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Research Article

Simultaneous Determination of Methamphetamine and Its Isomer N-Isopropylbenzylamine in Forensic Samples by Using a Modified LC-ESI-MS/MS Method

Yangxu Luo,[1] Juan Du,[1,2] Huadi Xiao,[1] Ling Zheng,[1] Xuncai Chen,[2,3] Ande Ma,[1,2] and Qizhi Luo[2,3]

[1]*Hygiene Detection Center, School of Public Health, Southern Medical University, Guangzhou, China*
[2]*Forensic Science Center, Southern Medical University, Guangzhou, China*
[3]*Department of Forensic Toxicology, School of Forensic Medicine, Southern Medical University, Guangzhou, China*

Correspondence should be addressed to Xuncai Chen; xche3815@smu.edu.cn, Ande Ma; mandmy@126.com, and Qizhi Luo; luoqizhi12@126.com

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Accurate identification and quantification of methamphetamine (MA) and its related substances are essential for the investigation and fair trial of drug offenses. In this study, a modified LC-ESI-MS/MS method for the simultaneous determination of MA and its isomer N-isopropylbenzylamine (N-IBA) in forensic samples was developed and validated. Optimum chromatographic separation of the target analytes was achieved on an Agilent Poroshell 120 SB-C18 column (4.6 × 100 mm, 2.7 μm) at 40°C with isocratic elution at the flow rate of 0.40 mL/min. The mobile phase was acetonitrile and 20 mM ammonium acetate solution containing 0.1% formic acid (80 : 20, *v*/*v*). Positive ESI-MS/MS detection was performed in multiple reaction monitoring (MRM) mode to identify and quantify the target analytes. Method validation showed excellent linearity in the range of 0.51 ng/mL~51 ng/mL for MA and N-IBA. The low limit of detection (LLOD) and low limit of quantification (LLOQ) reached 0.1 ng/mL and 0.3 ng/mL for both analytes. The method showed a satisfactory accuracy with an inter- and intraday-relative error (RE) <20%, and a precision of inter- and intraday relative standard deviation (RSD) less than 15%. The validated method was successfully applied in real forensic samples and resulted in the detection of MA and N-IBA in 8 suspected samples in drug cases that only deemed MA positive using our previous routine screening procedure, which avoided the misidentification of N-IBA as MA.

1. Introduction

Drug abuse has been increasingly becoming one of the most severe social problems all over the world. Among drugs of abuse, methamphetamine (MA) is the second most popular illicit drug worldwide which has been listed as a category I psychotropic substance under strict state control in most countries [1]. According to the United Nations Office on Drugs and Crime (UNODC), MA is one of the most dangerous drugs due to its central nervous excitatory effect, highly addictive, and numerous related disorders [2–4]. MA causes toxicity such as liver, brain neurological, cardiovascular, immune system injury, physical, and mental problems (violence, anxiety, and paranoia) [5–11]. Thus, accurate identification and quantification of MA in the forensic samples is particularly important for investigations and fair trials of drug crimes [12]. However, an isomer of MA, called N-isopropylbenzylamine (N-IBA), has often been used as the adulterant of MA in drug crimes due to their high similarity in structure (Figure 1), which easily resulted in the misidentification of N-IBA as MA in suspected samples [13]. For example, the forensic science laboratory of the United States Drug Enforcement Administration (DEA) reported several cases of counterfeiting MA hydrochloride with N-IBA hydrochloride from 2007 to 2008 [14]. Since 2011, the material identification center of the Ministry of State Security of the People's Republic of China has also reported several drug cases that N-IBA hydrochloride was mixed with MA

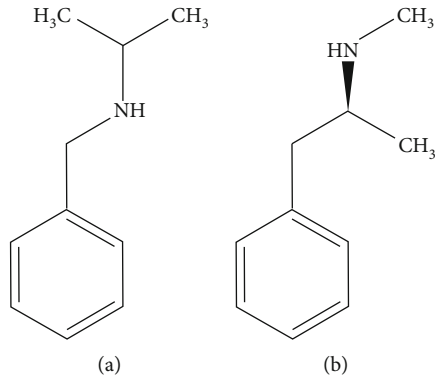

Figure 1: Chemical structure of (a) N-IBA and (b) MA.

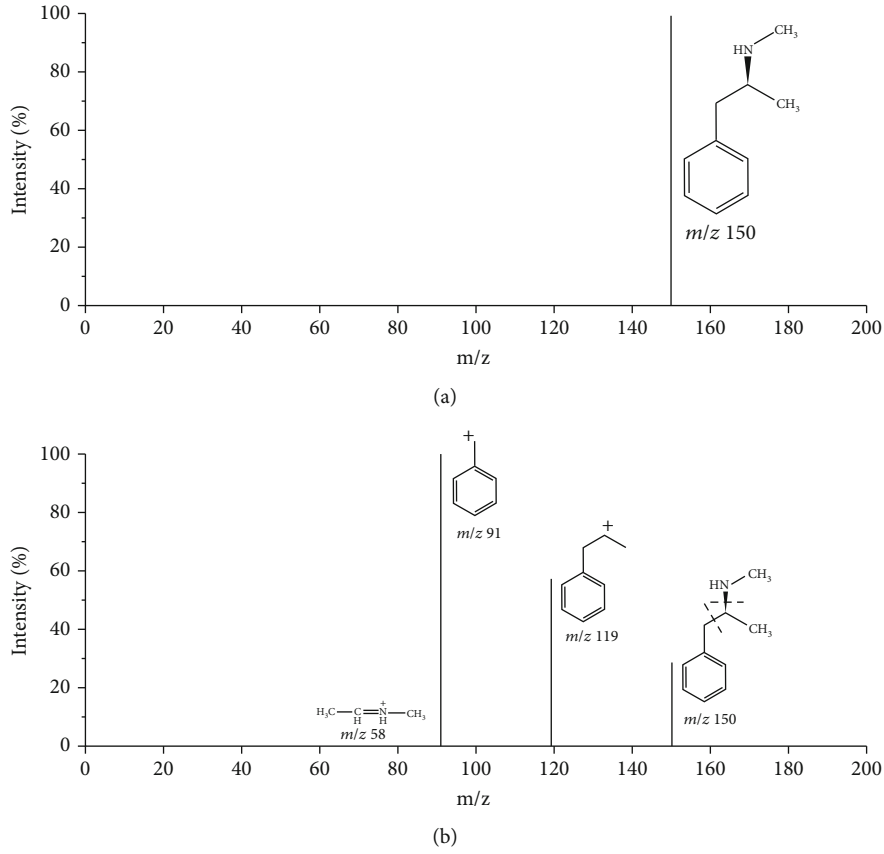

Figure 2: Mass spectra of methamphetamine (MA). (a) Molecular ion spectrum by (a) mass spectrum by ESI-MS (molecular ion $[M + H]^+ = 150$). (b) Product ion spectrum by ESI-MS/MS (precursor ion $[M + H]^+ = 150$, fragment ions = 119/91/58).

hydrochloride in China [14]. It is worth to mention that N-IBA with unknown toxicity is an important organic synthetic raw material commonly used in industrial productions [13, 15], which is not included in the illicit drug category. The misidentification of N-IBA in MA forensic samples may result in false-positive results, which would cause the misleading legal sentence to the suspects [14]. Therefore, it is necessary to establish an effective detection method for the discrimination of MA and I-NBA in suspected drugs.

At present, many researches have mainly focused on the detection of MA [16–23], while the simultaneous determination of MA and its isomer N-IBA in suspected drugs is rarely studied. The commonly used methods including infrared spectroscopy, colloidal gold-based immunoassays, and color tests are not suitable for the discrimination between MA and N-IBA since their poor specificity [24–28]. GC/MS analysis in full scan mode has been used to simultaneously quantify MA and N-IBA in suspected drugs [25, 27, 28]. However, the two compounds were hard to be effectively discriminated by GC/MS when there was a large concentration difference between them. Because the retention times for MA and N-IBA chromatographic separation were very close due to their high similar chemical structure, the compound with high concentration would

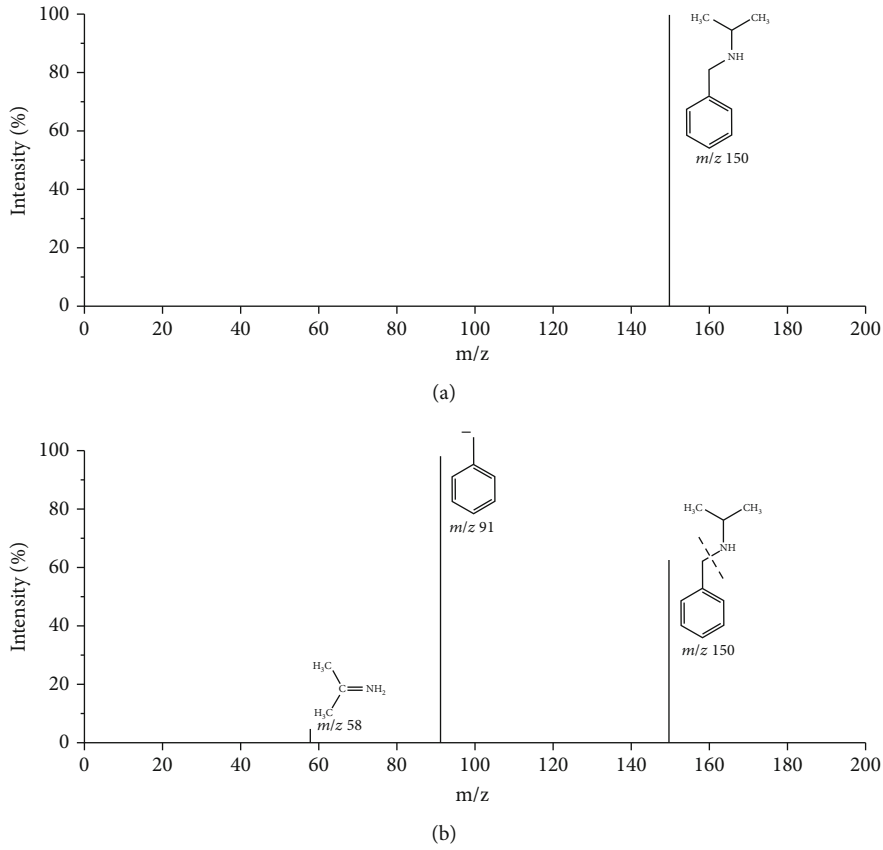

Figure 3: Mass spectra of N-isopropylbenzylamine (N-IBA). (a) Molecular ion spectrum by (a) mass spectrum by ESI-MS (molecular ion $[M + H]^+ = 150$). (b) Product ion spectrum by ESI-MS/MS (precursor ion $[M + H]^+ = 150$, fragment ions = 91/58).

interfere with another one with low concentration as the two compounds yield similar ion fragments for detection [25]. Liquid chromatography-mass spectrometry (LC-MS/MS) has fully proved itself as a powerful tool for detecting and confirming the presence of drugs in complex matrices [29, 30]. In consideration of the highly similar ion fragments between MA and N-IBA, the commonly used LC-MS/MS is insufficient to separate the MA from N-IBA. Hence, to develop a high efficient chromatographic separation technique in LC-MS/MS is highly desirable for the simultaneous determination of MA and its isomer N-IBA.

In this work, a modified LC-ESI-MS/MS method conducted with positive electrospray ionization (ESI) in multiple reaction monitoring (MRM) mode was subsequently developed and validated to discriminate MA and N-IBA in forensic science [24]. Optimum chromatographic separation of the target analytes was achieved on an Agilent Poroshell 120 SB-C_{18} column (4.6 × 100 mm, 2.7 μm) at 40°C with isocratic elution at the flow rate of 0.40 mL/min. The method was successfully applied to determine the MA and N-IBA with satisfactory selectivity, sensitivity, accuracy, and repeatability. In addition, 8 suspected samples in drug cases deemed MA positive were screened by using this new method, and the N-IBA was seized in all samples, illustrating the ubiquity of adulteration of N-IBA to MA.

2. Materials and Methods

2.1. Chemicals and Reagents. MA (purity 98%) was purchased from Merck Sigma-Aldrich (Darmstadt, Germany). N-IBA (purity 98%) was purchased from Energy Chemical (Shanghai, China). Methanol and acetonitrile of HPLC grade were purchased from Merck Millipore (Darmstadt, Germany). Ammonium acetate (purity 99.0%) was purchased from Aladdin Bio-Chem Technology Co., Ltd. (Shanghai, China). Formic acid of HPLC grade was from Tianjin Kemiou Chemical Reagent Co., Ltd. (Tianjin, China). Ultrapure water ($R > 18$ MΩ/cm) was obtained from a Millipore Milli-Q system (Merck Millipore, Darmstadt, Germany).

2.2. Solutions. Individual stock solutions of MA and N-IBA were prepared in methanol at the concentration of 1 mg/mL and stored at −20°C for up to 6 months. Appropriate concentrations of calibrator and QC working solutions were prepared by diluting the stock solutions with methanol. The mixed calibrator solutions were prepared with concentrations of 0.1, 0.2, 0.5, 1.0, 5.0, 10.0, 20.0, and 50.0 ng/mL. The QC samples were prepared in methanol at the low, medium, and high concentrations of 2.0 ng/mL, 10.0 ng/mL, and 20.0 ng/mL. The calibrator solutions, QC solutions were stored at 4°C prior to analysis.

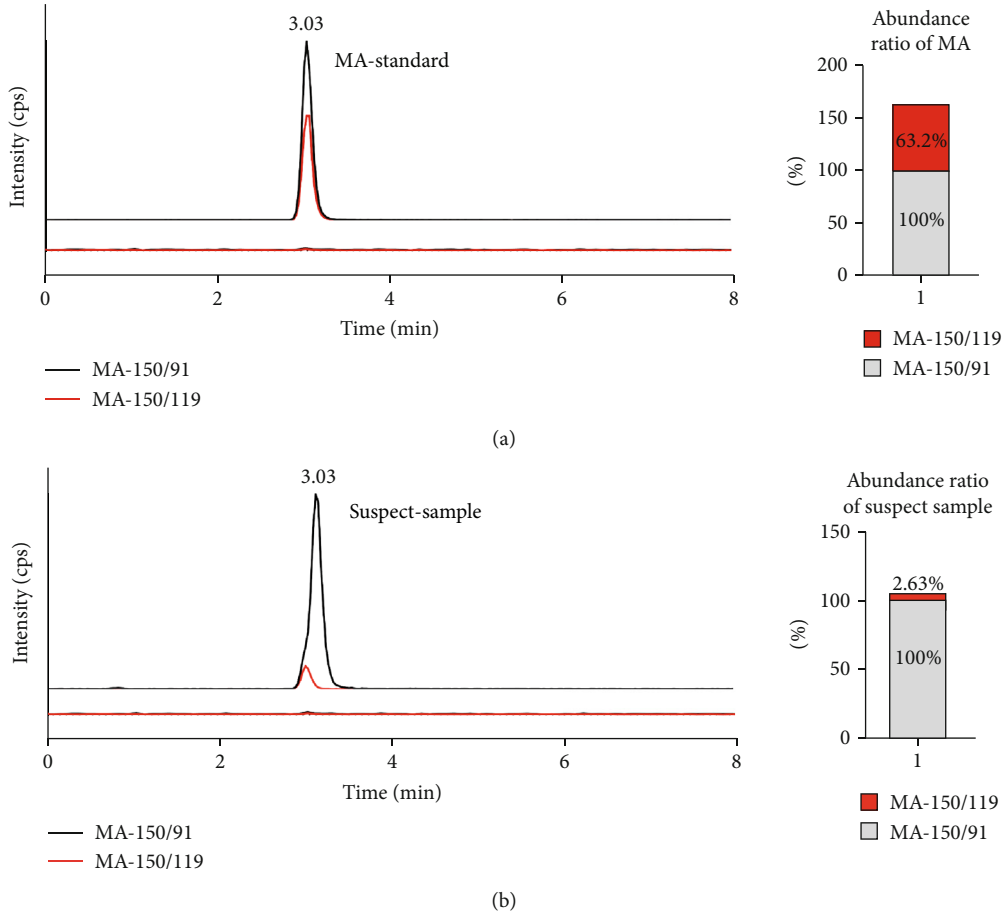

Figure 4: MRM chromatograms for the detection of MA and N-IBA by our routine screening procedures. (a) Upper: detected MA-Standard and the abundance ratio of MA-Standard between *m/z* 150/91and *m/z* 150/119, lower: blank. (b) Upper: detected suspect sample and the abundance ratio of suspect sample between *m/z* 150/91 and *m/z* 150/119, lower: blank.

2.3. Specimen Preparation. The specimens (white crystalline substances) were obtained from suspected cases of drug production and drug trafficking from 2017 to 2018 in Guangdong province, China. After homogenization, 50 mg of forensic crystal samples was accurately weighed, dissolved in methanol, and gradually diluted to a constant concentration of 1 mg/mL, 100 μg/mL, and 20 ng/mL. The sample solutions were then filtered by a 0.22 μm filter before further analysis.

2.4. Instrumentation. Experiments were performed on a Shimadzu HPLC system (Tokyo, Japan) consisting of two LC-20ADvp pumps, a CTO-20ACvp column heater, a SIL-20A autosampler, and a CBM-20A/20Alite controller. The HPLC was interfaced with an AB Sciex API 4000 QTRAP mass spectrometer (Foster City, USA) equipped with an ESI&Turb spray ionization source. All data were acquired and analyzed using the Analyst software (Version 1.5.2, AB Sciex, Foster City, USA).

2.5. LC-ESI-MS/MS Conditions

2.5.1. Routine Screening Procedures. The separation was performed on an RESREK Allure PFPP C_{18} column (2.1 × 100 mm, 5 μm) protected by RESREK Allure PFPP C_{18} column (2.1 × 10 mm, 5 μm) with isocratic elution at 0.40 mL/min, giving a total run time of 13.5 min. The column temperature is the same as the room temperature. The mobile phase consisted of methyl alcohol (eluent A) and 20 mM ammonium acetate solution containing 0.1% formic acid (eluent B) in a volume ratio of 70/30. The autosampler tray temperature was maintained at 15°C, and the injection volume was 20 μL.

The MS conditions were as follows: the ion polarity was positive, the ionspray voltage applied was 5500 V, the source temperature was set at 600°C, and the ion source gas1 (N_2), ion source gas2 (N_2), and curtain gas (N_2) were set at 65 psi, 65 psi, and 15 psi, respectively. The ion pairs of MA are 150/119 and 150/91.

2.5.2. Modified Detection Method. The separation was performed on an Agilent Poroshell 120 SB-C_{18} column (4.6 × 100 mm, 2.7 μm) protected by an Idec Health and Science precolumn (filter assay size: 2 μm) with isocratic elution at 0.40 mL/min, giving a total run time of 6.5 min. The column temperature was 40°C. The mobile phase consisted of acetonitrile (eluent A) and 20 mM ammonium acetate solution containing 0.1% formic acid (eluent B) in a volume ratio of 20/80 [14, 19]. The

autosampler tray temperature was maintained at 15°C, and the injection volume was 10 μL.

MS/MS parameters were optimized with standard solutions of each analyte infused into the ESI source at 10 μL/min via a syringe infusion pump. The MS conditions were as follows: the ion polarity was positive, the ionspray voltage applied was 5500 V, the source temperature was set at 600°C, and the ion source gas1 (N_2), ion source gas2 (N_2), and curtain gas (N_2) were set at 55 psi, 65 psi, and 15 psi, respectively. MRM scans were performed with each analyte utilizing one molecular ion and two most predominant fragments; of the two, the most sensitive transition was used as quantifier ions and the other transition as qualifier ions, the ratios of which are indicative of the analyte of interest. The selected MRM transitions, declustering potentials and collision energies for each analyte are given in Supplementary material Table 1.

2.6. Method Validation. This method was validated according to the CFDA drug analytical method validation guidelines with minor modifications [31]. The validation items include selectivity, the calibration curves and their linearity, LLOD and LLOQ, accuracy, precision, and stability.

2.6.1. Selectivity and Discrimination Capability. Method selectivity was tested by comparing the chromatograms obtained from six blank samples free of MA and N-IBA with those from standard solutions. The extracted ion chromatograms at the retention times of the target analytes were examined for interfering peaks.

The discrimination capability of the method was evaluated by analyzing low, medium, and high concentrations of mixed standard solutions of MA and N-IBA. The extracted ion chromatograms were then inspected for unequivocal identification of the two isomers.

2.6.2. Calibration Curves. The calibration curves were constructed by running six concentration levels of standard solutions ranging from 0.5 ng/mL to 50 ng/mL in three replicates. Determination coefficients (R^2) were determined by applying a weighted ($1/x^2$) least-squares linear regression. Calibrators and triplicate of QC samples at low, medium, and high concentrations were analyzed daily in each set of specimens.

2.6.3. LLOD and LLOQ. LLOD and LLOQ were evaluated using a triplicate of standard solutions at the lowest calibration concentrations of the target analytes. LLOD was defined as the concentration producing a Gaussian-shaped peak eluting within ±2% of mean calibration retention time, a minimum signal to noise (S/N) ratio of 3 : 1; LLOQ was defined as the concentration that met LLOQ criteria and quality control precision and accuracy within ±20% and had a minimum S/N ratio of 10 : 1.

2.6.4. Accuracy and Precision. Intraday precision was evaluated using five replicated of QC solutions at low, medium, and high concentrations (2.0, 10.0, and 20.0 ng/mL, respectively) on a single day, while interday precision was assessed on three separate days. The precision expressed as RSD,

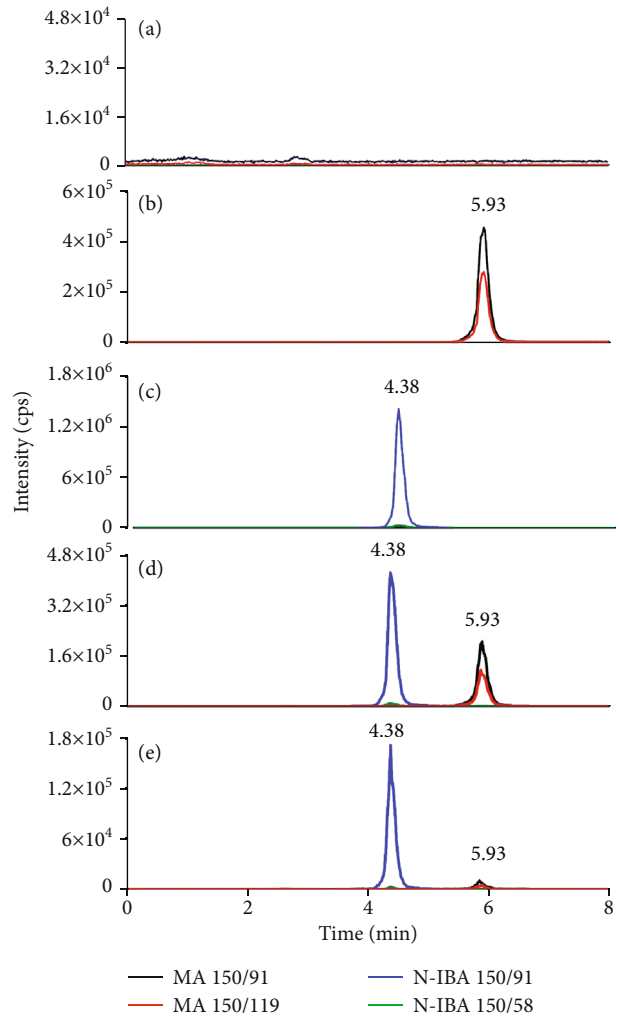

Figure 5: MRM chromatograms by LC-ESI-MS for detection of MA and N-IBA. (a) Blank. (b) Standard of MA. (c) Standard of N-IBA. (d) Mix-standard of MA and N-IBA (N-IBA standard at concentration of 9.14 ng/mL was mixed with MA standard at concentration of 10.27 ng/mL at equal volume ratio). (e) Actual forensic sample, SUM-1.

which was calculated as follows:

$$\text{RSD} = [\text{standard deviation (SD)}/C_{\text{mea}}] \times 100\%. \quad (1)$$

The accuracy was evaluated using triplicate of QC samples solutions at low, medium, and high concentrations; the relative error (RE) was calculated by the mean value of the measured concentrations (C_{mea}) from the theoretical concentration (C_{theo}).

$$\text{RE} = C_{\text{mea}}/C_{\text{theo}} \times 100\%. \quad (2)$$

2.6.5. Stability. Stability of suspected samples, calibrators, and QC samples was tested by reanalyzing them when these samples were kept for three days in an autosampler at 15°C, one week in room temperature at 20°C as well as one month in the freezer at 4°C. Stability was expressed as the relative

Table 1: Linearity, LLOD, and LLOQ for determining MA and N-IBA.

Compounds	LLOD (ng/mL)	LLOQ (ng/mL)	Linear range (ng/mL)	Equation	R^2
MA	0.1	0.3	0.51~51.30	$y = 3.24e^4x + 9.39e^4$	0.9959
N-IBA	0.1	0.3	0.51~51.20	$y = 8.49e^4x + 1.25e^4$	0.9973

Table 2: The accuracy and precision for quantification of N-IBA and MA.

Compounds	Nominal concentration (ng/mL)	Intraday ($n = 5$)		Interday ($n = 15$)	
		Accuracy (RE, %)	Precision (RSD, %)	Accuracy (RE, %)	Precision (RSD, %)
MA	2.05	110.1	5.4	121.9	10.0
	10.27	101.8	3.5	101.0	3.6
	20.53	101.1	4.0	110.0	9.9
N-IBA	1.83	96.2	5.1	110.7	11.6
	9.14	98.5	1.7	100.1	4.3
	18.28	97.2	2.7	103.6	6.4

average deviation (RAD) of the measured concentrations in different time.

3. Results

3.1. Development of Confirmatory Method. According to the structural data, N-IBA is an isomer of MA, normally exhibiting similar patterns of collision-induced dissociation (CID) and retention times with MA. Thus, the simultaneous identification for N-IBA and MA was easily confused.

The mass spectra for MA and N-IBA were obtained in full-scan MS and MS/MS mode. The predominant fragments of MA (Figure 2) were *m/z* 119 $[M\text{-}CH_4N]^+$, *m/z* 91 $[M\text{-}C_3H_8N]^+$, and *m/z* 58 $[M\text{-}C_7H_7]^+$, while the predominant fragments of N-IBA (Figure 3) were *m/z* 91 $[M\text{-}C_3H_8N]^+$ and *m/z* 58 $[M\text{-}C_7H_7]^+$. There are two overlapped fragments (*m/z* 91 $[M\text{-}C_3H_8N]^+$ and *m/z* 58 $[M\text{-}C_7H_7]^+$) between MA and N-IBA, besides the specific fragments at *m/z* 119 $[M\text{-}CH_4N]^+$ for MA. Thus, it is difficult to discriminate whether the N-IBA is adulterated in the MA forensic samples by mass spectra directly, especially when the quantity of the adulterated N-IBA is low. Based on our initial analysis of several suspected samples following our routine screening procedures for drug cases, three predominant fragments of MA were observed in the chromatogram of MA, but the unobservable difference was found in the abundance ratio between *m/z* 150/91 and *m/z* 150/119, comparing with the MA standard solution. As the analysis result of MA standard solution shown, the predominant fragments of MA were *m/z* 91, *m/z* 119, and *m/z* 58, and the abundance ratio between *m/z* 150/91and *m/z* 150/119 was approximately 1 : 0.63 (Show as Figure 4(a), supplementary material Table 2). The suspected samples showed the same fragments but the higher abundance ratio (1 : 0.026) (show as Figure 4(b)). To see this, we speculate that the existence of N-IBA in the suspected samples interfered the determination of MA. In order to simultaneously determine the N-IBA and MA in the forensic samples, a chromatographical separation was needed to well separate them, before testing them by mass spectra. Several different columns, mobile phases, and gradients were studied, and the isocratic separation using acetonitrile and 20 mM ammonium acetate solution containing 0.1% formic acid (20 : 80, *v/v*) as mobile phase on an Agilent Poroshell 120 SB-C_{18} column (4.6 × 100 mm, 2.7 μm) showed the best separation effect. Then, the MRM transition *m/z* 150/91 was selected as the quantitative ion pair for MA and N-IBA, and the predominant fragment *m/z* 119 of MA and *m/z* 58 of N-IBA was also utilized for the discrimination between MA and N-IBA. Compared to the reported GC/MS method, the excellent chromatographical separation and a minimum of two transitions in the MS/MS spectra for each analyte provide direct evidence to identify MA and N-IBA. The method was subsequently evaluated in compliance with the CFDA drug analytical method validation guidelines with minor modifications.

Table 3: Recoveries and RSDs of MA and N-IBA spiked in a suspected drug sample.

Compound	Background (ng/mL)	Sample Add (ng/mL)	Recovery (%)	RSD (%)
MA	0.48	2.05	87.8	2.8
		20.50	112.6	2.1
		51.30	112.1	1.9
N-IBA	5.28	1.83	96.7	0.5
		18.30	105.1	3.4
		51.20	108.1	0.9

3.2. Method Validation

3.2.1. Selectivity and Discrimination Capability. Selectivity of the method was examined by comparing chromatograms of individual standard solutions with blank sample. As Figure 5(a) showed that no interfering peaks were observed in the chromatogram of blank sample at the retention times of the target analytes.

The capability of the method to discriminate between MA and N-IBA was evaluated by analyzing individual standard

Table 4: The stability of MA and N-IBA under different conditions ($n = 3$).

Conditions	Samples	Compounds	Day 0 concentration (mean ± SD, ng/mL)	Measured concentration (mean ± SD, ng/mL)	RAD (%)
Autosampler for three days (15°C)	Suspect sample	MA	1.08 ± 0.05	1.15 ± 0.01	3.0
		N-IBA	15.27 ± 0.12	15.13 ± 0.15	0.4
	Mixed standard solution	MA	10.43 ± 0.23	10.23 ± 0.06	1.0
		N-IBA	9.39 ± 0.18	9.26 ± 0.20	0.7
Room temperature for one week (20°C)	Suspect sample	MA	1.45 ± 0.08	1.45 ± 0.05	0.1
		N-IBA	15.33 ± 0.12	15.00 ± 0.10	1.1
	Mixed standard solution	MA	10.53 ± 0.32	10.21 ± 0.24	1.5
		N-IBA	10.2 ± 0.20	9.38 ± 0.18	4.2
Freezer for 30 days (4°C)	Suspect sample	MA	1.16 ± 0.04	1.10 ± 0.10	2.4
		N-IBA	15.93 + 0.21	15.33 ± 0.15	1.9
	Mixed standard solution	MA	11.23 ± 0.125	10.43 ± 0.31	3.7
		N-IBA	9.91 ± 0.14	9.15 ± 0.06	4.0

solutions and mixed standard solutions at low (0.2 ng/mL), medium (20 ng/mL), and high (100 ng/mL). This range was chosen based on the LLODs and LLOQs. As Figures 5(b) and 5(c) shown, the retention times of MA and N-IBA were approximate 5.86 and 4.36 min, respectively. We also observed single peaks for each target analyte in the chromatograms of mixed standard solutions and suspected samples (Figures 5(d) and 5(e)), indicating the method with highly satisfactory capability of discrimination. Deserved to be mentioned, the routine screening procedure we had performed for psychotropics and narcotics screening more than a few thousands of samples cannot chromatographically separate MA and N-IBA; both compounds have the same retention time, which may result in misidentification. The routine screening samples that had been deemed MA positive were additionally tested, and some samples were ultimately confirmed as N-IBA or mixture of MA and N-IBA.

3.2.2. Linearity, LLOD, and LLOQ. Linearity was investigated by calculating the coefficient of determination (R^2) of calibration curve, and along with calibration curve equations, linear ranges, LLOD, and LLOQ of each target analyte are listed in Table 1. The calibration curves are in good linearity in the concentrations of 0.51~51.30 ng/mL for MA and 0.51~51.20 ng/mL for N-IBA with an R^2 above 0.99. The LLODs and LLOQs were 0.1 ng/mL and 0.3 ng/mL for MA and N-IBA.

3.2.3. Accuracy and Precision. Accuracy and precision were evaluated with five replicates of QC samples at low, medium, and high concentrations for each target analyte. The results of accuracy and precision of intraday ($n = 5$) and interday ($n = 5$) are shown in Table 2. The intra- and interday accuracy ranged from 96.2% to 110.1% and 100.1% to 121.9%. The intra- and interday precision ranged from 1.7% to 5.4% and from 3.6% to 11.6%. These results demonstrated that the modified LC-ESI-MS/MS method was reproducible and reliable for simultaneous determination of MA and N-IBA in forensic samples.

Table 5: Contents of MA and N-IBA in the suspected samples.

Sample ID	Quantity weighed (mg)	Contents of N-IBA (%)	Contents of MA (%)
SMU-1	54.7	80.4	7.31
SMU-2	54.0	74.5	0.21
SMU-3	54.7	75.4	Not detected
SMU-4	54.6	80.4	Not detected
SMU-5	55.0	0.5	Not detected
SMU-6	62.0	87.2	1.45
SMU-7	70.1	77.3	1.80
SMU-8	55.0	5.2	0.55

3.2.4. Recovery. Recovery was evaluated by adding low, medium, and high concentration levels of mixed standard solutions into known concentration samples. As shown in Table 3, the recovery value was in the range of 87.8-112.6%, and the RSD value was less than 10%.

3.2.5. Stability. Suspected samples and mixed standard samples were used to evaluate the stability of MA and N-IBA under different conditions. As shown in Table 4, the deviation values between two measured concentrations were less than 5% of RAD, indicating that MA and N-IBA were stable for three days in an autosampler at 15°C, for one week in room temperature at 20°C and one month in the freezer at 4°C. Matrix effect, recovery, and stability were within the acceptable range, indicating that the LC-MS/MS method was reliable.

3.3. Application to Real Forensic Drug Samples. During 2017-2018, 8 suspected samples were screened using our routine screening procedure and deemed MA positive, but the 8

suspected samples showed a quite unusual abundance ratio between m/z 150/91 and m/z 150/119 (show as Figure 4). Subsequently, the suspected samples were tested with the modified LC-ESI-MS/MS method, and the adulteration of N-IBA in MA forensic samples was confirmed (Table 5). The main components of these samples were N-IBA and a small amount of MA was found in some samples. Afterwards, all suspected samples in drug cases deemed MA positive were routinely analyzed by the modified LC-ESI-MS/MS method to prevent misidentification of MA and N-IBA.

4. Conclusions

According to the 2018 China drug report [32], MA is the most popular and easily accessible substances for drug addicts in China. In China, those suspected of producing, manufacturing, transporting, and possessing MA will face tough sentences. As we know, N-IBA is the positional isomer of MA, and both isomers have the same retention times using the previous routine screening procedure, which means that the identification of both isomers was easily confused. Since N-IBA is legal to use, misidentification of N-IBA for MA may result in serious consequences.

In the present work, a highly efficient modified LC-ESI-MS/MS method was developed and validated for the simultaneous determination of MA and N-IBA in forensic samples. The method exhibited excellent selectivity, linearity, accuracy, precision, and stability with a significantly lower LLOD and LLOQ than those in previous studies. When the method was applied to identify MA and N-IBA in several suspected samples in forensic cases, the MA and N-IBA were well separated and quantified. This work could offer new improvements for the forensic routine analysis of MA and N-IBA in suspected drug samples for clinical and forensic laboratories.

Data Availability

All data generated or analyzed during this study are included within the article.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Authors' Contributions

Yangxu Luo performed the conceptualization; Yangxu Luo, Juan Du, and Xuncai Chen performed the data curation; Yangxu Luo and Qizhi Luo performed the formal analysis; Juan Du and Ande Ma performed the investigation; Juan Du, Huadi Xiao, and Ling Zheng performed the methodology; Ande Ma and Qizhi Luo gather the resources; Ande Ma performed the validation; Yangxu Luo performed the visualization; Yangxu Luo wrote the original draft; Xuncai Chen and Qizhi Luo wrote, reviewed, and edited the manuscript. Yangxu Luo and Juan Du contributed equally to this work.

Acknowledgments

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Supplementary Materials

The following are available online at http://www.mdpi.com/xxx/s1. Table S1: modified detection method of multiple reaction monitoring parameters for determining MA and N-IBA. Table S2: abundance ratio of MA-Standard and suspect sample. *(Supplementary Materials)*

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