + 0.00356$	0.998
	Kidney	$y = 0.000352x + 0.00870$	0.999
	Pancreas	$y = 0.000380x + 0.0148$	0.999
	Adipose tissue	$y = 0.000923x + 0.0230$	0.997
5-Fluoro-AMB	Adipose tissue	$y = 0.000903x + 0.0169$	0.998

[a] If $y = 0$, the preexisting concentration (x) can be calculated as a minus value

Table 2 An example of intraday and interday repeatability for determination of diphenidine, AB-CHMINACA, and 5-fluoro-AMB in the adipose tissue of the deceased

Compound	Intraday ($n = 5$)		Interday ($n = 5$)	
	Concentration found (ng/ml)[a]	Repeatability (%RSD)	Concentration found (ng/ml)[a]	Repeatability (%RSD)
AB-CHMINACA	24.7 ± 2.48	10.0	24.8 ± 2.85	11.5
5-Fluoro-AMB	18.7 ± 1.10	5.9	20.0 ± 2.08	10.4
Diphenidine	9,100 ± 920	10.1	11,100 ± 1,870	16.9

RSD relative standard deviation

[a] Data given as mean ± standard deviation (SD)

of α-pyrrolidinovalerophenone in human specimens using solid-phase extraction with Oasis HLB cartridges coupled to LC–MS–MS analysis [8], we observed remarkable depressive matrix effects of 68–90 % (only 10–38 % detection) for stomach contents and all of the solid tissue specimens. However the depressive matrix effects were less than 3 % for whole blood and urine specimens. In a study on the postmortem distribution of α-pyrrolidinobutiophenone in human specimens using acetonitrile deproteinization extraction plus QuEChERS dispersive solid-phase extraction coupled to LC–MS–MS analysis [16], the depressive matrix effects were better; 15.8–60.3 % depression (39.7–84.2 % detection) for stomach contents and solid tissue specimens, and 1.8–12.4 % depression (87.6–98.2 % detection) for body fluid specimens such as whole blood, cerebrospinal fluid, and urine. Although the target compounds are different (not cathinones, but synthetic cannabinoids and diphenidine) in the present study, the depressive matrix effects for AB-CHMINACA, 5-fluoro-AMB, and diphenidine extracted

Table 3 An example of matrix effects and recovery rates for determination of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in the adipose tissue obtained from the deceased

Compound	Matrix effect (%)	Recovery (%)
AB-CHMINACA	91.4 ± 2.29	85.4 ± 7.22
5-Fluoro-AMB	94.1 ± 3.15	81.3 ± 2.81
Diphenidine	67.3 ± 2.40	109 ± 1.85

Data given as mean ± SD obtained by triplicate determinations

Table 4 Postmortem concentrations of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in body fluids and solid tissues of the deceased

Specimen	Concentration (ng/ml or g)		
	AB-CHMINACA	5-Fluoro-AMB	Diphenidine
Femoral vein blood	–[a]	–	715 ± 28.3
Right heart blood	–	–	707 ± 62.9
Left heart blood	–	–	923 ± 62.8
Urine	–	–	376 ± 23.7
Brain	15.6 ± 0.39	–	1,550 ± 49.1
Heart muscle	20.0 ± 3.00	–	2,070 ± 73.5
Lung	8.02 ± 0.71	–	1,600 ± 13.9
Liver	21.2 ± 1.30	–	2,960 ± 34.0
Spleen	7.55 ± 0.18	–	1,300 ± 31.9
Kidney	24.7 ± 1.63	–	2,510 ± 32.9
Pancreas	38.9 ± 4.55	–	1,910 ± 38.1
Adipose tissue	24.8 ± 2.48	18.7 ± 1.10	11,100 ± 1,120

[a] Below the lower limit of quantitation (<1 ng/ml or g)

from the adipose tissue were 5.9–32.7 % (67.3–94.1 % detection) using acetonitrile deproteinization plus QuEChERS dispersive solid-phase extraction plus passage through a Captiva ND Lipids cartridge coupled to LC–MS–MS analysis (Table 3). The depressive matrix effect for the brain specimen was also found to be less than 10 % by the present method (unpublished observation). The final treatment with the Captiva ND Lipids cartridge appears to be very useful in preventing the depressive matrix effects caused by ionized phospholipids.

Postmortem distribution of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in various specimens of the deceased

Table 4 shows the postmortem distribution of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in body fluids and solid tissues including the adipose tissue of the deceased. Quantitation of all three compounds could only be achieved for the adipose tissue. In many of the specimens, 5-fluoro-AMB or both 5-fluoro-AMB and AB-CHMINACA could not be quantitated, because they were below the lower limit of quantitation; however, in most such specimens, small peaks above the detection limit (0.1 ng/ml or g) were observed (magnified data not shown). Saito et al. [17] first reported a fatal case of MAM-2201 poisoning with a very high concentration (1,540 ng/g) of unchanged MAM-2201 in the adipose tissue of the cadaver. Their data and our result (Table 4) suggest that the adipose tissue is a good specimen to detect synthetic cannabinoids in their unchanged forms. Most probably, the synthetic cannabinoids are transported to the adipose tissues, and dissolve in the fatty tissues there, because of the high lipophilicity of the compounds. The compounds can remain in the adipose tissues in the unchanged forms for a long time, because the enzymes responsible for metabolism of synthetic cannabinoids may be absent or at low concentration in the the fat of the adipose tissue. These findings suggest that the adipose tissue is the specimen of choice when the involvement of synthetic cannabinoid(s) is suspected. Other than synthetic cannabinoids, lipophilic compounds such as diphenidine can also accumulate in the adipose tissue, as shown in Table 4. This point is also noteworthy for forensic toxicologists.

5-Fluoro-AMB could be quantitated only for the adipose tissue, while AB-CHMINACA could be quantitated for all solid tissues. This contrast between the two synthetic cannabinoids may be due to a minor difference in their structures; 5-fluoro-AMB contains a terminal methyl ester group, while AB-CHMINACA has an amide group at the same position.

Conclusions

We have presented the postmortem distribution of AB-CHMINACA, 5-fluoro-AMB, and diphenidine in body fluids and solid tissues obtained from a cadaver. To our knowledge, this is the first report to describe the concentrations of the three compounds in human specimens, although they have been disclosed for illegal products very recently [5, 10]. Our study showed that the adipose tissue is the best specimen to detect AB-CHMINACA, 5-fluoro-AMB, and diphenidine in their unchanged forms. From the extremely high level of diphenidine measured in the adipose tissue, we believe that diphenidine played a major role in the death of the victim in the present case.

Conflict of interest There are no financial or other relations that could lead to a conflict of interest.

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